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A new species of *Lobohalacarus* and other porohalacarids (Acari) in Windermere.

By

J. GREEN,

Bedford College, University of London.

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(With 2 figures in the text.)

CONTENTS.

INTRODUCTION.

Three species of the family Porohalacaridae are known to occur in Britain. Viets (1928) has listed the species, Turk (1953) shows no additions.

In July 1953 a series of seventeen mites was taken from a small sample of bottom deposit which was accidentally brought up in a plankton net from a depth of fifteen or twenty feet near Crow Holme in Windermere. There was a single specimen each of *Limnohalacarus wackeri* (Walter) and *Porohalacarus alpinus* (Thor.). The latter species represents an addition to the British list. The remaining fifteen specimens belong to an undescribed species of *Lobohalacarus* apparently allied to *L. gallicus* (Migot); the female of *L. gallicus* has recently been described by Angelier (1952). The specimens from Windermere differ from *L. gallicus* in several important respects.

LOBOHALACARUS DOLGARAE SP. N.*

Female. The general form of the body is given in fig. 1, in which the details of measurements made are given. Five females were examined in detail. In the following description the type specimen will be described first and then any variation found in the four paratypes will be noted.

The anterior dorsal plate is wider than long, the greatest width being behind the middle, thus differing from *L. gallicus* where the greatest width is in front of the middle. The posterior dorsal plate is longer than wide and has the border markedly sinuate at the level of the posterior angles of the ocular plates. In this respect it agrees with *L. gallicus* but differs from the other species in the genus which have evenly elliptical posterior dorsal plates. The ocular plates are truncate anteriorly and have the eyes near the anterior external angle; *L. gallicus* is the only species of the genus reported to have eyes. All the dorsal plates are evenly punctured; the spaces between the plates appear to be wrinkled.

The epimeral plates are fused with the genito-anal plate so that there is one large ventral plate which bears the genital orifice; the uropore opens posteriorly. The genital orifice bears three cupules on each lip and is surrounded by a number of fine setae (fig. 2A).

The form of the maxillary organ is shown in fig. 2 B. The rostrum is short, truncate anteriorly and bears two pairs of setae. The chelicerae are somewhat

* The trivial name *dolgarae* is derived from the Welsh christian name Dolgar.

pear-shaped in dorsal view; fig. 2 C shows one in lateral view. The palps consist of four podomeres. The first podomere is devoid of setae, the second bears two dorsally; the third has a short tooth-like appendage and the distal podomere has three setae. No variation was found in the number and disposition of these setae (*L. gallicus* has two setae on the second and two on the fourth podomere).

Each leg has six podomeres, the lengths of which are given in table 2 and the number of setae on each in table 3; all types of setae are included in this

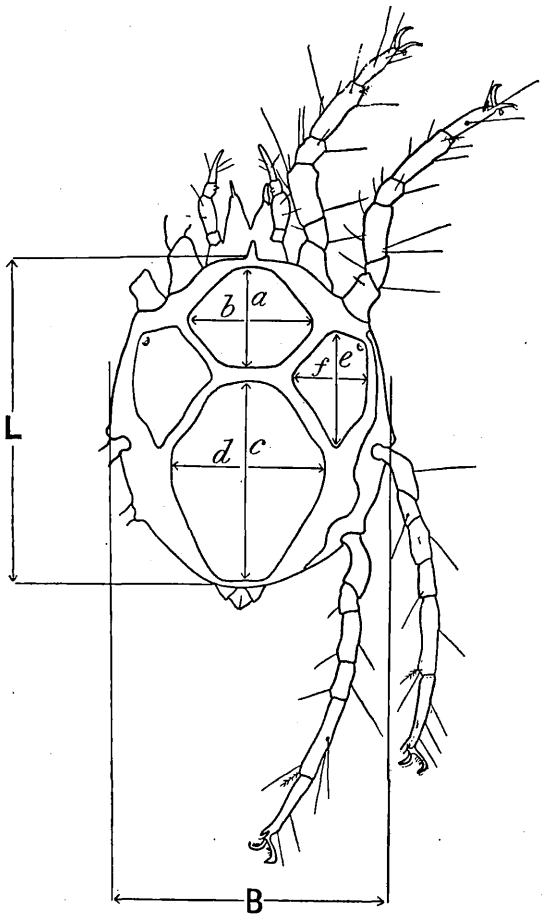

Fig. 1.—*Lobohalacarus dolgarae* sp. n., dorsal view to show details of measurements made.

table. The structure of the claws is shown in fig. 2 D and E. Legs III and IV have teeth on the claws whereas legs I and II do not. Leg I carries three plumose setae (fig. 2 F), the other legs carry only one plumose seta near the apex of the fifth podomere.

Male. Only a single male was found in the series; this agreed in most respects with the female. The size is slightly smaller than the average female and the podomeres of the legs somewhat shorter (tables 1 and 2). The chaetotaxy of the palps and limbs is identical with the female. The genital orifice is externally similar to that of the female, but in a cleared preparation the internal framework of the penis can be seen.

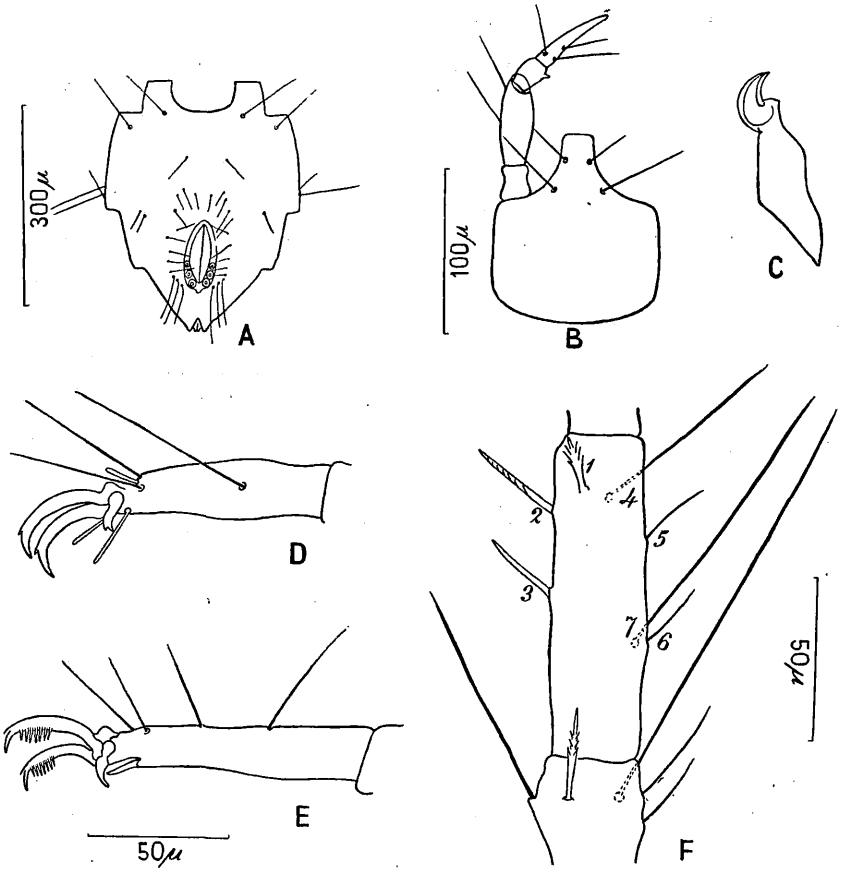

Fig. 2.—*Lobohalacarus dolgarae* sp. n.

A, ♀, ventral view. B, Maxillary organ and palp, ventral view. C, chelicera, side view. D, terminal podomere of leg II, side view. E, terminal podomere of leg III, side view. F, ventral view of podomeres four and five of leg I, when only six setae are present on podomere five it is seta No. 3 that is missing.

Table 1.

Dimensions of *Lobohalacarus dolgarae* sp. n. in μ.

(See fig. 1 for details of measurement.)

	L	B	a	b	c	d	e	f
Female, type	406	314	115	151	250	180	137	86
Range in paratypes	416–455	327–353	118–134	153–165	257–291	180–219	144–151	86–108
Male	403	330	111	147	275	198	144	86
Range in nymphs	377–390	288–327	90–108	126–147	162–223	126–158	122–126	90

TABLE 2.

Lengths of podomeres of legs.

	Leg 1						Leg II						Leg III						Leg IV					
Podomere	1	2	3	4	5	6	1	2	3	4	5	6	1	2	3	4	5	6	1	2	3	4	5	6
Female, type	57	57	79	39	94	65	51	54	75	36	90	62	79	43	62	43	101	83	75	43	65	46	105	82
Range in paratypes	54-57	54–62	79–82	39–43	94–101	62–75	46-54	51–57	76-79	36–39	90–98	65–75	72–79	39–46	54–65	39–43	101-108	83-86	79	39–46	62–72	46–51	108–111	82–90
Male	54	54	72	36	90	57	46	46	72	36	90	57	76	36	54	43	98	79	76	32	62	46	101	82
Range in nymphs	46	46	57–65	32	72–76	51–54	39–46	43–46	62	28–32	72	54–57	57–65	36	43–46	32–36	75–79	62–65	62–69	32–36	43–46	36	79	65–72

TABLE 3.

Numbers of setae on each podomere of the legs.

	Leg I						Leg II						Leg III						Leg IV					
Podomere	1	2	3	4	5	6	1	2	3	4	5	6	1	2	3	4	5	6	1	2	3	4	5	6
Female, type	1	2	5	4–5	7	7	1	2	5	4–5	7	6	1	2	3	2	5	4	0	2	3	3	5	3
Range in paratypes	1	2	5	5	6–7	6–7	1	2	5	4–5	7–8	6	1	1–2	2	2	5	4	0	2	3	3	5	3
Male	1	2	5	5	8	7	1	2	5	5	7	6	1	2	2	2	5	4	0	2	3	3	5	3
Range in nymphs	1	2	5	4	5–6	6	1	2	5	4	6	6	1	2	2	2	5	4	0	1	2	3–4	5	3

Nymph. Four nymphs were examined in detail. The most striking difference from the adult is that the epimeral and genito-anal plates are not fused. There is no definite genital orifice but two cupules are present on each side of the median line of the genito-anal plate. The dimensions and chaetotaxy of the limbs are somewhat different from the adult. The palps have an identical chaetotaxy with the adult. The type female will be retained in my collection; paratypes will be deposited at the British Museum (Nat. Hist.).

When first discovered in the bottom deposit all three species were covered with a dark brown felt and looked very similar to the figure of *Pachygnathus nigrescens* Brady, an inadequately described form from Crag Lake, Northumberland. It seems likely that *P. nigrescens* is in fact one of these species covered in felt. The presence of such a form indicates that these species will probably be found to be much more widespread in this country than our present knowledge indicates.

Summary.

Lobohalacarus dolgarae sp. n. is described from Windermere. *Limnohalacarus wackeri* Walter and *Porohalacarus alpinus* (Thor) are recorded from Windermere, the latter species being an addition to the British List.

Acknowledgments.

In the preparation of this paper I have received advice and criticism of a most helpful nature from Mr. A. R. Longhurst, Mr. H. C. Fountain, and Dr. F. A. Turk. Dr. E. Angelier provided me with a reprint of his paper on *Lobohalacarus gallicus* which saved a great deal of time.

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A NOTE ON THE FOOD OF *CHAETOGASTER DIAPHANUS*.

By J. GREEN,
Bedford College, University of London.

THIS paper is based mainly on the examination of the gut contents of 75 specimens of the oligochaete *Chaetogaster diaphanus* (Gruithuisen) taken from the shores of Crow Holme in Windermere in August, 1953.

Several workers have noted the general nature of the food of this species. Berg (1938) writes, " They creep about in the algae and live on *Chydorus*, *Pleuroxus*, *Cyclops* and Ostracoda ". Ude (1929) records " Krebschen und Nematoden im Darm ", whilst Cernosvitov (1945), who examined the species in Windermere, found small cladocerans, hydracarine larvæ, rotifers and nematodes in the gut. The relative numbers of each type of food taken do not appear to have been investigated.

The specimens studied were all taken from a restricted area. At the same time a sample of the other animals in the same immediate environment was taken. In this way it was possible to compare the frequency of the various animals in the gut of *Chaetogaster diaphanus* with their abundance outside. Table I gives the frequencies of the 1,059 organisms found in the worms taken from Windermere; and also the order of

TABLE I.

	In gut of *Chaetogaster diaphanus*.	In environment.
Cladocera	716	++++++
Copepoda	222	++++
Copepod nauplii	97	
Ostracoda	17	++
Chironomid larvæ	6	+
Hydracarine larvæ	1	++

abundance in the immediate environment. These latter estimates were purely subjective and were based upon the frequency with which the various forms appeared in small random samples of plant-material and water taken from the main sample. The relative abundance is indicated by the number of crosses.

The only disagreement between the two series is in the position of the hydracarine larvæ. This might indicate that they are distasteful and are rejected by the animal after it has captured them.

The cladocerans were all identified down to species and a separate table compiled (Table II). As an indication of the relative abundance given in the two tables it should be noted that chironomid larvæ (+ in Table I) were about as frequent as *Alonella nana* (++ in Table II). Again there is good agreement between the two series. *Alonopsis elongata* is too low down in the list, probably because this species carries at least one old carapace out of which it can escape if trapped. The absence of *Chydorus globosus* and *Peracantha truncata* from the gut of *Chaetogaster diaphanus* is attributed to chance.

TABLE II.

	In gut of *Chaetogaster diaphanus*.	In environment.
Bosmina obtusirostris Sars	654	++++++
Chydorus sphaericus (O. F. Müller)	26	++++
Acroperus harpae Baird	14	+++
Alonella nana Baird	6	++
Anchistropus emarginatus Sars	5	++
Alona guttata Sars	4	+
Alonopsis elongata Sars	3	+++
Ceriodaphnia pulchella Sars	2	+
Sida crystallina (O. F. Müller)	1	+
Eurycercus lamellatus (O. F. Müller)	1	+
Chydorus globosus Baird	*	++
Peracantha truncata (O. F. Müller)	*	+

* Not found in *Chaetogaster diaphanus*.

Three other small samples of *Chaetogaster diaphanus* have been examined. Three specimens from Regent's Park Lake in October had between them eaten 30 *Chydorus sphaericus*, 2 *Alona rectangula* and 1 *Acroperus harpae*. *Chydorus sphaericus* was the most abundant animal in the surrounding water. A sample of 10 *Chaetogaster diaphanus* from the Viaduct Pond, Hampstead Heath, taken in November, showed a much lower number of animals in the gut of each individual (Table III). Again, *Chydorus sphaericus* was the most abundant animal in the surrounding water.

TABLE III.

Viaduct Pond.	Pond in S. Wales.
17 *Chydorus sphaericus*.	44 *Stentor coeruleus*.
5 *Cyclops* sp.	2 *Paramecium* sp.
3 *Daphnia longispina* O. F. Müller.	4 Rotifers.
2 Ostracods.	2 *Chydorus sphaericus*.
2 *Pleuroxus trigonellus* (O. F. Müller).	1 *Cyclops agilis*.
1 *Peracantha truncata*.	

A more unusual sample was taken from a dish of material from a pond near the River Gwendraeth in South Wales in December. Most of the larger animals had died; protozoa were numerous, the large *Stentor coeruleus* being the most abundant. Eight specimens of *Chaetogaster diaphanus* were examined (Table III). The numbers of protozoa counted are probably not very accurate, for Slimm (1913, quoted in Stephenson, 1930) has shown that in the stomach of a starving *Chaetogaster* a living *Paramecium* is killed instantly and disintegrates in a few seconds. In a well fed animal the prey is passed on still alive into the intestine where it disintegrates slowly. The worms from South Wales were well fed. In one case a rotifer (*Colurus* sp.) was seen to be ejected through the anus still alive.

Out of the total of 96 worms examined only three were found with apparently empty guts.

The food taken by *Chaetogaster diaphanus* appears to be governed only by the largest size that it is capable of ingesting. Thus only young specimens of *Sida crystallina* and *Daphnia longispina* were found in the gut. Aquatic beetles (*Haliplus* spp.) and large adult hydracarines were present in the immediate environment of the worms in Windermere, but they were probably too large to fall prey to *Chaetogaster diaphanus*. The only departure noted from random feeding in the suitable size range was in the case of hydracarine larvæ, which were not found as frequently in the gut as their abundance in the environment would lead one to expect.

Summary.

(1) *Chaetogaster diaphanus* feeds on small invertebrates; protozoa, rotifers, nematodes, crustaceans, hydracarine larvæ and chironomid larvæ have been recorded in its gut.

(2) The numbers of each prey-species of suitable size taken appear to be directly related to the abundance of that species in the immediate environment of the worm.

(3) In Windermere hydracarine larvæ appear in smaller numbers in the gut of the worm than one would expect from their abundance in the worm's immediate environment. It is suggested that they are distasteful and are rejected by the worm after capture.

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THE STRUCTURE AND FUNCTION OF THE POST-ABDOMEN OF *CAMPTOCERCUS* (CRUSTACEA: CLADOCERA)

BY

J. GREEN

Bedford College, University of London

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(With 5 figures in the text)

CONTENTS

INTRODUCTION

There is great variation in the form of the post-abdomen in the Cladocera. In the Calyptomera there is usually a considerable amount of lateral compression, but when viewed from one side the post-abdomen may be very short and round or else show varying degrees of elongation up to the extremely narrow type found in the genus *Camptocercus*. The movements of the post-abdomen are usually restricted to simple antero-posterior flexions and extensions, with no lateral movement or rotation. The claws at the distal extremity are frequently used to clear the food groove between the thoracic limbs. In some mud-dwelling species, such as *Iliocryptus sordidus* (Liéven), vigorous thrusts of the post-abdomen help the animal to progress through its difficult environment. In the genus *Camptocercus* the post-abdomen is extraordinarily motile, as the author of the name clearly realized (cf. Baird, 1850).

While staying at the Freshwater Biological Laboratory of the University of Copenhagen, I had the opportunity of examining numerous living specimens of *Camptocercus lilljeborgi* Schödler and a smaller number of *C. rectirostris* Schödler. The account which follows is based entirely on observations made on living specimens and refers to *C. lilljeborgi*, though *C. rectirostris* was seen to perform similar movements.

MOVEMENTS OF THE POST-ABDOMEN

When swimming over the surface of the vegetable detritus, on which it feeds, *C. lilljeborgi* keeps its body horizontal, with the long open edges of the carapace valves on the underside. The post-abdomen is held in the position shown in Fig. 1 A. Occasional thrusts of the post-abdomen help the cladoceran

to clear small obstacles or to extract itself from tight corners. Sometimes the claws are used to clear the food groove. The abdomen is pushed forwards and the post-abdomen bent backwards to that the claws come to lie between the thoracic limbs (Fig. 1 B). One or more violent kicks of the post-abdomen serve to loosen any obstacle between the limbs.

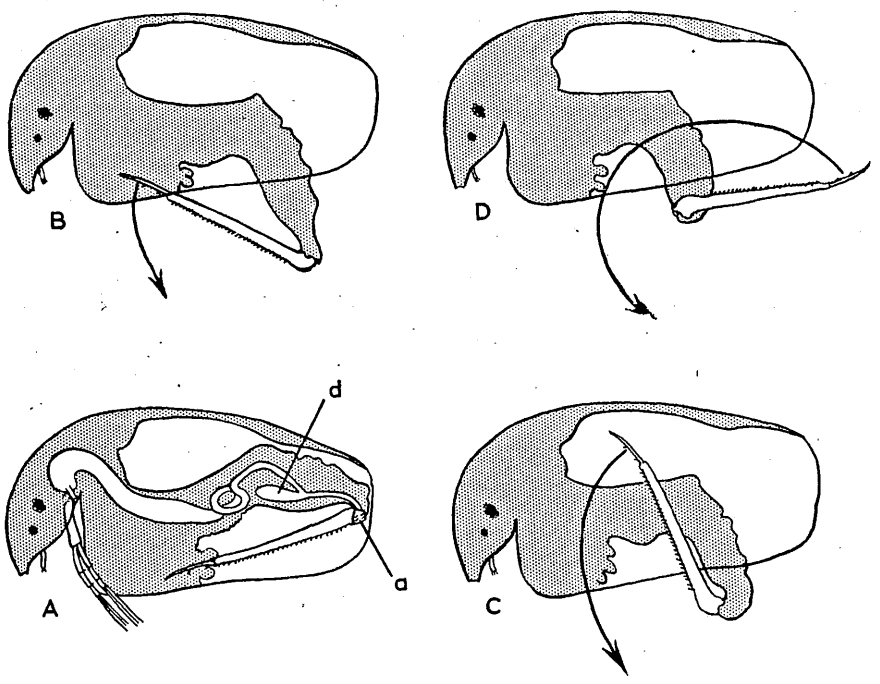

Fig. 1.—*Camptocercus lilljeborgi*, showing movements made by the post-abdomen: a, anus; d, diverticulum. The arrows indicate rapid thrusting movements of the post-abdomen.

More spectacular movements are illustrated in Figs. 1 C and 1 D. The post-abdomen can be slipped outside the carapace valves and bent backwards through an angle of 180° (Fig. 1 D) and then brought back to the normal position very rapidly. By bending back the post-abdomen through varying degrees and by moving the abdomen to various positions practically any part of the outside surface of the carapace can be scoured by the spines and the claws. It seems that these movements serve to keep the carapace free from epibionts. None of the specimens examined was carrying epibionts, although other small Cladocera from the same localities were found with various organisms on their carapaces. The movements might also help the cladoceran to escape from some comparatively small-mouthed predators, such as the oligochaete *Chaetogaster diaphanus* (Gruithuisen) which feeds to a large extent on Cladocera (Green, 1954). The vigour of the movement is such that if the post-abdomen catches against anything solid the cladoceran may be projected several times its own body length through the water.

THE STRUCTURE AND MUSCULATURE OF THE POST-ABDOMEN

The movements described above clearly necessitate a fairly complex musculature and a mobile joint between the rigid post-abdomen and the more flexible abdomen. Fig. 2 shows the general structure and armature of the post-abdomen. The spines on the dorsal border increase in size distally. The paired terminal claws bear two large teeth and two series of fine spinules. The articulation between the abdomen and post-abdomen is by means of a complex joint (Fig. 3). In the wall on each side of the abdomen there is a thickened bar or apodeme. At the distal end of the apodeme is a small thickened fulcrum. Between the fulcrum and the rigid post-abdomen is a region of flexible cuticle. The apodeme and fulcrum impart rigidity to the upper part of the abdomen while the flexible region allows great motility of the post-abdomen. The apodeme and fulcrum provide a resistance for the muscles which are attached to the post-abdomen to pull against and cause lateral bending of the flexible region.

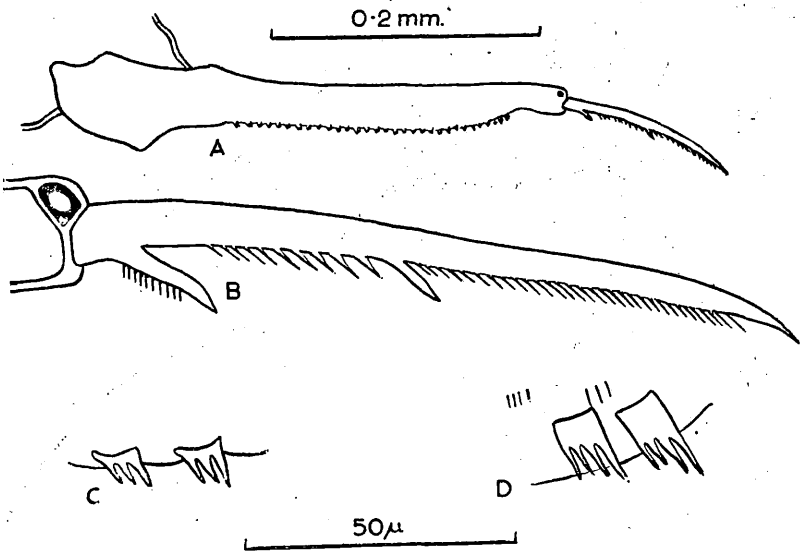

Fig. 2.—*Camptocercus lilljeborgi*, structure of the post-abdomen. A, lateral view of the post-abdomen; B, lateral view of one terminal claw; C, spines from the proximal part of the dorsal border; D, spines from the distal dorsal border.

A joint of a similar nature is found in certain other Cladocera, such as *Acroperus harpae* Baird, which do not perform such complex movements as *Camptocercus*. The thickenings of the cuticle serve to prevent buckling of the abdominal wall when the longitudinally disposed muscles contract. The reason why such thickenings are not found in *Daphnia* seems to be because here the abdomen is very much shorter. The last thoracic limb and its muscles probably impart enough rigidity to the base of the abdomen to provide sufficient resistance for the longitudinal muscles to bring about simple flexions and extensions of the post-abdomen. When a much longer abdomen is present,

as in *Acroperus* and *Camptocercus*, some form of mechanical support becomes necessary in the more distal region to prevent buckling of the abdominal wall.

Since Binder (1932) has given a detailed account of the musculature of *Daphnia magna* Straus it seems desirable to attempt to describe the muscles moving the post-abdomen of *Camptocercus*, using the same system of nomenclature.

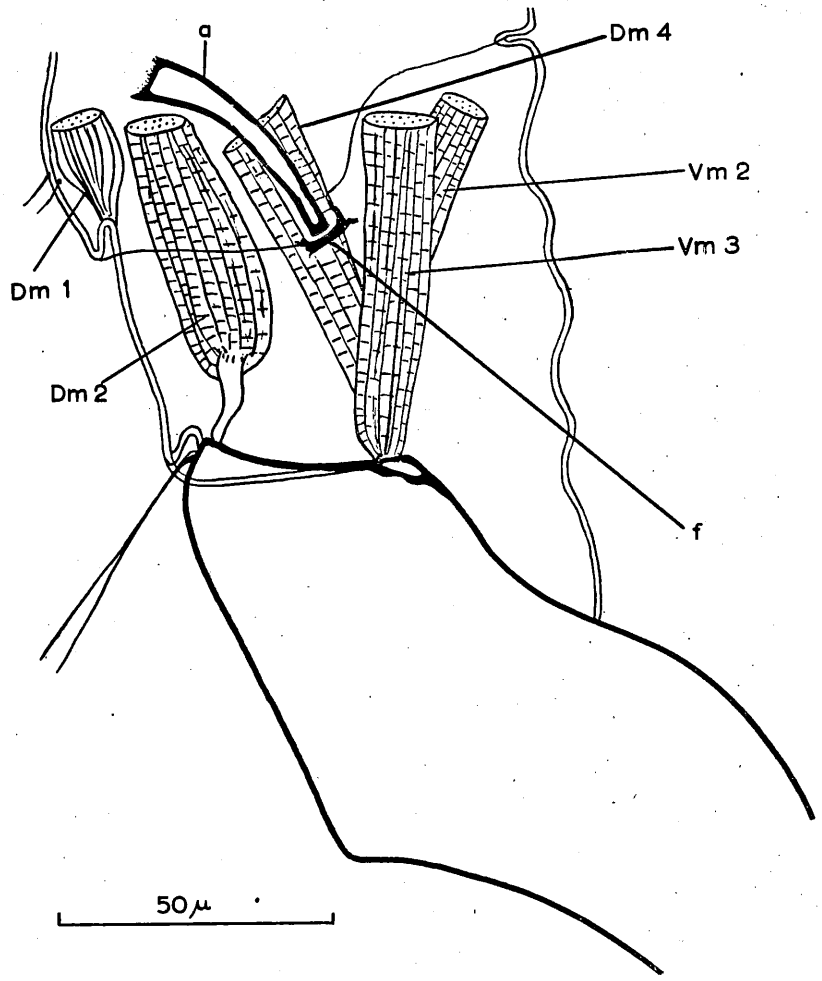

Fig. 3.—*Camptocercus lilljeborgi*, diagram of the joint between the abdomen and post-abdomen : a, apodeme ; f, fulcrum ; other letters as in Fig. 4.

The arrangement of the muscles moving the post-abdomen of *Daphnia* is comparatively simple (Fig. 4 A). The abdominal muscles, of which there are three on each side, originate in the abdomen and extend into the post-abdomen. The ventral muscles also number three on each side and extend from the region of the anterior thoracic limbs down to the base of the post-abdomen. There are four series of dorsal muscles on each side of the body extending from the

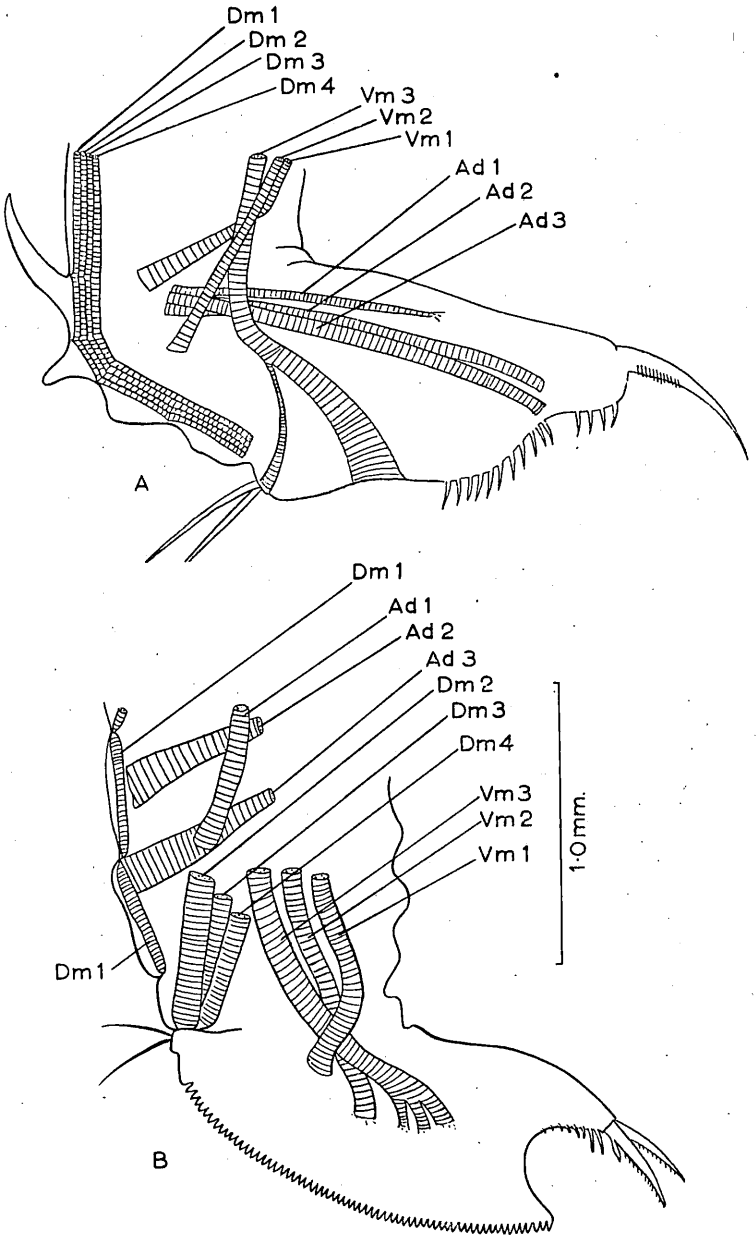

Fig. 4.—Muscles moving the post-abdomen of A, *Daphnia magna* and B, *Eurycercus lamellatus*. Dm 1–4, dorsal muscles; Vm 1–3, ventral mucsles; Ad 1–3, abdominal muscles.

heart to the base of the post-abdomen. There are dilator and constrictor muscles around the anus and rectum, but since these are not concerned in moving the post-abdomen they have been omitted from the diagram.

The direct homology of the muscles of *Camptocercus* with those of *Daphnia* presents certain difficulties, but if an intermediate stage is considered the task is made easier. *Eurycercus lamellatus* (O. F. Müller) provides such a stage

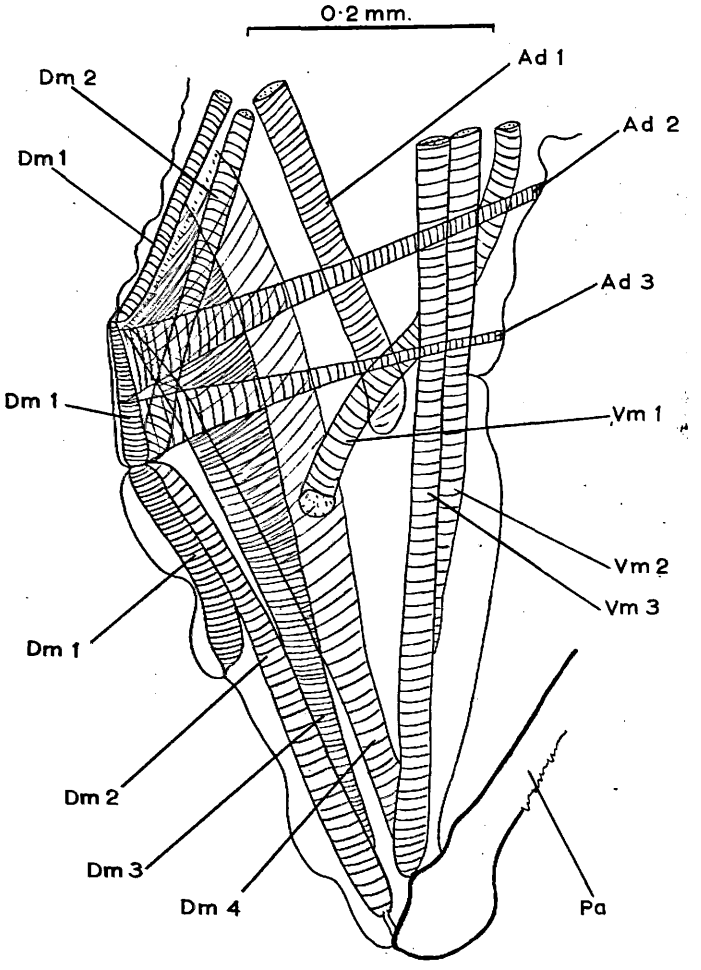

Fig. 5.—*Camptocercus lilljeborgi*, muscles moving the post-abdomen. Pa, post-abdomen; other letters as in Fig. 4.

(Fig. 4 B). The first difference from *Daphnia* is that the post-abdomen of *Eurycercus* is more heavily sclerotized. The first dorsal muscle (Dm1) is inserted higher than the second to fourth which are all inserted on the hind border of the post-abdomen. The abdominal muscles show the greatest change, although they originate in roughly the same position as in *Daphnia* they now pass obliquely forwards and do not enter the post-abdomen. The homology of these muscles with the abdominal muscles of *Daphnia* is not certain, but seems

probable by a process of elimination. The ventral muscles show very little difference from the condition in *Daphnia*.

The condition in *Camptocercus* is even more modified. There has been increased sclerotization of the post-abdomen and great elongation coupled with marked tapering of the abdomen. The elongation of the abdomen has increased the distance between the base of the post-abdomen and the last thoracic limb and has placed the post-abdomen in a position from which it can be slipped outside the carapace valves. For the sake of clarity the muscles in Fig. 5 are drawn narrower than they actually are. The whole of the lower part of the abdomen is filled with muscles. The gut in this region is very narrow and inconspicuous. The anus opens at the base of the post-abdomen (*a* in Fig. 1 A). Faeces pass very rapidly through the rectum from the diverticulum (*d*) which seems to be an adaptation for preventing the accumulation of food in the rectum where it might interfere with the movements of the muscles.

The first dorsal muscle of *Camptocercus* is inserted quite high in the abdomen at the level of the fulcrum. A fold frequently develops in the wall of the abdomen between the insertion of this muscle and the fulcrum. The fourth dorsal muscle has become attached to the ventral border of the post-abdomen leaving the second and third to pull on the hind (dorsal) corner. With the narrowing of the post-abdomen the insertions of ventral muscles 2 and 3 have moved anteriorly and now coincide with the insertion of the fourth dorsal muscle on the wall of the post-abdomen. These muscles pull the abdomen forwards and ventrally. The second and third abdominal muscles run from the dorsal surface to the ventral surface of the abdomen, they appear to modify the actions of the dorsal and ventral muscles. The first abdominal muscle runs obliquely forwards and dorsally, its action must to some extent counter that of the first ventral muscle.

The way in which these muscles bring about the complex movements shown in Fig. 1 is not fully understood, particularly the movement illustrated in Fig. 1 D. The process must involve rapid alternate contraction and relaxation of the various longitudinal muscles together with some rotation of the post-abdomen about its longitudinal axis due to differential pulls exerted by the muscles on the two sides of the body. The rapid movement back to the normal position seems to be effected entirely by the second and third dorsal muscles.

SUMMARY

Camptocercus lilljeborgi has a long, extremely motile post-abdomen. The movements of the post-abdomen keep the carapace free from epibionts, keep the food groove between the thoracic limbs clear, and may be of use in escaping from certain small predators. There is a complex joint between the abdomen and post-abdomen. The muscles moving the post-abdomen are described and homologized with those of *Eurycercus lamellatus* and *Daphnia magna*.

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HALACARID AND POROHALACARID MITES FROM SKOKHOLM ISLAND AND THE ISLE OF MAN.

By J. GREEN, Bedford College, University of London.

THE records in this paper are intended as a contribution to our knowledge of the distribution of certain aquatic mites. Comparatively little is known of the distribution of the marine Acari of the family Halacaridae around the British Coast. Even less is known about the distribution of their freshwater allies, the Porohalacaridae. Some authors (e.g. Turk, 1953) do not regard the latter group as a separate family and arrange its subdivisions as subfamilies of the Halacaridae. Others (e.g. Viets, 1936, 1939; Angelier, 1952) regard the Porohalacaridae as a separate family. This seems reasonable since the presence of cupules on the genital plate is a character which separates all the subfamilies of the Porohalacaridae from the Halacaridae, which do not have such cupules.

In the following lists the nomenclature of André (1946) is followed for the Halacaridae, except that subgeneric names have not been given to members of the genus *Rhombognathus*. The natural classification of the Rhombognathinae is the subject of a controversy between Viets (1952) and Newell (1953) and though the latter's system has much to recommend it, it has not been adopted here because certain changes in names would have to be explained in detail which would not be relevant to the purpose of this paper. The system of Viets (1939) is followed for the Porohalacaridae.

The first list records species collected during nine days in March 1953 and a further nine days in April 1955 on the south end of the Isle of Man.

Family **Halacaridae.**

Rhombognathus notops (Gosse). On *Pelvetia canaliculata*, Fleshwick Bay.

Rhombognathus magnirostris Trouessart. Abundant among barnacle scrapings and *Corallina* from the rocks below the Marine Biological Laboratory, Port Erin.

Rhombognathus setosus (Lohmann). On *Pelvetia canaliculata*, Fleshwick Bay.

Rhombognathus pascens (Lohmann). On *Botryllus* and *Corallina*, Dub Reef, Port Erin. In *Laminaria* holdfasts, Calf Sound. On *Halurus equisetifolius*, *Corallina* and *Cladophora*, Port St. Mary. On *Cladophora*, Derbyhaven.

Rhombognathus merrimani Newell. In intertidal rock crevices, Dub Reef, Fleshwick Bay and Port St. Mary. Not known previously outside N. America.

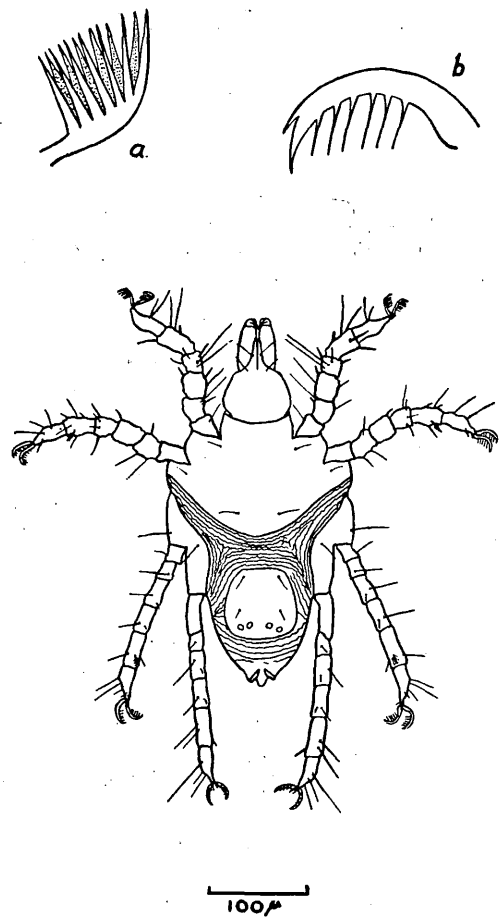
Rhombognathus seahami (Hodge). On *Cladophora*, Derbyhaven. On *Halurus equisetifolius*, Port St. Mary.

Rhombognathus armatus Lohmann. Among barnacle scrapings from the rocks below the lab., Port Erin. On *Enteromorpha*, Fleshwick Bay. These appear to be the first British records.

Halacarus (*Halacarellus*) *basteri* (Johnston). On *Corallina*, the breakwater, Port Erin.

Copidognathus rhodostigma (Gosse). In intertidal crevices, Port St. Mary. Dredged off Bradda Head.

Fig. 1.



Soldanellonyx monardi Walter. Nymph, ventral view. *a*, tarsal claw of leg I; *b*, tarsal claw of leg II.

Copidognathus fabriciusi (Lohmann). In *Laminaria* holdfast and on *Ceramium*, Derbyhaven. On *Cladophora*, Port St. Mary.

Simognathus minutus (Hodge). On *Cladophora*, Calf Sound. In intertidal crevices, Port St. Mary. This species is given by André (1946) as *S. sculptus* Brady, but Fountain (1953) has shown that *minutus* has priority.

Skokholm Island.

This second list records species collected on Skokholm during a week in August 1955.

Family **Halacaridae.**

Rhombognathus setosus Lohmann. Abundant on *Enteromorpha* on a ledge near Purple Cave.

Rhombognathus pascens Lohmann. In *Laminaria* holdfasts, South Haven.

Rhombognathus merrimani Newell. In *Laminaria* holdfasts, North Haven, Crab Bay, and in intertidal crevices, Peter's Bay.

Rhombognathus armatus Lohmann. In intertidal crevices, Crab Bay.

Halacarus (*Halacarellus*) *basteri* Johnston. A single specimen on *Corallina* in North Haven.

Agauopsis brevipalpus Trouessart. A single specimen was found among material from an intertidal crevice in Crab Bay. This species has not previously been recorded from Britain, although it is known from the French Channel coast (André, 1946).

Family **Porohalacaridae.**

Porohalacarus alpinus Thor. Five specimens were taken from East Pond. The pond was very shallow and the sample of detritus in which the mites were found was obtained by pressing down the vegetation underfoot and sweeping a small net through the water that oozed up. The only other record of this species in Britain is a single specimen from Windermere (Green, 1954).

Soldanellonyx monardi Walter. Two specimens were taken from East Pond. This species has not previously been recorded in Britain. One of the specimens was a nymph (figure 1). The main difference from the adult is the lack of a genital opening, and the presence of only two cupules on each side of the genital plate. The adult may have up to ten of these cupules on each side. The adult female from Skokholm had six on the right side and five on the left. The figure also shows the peculiar structure of the first tarsal claws. The addition of this species to the British list is the third since the publication of Turk's (1953) Synonymic catalogue of the British Acari. As there were only three porohalacarids in Turk's list a revised list of the British Porohalacaridae is given below together with the known localities.

Family **Porohalacaridae.**

Subfamily *Porohalacarinae.*

Porohalacarus alpinus Thor. Windermere, and East Pond, Skokholm.
Lobohalacarus dolgarae Green. Windermere.

Subfamily *Limnohalacarinae.*

Limnohalacarus wackeri Walter. Windermere, and the Isle of Man.
Soldanellonyx monardi Walter. East Pond, Skokholm.

Soldanellonyx parviscutatus (Walter) var. *transversaria* Viets. Loch Vennacher, Perthshire.

Subfamily *Porolohmanellinae*.

Porolohmannella violacea (Kramer). Weybridge; Epping Forest and the Isle of Man.

Summary.

Eleven species of the family Halacaridae are recorded from the Isle of Man. Two of these, *Rhombognathus armatus* Lohmann, and *R. merrimani* Newell, had not previously been recorded in Britain.

Six species of the Halacaridae and two of the Porohalacaridae are recorded from Skokholm Island. *Agauopsis brevipalpus* Trouessart and *Soldanellonyx monardi* Walter are recorded as British for the first time.

A revised list of the British Porohalacaridae is given.

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THE GROWTH OF *SCROBICULARIA PLANA* (DA COSTA) IN THE GWENDRAETH ESTUARY

By J. GREEN
Bedford College, University of London

(Text-figs. 1–3)

Scrobicularia plana (da Costa) is an abundant inhabitant of intertidal estuarine muds. Populations up to 1000 per m^2 have been recorded from the Tamar Estuary (Spooner & Moore, 1940). Similar population densities have been found in the Gwendraeth Estuary in South Wales. In one patch a population of 1025 per m^2 was found; the average length of the shells was 30 mm. Individual lengths up to 54 mm were found in other parts of the estuary. The large size of the specimens from the Gwendraeth prompted an investigation into the rate of growth and the age reached in this locality.

METHODS

Random samples of *Scrobicularia* were taken by digging up 1 m^2 to a depth of 30 cm in a region where the average population was 500 per m^2. A square metal frame with sides 0·5 m long and 15 cm deep was used to prevent mud slipping into the area being sampled. Four samples taken using such a frame made up the total sample of 1 m^2. Samples were collected in December 1953, April 1954, August 1954, April 1955 and July 1955.

The large specimens were sorted by hand and the small specimens by sieving. A sieve with a mesh of about 6 mm was used for the early samples, but for the last sample a 1 mm-mesh sieve was used. As there were some empty shells in the mud, care was taken to collect only the living animals; these were recognized in the field by the slightly protruding mantle lobes and by the presence of an oxidized layer of soil immediately next to the shell even when the animals were taken from a black layer. The specimens were washed and then boiled until the shells gaped widely and the animals dropped out. The shells were then washed and dried; one valve was kept from each. In this way a large number of shells could be stored and their lengths measured at leisure. The length of each shell, to the nearest mm, was written in pencil on its inner surface for future reference and size grouping. In all some 2500 shells were sorted and measured.

Like many other bivalved molluscs, *S. plana* has growth rings on its shell, but often only the last one or two are visible. The earlier growth rings become obliterated and only those left by the last few winters can be found. It is thus

not possible to determine the age of a shell directly from its growth rings. It is, however, possible to deduce an approximate age by using a series of shells such as that shown in Fig. 1. By homologizing the rings on each shell with those on larger and smaller shells, and by working back to progressively smaller shells an estimation can be made of the age of any shell in the series. Examination of the growth rings on some of the large specimens was found to be facilitated by immersion in dilute hydrochloric acid for a short period. Immediately after such treatment the growth rings were very clear, but when the treated shells were stored the rings tended to disappear, and, of course, the treated shells were much more fragile than the untreated shells.

The environmental conditions in the sampling area were studied by measuring the salinity of the water over the area during a tide and by a grade analysis of the soil.

The salinity of the water was estimated with a glass hydrometer; the temperature of the water was measured at the same time and the density reading converted to salinity in ‰ using the graph given by Harvey (1945, fig. 12).

The sample of soil for analysis was taken by pushing a stout glass tube into the ground and then lifting it out after clearing the soil away from its sides. The ends of the tube were then closed with rubber bungs. The sample was taken about 3 h after the tide had uncovered the sampling area so that most of the superficial water had drained away. In the laboratory the soil was pushed out of the tube and chopped into 2 cm lengths which were weighed and then dried for 16 hr at 105° C. The subsequent treatment of the samples was identical with that of Holme (1949) except that the treatment with hydrochloric acid to remove carbonates was omitted and sieves with the following meshes were used: 30, 60, 90 and 100 I.M.M. In practice it was found that over 95% of the soil passed through the 100 I.M.M. sieve.

CONDITIONS IN THE SAMPLING AREA

The area from which the samples were taken was about 70 m long, with the length parallel to the edge of the tide, and about 4 m wide. Neap tides covered the area for about 2 h; on a spring tide the time was increased to about 4 h. The depth of water over the surface of the mud on a neap tide was 0·5 m. The depth over the mud on a spring tide was not measured but is estimated at 2·0 m. The times given for coverage by the tide do not represent the limits of the time available to *Scrobicularia* for feeding since the surface is poorly drained and still wet for several hours after the tide has receded. The inhalant siphons of *Scrobicularia* can be seen actively sucking up the surface mud 3 or 4 h after being uncovered by the tide.

The salinity of the water over the area during a neap tide on 29 July 1955 is shown in Table 1. The salinity is probably somewhat higher than usual, due to the hot dry summer and the low state of the river. It was not practic-

able to measure the salinity during a spring tide, but a few measurements made during the winter 1952–53 showed a salinity of 18‰ at a level just above the sampling area about an hour before high tide. This somewhat lower salinity on a spring tide in winter when compared with a neap tide in summer indicates that the main factor governing salinity over the sampling area is the strength of flow of the river. Other measurements made during winter, as the tide was just uncovering the area, showed a salinity as low as 2‰.

TABLE 1. SALINITY OF THE WATER COVERING THE SAMPLING STATION DURING A NEAP TIDE IN JULY 1955

The tide covered the area at 12.44 G.M.T. and uncovered it at 14.45 G.M.T.

Time (G.M.T.)	Salinity (‰)	Time (G.M.T.)	Salinity (‰)
12.30	25	14.15	12
13.00	25	14.30	11
13.30	25	15.00	11
14.00	15		

TABLE 2. GRADE ANALYSIS OF THE SOIL IN THE SAMPLING AREA

Depth (cm.)	% water content	Fine sand as % dry weight	Silt + clay as % dry weight
1–2	31	75	24
2–4	28	78	21
4–6	30	80	19
6–8	29	81	17
8–10	27	84	13
10–12	26	88	10
12–14	24	90	8
14–16	24	90	8

Grade analysis of the soil showed that most of the particles passed through the 100 I.M.M. sieve and that of the total dry soil between 8 and 24% by weight belonged to the silt + clay fraction (Table 2). The most significant variation with depth is in the amount of silt + clay; the surface layers contain three times as much as the soil at a depth of 14–16 cm. The water content, as estimated by the present method, may not be quite the same as that of the mud *in situ*, but it does give a measure of the water-holding capacity of the soil. It is clear from Table 2 that this capacity increases with increasing silt + clay content, so that the surface layers have a higher water content.

AGE ESTIMATED FROM GROWTH RINGS

From a series such as that shown in Fig. 1 it is possible to construct a growth curve. Such a curve, based on the specimens in Fig. 1, is given by the broken line in Fig. 2. The middle part of the curve is based on several specimens at each length (Table 3). Other series of shells give slightly different curves, and if the whole range of variation is considered it is possible to draw up a table giving the upper and lower limits of length for a given year group

(Table 5). This range of variation, based on the December 1953 sample, is shown in Fig. 2. Attempts to gain greater precision in defining the growth curve do not seem to be very profitable. The details of the curve can only apply to this locality, and further examination of the shells with several

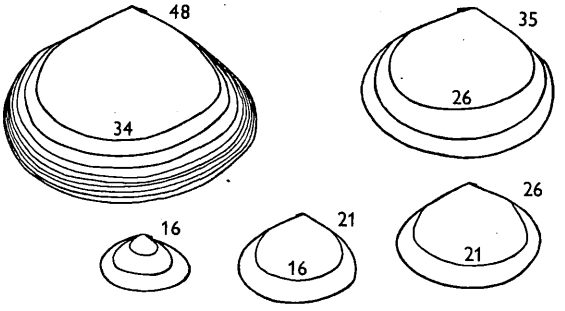

Fig. 1. *S. plana.* Series of shells used to construct the growth curve given by the broken line in Fig. 2. All are drawn to the same scale. The numbers above each shell and on some of the rings give the length in mm.

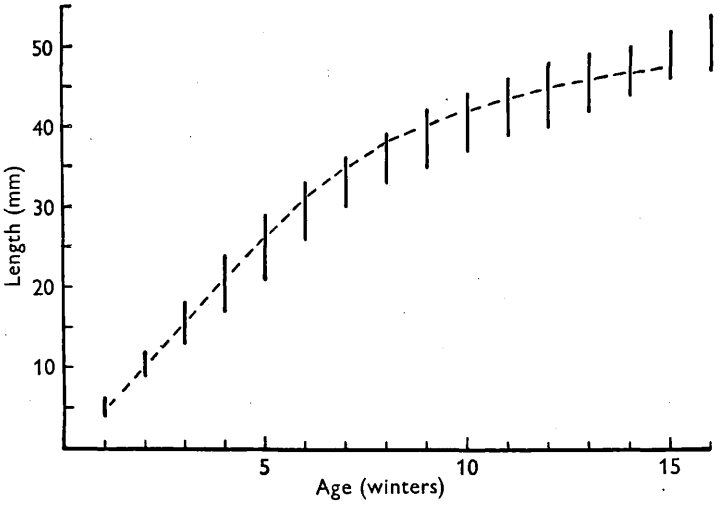

Fig. 2. *S. plana.* Growth curve constructed using the specimens in Fig. 1 (broken line), and the range of variation (shown by vertical lines) found in other series of shells.

growth rings often reveals a narrow interval between two wider ones, indicating that some years are better for growth than others.

If the mid-point in the range of variation given for each year group in Table 5 is taken as representative of an 'average' animal of that year the following summary can be given of the growth of *Scrobicularia.* A length of

5 mm is reached by the first winter. During each of the next two years another 5 mm is added to the length. Between the third and fourth winters 6 or 7 mm are added, thereafter the growth rate steadily decreases until the ninth or tenth winter, after which a fairly steady addition of about 2 mm per year is made.

TABLE 3. GROWTH RINGS ON MEDIUM-SIZED *SCROBICULARIA PLANA*

Sample collected in December 1953

Length of shell (mm)	No. examined	Mean length of growth rings (mm)	
		Last	Penultimate (when visible)
21	10	15·4	—
26	10	21·0	—
30	10	26·4	21·5 (4)*
35	10	32·2	26·5 (7)
38	5	35·0	30·0 (2)

* The numbers in parentheses give the number with two growth rings visible.

TABLE 4. GROWTH RINGS ON LARGE *SCROBICULARIA PLANA*

Sample collected in December 1953

Length 46 mm with 7 rings between one at 36 mm and the shell edge
Length 46 mm with 5 rings between one at 38 mm and the shell edge
Length 46 mm with 5 rings between one at 39 mm and the shell edge
Length 46 mm with 6 rings between one at 35 mm and the shell edge
Length 46 mm with 6 rings between one at 33 mm and the shell edge
Length 50 mm with 7 rings between one at 36 mm and the shell edge
Length 50 mm with 5 rings between one at 43 mm and the shell edge
Length 50 mm with 6 rings between one at 37 mm and the shell edge

TABLE 5. VARIATION IN LENGTH WITH AGE IN *SCROBICULARIA PLANA* FROM THE GWENDRAETH ESTUARY

Age (winters)	Length (mm)	Age (winters)	Length (mm)
1	4–6	9	35–42
2	9–12	10	37–44
3	13–18	11	39–46
4	17–24	12	40–48
5	21–29	13	42–49
6	26–33	14	44–50
7	30–36	15	46–52
8	33–39	16	47–54

Raymont (1955) has studied the early growth of *S. plana* in Kyle Scotnish, Scotland. Here the shells reach a length of 6 mm by the first autumn and 16 mm by the second. This is clearly a much greater increase during the second season than that found in the Gwendraeth, and may be due to the artificial fertilization of Kyle Scotnish.

Table 5 and Fig. 2 indicate that the age of shells above 20 mm in length cannot be determined accurately. For instance, a shell with a length of 33 mm might be 6, 7 or 8 years old. When even larger shells are considered the accuracy diminishes still further. A shell with a length of 46 mm may be anywhere between 11 and 15 years old. Some idea of the variation in large shells can be deduced from Table 4. Some individuals appear to live for about 18 years.

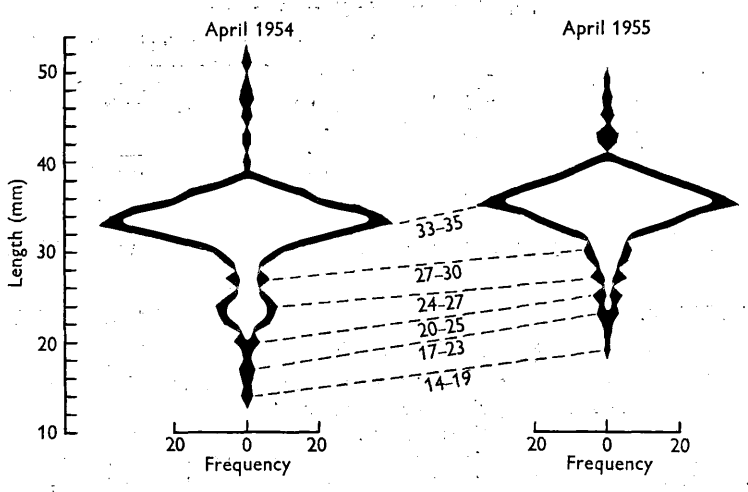

Fig. 3. *S. plana.* Shift in the peaks of length frequency distribution during one year.

LENGTH FREQUENCY DISTRIBUTION

In the preceding section it was assumed that the growth rings on the shell are annual rings. In order to test this assumption two samples were taken at a year's interval (April 1954 and 1955) to see how the peaks of the length frequency kites moved (Fig. 3). The shifts of the peaks are in fair agreement with the intervals between growth rings and all lie within the range of variation given in Table 5. If the mean size of the whole sample is considered there is a shift from 32·1 to 34·6 mm which is of a similar order to the increase in length between the seventh and eighth winters.

The earlier samples give no indication of the numbers of small shells so that no idea could be gained of the numbers of spat surviving. The July 1955 sample, taken with a 1 mm-mesh sieve, and sieved very gently so as not to destroy the delicate shells, showed a very small percentage of small shells. Only seven of the 483 shells in the sample were below 10 mm in length. This indicates that there is at present very little successful settlement of spat in the area. The dense adult population is probably the cause of this. Five

hundred inhalant siphons working in a square metre would make the area a most difficult one for a recently metamorphosed *Scrobicularia* to survive in. Nevertheless, a few do survive, and it may be that when the present population bulge at a length of 30–40 mm dies out there will be heavier successful settlements of spat.

SUMMARY

Populations of *Scrobicularia plana* with densities up to 1025 per m^2 are found in the Gwendraeth Estuary.

In a region where the population density was about 500 per m^2 it was found that some specimens lived for 16–18 years and reached a length of 54 mm. An approximate growth curve is given.

There is very little successful settlement of spat in the sampling area; less than 2% of the shells were under 10 mm in length in July 1955. This low figure is attributed to the feeding activities of the dense adult population making the area difficult to settle in.

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Osmoregulatory Function of the Contractile Vesicle of *Asplanchna*

THE contractile vesicle of the rotifer *Asplanchna priodonta* is a thin-walled muscular sac. It receives fluid from two ducts, each of which is headed by four flame bulbs. When the vesicle contracts it forces the fluid through a third duct which opens on the surface of the animal. Previous records of the rate of contraction of this vesicle have been in disagreement. Masius[1] states that there are ten contractions per min., while Nachtway[2] says that there are only one or two per min. My own observations on freshly collected specimens from Regent's Park Lake show a mean time of 2 min. 6 sec. for one contraction, in lake water at 20° C. Calculation of the rate of output in lake water gives a figure of 1,972μ^3 per sec., which is more than twice as high as the rate of vacuolar output by the largest freshwater ciliates[3]. The time taken to eliminate a volume equal to that of the body is about 300 min., which is much longer than in most freshwater ciliates and is approached only by *Frontonia leucas*[4].

When the salinity of the external medium is increased to 1‰ the time per contraction increases to 3 min. 8 sec., and when the salinity is increased

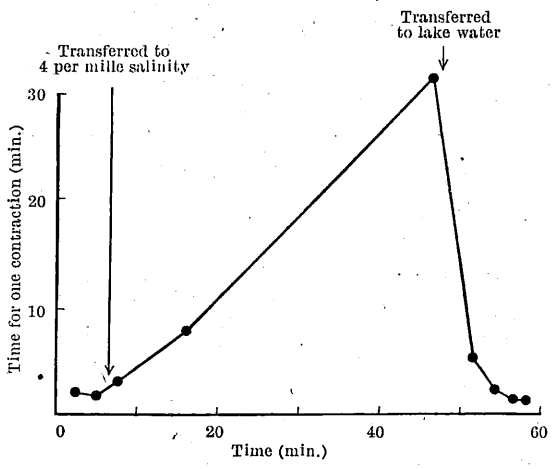

Fig. 1

to $2^0/_{00}$ the time per contraction becomes 8 min. 23 sec. During these experimental changes of the external medium there were no measurable changes in the ultimate diameter of the vesicle. At still higher salinities the interval between contractions becomes too long to observe conveniently. This effect of increase in salinity is reversible. Fig. 1 shows the effect of transferring an individual to a salinity of $4^0/_{00}$ and then back to lake water. Although the contractile vesicle is a complex syncytial organ, its behaviour in relation to the salinity of the external medium is strikingly similar to the contractile vacuole of the Ciliata and indicates a similar osmoregulatory function.

J. GREEN

Bedford College,
Regent's Park,
London, N.W.1.
Nov. 27.

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Parasites and epibionts of Cladocera in rock pools of the Tvärminne archipelago

J. GREEN

The small pools on the rocky islands of the Baltic coast have attracted a good deal of attention from zoologists; the fauna is well known. Less attention has been paid to the parasites and epibionts of the animals living in the pools. The work described in this paper is intended to remedy this with regard to the Cladocera.

LAGERSPETZ (1955) has studied the distribution of three species of *Daphnia* in rock pools of the Tvärminne archipelago. He gives references to general descriptions of the locality and to earlier studies on the rock pools, so that there is no need to repeat the same information here. All that need be said is that the area has numerous small islands with rock pools in abundance, making it ideal for a survey of the present type.

Methods

Collections were taken from the rock pools with a small, fine-meshed plankton net. If any detritus was present in the pool it was stirred up and a sample taken to search for bottom-dwelling Cladocera. All the material was examined fresh, mostly on the day of collection, occasionally on the following day. The dimensions of each pool from which a sample was taken were recorded, and the salinity of the water determined by titration with silver nitrate, potassium chromate being used as indicator.

The routine examination for parasites and epibionts consisted of a search through a variable number of specimens, but before a negative result was recorded at least 50 specimens were examined. Previous experience had shown that the search for parasites was more likely to be successful if large (old) specimens were examined. The search for epibionts was aided by selecting females with well developed embryos in the brood pouch, that is females near the end of an instar. As one might expect, epibiotic populations are better developed on such specimens than on those which have recently moulted. When estimates of the incidence of infection were made, the animals to be examined were, of course, taken at random.

In giving details of the distribution of certain of the parasites and epibionts I have used the names of islands that are in current use at the Zoological Station, Tvärminne.

Analysis of distribution

The survey was made during the months of July and August 1956. A total of 67 pools containing Cladocera were examined. These pools were all small, being at most 3 or 4 metres in diameter and less than 40 cm. deep. All the pools could be classified into LEVANDER's (1900) types 3, 4 and 6. The salinity in the pools ranged from less than 0.1 $^0/_{00}$ to 6.4 $^0/_{00}$.

The parasites (P) and epibionts (E) and the number of pools in which each was found, are listed in the following.

BACTERIA: *Spirobacillus cienkowskii* Metchnikoff P. 6, White fat cell bacterium P. 4, Filamentous bacterium E, 7.
TRICHOMYCETES: *Amoebidium parasiticum* Cienkowski E. 3.
ALGAE: *Characiopsis* sp. E. 7, *Stylosphaeridium inhaerens* (Bachman) Pascher E. 7, *Colacium vesiculosum* Ehrenberg E. 21.
SPOROZOA: *Thelohania cladocera* Jirovec P. 1, *Octosporea bayeri* Jirovec P. 4.
PERITRICHA: *Rhabdostyla pyriformis* Perty E. 3, *Rhabdostyla conipes* Kahl E. 1, *Epistylis helenae* nom. nov. E. 19, *Intrastylum invaginatum* Stokes E. 2, *Vorticella octava* Stokes E. 32.
ROTIFERA: *Brachionus rubens* Ehrenberg E. 6.

One additional epibiont has been omitted from this list. This is the flagellate *Cephalothamnium caespitosa*, which was found attached to *Daphnia magna* in one pool, but was probably overlooked in several others because it is very small and not easy to find.

Table 1. Occurrence of parasites and epibionts on Cladocera. The figures in brackets give the number of pools in which each cladoceran was found.

	Daphnia magna (48)	*Daphnia pulex* (23)	*Daphnia longispina* (12)	*Scapholebris mucronata* (11)	*Chydorus sphaericus* (59)	*Alona rectangula* (19)
Spirobacillus cienkowskii	3	1	—	—	4	—
White fat cell bacterium	4	—	—	—	—	—
Filamentous bacterium	4	4	—	—	2	—
Amoebidium parasiticum	1	1	—	—	—	—
Characiopsis sp.	5	3	—	—	—	—
Stylosphaeridium inhaerens	4	2	1	—	—	—
Colacium vesiculosum	14	11	1	—	4	—
Thelohania cladocera	—	—	1	—	—	—
Octosporea bayeri	4	—	—	—	—	—
Rhabdostyla pyriformis	1	—	2	—	—	—
Rhabdostyla conipes	—	—	1	—	—	—
Epistylis helenae	14	8	1	—	2	—
Intrastylum invaginatum	2	—	1	—	2	—
Vorticella octava	28	8	2	1	9	—
Brachionus rubens	4	—	2	—	—	—

Table 1 gives the number of occurrences of each organism on the six most abundant Cladocera. Five other species of Cladocera were found in the rock pools. Of these, *Alona affinis* (Leydig), *A. quadrangularis* (O. F. Müller) and *Alonella excisa* (Fischer) were each found only once, *Alona tenuicaudis* Sars was found twice, and *A. guttata* Sars three times. On account of their infrequency of occurrence these species have been omitted from the general analysis.

Several interesting features are brought out by Table 1. The first is that no parasites or epibionts were found on *Alona rectangula*. Secondly, the number of times that *Chydorus sphaericus* was found with epibionts is much lower than one would expect from its abundance and frequency of occurrence, particularly when it is noted that it was found together with *Daphnia magna*, the most infested species, in 36 of the pools. It is evident that there is some selection exercised by the epibionts when finding their host. The sparseness of epibionts on essentially bottom-dwelling animals like *Alona* and *Chydorus* is surprising because the general experience of workers on epibiotic peritrichs is that bottom-dwelling species are usually more heavily infested than related free-swimming species (cf. PRECHT 1935).

Scapholebris mucronata was almost completely free from epibionts. This may be due to it being a surface-film-dweller and having at least part of its surface with hydrofuge properties.

It was not possible to determine the distribution of epibionts in relation to salinity with any precision. Only a few pools were found with a salinity over 4 $^0/_{00}$ and containing Cladocera. *Vorticella octava* was the only epibiont found in salinities above 4 $^0/_{00}$.

The three most frequent epibionts, *Vorticella*, *Epistylis* and *Colacium*, show certain peculiarities in their distribution (Table 2). Only four pools contained all three. *Vorticella* was found in 32 pools, and in 22 of these it was not accompanied by either of the other two. In contrast with this, both *Epistylis* and *Colacium* were only found alone five times each. This indicates that *Vorticella* is most frequently found in situations where *Colacium* and *Epistylis* do not occur. There is probably some competition for space on the surface of the host between *Vorticella* and the other epibionts. When *Vorticella* was found together with *Epistylis* it was noticed that they tended to occupy different parts of the host's surface. *Vorticella* occupied the carapace and was particularly dense on its margins, while *Epistylis* was found mainly on the antennae. When only one of these peritrichs was present it tended to cover the whole of the host's surface.

Table 2. The numbers of pools in which *Vorticella*, *Epistylis* and *Colacium* were found as epibionts on Cladocera.

Vorticella octava	*Epistylis helenae*	*Colacium vesiculosum*	found with
22	6	8	*Vorticella*
	5	12	*Epistylis*
		5	*Colacium*

There may also be competition between *Colacium* and *Vorticella*. It has been shown elsewhere that competition does occur between peritrichs and *Colacium* (GREEN 1955). One difficulty with the data from Tvärminne seems to be the absence of any evidence of competition between *Colacium* and *Epistylis*. The explanation of this difference in the occurrence of *Vorticella* and *Epistylis* with *Colacium* might be as follows. *Epistylis* has a non-contractile stalk which elongates during growth and carries it above the general level of *Colacium*. Thus in competing for space *Epistylis* only needs enough room to attach its stalk. *Vorticella* has a contractile stalk, which again is long enough to carry the body

above the level of *Colacium*, but when the stalk contracts it forms a spiral which takes up much more space than the diameter of the stalk. The frequent contractions of the stalk of *Vorticella* will bring the body into collision with *Colacium*, so causing a mechanical disturbance to both species. The outcome of any competition between *Vorticella* and *Colacium* will depend to some extent on the amount of light reaching the pool, for it has been shown that light is an important factor in the competition between *Colacium* and peritrichs (GREEN 1955).

Systematic notes on parasites

Spirobacillus cienkowskii Metchnikoff 1889. – The disease caused by this bacterium can often be recognised in the field because the affected individuals become scarlet in colour. If an infected individual is examined with a microspectroscope, two absorption bands are found; one at 546 – 550 mμ and the other at about 500 mμ. The latter band is broader than the former. According to Metchnikoff, the bacterium goes through a cycle which lasts for about five days and ends with the death of the host. JIROVEC (1932) described *Spirobacterium daphniae*, which may be the same as the species described by Metchnikoff.

The records made during the present survey are as follows.

1) In *Chydorus sphaericus* on the unnamed island W. of Rovholmen.
2) In *Daphnia magna* on the unnamed island S.W. of Rönnharukobben.
3) In *Chydorus sphaericus* on Storsundsharun.
4) In *C. sphaericus* and *Daphnia magna* on Spikarna.
5) In *D. magna* and *D. pulex* in separate pools on the unnamed island W. of Rovholmen.

In all these pools the frequency of infection was very low, of the order of 0.5 % infection.

White bacterial disease. – A bacterium which infects the fat cells of *Daphnia* was found in four pools; two on the unnamed island W. of Rovholmen and two on Brännskär. *Daphnia magna* was the host in all the pools. This bacterium is easily detected because it causes the fat cells to appear white by reflected light.

The population of *Daphnia magna* in one of the pools on Brännskär was examined in some detail on three separate occasions. Table 3 gives the composition of the population on the three dates. On the first occasion the population was in fairly good condition; 60 % were producing parthenogenetic eggs, and only 4 % were infected with the bacterium. Twelve days later there was a great drop in the % of females with parthenogenetic eggs and an increase to 20 % infected. After a further 8 days the % infected had fallen slightly, but the % with parthenogenetic eggs decreased still further. During the period of observation there was a considerable increase in the % of females with ephippia and in males.

Table 3. Percentage composition of a population of *Daphnia magna* infected with the White fat cell bacterium.

	parthenogenetic ♀♀	♀♀ with ephippia	♀♀ of mature size without eggs	immature ♀♀	♂♂	% of total population infected
28th July	60	5	7	21	7	4
9th August	15	14	8	50	13	20
16th August	7	23	15	30	25	18

Table 4. Percentage infection of *Daphnia magna* with White fat cell bacterium.

	parthenogenetic ♀♀	♀♀ with ephippia	♀♀ of mature size without eggs	immature ♀♀	♂♂
9th August	30	50	87	7	9
16th August	10	28	66	1	1

The effect of the bacterium on egg production was also shown by examining the % infection of females in various reproductive states (Table 4). It is clear that the % infection of females with parthenogenetic eggs is much lower than in ephippial females, which in turn is lower than the % infected

in females of mature size without eggs. It was also found that of the females with parthenogenetic eggs those which were infected carried fewer eggs than those which were free from infection. Random samples of 20 females from each group were examined on August 9th. The infected females were carrying an average of 5.2 eggs each, while those which were free from infection carried 8.6 eggs each.

Octosporea bayeri Jirovec 1936. – This species was originally described from Czechoslovakia, where it was found in the fat body of *Daphnia magna*. Each pansporoblast produces eight spores which vary in length from 5 – 9 μ.

In the present survey this microsporidian was found on four islands: Långskärsstorsgrundet, Storsundsharun, the unnamed island W. of Rovholmen and the unnamed island S.W. of Rönnharukobben. The host was *Daphnia magna*.

A random sample of 200 *D. magna* from the pool on the unnamed island was examined to estimate the degree of infection. The results are given in Table 6. There is clearly a higher incidence of infection among the females without eggs than among those with eggs. It seems that infection reduces egg production. The infected females with eggs only had 4 – 6 eggs in the brood pouch while those which were free from infection carried 7 – 11 eggs.

Table 5. Frequency of infection of *Daphnia magna* by *Octosporea bayeri* (200 examined).

	parthenogenetic ♀♀	♀♀ of mature size without eggs	immature ♀♀	♂♂
Free	20	28	40	74
Infected	2	18	2	16

Thelohania cladocera Jirovec 1936. – This species was first described from *Daphnia magna* and later recorded from *D. pulex* (Weiser 1947). The spores are pear-shaped, 3.0 – 3.4 μ long and 1.5 – 1.8 μ wide. Eight spores are formed by each pansporoblast. This species was found only once, infecting *D. longispina* in a small pool on Skarvkyrkan.

Systematic notes on epibionts

Filamentous bacteria. – A member of the Chlamydobacteriales was found attached to Cladocera in 7 pools; 6 of these were on the unnamed island W. of Rovholmen, the other was on Storsundsharun. The hosts were *Daphnia magna*, *D. pulex* and *Chydorus sphaericus*. In one of the pools on the unnamed island W. of Rovholmen this was the only epibiont found.

Amoebidium parasiticum Cienkowsky. – A detailed description of the life history and systematic position of *Amoebidium* has been given by Tuzet & Manier (1951). The stage most frequently found is the vegetative filament, which is colourless and up to 0.5 mm long with a diameter of 10 μ. This epibiont was found three times during the present survey. The records are as follows.

1) A few filaments on *Alona guttata* from a pool on Skarvkyrkan.
2) Abundant on *Daphnia magna* from a pool on the unnamed island S.W. of Rönnharukobben.
3) A few filaments on *Daphnia pulex* from a pool on the unnamed island W. of Rovholmen.

Characiopsis sp. – This alga was found in seven pools on four islands; the hosts were *Daphnia magna* and *D. pulex*. The specimens belong to a large species of *Characiopsis*. There is no distinct stalk and the cell does not become narrower towards the base. The cells reach a length of 63 μ, and have the apex rounded. Further work will be needed to determine which species these specimens belong to.

Stylosphaeridium inhaerens (Bachman) Pascher. – In seven pools on four islands this small green alga was found attached to the three species of *Daphnia*. It is a small species, about 10 μ long with a short gelatinous stalk. In the cell near to the stalk are two small vesicles. Pascher (1927) says that this species usually lives a tached to the blue-green alga *Anabaena*. It was very sparse on *Daphnia* in the rock pools.

Colacium vesiculosum Ehrenberg. – The systematics of the genus *Colacium* are in need of study. The problem has been discussed by Pringsheim (1953). I adopt his view that this species includes *C. sideropus* Skuja (1939) and *C. cyclopicola* (Gicklhorn 1925).

This was one of the most widespread epibionts in the rock pools; it was found in 21 pools on 9 islands. The hosts were *Daphnia magna*, *D. pulex*, *D. longispina* and *Chydorus sphaericus*. The density of infestation was very variable, though usually rather sparse. Two populations of *D. magna* were found with very dense coatings of *Colacium*.

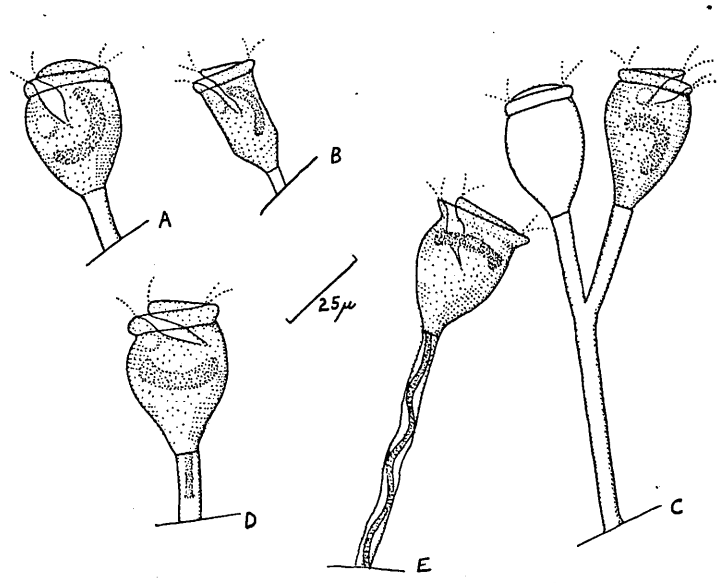

Fig. 1. Peritrichs found on Cladocera in rock pools of the Tvärminne Archipelago. A – *Rhabdostyla pyriformis*; B – *R. conipes*; C – *Epistylis helenae*; D – *Intrastylum invaginatum*; E – *Vorticella octava.*

Rhabdostyla pyriformis Perty. – The peritrichs found attached to Cladocera in the Tvärminne Archipelago are illustrated in Fig. 1. *R. pyriformis* was found in three pools, two on Skarvkyrkan and one on Långskärsstorgrundet. On the first island the host was *Daphnia longispina*, and on the second *D. magna* was the host.

Rhabdostyla conipes Kahl. – This peritrich was found in one pool on Brännskär. The host was *Daphnia longispina.*

Epistylis helenae nom. nov. – In 1905 FAURE FREMIET gave the name *Epistylis daphniae* to a species which he presumably found on a species of *Daphnia.* His description was brief and was concerned only with the stalk. In 1935 KAHL included *E. daphniae* in his monograph of the peritrichs, but he included under the same species a form which he figured and of which he says 'sie entspricht aber kaum dieser Art'. Later, NENNINGER (1948) confused the situation by describing *E. daphniae* Faure Fremiet var. *infundibulata*, which she says 'weist mit der von Kahl unter *E. daphniae* abgebild. Art Ähnlichkeit auf, dagegen nicht mit der Beschreibung der *E. daphniae* von FAURE FREMIET'. The confusion arises from her also giving *E. daphniae* Kahl as a separate species.

It seems clear from FAURE FREMIET's description of the stalk of his species that it is quite distinct from the species figured by KAHL, but KAHL did not give a separate name to his species. NENNINGER's name is clearly not acceptable owing to her confused presentation. I propose to use the name *Epistylis helenae* nom. nov. for the species found on *Daphnia* (and other Cladocera) and having the following characters. Body length 35 – 45 μ, stalk up to 120 μ long. Body shaped as in Fig. 1 C, with a flat disc and a curved sausage-shaped macronucleus. The contractile vesicle lies below the peristome. The surface of the body is faintly cross-striated. The stalk is simple and smooth. The colonies are small, usually with only 2 – 4 individuals.

Epistylis helenae was found in 19 pools on seven different islands. The hosts were *Daphnia magna, D. pulex, D. longispina* and *Chydorus sphaericus.* There was a marked tendency for *E. helenae* to occur most frequently on the antennae of its host.

Intrastylum invaginatum Stokes. – This species was found in two pools on Allgrundet. *Daphnia magna, D. longispina* and *Chydorus sphaericus* were the hosts.

Vorticella octava Stokes. – I have included the specimens found on Cladocera in the rock pools in this species because the body size (length 32 – 45 μ) and the position of the nucleus (Fig. 1 E) together with the general form of the body indicate that the specimens are, at least, closely related to *V. octava.*

This was the most widespread and frequent of all the epibionts; it was found in 32 pools on 9 different islands, and it was found on 5 of the 6 most frequent Cladocera (see Table 2). The stalk of this species grows to a length of 230 μ. The length of the stalk varies with the position of the epibiont on the body of its host, and is no doubt influenced by the currents produced by the swimming movements of the host.

One sample of *Daphnia magna* from a pool on the unnamed island W. of Rovholmen had a very dense coating of this epibiont. Three females of *D. magna* were found with chironomid larvae clinging to the tail spine and browsing on the bodies of *Vorticella*. The even contour formed by the bodies of *Vorticella* was broken up by the activities of the chironomid larva. This is an unusual occurrence and does not appear to have been reported previously.

Bracihonus rubens Ehrenberg. – The rotifers of the genus *Brachionus* have been monographed by AHLSTROM (1941), who gives a key for the separation of all the species.

During the present survey *B. rubens* was found in six pools: 3 on Skarvkyrkan, 1 on the unnamed island S.W. of Rönnharukobben, 1 on Spikarna and 1 on Segelskär. In 2 of the pools on Skarvkyrkan the host was *Daphnia longispina*; in all the others *D. magna* was the host. The numbers per individual host did not exceed 10.

This study was made while staying at the Tvärminne Zoological Station of the University of Helsinki. I am grateful to Professor PONTUS PALMGREN, the Director, for the facilities he provided and to the other Finnish zoologists at the station, particularly Mr. KARI LAGERSPETZ, for their help and hospitality. This work was aided by a grant from the Central Research Fund of London University.

Summary

A survey has been made of the parasites and epibionts of Cladocera in 67 rock pools on islands of the Tvärminne Archipelago.

The following parasites were found: *Spirobacillus cienkowskii*, another bacterium which makes the fat cells appear white, *Octosporea bayeri*, and *Thelohania cladocera*. The White fat cell bacterium and *Octosporea bayeri* were found to inhibit egg production by *Daphnia magna*.

Twelve species of epibionts were found, including filamentous bacteria, algae, protozoa and rotifers. The new name *Epistylis helenae* is given to a form which was originally included under *Epistylis daphniae* Faure Fremiet by KAHL (1936).

An unidentified chironomid larva was found clinging to *Daphnia magna* and eating the bodies of *Vorticella octava*, which was attached to the cladoceran.

An analysis of the distribution of the epibionts indicates that *Vorticella octava* competes with *Epistylis helenae* and *Colacium vesiculosum* for space on the surface of Cladocera.

Bedford College,
University of London

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EUDACTYLINA RACHELAE N.SP., A COPEPOD PARASITIC ON THE ELECTRIC RAY, *TORPEDO NOBILIANA* BONAPARTE

By J. GREEN
Bedford College, University of London

(Text-fig. 1)

Five species of the genus *Eudactylina* are known from British waters (Scott & Scott, 1913), and of these only one, *E. acuta* Van Beneden, is recorded in the *Plymouth Marine Fauna* (Marine Biological Association, 1957, p. 181). The species described below was found among the gill filaments of a large specimen of *Torpedo nobiliana* Bonaparte (det. P. G. Corbin) taken in trawl by R.V. *Sula* off Plymouth in July 1957. There were five females.

Eudactylina rachelae n.sp.

Description of the female

The body is elongated, maggot-shaped. The length, excluding the egg strings, is 2·3–2·5 mm. When fresh the colour is cream, with the gut showing through brown. The first pedigerous segment is fused with the head, the whole being covered by a single dorsal shield which is one and a third times as long as wide, and somewhat narrowed anteriorly. The second and third pedigerous segments have dorsal shields which are similar in length, while that of the fourth pedigerous segment is longer. The dorsal shield of the fifth pedigerous segment is smaller than those of the second and third. The genital segment is narrower than the preceding segments. The urosome (excluding the genital segment) has two segments; the first being roughly twice as long as the second. The dorsal surface of the body bears numerous crescent-shaped lamellar processes, which look like minute spinules when viewed from the side.

The antennules (Fig. 1B) are stout, of five podomeres, the last indistinctly separated from the fourth. The armature is similar to that of *E. acuta* and *E. similis*. The two principal spines lack the fringe of minute spinules along the upper edge, which is found in *E. similis*. The large spine on the second podomere has a series of minute crescent-shaped depressions, each of which appears to contain a spinule lying almost parallel to the border of the spine. The large spine on the third podomere has three shallow oblique teeth on each side (Fig. 1C).

The antennae (Fig. 1D) have four podomeres. The second podomere has a strong spine-like process on the inner side, while the third has two such processes and a seta. The third podomere also bears numerous crescent-shaped processes similar to those on the dorsal surface of the body. The

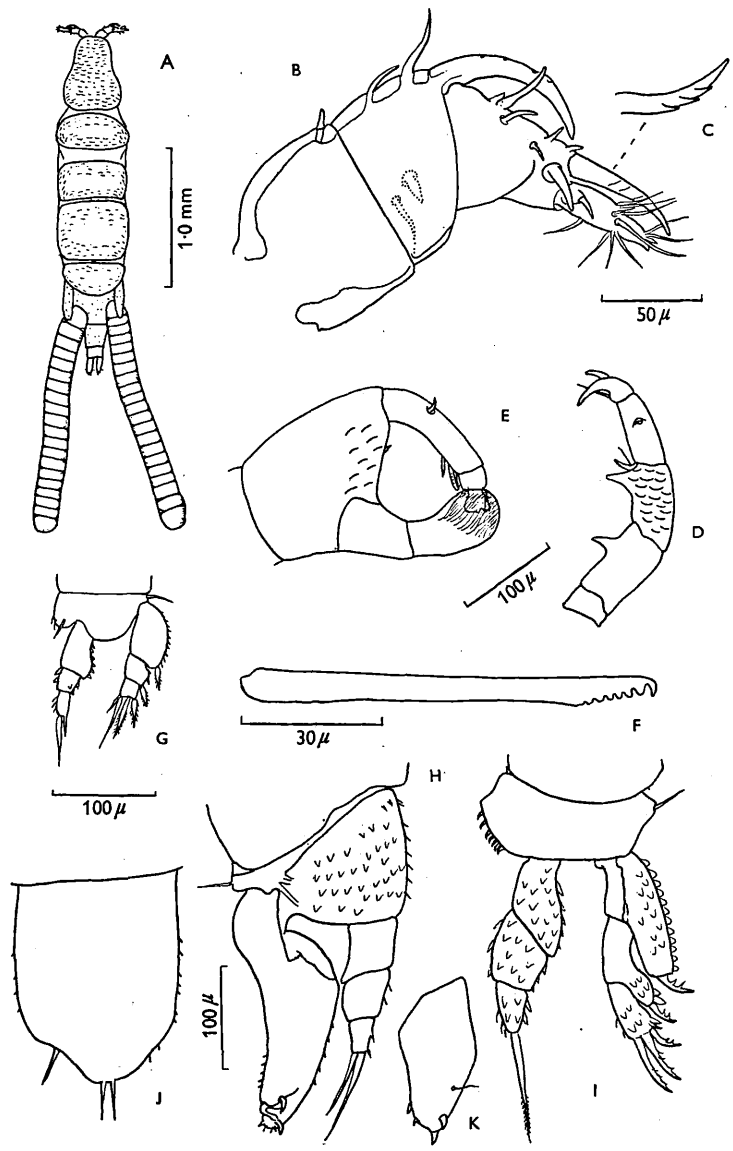

Fig. 1. A–K, *Eudactylina rachelae* n.sp., adult female. A, dorsal view; B, antennule; C, spine on third podomere of antennule; D, antenna; E, second maxilliped; F, mandible; G, first leg; H, second leg; I, third leg; J, fifth leg; K, ventral view of left caudal ramus. G–K are all drawn to the same scale.

fourth podomere bears what appears to be a sensory pit. The antenna ends in two stout spines and a seta.

The mandible (Fig. 1F) is long and straight, with a terminal hook and seven or eight short blunt teeth.

The maxilla, like that of *E. similis*, has two setae on the principal lobe and one on the secondary lobe.

The first maxilliped has three podomeres and terminates in a stout spine with two tufts of setae at its base.

The second maxillipeds (Fig. 1E) are large and chelate. The movable branch of the chela ends in an expanded cup-like structure which fits inside the thin, expanded and minutely fluted end of the immovable branch.

The first pair of legs is much smaller than the second to fourth pairs. The second basal podomere has one moderate and two small spines on its inner border. The endopod and exopod each have three podomeres.

The second legs have strongly modified exopods (Fig. 1H), while the endopod has three normal podomeres.

The third and fourth legs are similar in structure (Fig. 1I). The surface of the podomeres is drawn out into flattened spine-like processes which appear as short triangular lamellae when viewed from the side. The strong spines on the exopods are distinctly toothed.

The fifth leg consists of a single plate-like podomere, with three terminal setae and a few spinules along the lateral borders.

The caudal rami are one and a half times as long as the anal segment. Each ramus has two short terminal spines, two weaker inner spines and one feeble outer seta.

Remarks

This species is closely related to *E. similis* T. Scott, but can be separated by the differences given in Table 1.

TABLE 1

E. similis T. Scott	*E. rachelae* n.sp.
Large spine on third podomere of antennule with minute spinules along upper border	Minute spinules not present on this spine, but three oblique teeth present on each side
Mandible curved	Mandible straight
Movable branch of second maxilliped ends in a spine	Movable branch of second maxilliped ends in an expanded cup
First legs not much smaller than others. The inner border of the second basal podomere with two stout spines	First legs much smaller than others. Inner border of the second basal podomere with one moderate and two small spines
Spines on third and fourth legs not toothed	Spines on third and fourth legs toothed
Caudal rami without two inner spines	Caudal rami with two inner spines

Type material. The holotype and one paratype will be deposited at the British Museum. Three paratypes will be retained in my collection.

These specimens were collected while working at the Plymouth Marine Laboratory; my thanks are due to the Director and Staff for the facilities provided. My thanks are also due to Dr J. Llewellyn for bringing these copepods to my notice.

SUMMARY

Eudactylina rachelae n.sp. is described and figured. This copepod was found among the gill filaments of *Torpedo nobiliana* Bonaparte caught off Plymouth in July 1957.

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DACTYLOPUSIOIDES MACROLABRIS (CLAUS) (COPEPODA: HARPACTICOIDA) AND ITS FROND MINING NAUPLIUS

BY

J. GREEN

Bedford College, University of London

[Accepted 12th November 1957]

(With 13 figures in the text)

CONTENTS

INTRODUCTION

During an examination of some fronds of the brown seaweed *Dictyota dichotoma* Lamour, collected from Torquay in July 1957, several harpacticoid nauplii were found in small galleries, or mines, between the two epidermal layers of the alga. The purpose of this paper is to describe these nauplii and to determine their systematic position.

DESCRIPTION OF THE NAUPLIUS

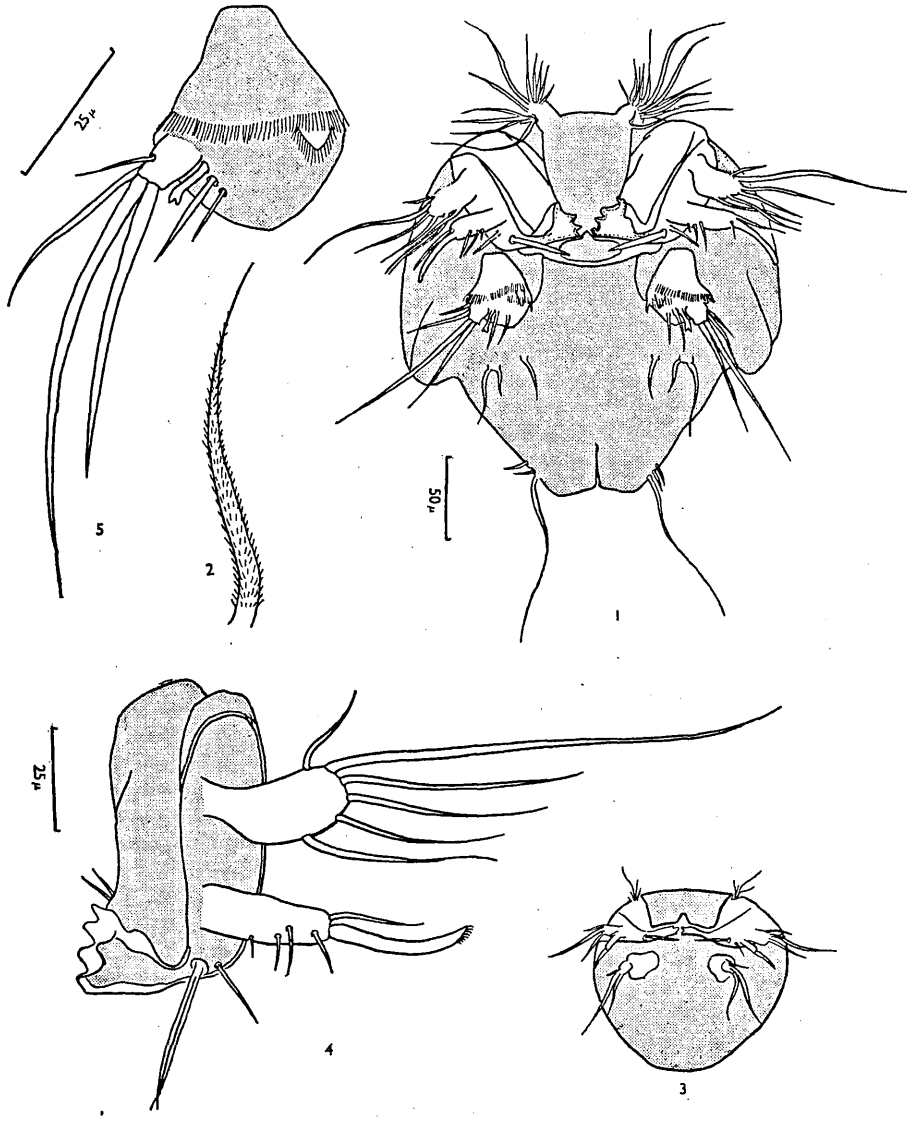
The nauplii vary in length from 103 to 234μ. The description which follows applies to the largest nauplius; the smaller nauplii were similar in structure, but had fewer setae on the appendages and body. Several specimens were mounted in polyvinyl-lactophenol and stained with lignin pink or chlorazol black.

The body of the nauplius is rounded, with the posterior part narrowed. In the largest specimen the narrower hindpart is clearly demarcated (Fig. 1), but in the smaller specimens the body is more evenly rounded.

The first antenna is a simple projection with a number of fine setae, some of which, when viewed under an oil immersion objective, have very fine processes projecting from all over their surfaces (Fig. 2). The second antennae are modified to form biting appendages; they have strong biting gnathobases. A large seta projects from each gnathobase towards the mouth. At first sight this seta appears to be blade like, but in fact it has two branches which lie side by side, or it might even be two setae lying side by side. However, there appears to be a common pit at the insertion, and I cannot separate two setae at the base. There are two small setae on the medial border, just inside the biting part. The endopod and exopod are each formed from a single podomere.

The exopod has six setae, while the endopod also has six, but more variable in size. One of the terminal setae on the endopod is large, and has fine unilateral feathering at its tip.

The third appendage, which must be the morphological equivalent of the adult mandible, consists of two podomeres. There is a large basal podomere, with a curious triangular flap near its medial border, and a ring of microtrichs, and a smaller, distal podomere bearing four setae, two of which are very



Figs. 1–5.—Nauplius of *Dactylopusioides macrolabris*. 1, largest nauplius, ventral view. 2, one seta from the first antenna, greatly enlarged. 3, smallest nauplius, drawn to the same scale as Fig. 1. 4, second antenna, ventral view. 5, third appendage, or mandible, ventral view.

large. Alongside the distal podomere, on the basal podomere, are three setae, one of which has a forked end.

Three setae are found on the body just behind the mandible ; these probably represent the maxillule. A further three setae are found in the position of future furcal setae.

DESCRIPTION OF THE ADULT

A single copepodid and a single adult female were found inhabiting mines on the same frond as the nauplii. They were both of the same brilliant crimson colour as the nauplii.

Using the key in Lang's monograph (1948) the adult female was identified as *Dactylopusioides macrolabris* (Claus). The original description by Claus (1866) and the description of *D. stampaliae* by Brian (1928) have also been examined. This has confirmed Lang's view that *D. macrolabris* and *D. stampaliae* are synonymous. In order to establish the validity of the identification of the single female as *D. macrolabris* most of the important taxonomic features are illustrated in Figs. 6–13. These figures also shows some features which have not previously been adequately illustrated. Figure 6 shows that some of the setae on the second podomere of the first antenna are stouter editions of those on the first antenna of the nauplius. This strengthens the view that the female really is the adult of the nauplius. The only really conclusive way of establishing this would be to find a female with fully developed nauplii in its egg sac, as Harding (1954) did with *Thalestris rhodymeniae.*

The distal podomere of the exopod of leg 2 had two setae on its inner border (Fig. 10), a point which agrees with Lang's view that Monard's (1935) figure showing three setae in this position is incorrect.

Monard's figure of the first leg shows a strong straight seta on the inner border of the second podomere of the exopod. This is very difficult to see in the specimen from Torquay, in which it is weak, curved and transparent.

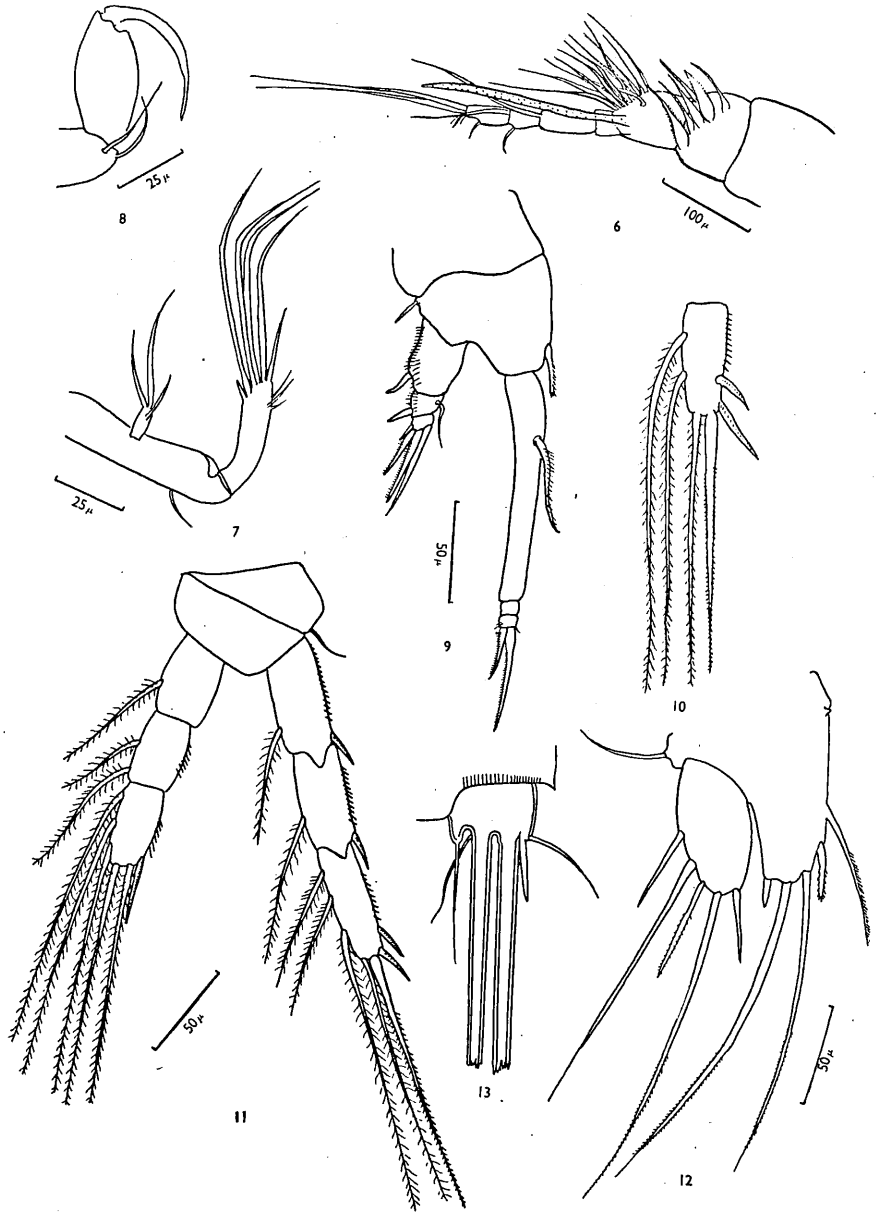
DISCUSSION

The habit of burrowing between the epidermal layers of an alga, as done by the nauplii of *Dactylopusioides*, is reminiscent of the habits of various larval insects which burrow in a similar manner in the leaves of phanerogamous plants. These insects are known as leaf miners (see Hering, 1951, for a general account). The term frond miner seems a reasonable parallel to apply to the nauplii described above.

The present record of *Dactylopusioides macrolabris* appears to be the first for Britain, and is the first record of a miner in the fronds of a phaeophycean alga. Three other species of harpacticoid copepods are known to have similar habits, but mining in Rhodophyceae instead of Phaeophyceae.

Bocquet (1953) described *Diarthrodes feldmanni*, which is found in the fronds of several species of Rhodophyceae. All stages of this copepod, from the first nauplius to the fifth copepodid are found in galleries in the thallus of the alga. The copepod eats the interior of the thallus, leaving only the cuticle. The early stages form a linear gallery, but the later stages widen out a terminal chamber.

Fahrenbach (1954) described *Diarthrodes cystoecus*, the adults of which live in the water-filled, bladder-like thalli of the red alga *Halosaccion glandiforme*. The nauplii burrow into the wall of the bladder, and produce small cysts which project above the outer surface of the algal thallus. The copepod



Figs. 6–13.—Adult female of *Dactylopusioides macrolabris*. 6, first antenna. 7, second antenna. 8, maxilliped. 9, first leg. 10, third podomere of the exopod of the second leg. 11, third leg. 12, fifth leg. 13, caudal ramus ; the two long setae are shown as broken off.

becomes mature within the cyst, and escapes to the inside of the bladder through a small hole.

Both Bocquet and Fahrenbach state that they found the nauplii of their respective species, but they did not describe them.

Harding (1954) redescribed the nauplius originally described by Brady (1894) as *Fucitrogus rhodymeniae*, and described the adult, showing it to be a member of the genus *Thalestris*, Harding's description of the nauplius clarified the homologies of the appendages, and shows marked similarities to the nauplius described in the present paper, particularly in the presence of a powerful biting gnathobase on the second antenna. The first antenna of the nauplius of *T. rhodymeniae* bears peculiar fir cone-like processes (presumably modified setae) ; these are absent from the nauplius of *Dactylopusioides*, but certain of the setae show a slight tendency in this direction in their structure (Fig. 2).

The genera *Thalestris*, *Diarthrodes* and *Dactylopusioides* all belong to the family Thalestridae, many members of which are recorded in Lang's monograph as living among algae. It seems probable that other members of this family will be found to have frond mining habits, and it will be of great interest to see if all the species have the second antennae of their nauplii as strongly modified as the second antennae of *Thalestris rhodymeniae* and *Dactylopusioides macrolabris*.

ACKNOWLEDGMENTS

The specimens described above were collected while working at the Plymouth Marine Laboratory ; my thanks are due to the Director and staff for the facilities which they provided. Dr Mary Park kindly identified *Dictyota* for me. I am most grateful to Dr J. P. Harding for some helpful discussion and for reading the manuscript.

SUMMARY

The nauplius of *Dactylopusioides macrolabris* (Claus) is described and figured. The second antenna is modified to form a biting appendage, while the mandibles are not capable of biting.

The nauplii and an associated adult female were found in galleries in the fronds of the brown alga *Dictyota dichotoma*, from Torquay, Devon. This is the first British record of this copepod, and the first record of a copepod mining the fronds of a member of the Phaeophyceae.

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COPEPODA PARASITIC ON BRITISH AMPHIPODA (CRUSTACEA), WITH A DESCRIPTION OF A NEW SPECIES OF *SPHAERONELLA*

BY

J. GREEN

Bedford College, University of London

[Accepted 11th February 1958]

(With 26 figures in the text)

CONTENTS

INTRODUCTION

In July 1957 a parasitic copepod was found in the brood pouch of the amphipod *Erichthonius brasiliensis* (Dana) at Plymouth. While the systematic position of this copepod was being investigated, Dr J. P. Harding suggested that I should also examine the unnamed copepods found as parasites on amphipods in the collection at the British Museum (Natural History). This paper is a report on this examination.

MATERIAL

Two specimens were obtained from Plymouth.

A single specimen, from the Norfolk coast was received from Mr R. Hamond.

The British Museum material was in seven tubes. One tube, from the Norman collection, contained five species of amphipods. Four of the specimens were infected with copepods, while the fifth, *Perioculodes longimanus* (Bate & Westwood) was not infected, but had several abnormally large eggs in its brood pouch. One of the specimens in the tube had lost most of its appendages, making determination by ordinary methods difficult. This specimen was sent to Mr G. M. Spooner, who determined it as a female *Aora typica* Kroyer. The remaining tubes each contained a single host species, or copepods collected from a single host species.

Family HERPYLLOBIIDAE

Rhizorhina ampeliscae Hansen, 1892

Material examined. (1) One adult female from the brood pouch of *Ampelisca diadema* (Costa), Guernsey 1906 (Norman Collection). (2) Two adult females and a male from *Ampelisca brevicornis* (Costa), Garroch Head, Clyde area, July 1954 ; collected by Dr R. B. Pike.

These appear to be the only records of this species since Hansen's (1892) original description. Leigh-Sharpe (1926), in his account of the Herpyllobiidae, gave figures of this species, taken from Hansens original drawings. The copy of the male is inaccurate ; the setae on the second abdominal segment have been omitted, and the large flattened aesthetasc on the first antenna is shown as being terminal, resembling a podomere. These mistakes have been reproduced by Oorde-de-Lint and Schuurmans Stekhoven (1936). The abdominal setae were correctly figured by Hansen, and he also showed the aesthetasc in its correct position, coming from the middle of the third podomere.

Distribution. Norway, Denmark (Hansen) ; now recorded from the Clyde and Guernsey.

Hosts. *Ampelisca brevicornis* (Costa) (Hansen, as *A. laevigata*, and present paper), *A. diadema* (Costa) (present paper).

Family CHONIOSTOMATIDAE

Sphaeronella leuckarti Salensky, 1868

Syn. *S. elegantula* Hansen, 1897.
? *S. aorae* Scott, 1905.

Material examined. (1) One adult female and two pupae on *Cheirocratus sundevalli* (Rathke), E. Little Cumbrae Island, Clyde, 4–6 m. August 1954. Collected by Dr R. B. Pike. (2) One adult female and one male on *Aora typica* Kröyer, Guernsey 1906 (Norman Collection).

When Hansen wrote his monograph (1897) he described eight species of the *leuckarti* group of *Sphaeronella*, and said ' I must consider *Sphaer. Leuckarti* Sal. as a ninth (to me unknown) species '.

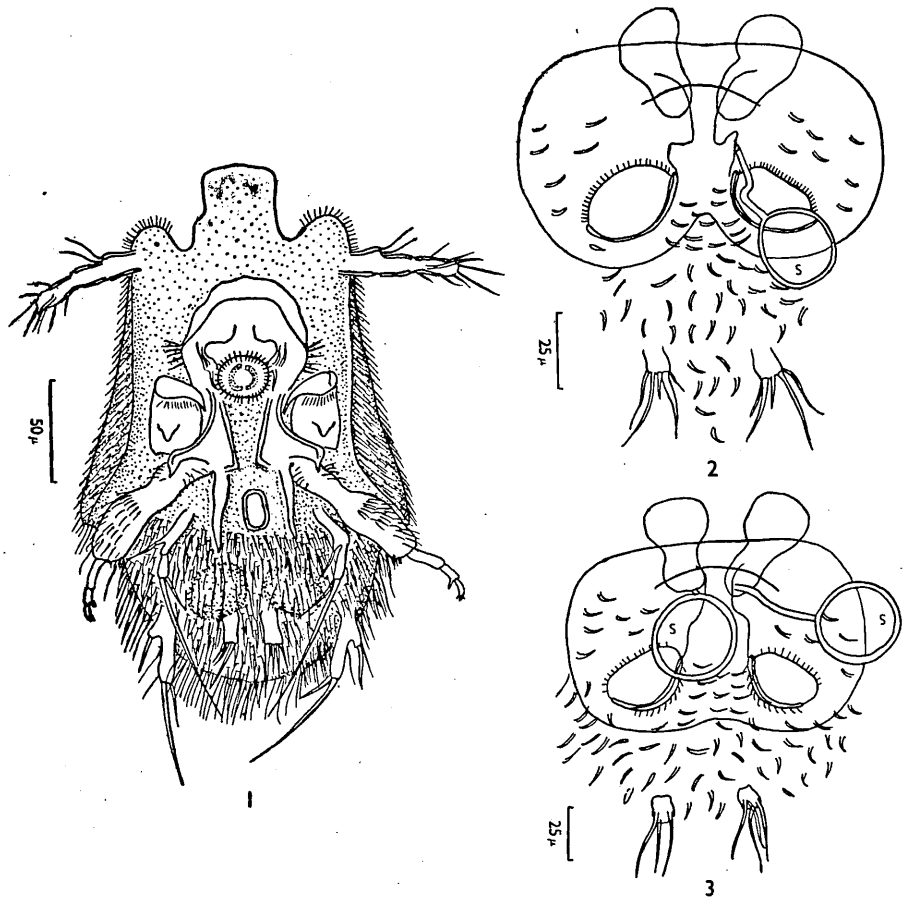
The specimens from *Cheirocratus sundevalli* were at first identified as *Sphaeronella elegantula* ; they agreed in detail with Hansen's description. When the specimens from *Aora typica* were examined they were also found to agree with the description of *S. elegantula*. *Aora* belongs to the same family as *Microdeutopus*, from which Salensky described *S. leuckarti*, and when Salensky's figures were examined it became evident that *elegantula* is a synonym of *leuckarti*.

This synonymy is being based primarily on the males, which are more characteristic than the females. The points in common between Hansen's description of *elegantula*, Salensky's description of *leuckarti*, and the male which I have examined (Fig. 1) are as follows.

All have the characters of the *leuckarti* group as given by Hansen, and in addition the following detailed similarities.

The sub-median skeleton is produced backwards behind the base of the maxilla to form a short spike over the outer edge of the base of the maxillipede. The processes at the inner margins of the bases of the maxillipedes are very large, and there is an irregular ring-like structure between them. There is a distinct triangular projection on the basal podomere of the maxilla. The caudal rami are longer than broad, and have about four terminal setae. These characters will serve to distinguish the male of *leuckarti* from any other species in its group.

The females described by Hansen and Salensky agree in shape and size, and in the general form of the genital area. There is some variation in the degree of concavity of the posterior border of this area. Hansen figures a specimen in which this border is almost straight, but says that in another specimen the border was slightly more concave. The specimen figured by Salensky had a posterior border similar to that of the female from *Aora* (Fig. 2).

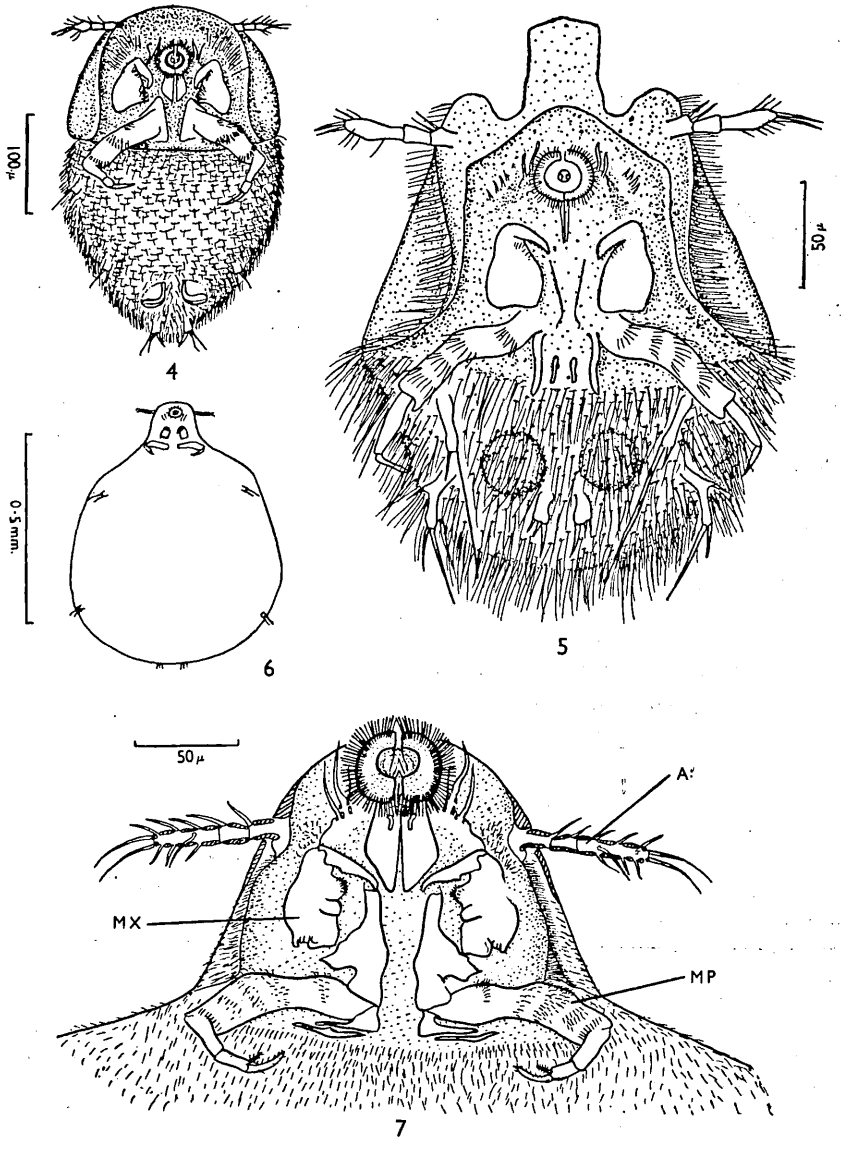


Figs. 1–3.—*Sphaeronella leuckarti.* 1, male, ventral view. 2, genital area of female from *Aora typica.* 3, genital area of female from *Cheirocratus sundevalli.* s–spermatophores.

The female from *Cheirocratus* (Fig. 3) had a less concave border. Both females examined by me had a number of peculiar membranous hairs on and behind the genital area ; similar hairs were described by Hansen from one of his specimens. Both Hansen and Salensky figure only two setae on the caudal rami of the female. Figs. 2 and 3 show that four or even five terminal setae may be present. This brings the caudal ramus of the female into line with that of the male. However, these setae are easily broken off, and the rami may present a very different appearance.

Scott (1905) described *Sphaeronella aorae* from '*Aora gracila* (Bate)'. This is probably a synonym of *leuckarti* ; it is certainly a member of the *leuckarti* group, but the figures and description are quite inadequate to decide which species it should be assigned to.

Distribution. Naples (Salensky), Denmark (Hansen). Now recorded from the Clyde and Guernsey.



Figs. 4–7.—*Sphaeronella danica.*—4, young female, ventral view. 5, male, ventral view. 6, female, ventral view. 7, head of female, ventral view, A–antennule ; MX–maxilla ; MP–maxillipede.

Hosts. *Microdeutopus gryllotalpa* Costa (Salensky), *Cheirocratus sundevalli* (Rathke) (Hansen and present paper), *Aora typica* Kröyer (present paper).

Sphaeronella danica Hansen, 1897

Material examined. (1) Two adult females, one very young female, one male and a pupa from the brood pouch of *Corophium crassicorne* Bruzelius, Guernsey 1906 (Norman Collection). (2) Two females, one of which has fully developed larvae in its egg sacs, from brood pouches of *Erichthonius brasiliensis* (Dana), Asia Shoal, Plymouth, July 1957.

The adult female from *Corophium crassicorne* was quite typical of the species, agreeing with Hansen's original description in most details. The only exception was a detail of the genital area, which is discussed later. Fig. 4 shows a very young female, which contrasts greatly with the adult. The male (Fig. 5) was not figured by Hansen. The only differences which Hansen gives from the male of *elegantula* (*leuckarti* Salensky in this paper) are the more clumsy shape and the slightly shorter caudal stylets. There are certain other differences. The processes at the bases of the maxillipedes are shorter and weaker in *danica*, and there is a pair of projections between these, instead of the ring-like structure which is present in *leuckarti*. The maxillule lacks a posteriorly directed process, and there is no spine formed on the outer side of the base of the maxillipede.

The females from *Erichthonius brasiliensis* may be described as follows. The body is sub-globular, with a well defined head (Figs. 6 and 7). The length is about 0·7 mm. and the maximum width 0·52 mm. In life the body was coloured orange.

The antennules have two sub-equal basal podomeres and a much longer distal podomere. Eleven setae are present on the whole antennule, the terminal pair being the longest.

The rostrum is of the usual pattern for the genus, with moderately long setae around the border.

The antennae could not be found.

The maxillules have three forwardly directed branches, but no posteriorly directed branch (Fig. 8).

The maxilla is well developed, and the maxillipede normal for the genus with groups of fine hairs on the basal podomore. The terminal spine is finely spinulated. Two pairs of trunk appendages are present ; each is a single podomere with two terminal setae.

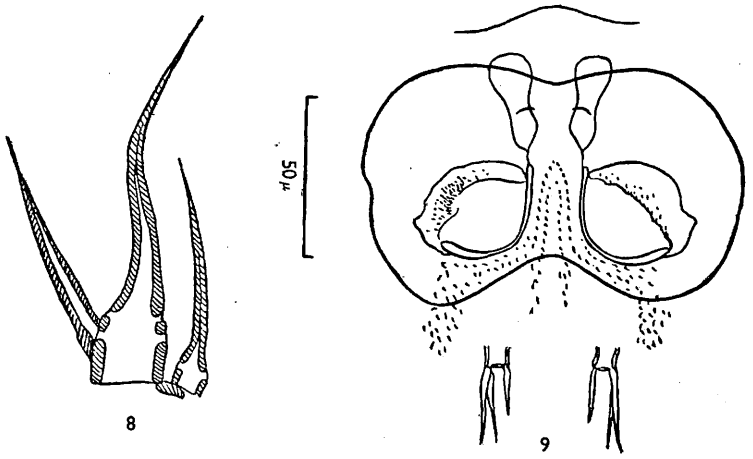
The genital area (Fig. 9) is wider than long, slightly concave anteriorly and more strongly concave posteriorly. The genital openings are curved. There is a scattered pattern of minute hairs on the posterior part of the area. Some of the hairs extend beyond the area as far back as the caudal rami. The paired receptacula seminis extend from the anterior edge of the genital openings to a short way in front of the genital area.

The caudal rami lie behind the genital area ; each is provided with three setae.

A spermatophore was found attached to one female. The body of the spermatophore was 38 μ long, and the stalk was 46 μ long.

The ovisacs are oval, measuring about 0·66 × 0·58 mm. One female had six ovisacs, each of which contained 70 to 80 eggs.

The above description differs in some slight details from that given by Hansen, but I believe that the specimens belong to his species, for the following reasons. The body is of the same size and shape, and the maxillules lack a posteriorly directed process. This was also the case in the undoubted female



Figs. 8–9.—*Sphaeronella danica*. 8, maxillule of female, greatly enlarged. 9, genital area of female.

danica from *Corophium crassicorne*. The genital area agrees with Hansen's description in all particulars except one. Hansen's figure shows the hind border projecting backwards in the middle. I believe this to be an error. Fig. 9 shows that the minute hairs on the posterior part of the genital area continue backwards in two series, and this might easily give the impression of the genital area projecting backwards between them.

The larva of *S. danica* was not described by Hansen. Fully developed larvae were found in the egg sacs of one of the females from *Erichthonius*. The general form of the body is shown in Figs. 10 and 11.

The antennules have three podomeres, the last with two strong and very long setae (Fig. 12).

The antennae have three distinct podomeres armed with minute setae, and two larger terminal setae.

The maxilla has a large basal podomere similar to that of the adult, but a terminal claw is clearly demarcated from the terminal podomere.

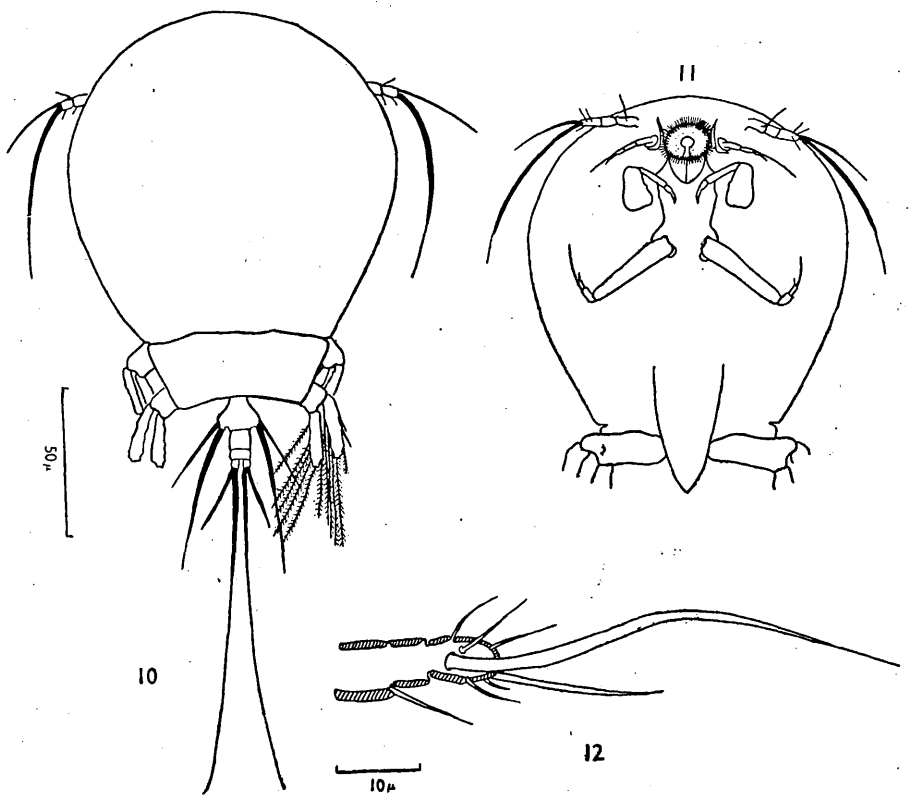
The maxillipede has a long basal podomere, followed by a smaller podomere and a terminal spine.

The two legs are well developed, the endopod and exopod of each consist of a single podomere armed with plumose setae.

The abdomen is armed with thick spiniform setae ; the stronger setae on the first abdominal segment reach beyond the tips of the weaker setae on the caudal rami. The caudal rami are about as long as wide, with two terminal setae, one of which is much longer than the other.

Distribution. Denmark (Hansen). Now recorded from Guernsey and Plymouth.

Hosts. Corophium crassicorne Bruzelius (Hansen and present paper), *Erichthonius brasiliensis* (Dana) (present paper).



Figs. 10–12.—Larva of *Sphaeronella danica.* 10, dorsal view, setae have been omitted from the legs on the left side. 11, ventral view of anterior end. 12, antennule.

Sphaeronella longipes Hansen, 1897.

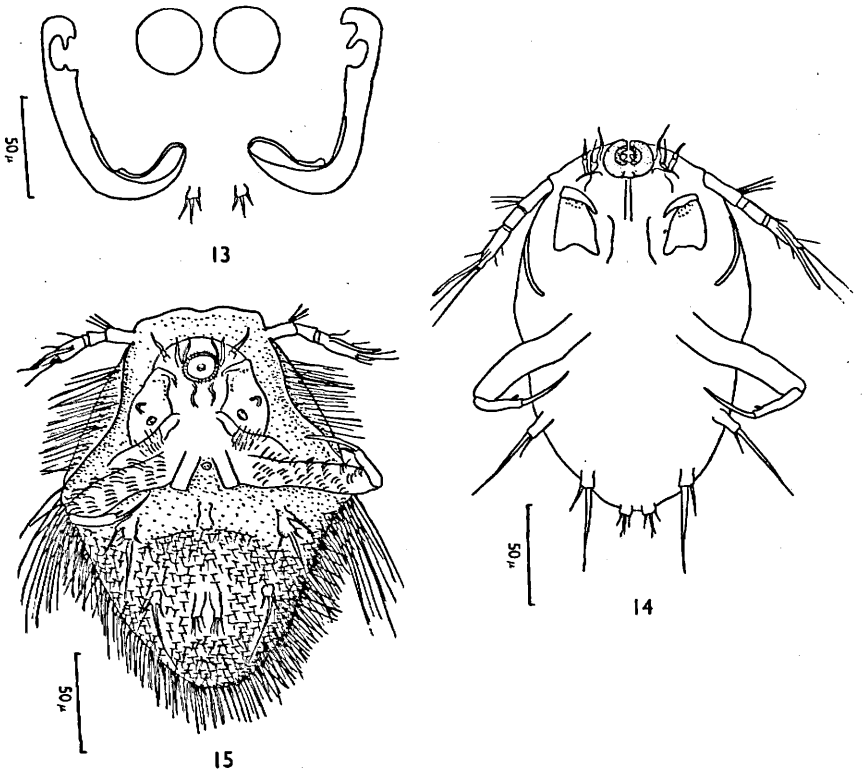
Material examined. (1) One adult female, one very young female, one male and a larva, from *Ampelisca tenuicornis* Lilljeborg, St. Brelades Bay, Jersey, 1 fthm, August 1955. Collected by Dr R. B. Pike.

The principal diagnostic character of this species is the very long terminal seta on the trunk legs of the female.

Hansen was unable to give any information about the genital area ; this is shown in Fig. 13.

A very small female is shown in Fig 14 ; this is smaller than any found by Hansen, and shows certain differences in the relative widths of the head and trunk when compared with Hansen's small females. These differences can be explained by the allometric growth of the trunk during increase in size of the female.

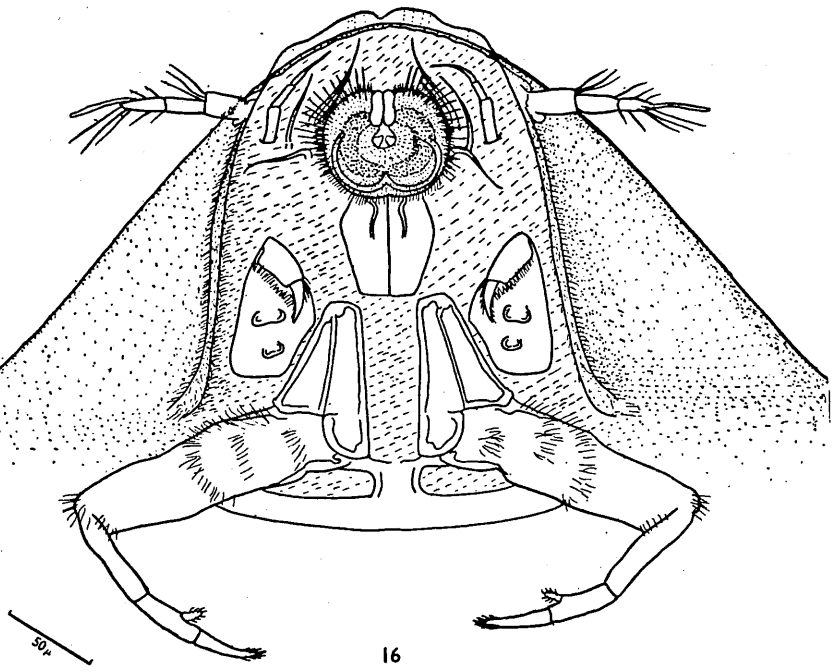
The male was unknown to Hansen. The specimen found during the present investigation is shown in Fig. 15. The length was 180 μ. The antennules have three podomeres, the basal one with three terminal setae, and the third with a distinct aesthetasc and six or seven setae. The antenna is distinct, with three small podomeres. The maxillule has only two forwardly directed processes. The maxilla has a series of blunt teeth and two larger protuberances on the basal podomere. The maxillipedes are of normal pattern, with stiff setae on the basal podomere. The processes from the sub-median skeleton which project over the bases of the maxillipedes are rectangular in shape, with truncated ends. This appears to be a good diagnostic character for the male of this species. The trunk legs are small ; the first has an outer branch with a stout terminal seta, and an inner branch with one large and three small terminal setae. The second trunk limb is unbranched and has three terminal setae. The caudal rami are elongated and have four terminal setae, one of which is complex.



Figs. 13–15.—*Sphaeronella longipes*. 13, genital area of female. 14, very young female, ventral view. 15, male, ventral view.

Distribution. Denmark (Hansen), now recorded from Jersey.

Host. Ampelisca tenuicornis Lilljeborg (Hansen and present paper).



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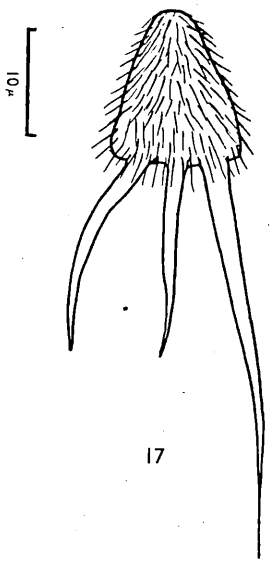

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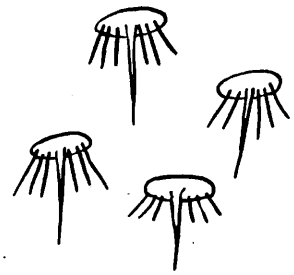

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Figs. 16–18.—*Sphaeronella frontalis.* 16, head of a large female, ventral view, showing appearance when the frontal cup is reflected dorsally. 17, first trunk limb. 18, detail of body hairs of male.

Sphaeronella frontalis Hansen, 1897

Material examined. (1) One adult female from *Ampelisca tenuicornis* Lilljeborg, Garroch Head, Clyde, 70 fthms., March 1949. (2) One adult female, with nine egg sacs, from *A. tenuicornis*, Garroch Head, 70 fthms., January 1950. (3) Three adult females and two males, from *Ampelisca macrocephala* Lilljeborg, Garroch Head 110 m., February 1950. All the specimens were collected, and the hosts determined by Dr R. B. Pike.

Some of the large females were difficult to determine at first, because the characteristic flat cup on the frontal margin was not visible from the ventral surface (Fig. 16). Once it was realised that this cup is sometimes reflected dorsally the determination was simplified. Fig. 16 is also shows the strongly developed ventral skeleton and the well marked transverse bar behind the maxillipedes. Hansen states that the trunk legs are 'normal for the genus'; this might be taken to mean that they have two terminal setae. All the specimens which I have examined had three terminal setae and a covering of fine hairs (Fig. 17).

The male has a characteristic covering of multiple hairs. Hansen says that each of these hairs has about ten processes springing from the base, but in my specimens there were only six or seven, and the median process was longer and stronger than the others (Fig. 18).

Distribution. Denmark (Hansen), now recorded from the Clyde.

Hosts. *Ampelisca macrocephala* Lilljeborg (Hansen and present paper), *A. tenuicornis* Lilljeborg (present paper).

SPHAERONELLA PIKEI, sp. n.

Material examined. One adult female from the brood pouch of *Pontocrates arenarius* (Bate), Guernsey 1906 (Norman Collection).

This species is named after Dr R. B. Pike, in recognition of his industry in collecting so many of the specimens described in this paper. The shape of the body is shown in Fig. 19. The length is 0·82 mm. The head is relatively very small. There is a fringe of fine hairs along the frontal border, and a few very small hairs on the lateral borders.

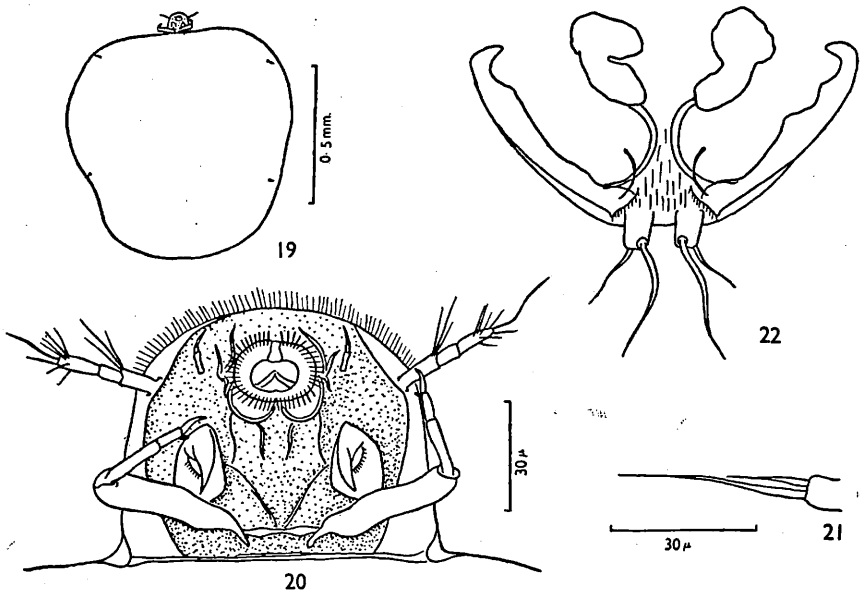
The antennule has three podomeres, with a total of twelve setae, four of which are on the basal podomere, and the remainder on the distal podomere. The antennae are small, but distinct, with three podomers. The mouth cone is normal for the genus, and the maxillules each have three processes, one of which is directed posteriorly. The maxilla has a sub-triangular basal podomere with a crescentic flap bearing a row of short hairs. The distal podomere and claw of the maxilla are fused. The maxillipedes have an elongated basal podomere, with two slender distal podomeres and a terminal spine. The ventral skeleton of the head is weakly developed, but there is a thickened ridge running transversely between the bases of the maxillipedes and a narrow bar separating the head from the trunk.

The trunk is devoid of hairs. The trunk legs are minute (Fig. 21); each consists of a single podomere with two terminal setae, one of which is twice as long as the other.

The genital area is triangular in shape (Fig. 22), with the caudal rami inserted within the posterior margin. The caudal rami bear two setae, one twice as long as the other.

Two egg sacs were found with the specimen ; they were oval in shape, measuring 0·48 × 0·34 mm.

The holotype is in the British Museum (Natural History) registered number B.M. 1958.1.2.4.



Figs. 19–22.—*Sphaeronella pikei* sp. n. 19, female, ventral view. 20, female, ventral view of head. 21, second trunk limb of female. 22, genital area of female.

This species is fairly closely allied to *S. giardi* Hansen and *S. bonnieri* Hansen ; in Hansen's key (1897) to the genus it would run down to *S. bonnieri*. However, it differs from *S. bonnieri* in the shape of the genital area, and in the presence of a distinct fringe of hairs along the frontal border. The trunk legs are also unusual, having one seta much longer than the other. This is a similarity to *S. longipes*, but the latter species has much larger trunk legs, and a very different genital area.

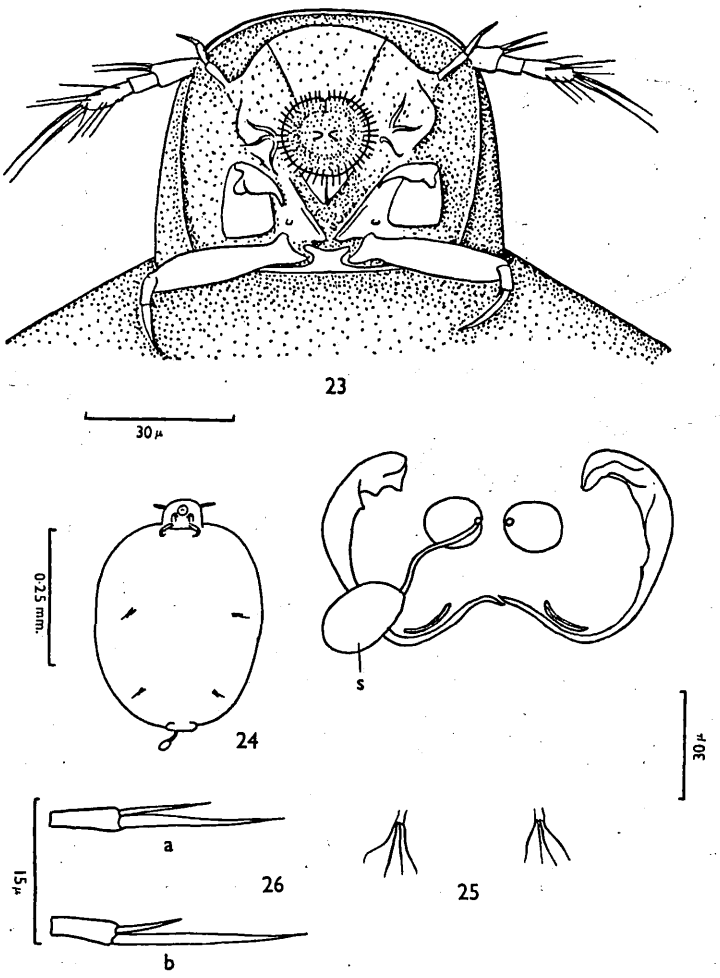
Sphaeronella valida Scott, 1905

Syn. *Sphaeronella minuta* var. *valida* Scott, 1905.

Material examined. One female from the brood pouch of *Megamphopus cornutus* Norman, taken off the Norfolk coast (53° 04 N, 01° 00 E) 12 fthms., September 1957. Collected by Mr R. Hamond.

The specimen from Norfolk is illustrated in Figs. 23–26. The total length of the specimen is 0·45 mm. The most important characters to be noticed are as follows. The head is naked, without any fringing hairs. The antennules are large, with three podomeres bearing a total of eleven setae, two of which are on the proximal podomere and nine on the distal podomere. The antennae

are distinct, with three podomeres. The maxillules have three processes, one of which is directed posteriorly. The maxillae have a large basal podomere which appears to be smooth and lacks the projections which are often found in other species of the genus. The maxillipedes are of the usual pattern, but are smooth, lacking setae. The ventral head skeleton is well developed,



Figs. 23–26.—*Sphaeronella valida*. 23, head of female, ventral view. 24, female, ventral view. 25, genital area and caudal rami of female ; s–spermatophore attached to entrance of spermatheca. 26, trunk limbs of female ; a–first, b–second.

and a bar, which bears a thickening in the middle, separates the head from the trunk. The trunk lacks hairs, and its limbs are placed more posteriorly than in most species of the genus. The genital area is surrounded by an incomplete ring of chitin, with a concave posterior border (Fig. 25). The caudal rami are placed well behind the genital area ; each ramus is slightly longer than wide and bears four setae.

Scott's drawings and description of *Sphaeronella minuta* var *valida* are inadequate to distinguish it from many other species in the genus. He gives no information about the antennae, maxillules, trunk limbs or the genital area, all of which must be examined before a species can be properly diagnosed.

The description given above of the Norfolk specimen should be regarded as a redescription, and the specimen regarded as a neotype—at least until such time as Scott's original specimen is located and redescribed. The status of *valida* has been raised to specific rank because Scott's drawings raise considerable doubts about *valida* being a mere variety of *minuta*.

The Norfolk specimen has been assigned to *valida* because the body shape is similar, and the caudal rami are located some distance behind the genital area (this is shown in side view in Scott's fig. 18). Further Scott's drawing of the maxillipede does not show any setae, which agrees with the description given above. Finally, the specimen was taken on the same host (Scott gives the host as *Melamphopus cornutus* Norman).

Distribution. Scotland, Firth of Forth (Scott); now recorded from Norfolk.

Host. *Megamphopus cornutus* Norman (Scott and present paper).

ACKNOWLEDGMENTS

I am most grateful to Dr J. P. Harding for his suggestion that I should examine this material, and for his helpful criticism of the manuscript.

SUMMARY

Rhizorhina ampeliscae Hansen is recorded from Guernsey and the Clyde area.

Five species of the genus *Sphaeronella* are recorded from British waters for the first time. One of these, *S. pikei*, is a new species.

Sphaeronella elegantula Hansen is shown to be a synonym of *S. leuckarti* Salensky.

The larva of *S. danica* and the male of *S. longipes* are described for the first time.

The following new hosts are recorded. *Aora typica* parasitized by *S. leuckarti*. *Erichthonius brasiliensis* parasitized by *S. danica*. *Ampelisca tenuicornis* parasitized by *S. frontalis*. *Pontocrates arenarius* parasitized by *S. pikei* sp. n.

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Carotenoid Pigment in *Spirobacillus cienkowskii* Metchnikoff, a Pathogen of Cladocera

CLADOCERA are often coloured red by hæmoglobin in their blood[1]; another red pigment has been found in specimens infected by *Spirobacillus cienkowskii* which Metchnikoff[2] described from *Daphnia magna* Straus. The bacterium passes through a series of stages within the cladoceran and causes it to assume a bright red colour.

As a general rule less than 1 per cent of a cladoceran population shows signs of infection, but I have recently found a population of *Simocephalus vetulus* O. F. Müller in which up to 2 per cent of the individuals were infected. Several specimens of *Sida crystallina* (O. F. Müller) from the same body of water were also found to be infected with the same bacterium.

About three hundred infected specimens of *Simocephalus* were ground up in water and then centrifuged at low speeds to leave a cloudy red suspension which consisted almost entirely of bacteria. When this suspension was centrifuged at 9,000 r.p.m. the bacteria were all spun down and a colourless fluid was left. This demonstrates that the pigment is confined to the bodies of the bacteria and is not a host reaction. The bacterial sludge was treated with acetone, when the pigment was easily extracted, then

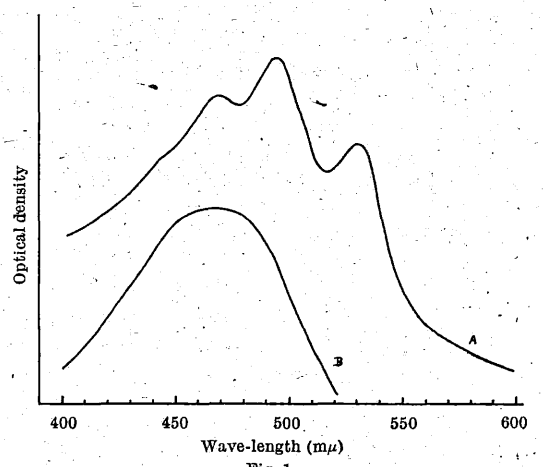

Fig. 1

taken into petrol ether (boiling point, 60–80° C.) after dilution of the acetone with water. The solution in petrol ether was dried over anhydrous sodium sulphate and examined in a spectrophotometer. Absorption peaks were found at 470, 495 and 530 mμ (Fig. 1,*A*). The last two peaks are in fair agreement with those of rhodoviolascin and those of α-bacteriopurpurin, which is probably a demethylated form of rhodoviolascin[3]. The first peak is about 10 mμ too far towards the red end of the spectrum. The explanation for this was found when uninfected specimens of *Simocephalus* were treated in the same way as the infected specimens. An olive-green supernatant fluid was left after centrifuging; when this was treated with acetone it turned orange. The orange pigment was taken into petrol ether after dilution of the acetone water phase with more water. The absorption curve of the petrol ether solution is shown in Fig. 1,*B*. The single peak at 470 mμ agrees with that of astacene derived from astaxanthin which was present in the form of a green carotenoprotein. The presence of a small amount of astacene in the extract from the infected specimens would account for the apparent bathochromic displacement of the first peak of the bacterial pigment.

It is evident from these observations that the red pigment is a carotenoid of the α-bacteriopurpurin type, and that it is confined to the bacteria. There is an indication that the pigment is at least partially derived from carotenoid present in the host, because the patches of carotenoid protein which normally occur in healthy *Simocephalus* are changed to red when the pigment-bearing cells are infected with the bacterium.

J. Green

Zoology Department,
Bedford College, Regent's Park,
London, N.W.1. Oct. 27

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Haemoglobin and the Habitat of the Harpacticoid Copepod *Elaphoidella gracilis* (Sars)

HÆMOGLOBIN has only recently been found in free-living copepods. Munro Fox[1] found that certain mud-dwelling harpacticoids contained this respiratory pigment, while allied species living in moss or open water lacked it. The correlation between hæmoglobin and habitat can now be extended by the discovery of hæmoglobin in *Elaphoidella gracilis*, a species which inhabits burrows in decaying aquatic vegetation. The red pigment in this species is easily visible under the microscope, and its identity was established spectroscopically.

Elaphoidella gracilis is not very often recorded because of its burrowing habits. Gurney[2], writing of its seasonal occurrence, says "the capture of it is so capricious that nothing certain is known". Donner[3] regards it as a summer form, but I have found this species to be present and active in the Long Water at Hampton Court throughout the whole of the last winter.

J. GREEN

Zoology Department, Bedford College,
Regent's Park, London, N.W.1.

April 28.

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SPHAERONELLA SEROLIS MONOD, AND A NEW SPECIES OF *RHIZORHINA*, COPEPODS PARASITIC ON THE ISOPOD *SEROLIS BROMLEYANA* SUHM (CRUSTACEA)

BY

J. GREEN

Bedford College, University of London

[Accepted 14th October 1958]

(With 16 figures in the text)

CONTENTS

INTRODUCTION

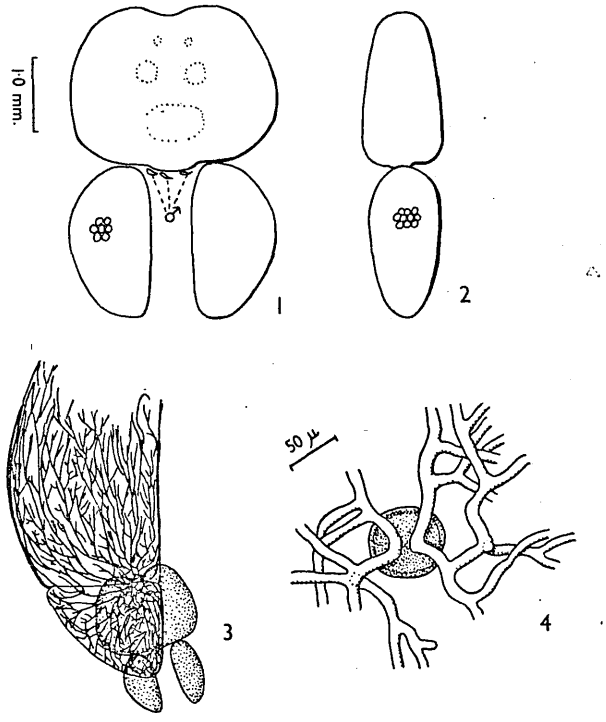
The genus *Rhizorhina* has hitherto been known from a single species, *R. ampeliscae* Hansen (1892), which was originally described from Denmark and Norway, and had not been recorded elsewhere until recorded by Green (1958) from Guernsey and the Clyde. The genus is remarkable because it is parasitic on Crustacea, while all the other members of the family to which it appears to belong, the Herpyllobiidae, are parasites on polynoid polychaetes. *Rhizorhina ampeliscae* is a parasite of members of the amphipod genus *Ampelisca* ; the new species described here parasitises an isopod, and was collected by the New Zealand Expedition to Chatham Island.

Family HERPYLLOBIIDAE

RHIZORHINA SEROLIS, sp. n.

Description of the female. The body is bean shaped (Figs. 1 and 2), about 2·0 mm. long, 2·5 mm. wide, and 1·1 mm. thick. The dotted areas in Fig. 1 indicate slight depressions of the surface. There are no appendages. A small sclerotised ring is present around the mouth, from which issue two pairs of root-like processes. These roots, which are 10–14μ in diameter, pass into one of the pleopods of the host and ramify greatly (Fig. 3). The egg sacs are variable in size, but when fully developed they are pear shaped, about 1·95 mm. long and 1·20 mm. at the greatest width. Each egg has a diameter of about 0·13 mm. The males attach to the females, at the posterior end between the egg sacs.

Description of the male. The male is very similar to the male of *R. ampeliscae*. The body (Fig. 5) is of a typical copepod shape, but there are



Figs. 1–4.—*Rhizorhina serolis*. 1, female, dorsal view. 2, female, lateral view. 3, pleopod of *Serolis bromleyana* with female attached, showing the extent of the rooting system from the mouth. 4, detail of the rooting system near the mouth.

only two pairs of swimming legs. The size is only a small fraction of that of the female ; the total length, excluding setae, is about 0·2 mm.

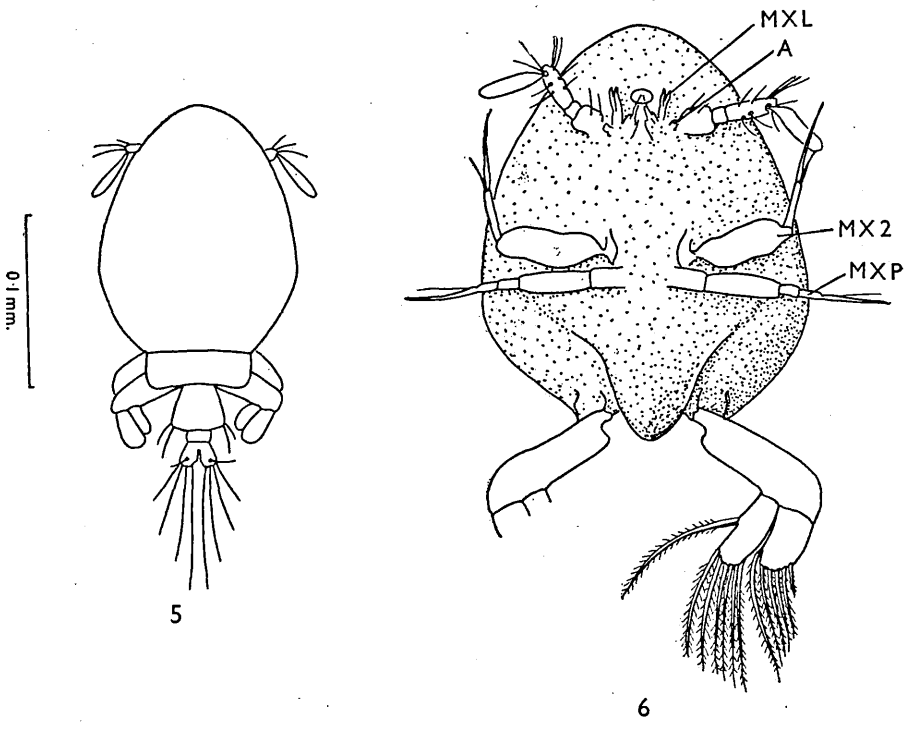
Each antennule has three podomeres, the first with two setae, the second without setae, and the third with nine setae and a large flattened aesthetasc which is practically as large as the podomere. This aesthetasc is inserted sub-terminally and its long axis is usually inclined at an angle to the long axis of the third antennulary podomere.

The antennae are reduced to small projections near the bases of the antennules.

The mandibles are thin and stylet shaped, projecting slightly through a small sclerotised ring which indicates the position of the mouth.

The maxillules are each represented by two small forwardly projecting processes which lie between the bases of the antennules. Hansen interprets these structures in *R. ampeliscae* as maxillae, but by comparison with the larvae of the Choniostomatidae it is clear that the same sequence of appendages can be made out and that the maxillae are the larger appendages placed more posteriorly.

The maxillae have a large basal podomere and a much narrower terminal podomere which bears a long stout spine and a slender seta.



Figs. 5–6.—*Rhizorhina serolis*. 5, male, dorsal view, the setae on the swimming legs have been omitted. 6, thorax and head of male, ventral view. A—antenna ; MXL—maxillule ; MX2—maxilla ; MXP—maxilliped.

The maxillipeds each have three podomeres, the last bearing a strong spine and a slender seta.

The two pairs of swimming legs are similar in structure. The endopods bear seven plumose setae, one of which is inserted on the inner border, and the exopods bear six setae which decrease in length towards the outer margin.

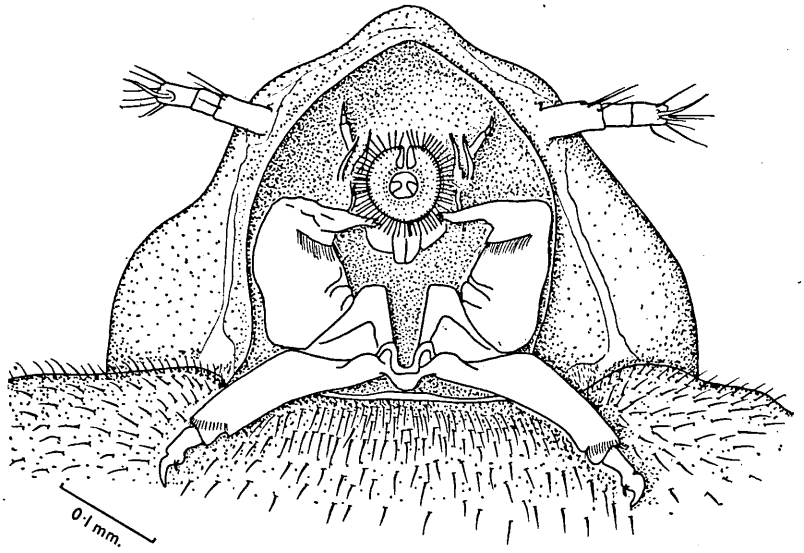
The caudal rami bear four terminal setae, of which the median ones are the longest. There is also a small seta on the dorsal surface of each ramus.

Occurrence. This species was numerous on the pleopods of *Serolis bromleyana* Suhm. As a rule only one female was attached to each host. The hosts were collected by the New Zealand Chatham Island Expedition from a depth of 403 metres on Chatham Rise (Station 6, 43° 40′ S., 179° 28′ E.).

Family CHONIOSTOMATIDAE

SPHAERONELLA SEROLIS Monod

This species was described by Monod (1930), but owing to the limitations of his material certain details of anatomy were not described. The material described in the present paper was obtained from a single female of *Serolis bromleyana*, which was also infected with *Rhizorhina serolis*. The brood pouch of the isopod contained four large females of *Sphaeronella*, one small female,



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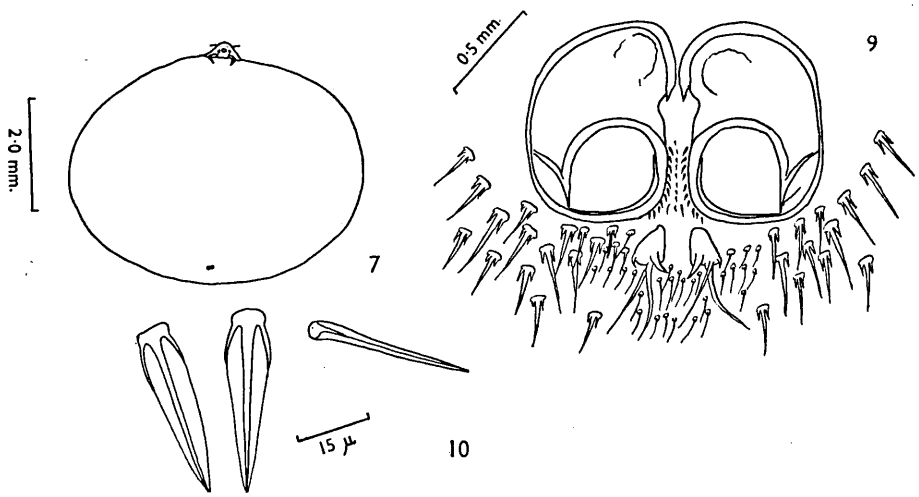

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Figs. 7–10.—*Sphaeronella serolis*. 7, adult female, ventral view. 8, head of adult female, ventral view. 9, genital area of adult female. 10, setae from near the genital area of an adult female.

two adult males, one male pupa with a cast larval skin by its side, and seventy-two egg sacs ! Assuming that no females had been lost from the brood pouch (there is no reason for believing that any were missing, since the oostegites were quite firmly in position) this implies an average production of eighteen egg sacs by each of the large females.

The females are much larger than any other choniostomatid, but have a relatively minute head. My examination of the appendages reveals some differences from those described by Monod, but I do not regard them as sufficient grounds for establishing a new species.

The first podomere of the antennule bears three setae, the second lacks setae, while the third bears seven setae and an aesthetasc.

Monod shows the maxillules as having but a single process, but my specimens were found to have three processes forming each maxillule, one of the processes was directed backwards, the other two forwards.

No appendages were found on the trunk.

The genital areas (Fig. 9) on my specimens were much better developed than on Monod's. The caudal rami lie immediately behind the genital area and bear two long setae as well as a stout inner spine and a small outer spine. The setae at the sides and behind the genital area are peculiar in structure. When viewed with a 1/6th inch objective they appear to have a long central prong and two small outer prongs, but critical observation using an oil immersion objective shows that the outer prongs are united with the central prong by a very thin membranous structure (Fig. 10).

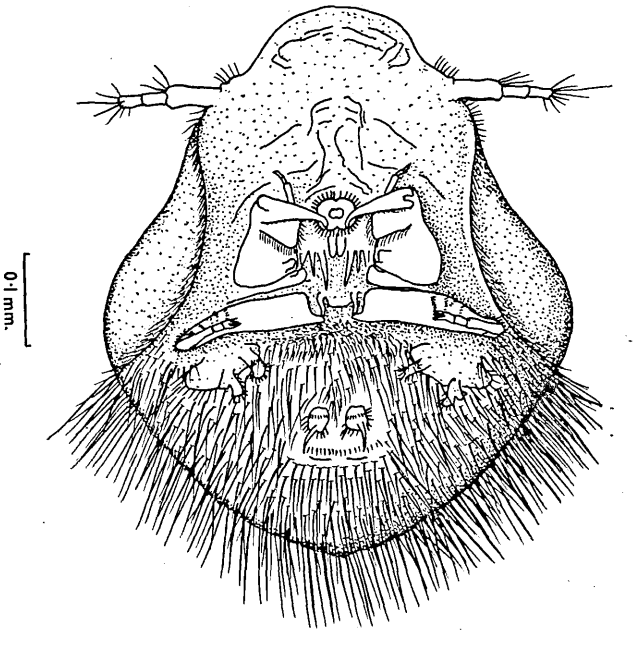
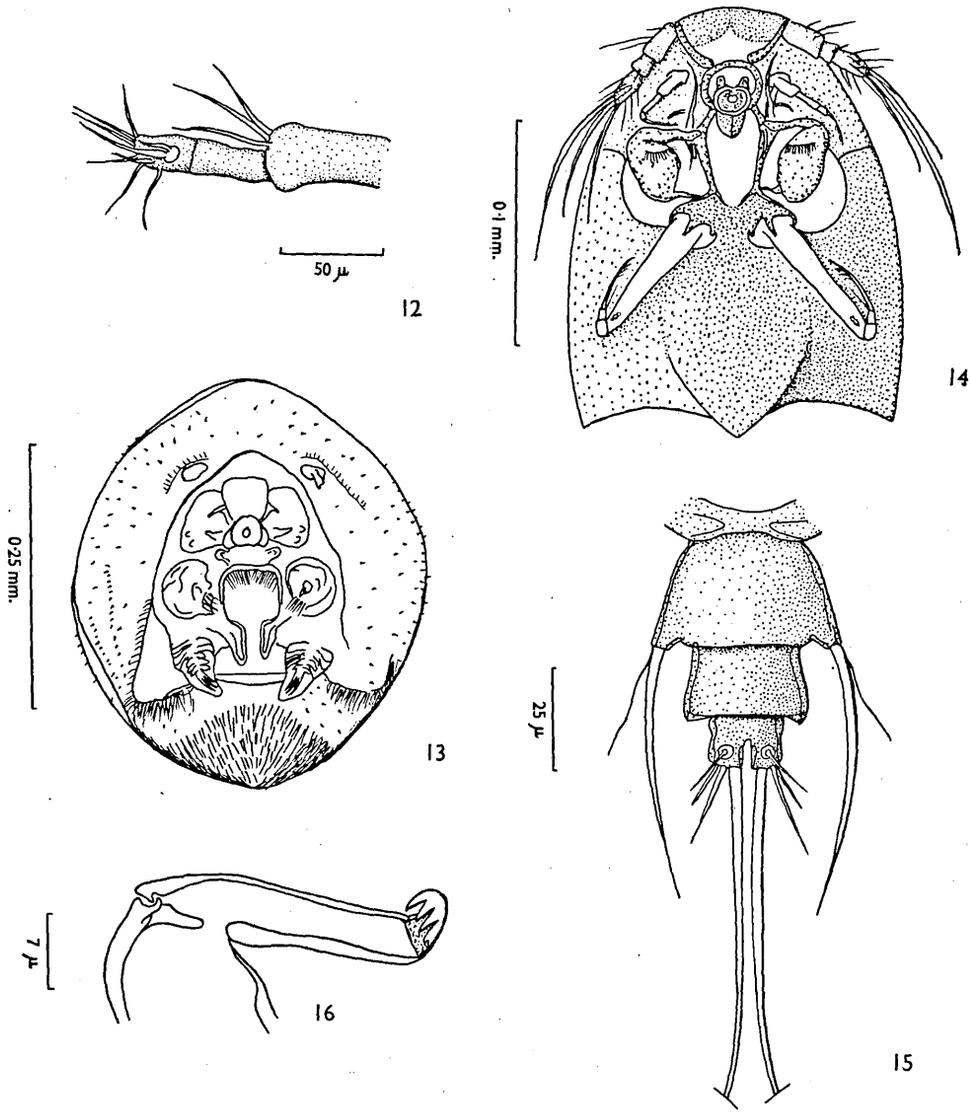


Fig. 11.—*Sphaeronella serolis*, adult male, ventral view.



Figs. 12–16.—*Sphaeronella serolis*. 12, left antenna of adult male. 13, pupal male, ventral view. 14, larva, ventral view of thorax and head. 15, larva, dorsal view of abdomen. 16, larva, terminal podomere of maxilla, ventral view.

The male (Fig. 11) is much smaller than the female, only reaching a length of 0·65 mm. The frontal border projects in front of the antennules, forming a flat, roughly semicircular, structure in front of the globular body.

The structure of the trunk limbs is very difficult to see because the hind part of the trunk is so densely covered with setae.

The maxillules of the male could not be found.

The male pupa is shown in Fig. 13 ; the total length was 0·40 mm. The pupal skin bore a few setae which were irregularly distributed.

A cast off larval skin was found attached to the pupa. This was 288μ long, excluding the long setae on the caudal rami, which added a further 169μ to the length.

The larval appendages are of the usual choniostomatid pattern. The terminal podomere of the antennule bears ten setae, two of which are long and terminal, and a very long aesthetasc. The antennae are well developed, of three podomeres, the last bearing a long plumose setae. The maxilla shows an unusual modification of the tip, which is bent downwards and produced to form three strong teeth (Fig. 16).

The first abdominal segment bears a large spiniform seta and a much finer shorter seta on each side. The caudal rami each bear a very long median seta, two shorter outer setae and a small dorsal seta (Fig. 15).

Occurrence. This species was described by Monod (1930) from the brood pouch of *Serolis pagenstecheri* Pfeffer, from Westcumberland Bay, South Georgia. It is now recorded from the brood pouch of *Serolis bromleyana* Suhm from a depth of 403 metres on Chatham Rise.

DISCUSSION

One striking feature of these records is the erratic occurrence of these species in Nature. As far as I am aware, *Sphaeronella serolis* has not been recorded since Monod's original description ; and now it is found on another species of *Serolis* from a locality about five thousand miles from the original one. It might be thought that this is a widespread species that has been overlooked, but I have searched through the large collection of *Serolis* species in the British Museum (Natural History), and have not found any specimens of *Sphaeronella* or of *Rhizorhina*.

A point of general interest is the great resemblance between the male of *Rhizorhina* and the larva of *Sphaeronella* ; indeed the male of *Rhizorhina*, if found alone, could easily be mistaken for a larval *Sphaeronella*, except when it has spermatophores formed within the body. There are differences in the appendages, as can be seen when Figs. 6 and 14 are compared. But the overall resemblance is so great that one reaches the conclusion that the male of *Rhizorhina* is neotenic, and that the Herpyllobiidae have advanced further in their modification than the Choniostomatidae. This is borne out by the females ; in the Herpyllobiidae they are generally without any appendages at all, while the choniostomatids have well developed head appendages and often have diminutive trunk appendages as well.

ACKNOWLEDGMENTS

I am grateful to Dr R. B. Pike and Dr J. P. Harding for the opportunity to examine these specimens, also to Dr Harding for reading and criticising the manuscript.

SUMMARY

The male and female of *Rhizorhina serolis* sp. n. are described and figured. This copepod is parasitic on the isopod *Serolis bromleyana*, from Chatham Rise, between Chatham Island and New Zealand.

Sphaeronella serolis Monod is recorded from the same host and locality. The hitherto unknown larva and male pupa of *S. serolis* are figured.

The male of *Rhizorhina* shows a great similarity to the larva of *Sphaeronella*, and is considered to be neotenic.

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PIGMENTATION OF AN OSTRACOD, *HETEROCYPRIS INCONGRUENS*

BY J. GREEN

Bedford College, University of London

(*Received* 30 *April* 1959)

INTRODUCTION

The information available concerning the pigments of ostracods is meagre. Fox (1948) records the presence of haemoglobin in the blood of *Cypria ophthalmica* (Jurine) and later (1957) in the blood of *Pseudocypris*. Fox (1955) also records the presence, in *Cypris pubera* (O. F. Müller), of a green pigment showing a reversible colour change to brown in alkaline conditions, and a similar change when reduced with sodium dithionite. The chemical nature of this pigment is unknown.

There are numerous references to the colours of ostracods in the systematic literature, and some particularly fine coloured plates in the works of G. O. Sars, but there is no indication of the chemical nature of the pigments involved. In one of Sar's papers (1894) there is a coloured figure of a *Heterocypris* species (plate IV, under the name *Cypris sydneia* King) which gives a very good impression of the living animal. In a later paper (1924) Sars remarks that the genus *Heterocypris* is notable for its yellow colour; this being a feature by which it may be distinguished from most other ostracods.

Because of this remarkable coloration, and because the species sometimes occurs in numbers large enough to permit the application of chemical methods, it was decided to investigate the pigments of *H. incongruens* (Ramdohr).

MATERIAL AND METHODS

The stock of ostracods came from a large aquarium tank at Bedford College. This tank contained mud from an unknown locality, and the only other animals in the tank were a few small copepods and rhabdocoels. In the spring of 1958 *H. incongruens* became very numerous, and samples were taken in order to extract pigments. The ostracods remaining in the tank were fed with various algae which grew in other aquarium tanks containing axolotls and catfish.

When controlled feeding was desired the unicellular green alga *Chlorella vulgaris* Beij was used. This alga was grown in pure culture on agar slopes in front of a mercury vapour lamp. In certain other experiments the filamentous blue-green alga *Anabaena cylindrica* Lemm. was used; this was also grown in pure culture on agar slopes.

A number of the ostracods were reared individually in standard conditions similar to those used in a previous study of the growth of *Daphnia* (Green, 1956).

The only difference was that the standard suspension of *Chlorella* was allowed to settle on the bottom of the culture tube and was not stirred as it was in the study of *Daphnia*.

When a sample of ostracods was taken the animals were passed through several changes of clean water over a period of about half an hour. This enabled a substantial amount of material to be eliminated from the guts of the ostracods so that the pigment analysis was not complicated by pigments from the plant material in the gut. Various methods of extracting pigments were tried; details are given in the relevant section. Absorption spectra were examined in a Unicam S.P. 500 spectrophotometer with an ultra-violet attachment.

LOCATION OF PIGMENTS

The general colour of *Heterocypris incongruens* is orange yellow, with a tendency towards brownness on the upper parts of the shell valves. The gut often appears as a brownish mass inside the body, while the gut diverticula, one in each shell valve, appear as dark streaks against the general yellow background. There is much orange yellow pigment in a granular form in the epidermal cells of the shell valves, and finer granules of a similar pigment in the locomotory limbs and furca. The blood is also orange yellow; here the pigment is in solution and not granular. The eggs, which can sometimes be seen in the ovary, vary in colour from pale orange to bright scarlet. When the eggs are scarlet the pigment can be seen to be located in two different types of droplets. The larger droplets are orange in colour and are about 2 or 4 μ in diameter, while the smaller droplets are only about 0·5 μ in diameter and are bright scarlet in colour. It has not been possible to investigate the pigments of the eggs separately, but it is probable that they are carotenoids, because they give a blue colour with sulphuric acid and are soluble in acetone and petrol ether.

When *H. incongruens* is reared at a temperature of 22° C. and fed on *Chlorella* the ovary begins to appear orange or red after the animal has passed through seven moults and is about 16 days old. The first eggs are cast off attached to the shell valves at the next moult. After this the animal does not moult again and subsequent batches of eggs are carried within the shell valves for a variable period or else laid on some solid object near the surface of the water. The first eggs remain attached to the shell valves for about a week until the young ostracods emerge. The main difference from the adult pigmentation is that the young are paler, and the gut diverticula do not appear in the shell valves until the fifth instar (Schreiber, 1922).

LACK OF HAEMOGLOBIN

Examination with a microspectroscope has failed to detect haemoglobin in *Heterocypris incongruens*. This was so even when the ostracod was well fed and kept in water containing less than 1·0 ml./l. of oxygen for 2 weeks.

Attempts were also made to produce a pyridine haemochromogen, by the addition of sodium dithionite and pyridine. The microspectroscopic technique used was sensitive enough to demonstrate the presence of haem in a single egg of a pale

cladoceran such as *Diaphanosoma*, but it failed with over twenty specimens of *Heterocypris*. It is clear that haem pigments are absent, or very scarce, in this ostracod. This is in contrast with the species examined by Fox (1948, 1957) where haemoglobin is easily detectable in a single specimen.

The ability of *H. incongruens* to survive, and indeed flourish, at low oxygen concentrations has been demonstrated by Fox & Taylor (1955). One of my own experiments, originally designed for another purpose, has accidentally demonstrated that this ostracod can endure anaerobic conditions for a considerable period. The oxygen concentration in one of the experimental flasks fell so low that it became undetectable by the micro-Winkler technique of Fox & Wingfield (1938), and the water smelt strongly of hydrogen sulphide. In spite of this the ostracods flourished, and after 2 weeks one specimen was found with 32 eggs within its shell valves. Numerous eggs had been deposited on the wall of the flask just at the surface of the water. It is clear that *H. incongruens* can live and reproduce at extremely low oxygen concentrations without the aid of any detectable concentration of haem pigments. This point will be taken up again in the discussion.

CAROTENOIDS

Acetone extracts from several thousand ostracods yielded an orange solution, from which the pigment was taken into petrol ether after dilution of the acetone with water. After drying the petrol ether solution over anhydrous sodium sulphate, a phase test with 90% methanol showed the orange pigment to be entirely epiphasic. When chromatographed on an alumina column the pigment resolved into two bands, both of which passed through the column with petrol ether, but the second took much longer than the first. The absorption spectrum of the first fraction in hexane showed a shoulder at 426 mμ and distinct peaks at 450 and 477 mμ. This is in good agreement with the absorption spectrum of β-carotene given by Karrer & Jucker (1950). The second fraction showed a single absorption peak at 468 mμ in hexane. This suggests an astaxanthin ester. When the fraction was saponified with potassium hydroxide in methanol the pigment became hypophasic when tested against petrol ether, and remained hypophasic in the presence of excess water. When the hypophase was acidified with glacial acetic acid the pigment passed rapidly to the epiphase. This behaviour is characteristic of astacene liberated from an astaxanthin ester.

The diet of the ostracod included green and blue-green algae. The food would thus contain a variety of xanthophylls; for instance: *Chlorella vulgaris* has about 75% of its carotenoids in the form of lutein and violoxanthin, and a further 10% in the form of neoxanthin (Goodwin, 1954), *Anabaena cylindrica* has 17% of its carotenoids in the form of myxoxanthophyll (Goodwin, 1957). There was no indication of the presence of any of these hypophasic pigments in the extracts from the ostracods.

PTERIDINE

After repeated acetone extraction the residue was still yellow in colour; when it was treated with an ammonia solution a pale yellow pigment was extracted but faded rapidly and became colourless. The pale yellow solution and the colourless solution resulting from it had a brilliant blue fluorescence when exposed to ultraviolet light (365 mμ). This fluorescence was quenched by the addition of sodium dithionite, but returned when the solution was shaken with air. The fluorescence was destroyed by the addition of strong acid or alkali.

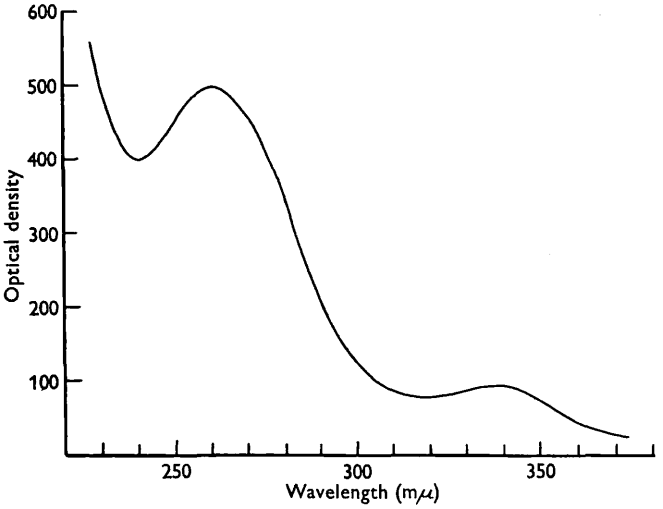

Fig. 1. Absorption spectrum of blue fluorescent ammonia extract from *Heterocypris incongruens*.

An equal volume of absolute ethanol (spectroscopically pure) was added to the ammonia extract and the mixture was filtered. The ultra-violet absorption spectrum of the filtrate was examined, using a mixture of ammonia and absolute ethanol as the blank. The result is shown in Fig. 1. There is a distinct absorption maximum at 260 mμ and a low rounded hump with maximum absorption at 338 mμ. This absorption curve is consistent with the presence of a pteridine (cf. Mason, 1954).

The colourless solution was chromatographed on paper using a 4:1:5 butanol–acetic acid–water mixture as the developing solvent. This usually resulted in a single fluorescent spot with an R_F value of 0·4, but on some occasions traces of a second spot were seen; this may have been an artifact due to trailing of the fluorescent substance in the developing mixture.

It was thought that the colourless fluorescent substance might be 2-amino-4-hydroxypteridine-6-carboxylic acid (pterincarbonic acid), which was obtained by Forrest & Mitchell (1954) from a photolabile yellow pigment in *Drosophila melanogaster*, and which Ziegler-Günder (1956) found as a derivative from a yellow pteridine in the skin of *Rana temporaria*. This possibility was tested by a series of

chromatograms, using ammonia extracts of frog skin which had been exposed to light before starting the chromatogram. In this way it was found that the ostracod pteridine migrated more quickly than the spot which Ziegler-Günder identified as pterincarbonic acid. Further, the fluorescence of the substance from the ostracod is somewhat more violet in colour. The precise identification of the fluorescent pigment of *Heterocypris* must await the availability of much larger amounts of material, but meanwhile the evidence presented above indicates that it is a pteridine.

The distribution of the fluorescent pigment in the body of the ostracod was examined by dissecting the animals in an ammonia solution under an ultra-violet lamp. It was found that the pigment is distributed throughout the body, even in the limbs and blood, but is absent from the eggs.

BILADIENE IN THE GUT WALL

The ostracods from the aquarium tank were found on dissection to have dark blue granules in the cells of the gut epithelium, both in the central portion of the gut and in the diverticula extending into the shell valves. The granules were more or less rounded, with a diameter varying from 0·5 to 3 μ. When treated with yellow concentrated nitric acid a brilliant Gmelin reaction resulted. The granules became a brighter blue, then successively purple, red, orange and yellow. This is a clear indication that a bile pigment is accumulated in the gut wall.

A large sample of the ostracod from the aquarium tank was ground up with a 19:1 methanol–sulphuric acid mixture, giving a bluish solution. The blue pigment was taken into chloroform after dilution of the methanol with water. The solution in chloroform had a red fluorescence in ultra-violet light, but unlike the red fluorescence of porphyrins this was quenched by the addition of acid. The addition of zinc acetate and a little iodine in methanol did not alter the fluorescence, but when more iodine was added the fluorescence became green. This behaviour of the pigment is consistent with that of a biladiene (Lemberg & Legge, 1950; Comfort, 1950). Unfortunately there was not enough of the pigment to measure its absorption spectrum.

A series of experiments was made to see what factors might influence the accumulation of the biladiene in the gut wall. Groups of ostracods were separated into dishes containing *Chlorella* as food. Some dishes were kept in the dark, some in front of a mercury vapour lamp, and some of the ostracods were kept in conical flasks in the dark to reduce the oxygen content of the water. The same clear result was obtained in each group. After 6 days the amount of biladiene in the gut wall had decreased to about half the original amount, and after 9 days the gut wall was colourless. The biladiene is evidently not derived from green algae, and continued feeding on such algae leads to the eventual disappearance of the bile pigment.

The next step was to investigate the mud of the aquarium tank to see if any blue-green algae were present; the search resulted in the finding of small amounts of a dark blue species of *Oscillatoria*. The ostracods were evidently accumulating the bile pigment from the phycobilin in this alga. Final proof of this was given when

specimens of the ostracod were reared from birth on a diet of *Chlorella*, and then transferred to a diet of the blue-green *Anabaena cylindrica*. The gut wall of the animals feeding on *Anabaena* became a bright blue green, while the gut wall of control specimens feeding on *Chlorella* remained colourless.

A significant point which emerged from this experiment was that the colour of the gut wall varied with the species of blue-green alga on which the ostracods were feeding. When the dark blue *Oscillatoria* was eaten the gut wall was dark blue, and when the lighter blue-green *Anabaena* was eaten the gut wall was correspondingly paler. It seems that the phycobilins of the blue-green algae are accumulated in the gut wall of the ostracod without much change in their constitution.

Experiments were also made to see if other species of ostracods could accumulate phycobilins. The ostracods were kept in dishes with a mixed diet of *Chlorella* and *Anabaena*. Samples of each species were examined at intervals to see if the gut wall was coloured; if a blue green colour was present its identity was confirmed with the Gmelin reaction. In this way it was found that the following species can accumulate phycobilins: *Cypridopsis vidua* (O. F. Müller), *Herpetocypris reptans* (Baird), *Cyclocypris ovum* (Jurine), and an unidentified species of *Candona*.

DISCUSSION

The absence of haemoglobin from this ostracod, coupled with its ability to survive in anaerobic conditions indicates that the intermediate pathways of oxygen transfer differ from the usual haemoglobin-cytochrome system. The form that the alternative pathway may take is unknown, but it may possibly involve the fluorescent pigment which shows a reversible reduction on the addition of sodium dithionite. This property of reversible oxidation and reduction might well play some part in the intermediate metabolism of the ostracod under anaerobic conditions.

The occurrence of pteridines in decapod crustaceans has been surveyed by Busnel & Drilhon (1948). They found fluorescent pigments associated particularly with the darker pigments, both melanins and ommatines. It is possible that the brownness which sometimes appears on the upper parts of the shell valves of *Heterocypris* is due to an ommatine. The method used to extract the blue fluorescent substance would also extract ommatines, and the ultra-violet absorption of such pigments would reinforce the peak found at 260 mμ.

There does not appear to be any information available in the literature concerning the presence of pteridines in the Entomostraca. My own preliminary investigations have failed to find any easily detectable amounts of such substances in the cladoceran, *Daphnia magna*, or in an anostracan, *Artemia salina*. This was in spite of using numbers of these species greatly in excess of the numbers of *Heterocypris* which gave the brilliant blue fluorescence.

There does not seem to be any previous literature on the carotenoids in ostracods. The occurrence of β-carotene and astaxanthin esters is rather what might be expected from the little knowledge we have of carotenoids in other Entomostraca. The cladoceran *Daphnia magna* contains both β-carotene and astaxanthin (Green,

1957), and both these pigments have been found in certain copepods (Goodwin & Srisukh, 1949; Batham, Fisher, Henry, Kon & Thompson, 1951).

Bile pigments are of infrequent occurrence in the Crustacea: biliverdin is the only one which has been identified. It is found in the liver of the North American crayfish *Cambarus* (Bradley, 1908), and in the roots of certain parasitic cirripedes (Raphael, 1948; Fox, 1953). It might be thought that various other small crustaceans would have the ability to accumulate phycobilins from blue-green algae in their food, but I have not noticed any such accumulation in any freshwater cladoceran or copepod collected over a period of several years. The small cladoceran *Chydorus sphaericus* has been reared through several generations on a pure diet of *Anabaena cylindrica*, which proved to be an excellent food, but no trace of phycobilin accumulation in the gut wall has been found.

SUMMARY

1. *Heterocypris incongruens* contains at least three different types of pigment: carotenoids, a pteridine, and a bilin. Haemoglobin and other haem pigments appear to be lacking in this species.

2. Astaxanthin and β-carotene are the only carotenoids found, even when the ostracod is feeding on algae containing abundant xanthophylls of various types.

3. A yellow pteridine, which rapidly decolorizes after extraction, is widespread in the body of the ostracod, but not in its eggs. It is suggested that this substance may play a part in the intermediate metabolism of the ostracod when in anaerobic conditions. *Heterocypris* can live and reproduce in anaerobic conditions for at least 2 weeks.

4. Biladiene pigments accumulate in the gut wall when the ostracod feeds on blue-green algae. These pigments can be made to disappear from the gut wall by restricting the diet to green algae, then made to reappear when a blue-green alga is given as food.

My thanks are due to Dr G. E. Fogg for his generous gift of a pure culture of *Anabaena cylindrica*, from which my own cultures were started. Prof. H. Munro Fox, F.R.S., and Dr Barbara M. Gilchrist have kindly read and criticized the manuscript.

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A CHECK-LIST OF BRITISH MARINE HALACARIDAE (ACARI), WITH NOTES ON TWO SPECIES OF THE SUB-FAMILY RHOMBOGNATHINAE

By J. Green
Bedford College, University of London

(Text-figs. 1–4)

Mites of the family Halacaridae are abundant on the sea shore, and in the sea down to considerable depths. Their identification is not an easy matter; there is no one account which can be used to identify all the British species, and the use of any one of the general accounts of European species, such as André (1946) or Viets (1936), by itself might easily lead to misidentification.

The purpose of the present list is to record the species which have so far been found around the British Isles, and to indicate sources of reliable descriptions of each species. A list of the British species of this family was given in a general list of British Acari by Turk (1953), and this has provided the starting point for the present list; no reference to occurrence in Britain before Turk's list is given, apart from one by Halbert (1920) which was left out by Turk. Twelve of the thirty-six species in the present list were not recorded by Turk. One species, *Halacarus zosterae* (Fab.), recorded by Turk has been omitted because the original description is quite inadequate to decide even which genus the creature should belong to. Eight species are known from British fresh water; these have not been included in the present list.

Only full species are given in this list, infra specific categories have been omitted. When the specific name is followed only by the author and date it indicates that the species was described from British material, and has not appeared in a general account of the group. The references in square brackets following the date indicate the source of a reliable description, while the succeeding references in rounded brackets give the authority for including the species as British.

The names in this list generally follow those given by Viets (1956) except that I have adopted Newell's (1947) classification of the Rhombognathinae, and the genus *Copidognathus* has not been divided into subgenera.

FAMILY HALACARIDAE Murray 1876

SUBFAMILY HALACARINAE Viets 1927

Gen. *Halacarus* Gosse, 1855

Subgen. *Halacarus* Gosse, 1855 s.str.

ctenopus Gosse, 1855 [Newell, 1947] (Turk, 1953)
actenos Trouessart, 1889 [Newell, 1947] (Turk, 1953)
bisulcus Viets, 1927 [Viets, 1936] (Spooner, 1959)

Subgen. *Thalassarachna* Packard, 1871 (= *Halacarellus* Viets, 1927)

basteri (Johnston, 1836) [Newell, 1947] (Turk, 1953)
southerni (Halbert, 1915)
areolatus (Halbert, 1915)
subterraneus (Schulz, 1933) [Newell, 1947][1]

Gen. *Copidognathus* Trouessart, 1888

granulatus (Hodge, 1863) [André, 1946, as *C. glyptoderma* Trs.] (Turk, 1953)
rhodostigma (Gosse, 1855) [André, 1946] (Turk, 1953)
loricifer André 1946 [André, 1946] (Turk, 1953)
fabricii (Lohmann, 1889) [André, 1946] (Turk, 1953)
tabellio (Trouessart, 1894) [André, 1946] (Turk, 1953)
lamellosus (Lohmann, 1893) [André, 1946] (Turk, 1953)
oculatus (Hodge, 1863) [André, 1946] (Turk, 1953)
gracilipes (Trouessart, 1889) [André, 1946] (Turk, 1953)
gibbus (Trouessart, 1889) [André, 1946] (Turk, 1953)

Gen. *Agauopsis* Viets, 1927

brevipalpus (Trouessart, 1889) [André, 1946] (Halbert, 1920)

SUBFAMILY POROHALACARINAE

Gen. *Caspihalacarus* Viets, 1928

hyrcanus Viets, 1928 [Viets, 1928*a*] (Green, 1956)

SUBFAMILY RHOMBOGNATHINAE

Gen. *Rhombognathus* Trouessart, 1888

notops (Gosse, 1855) [Newell, 1956] (Turk, 1953)
magnirostris Trouessart, 1889 [Newell & André, 1959][2]
lionyx Trouessart, 1900 [Newell & André, 1959][2]

[1] This species has not previously been recorded from Britain. I have identified a single specimen from the Essex coast, near the mouth of the Thames.

[2] I have British specimens which agree with Newell and Andrés' recent redescription of these species.

Gen. *Isobactrus* Newell, 1947

setosus (Lohmann, 1889) [Newell, 1947] (Turk, 1953)
levis (Viets, 1927) [Newell, 1947] (Green, 1956*a*)
uniscutatus (Viets, 1939) [Viets, 1939] (Green, 1956*a*)

Gen. *Rhombognathides* Viets, 1927, emnd. Newell, 1947

pascens (Lohmann, 1889) [Newell, 1947] (Turk, 1953)
spinipes (Viets, 1933) [Willmann, 1952] (Green, 1956*a*)
seahami (Hodge, 1860) [Newell, 1947] (Turk, 1953)
trionyx (Trouessart, 1899) [André, 1946] (Turk, 1953)
merrimani Newell, 1947 [Newell, 1947] (Green, 1956*b*)
mucronatus (Viets, 1927) [Newell, 1947][1]

Gen. *Metarhombognathus* Newell, 1947

armatus (Lohmann, 1893) [Newell, 1947] (Green, 1956*b*)
nudus (Viets, 1928*b*) [Sokolov, 1952] (see below)

SUBFAMILY SIMOGNATHINAE

Gen. *Simognathus*

minutus (Hodge, 1863) [André, 1946 as *S. sculptus*] (Turk, 1953)

SUBFAMILY LOHMANELLINAE

Gen. *Lohmanella* Trouessart, 1901

falcata (Hodge, 1863) [André, 1946] (Turk, 1953)

Gen. *Scaptognathus* Trouessart, 1889

tridens Trouessart, 1889 [André, 1946] (Spooner, 1959)
trouessarti Halbert, 1915

Two of the mites recorded in the above list are of interest for two distinct reasons. Their occurrences in Britain represent considerable extensions of their known ranges, and their systematic status needs clarification, particularly in relation to the subdivision of the subfamily Rhombognathinae.

Metarhombognathus nudus (*Viets*)

In 1928*b* Viets described two species of the genus *Rhombognathus* from the Murmansk coast. Both species were found on the same alga, and one, *R. nudus*, was found only as the adult, while the other, *R. contectus*, was found

[1] Not previously recorded from Britain. I have a specimen from the Tees estuary, collected by Miss E. Clay.

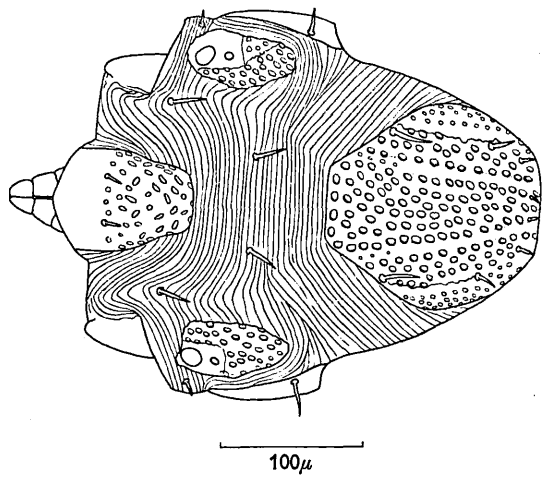
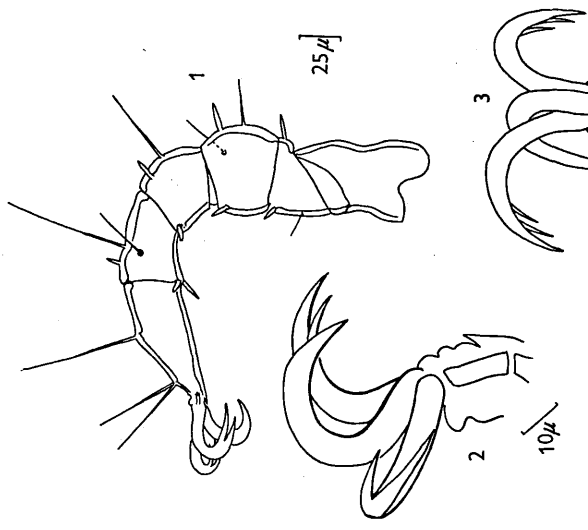


Fig. 4. Deutonymph of *Metarhombognathus nudus*, dorsal view of body. This specimen has a squarer posterior border to the anterior dorsal plate than most of the other specimens examined.



Figs. 1–3. *Metarhombognathus nudus*. 1, Leg IV of adult. 2, terminal claws of leg I of adult. 3, terminal claws of leg I of larva.

only as the deutonymph. Newell (1947), on the basis of his experience with a similar pair of forms, suggested that the two were in fact only one species. As Newell puts it 'It is highly improbable that two extremely closely allied forms (as shown by the unique structure of the lateral claws) could occupy the same habitat, both forms being numerous, and yet one be known only as the adult and the other only as the deutonymph'.

The two species have not apparently been recorded in the literature since the original descriptions and a repetition of these by Sokolov (1952), who still regards them as separate species. In Viet's (1956) register of the Halacaridae the two forms are maintained as separate species.

In March 1959 specimens agreeing with the description of *R. nudus* were found in small splash pools at the bottom of the cliffs at Cullercoats, Northumberland. The mites were very numerous in these pools, and all stages from larvae to adults were found.

The principal difference between *R. nudus* and *R. contectus* is that the dorsal plates of the former are small while those of the latter are large. When a random sample of twenty-five specimens from Cullercoats was examined in detail after clearing in lactic acid three specimens were found to be larvae. These had very small dorsal plates; the body length, from the front of the anterior dorsal plate to the posterior tip of the body, ranged from 216 to 223 μ. Five of the series were protonymphs, with small dorsal plates, and body lengths ranging from 223 to 295 μ. Seven deutonymphs, ranging in length from 350 to 380 μ, were found; these all had large dorsal plates and would be identifiable as *R. contectus*. Ten adults, with small dorsal plates, and having body lengths of 320–500 μ, completed the series.

This result of examining a random sample clearly indicates that *R. contectus* is the deutonymph of *R. nudus*. The latter name takes precedence owing to page priority. The finding that the protonymph has small dorsal plates like the adult is in agreement with Newell's description of the protonymph of *Metarhombognathus armatus* Lohmann, which has deutonymphs with large dorsal plates and adults with small dorsal plates. Newell has produced conclusive evidence that the deutonymphs of *M. armatus* with large dorsal plates are followed by adults with small dorsal plates by dissecting an adult from a quiescent deutonymph.

Further evidence that *R. contectus* is the deutonymph of *R. nudus* can be taken from the structure of the lateral claws. In the larvae, protonymphs and deutonymphs the lateral claws each carry two teeth on their inner borders (Fig. 3). This agrees with Viets figure of the claws of *contectus*. Nine of the adults had lateral claws with a single tooth on the inner border (Fig. 2), while one had two of its lateral claws with two teeth on their inner borders; it is noteworthy that this was the smallest of the adults. The loss of one of the teeth on the lateral claws appears to be normal when the adult stage is reached.

A further question is raised by the generic, or subgeneric, position of this

species. The difficulty is that two different systems of classification have been proposed. Viets (1936, 1952, 1956) regards the subfamily Rhombognathinae as consisting of a single genus *Rhombognathus* Trouessart, which he subdivides into three subgenera. Newell (1947, 1953) regards the subfamily as comprising four genera. Using Viet's system the present species would be called *Rhombognathus* (*Rhombognathopsis*) *nudus*. I have, however, adopted Newell's system for reasons which will become apparent in the discussion of the next species.

Rhombognathides merrimani *Newell*

This species has been recorded in Britain from the following localities: Isle of Man (Green, 1956*a*); Skokholm Island, Pembrokeshire (Green, 1956*a*); Gwendraeth Estuary, Carmarthenshire, on *Cladophora*, and in the gut of *Gobius minutus* (Green, unpublished records). Previous to these records it was known only from North America.

Newell (1947) has given an admirably detailed description of this species, so that no further description is necessary here. The most important feature of this species, from a systematic point of view, is its occurrence in two varieties: *merrimani* having two claws on all its legs, and *needleri* having three claws on legs I and II, and two claws on legs III and IV. Apart from this difference in claw number the two varieties are identical. My British specimens include both varieties, and I am unable to find any other difference between them, so that I am convinced with Newell that they belong to the same species. Viets (1950, 1952, 1956) does not accept this; he regards the two forms as not only separate species, but belonging to different subgenera. This is necessary if Viet's classification of the Rhombognathinae is followed, since the subgenera are separated entirely on claw characters, as follows:

Leg	I	II	III	IV
Rhombognathus	2	2	2	2
Rhombognathides	3	3	2	2
Rhombognathopsis	3	3	3	3

If the two forms of *R. merrimanni* are conspecific, as Newell and I believe, then Viet's classification must fall, and a substitute be found. I have adopted Newell's system because it is based upon an analysis of a considerable number of characters and it groups together species which are obviously closely allied although they may differ in claw number. The alternative to Newell's system would be to keep the single genus *Rhombognathus* and not divide it further, this would be a negative attitude, resulting in a genus of some thirty species with no expression of affinities within the group.

The specimens of *Metarhombognathus nudus* were collected while running a marine biology course for students at the Dove Marine Laboratory, Cullercoats. It is a pleasure to thank Dr H. O. Bull and his staff for the facilities which they made available to us.

SUMMARY

A check list of British marine mites of the family Halacaridae is given.

Metarhombognathus nudus (Viets), hitherto known only from the Barents Sea, is recorded from Cullercoats, Northumberland. Evidence is presented which indicates that the form described as *Rhombognathus contectus* Viets is the deutonymph of *Metarhombognathus nudus*.

The known distribution of *Rhombognathides merrimani* Newell in Britain is summarized, and its systematic status in relation to the classification of the Rhombognathinae is discussed. It is recommended that Newell's classification of this subfamily be adopted.

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ZOOPLANKTON OF THE RIVER SOKOTO. THE ROTIFERA

By

J. GREEN

Bedford College, University of London

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(With 20 figures in the text)

Forty-one species of Rotifera are recorded from the River Sokoto, North Nigeria, and details of their seasonal abundance are given. There is a marked change in the composition of the rotifer fauna of the river when it floods in the single wet season. The cyclomorphosis of *Keratella tropica* begins with the appearance of the species in December. At this stage most individuals lack a left posterior spine. The left posterior spine then grows longer in successive samples until February or March and then decreases to very short lengths in June. The right posterior spine does not vary much throughout the cycle and is always positively correlated with the length of the lorica. The length of the left spine shows varying degrees of correlation with the length of the lorica according to the rate of change of length of the left spine. *Brachionus caudatus* shows synchronous changes of both its posterior spines, the lengths of which are positively correlated with the length of the lorica. The relation of these findings to the general theories of cyclomorphosis is discussed. The flotation theory of Wesenberg Lund is rejected from application to the species studied. The necessity of considering cyclomorphic phenomena in relation to body size is emphasized.

CONTENTS

INTRODUCTION

The material on which this and subsequent papers will be based was collected by Mr M. J. Holden during a four-year tour of duty as a fisheries research officer in the North Nigerian province of Sokoto. A general description of the hydrology of the River Sokoto and a study of the fluctuations of the major groups of planktonic organisms has been given by Holden &

Green (1960). The dominating feature of the hydrology is the flooding of the river during the single wet season (Fig. 1). This has great effects on the abundance and composition of the zooplankton.

Temperatures in the River Sokoto are generally high, but during December and January a cold wind, the harmattan, blows southwards from the Sahara, and cools the river (Fig. 1). The annual cycle in pH is also shown in this figure, the main feature is a marked lowering of pH when the river is in flood and more acid water is washed out from bordering swamps.

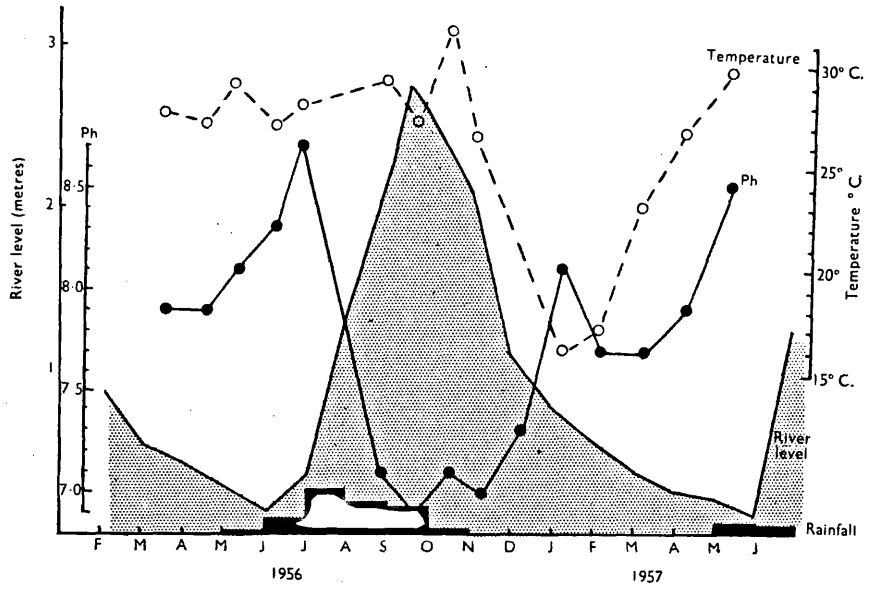

Fig. 1.—Some features of the environment. Temperature and pH are given for the pool Fesafari, the data for the river are very similar (*cf.* Holden & Green, 1960).

When the floods subside a pool called Fesafari becomes isolated from the main channel of the river. The seasonal changes in form of *Keratella tropica* and *Brachionus caudatus* have been followed in this pool in preference to the river because the populations in the pool were more likely to be homogeneous than the constantly changing populations swept down by the river.

The seasonal cycles of temperate zone rotifers are now well known (*cf.* Kofoid, 1908 ; Wesenberg Lund, 1930 ; Carlin, 1943), but very little is known of their cycles in tropical countries. The work described below, while imperfect in details, provides a general account of the seasonal cycle of rotifers in a tropical river.

METHODS

The plankton was collected by Mr Holden, using a fine meshed plankton net which was towed for a fixed distance just below the surface of the water. Details of the sampling stations in the main channel and in the pool Fesafari are shown on a map in Holden & Green (1960).

The samples were preserved in 5 per cent formalin, and were in excellent condition when I received them for study. The volume of each sample was made up to 30 ml. and sub samples were taken after mixing each sample thoroughly. All the animals in a sub-sample of 0·5 ml. were counted and assigned to major groups, such as Cladocera, Calanoida, Rotifera, etc. This gave a measure of the actual abundance of each group. The relative abundance of each species within the major groups was estimated by another series of samples in which 100 specimens of each group were identified to specific level.

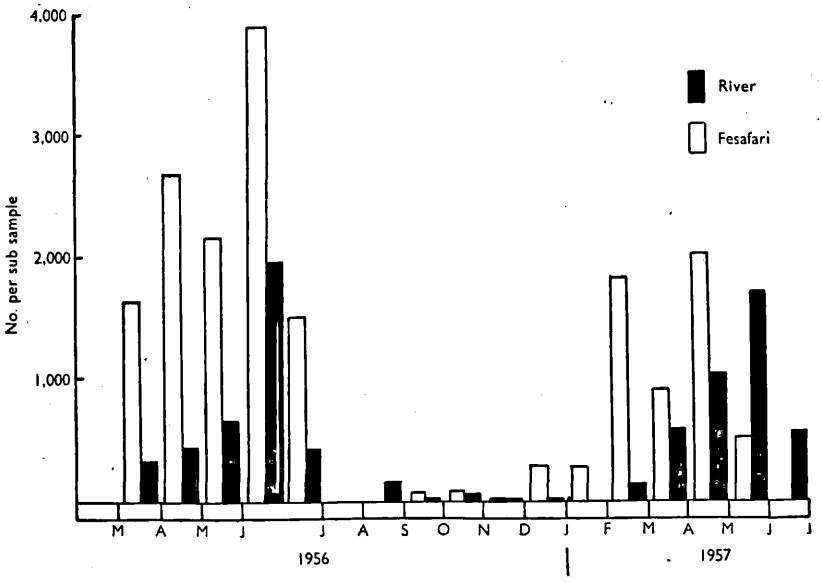

Fig. 2.—Seasonal variation in total rotifers in the River Sokoto and the pool Fesafari. Notice in this figure that two samples were taken in June 1956 but no sample was taken in July ; this also applies to the other diagrams showing seasonal occurrence. Dates of samples are given in Tables 1 and 2. Each sub sample represents the number of individuals in approximately one-third of a cubic metre.

The technique used for this series of counts varied slightly with the different groups. Rotifers were identified and counted after they had been mixed with a little lactic acid containing lignin pink. This stained the loricas of many of the species and the refractive index of lactic acid is such that the sculpturing of the lorica was easy to see. Preparations made in this way were examined on a specially ruled glass slide with lines at intervals just a little under the field diameter of a monocular microscope with a 16 mm. objective. The slide was moved with a mechanical stage and each row was systematically searched for rotifers. The sequence in which rotifers were encountered was essentially random, and the counts can be regarded as reasonable estimates of the relative abundance of each species.

A parfocal higher powered objective was used to identify any small or difficult specimens which were not recognizable under the lower power.

The figures showing seasonal occurrence were drawn using data calculated from the relative abundance of each species and the total rotifers in each sub sample.

SYSTEMATIC SURVEY AND SEASONAL OCCURRENCE

Order BDELLOIDEA

Many of the species belonging to this group are not identifiable in preserved material. Several specimens were found in samples from the river taken between July and December, which appeared to belong to the genus *Habrotrocha*. These would not be true planktonic species, and their occurrence in the samples is related to the flooding of the river.

One species of this order was identifiable as follows.

Family PHILODINIDAE

Rotaria neptunia Ehrenberg

A few specimens were found in samples taken from the river on September 12th, 1956, and April 6th, 1957. Most of the species in this genus are bottom dwellers, but the present species has been recorded in the plankton by other authors, for instance Kofoid (1908) in the River Illinois, and Beauchamp (1932) in the East African Lakes.

Order MONOGONONTA

Sub-order *PLOIMA*

Family BRACHIONIDAE

This family is by far the best represented of all the rotifer families in the samples ; no less than twenty of the species are brachionids. The relative importance of the members of this family can be seen in Tables 1 and 2.

The taxonomy of this family is complex, particularly within the genera *Brachionus* and *Keratella*. Both these genera were revised by Ahlstrom (1940, 1943), but since these revisions there have been additions, and changes in the status of various forms have been proposed (Gillard, 1948 ; Hauer, 1953, 1957 ; Carlin, 1943 ; Berzins, 1954, 1955).

Anuraeopsis navicula Rousselet (Fig. 3 a)

The specimens from the Sokoto were about 93 μ long and 46 μ wide, and they agree with the form described from Lake Nakavali by Beauchamp (1932) as the variety *coelata*. Gillard (1948) believes that this may prove to be a geographical race, and places *A. punctata* Evens (1947) as a synonym of Beauchamp's variety.

This species was not found in the main channel of the river, but was found in small numbers in Fesafari in April and June.

Brachionus bennini (Leissling) (Fig. 3 f)

A single specimen was found in the sample taken from the river on March 4th, 1957. It has not previously been recorded in West Africa, but it has been found in East Africa.

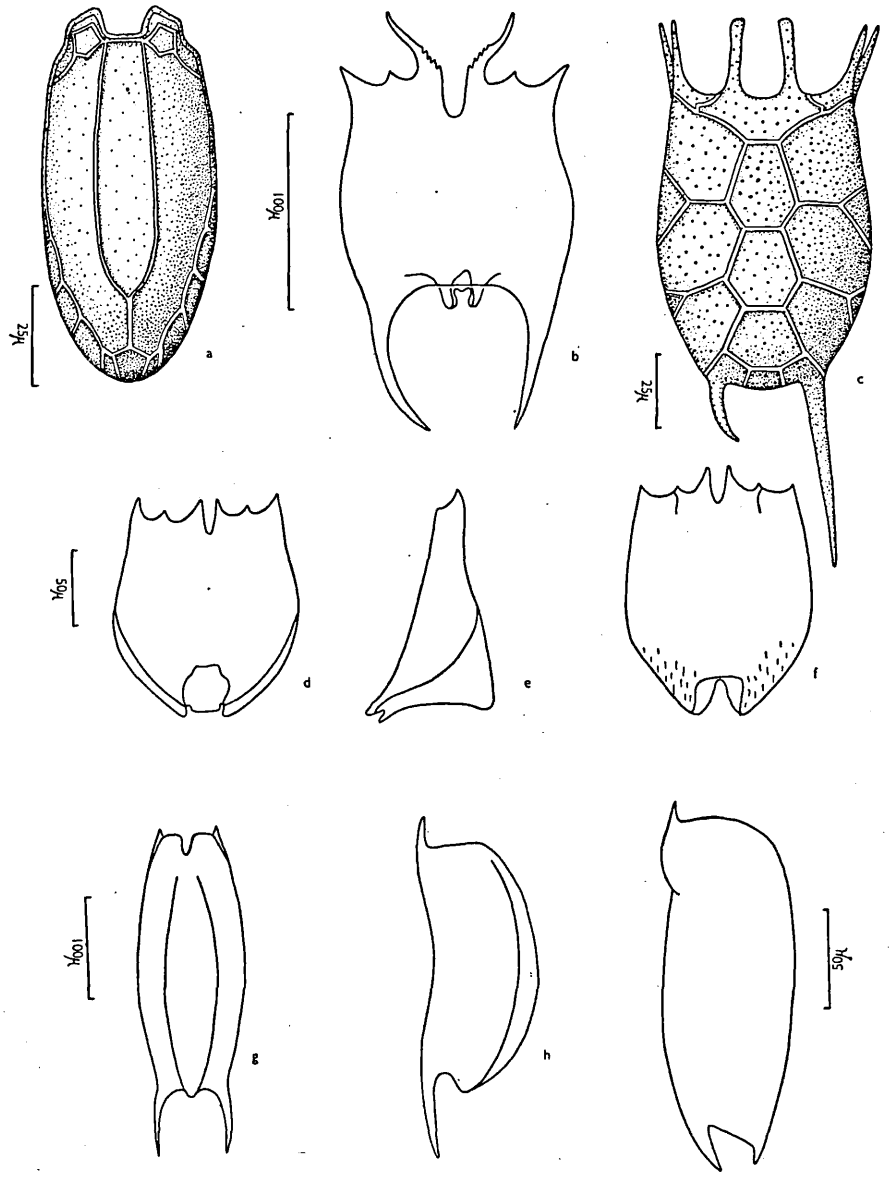

Fig. 3.—Brachionidae from the River Sokoto and the pool Fesafari. a, *Anuraeopsis navicula*, dorsal view. b, *Brachionus quadridentatus*, dorsal view. c, *Keratella tropica*, dorsal view. d, *Brachionus bidentatus*, dorsal view. e, *B. bidentatus*, lateral view. f, *Brachionus bennini*, dorsal view. g, *Mytilina ventralis*, dorsal view. h, *M. ventralis*, lateral view. i, *M. ventralis* f. *brevispina*, lateral view. Note that i is drawn to twice the scale of g and h.

Brachionus bidentata Anderson (Fig. 3 d, e)

A single specimen was found in the sample taken from the river on June 10th, 1957. I have not seen any previous records of this species in Africa.

TABLE 1

Family Brachionidae. Relative numbers of each species in the River Sokoto, as percentage of the total rotifers in each sample.

	14 Mar	10 Apr	5 May	6 Jun	26 Jun	14 Aug	12 Sep	9 Oct	7 Nov	1 Dec	1 Feb	4 Mar	6 Apr	16 May	10 Jun
Brachionus															
bennini	–	–	–	–	–	–	–	–	–	–	–	1	–	–	–
bidentata	–	–	–	–	–	–	–	–	–	–	–	–	–	–	1
budapestinensis	–	3	3	–	1	–	–	–	–	–	–	–	–	4	–
calyciflorus	+	14	3	4	4	1	–	–	1	4	1	5	3	+	1
caudatus	78	40	42	32	27	6	10	2	7	26	58	73	55	32	12
diversicornis	–	1	4	–	–	–	–	–	2	–	–	–	2	1	–
falcatus	2	6	11	5	15	1	–	12	4	6	2	3	5	10	4
quadridentatus	4	1	1	–	–	4	2	4	–	–	3	1	1	–	8
Dipleuchlanis															
propatula	–	–	–	–	–	5	–	–	–	–	–	–	–	–	–
Euchlanis															
triquetra	–	–	–	–	–	1	–	–	–	–	–	–	–	–	–
Epiphanes															
macroura	–	–	–	+	–	–	–	–	–	–	–	–	–	+	–
Keratella															
tropica	1	–	1	–	1	1	–	–	–	2	32	14	3	–	5
Macrochaetus															
collinsi	–	–	–	–	–	8	2	2	8	6	–	–	–	–	–
Mytilina															
ventralis	–	–	–	–	–	1	–	–	1	2	–	–	–	–	–
Notholca															
acuminata	–	–	1	–	–	–	–	–	–	–	–	–	–	–	–
Platyias															
patula	12	5	–	–	–	11	–	–	–	–	–	1	1	–	5
quadricornis	+	3	–	–	–	6	1	–	–	4	–	–	–	–	6
Trichotria															
tetractis	–	–	–	–	–	1	–	–	–	–	–	–	–	–	–

+ indicates present, but less than 1 per cent of total rotifers.

TABLE 2

Family Brachionidae. Relative numbers of each species in Fesafari, as percentage of the total rotifers in each sample.

	23 Mar	18 Apr	7 May	7 Jun	25 Jun	19 Sep	13 Oct	10 Nov	8 Dec	8 Jan	3 Feb	7 Mar	5 Apr	17 May
Anuraeopsis														
navicula	–	2	–	–	+	–	–	–	–	–	–	–	–	–
Brachionus														
calcyciflorus	1	1	2	–	3	–	–	–	+	–	–	1	–	1
caudatus	35	66	77	47	66	6	4	18	9	21	75	69	61	10
falcatus	20	6	15	9	7	–	2	12	5	13	5	16	26	25
quadridentatus	–	–	–	–	–	1	–	–	–	–	–	–	–	–
Dipleuchlanis														
propatula	–	–	–	–	–	–	7	–	–	–	–	–	–	–
Diplois														
daviesiae	–	–	–	–	–	12	10	–	–	–	–	–	–	–
Euchlanis														
dilatata	–	–	–	–	–	–	–	1	–	–	–	–	–	–
Keratella														
tropica	8	4	5	+	–	–	–	–	3	60	20	14	10	–
Macrochaetus														
collinsi	–	–	–	–	–	–	1	3	–	–	–	–	–	–
Mytilina														
ventralis	–	–	–	–	–	1	1	–	–	–	–	–	–	–
Platyias														
patula	–	–	–	–	–	1	–	–	6	–	–	–	–	–
quadricornis	–	–	–	–	–	5	1	3	–	–	–	–	–	1

\+ indicates present, but less than 1 per cent of total rotifers.

Brachionus budapestinensis Daday

This species was present in small numbers in the river in April and May 1956, and a single specimen was found in June of the same year. Small numbers again appeared in the river in May 1957. It was not found in the pool Fesafari. Kofoid (1908) found this species in the River Illinois from the end of June until the early part of October, and regarded this as a mid-summer planktont, with its maximum near the maximum summer temperature.

Brachionus calyciflorus Pallas

The seasonal occurrence of this cosmopolitan species is shown in Fig. 4. It was generally more abundant in the main channel than in Fesafari.

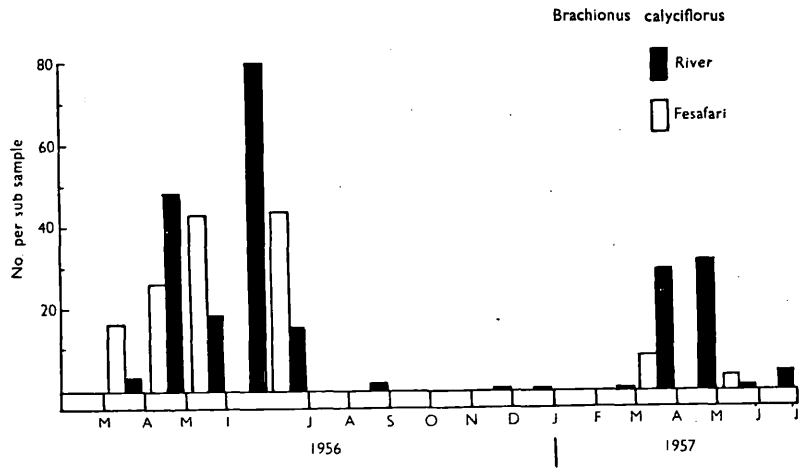

Fig. 4.—Seasonal occurrence of *Brachionus calyciflorus*.

Brachionus caudatus Barrois & Daday (Fig. 6)

This is the dominant rotifer in the river. The seasonal occurrence is shown in Fig. 5. The cyclomorphosis of the species is dealt with on pages 516-519.

Some workers regard this species as a mere variety of *B. angularis* ; Kofoid (1908) goes so far as to say that he does not regard it as worthy of even varietal distinction. Fig. 6 shows that in the Sokoto a whole series exists, from forms with very long spines to much smaller forms, which, although mature and carrying eggs, lack any clearly developed spines. I have retained the name *B. caudatus* because I believe that the small specimens from the Sokoto are by no means the same as *B. angularis*, which is typically a much larger form lacking any trace of posterior spines. Gillard (1948), Hauer (1953, 1957) and Voigt (1957) all retain *B. caudatus* as a distinct species.

All the specimens from the Sokoto had only two spines on the anterior border of the lorica. It is of interest that in North-East Brazil Hauer (1953) found a series of forms of *B. caudatus* showing the same variation in the posterior spines as that found in the Sokoto, but all having six spines on the anterior border of the lorica.

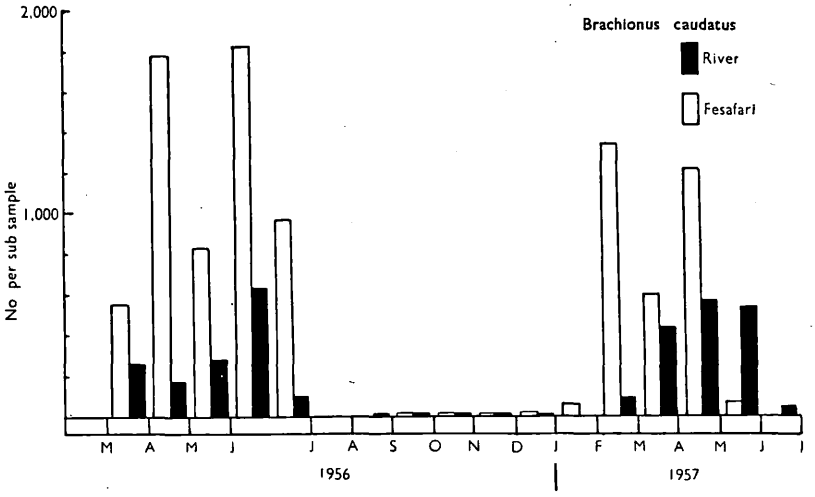

Fig. 5.—Seasonal occurrence of *Brachionus caudatus*.

Brachionus diversicornis (Daday)

This species has long been known under the name *Schizocerca diversicornis*, but most recent authors agree that it should be placed in the genus *Brachionus*. In Europe it is generally regarded as a summer form. It has been recorded from the summer plankton of the Rhine (Lauterborn, 1898) and the Elbe (Schorler, 1900). It occurred in the River Sokoto in April and May in 1955, 1956 and 1957. A few specimens were also found in November 1956, indicating that the species might have a second pulse when conditions are suitable. No specimens were found in the samples from Fesafari.

A specimen from the sample taken on April 6th, 1957, was found to be infected with *Dimoerium hyalinum* Przesmycki, a parasite of uncertain systematic position. The form in *B. diversicornis* consisted of numerous oval bodies measuring $32 \times 14\,\mu$.

Brachionus falcatus Zacharias

This species is only found where the water is warm ; in Europe it is a summer form, and it is widespread in the tropics. The seasonal occurrence is shown in Fig. 7. This species was always more abundant in the pool Fesafari than in the main channel. In Fesafari it sometimes formed as much as 45 per cent of the total rotifers.

Although no detailed measurements have been made there does not seem to be much seasonal variation in the length of the posterior spines of this species in the Sokoto. The variation in spine length in a single sample is illustrated in Fig. 8.

In the sample taken from Fesafari on December 8th, 1956, a newly-hatched specimen was found. The long anterior spines were still curved ventrally around the lorica which was distinctly rounded and egg-shaped. The length of the lorica was $98\,\mu$ and the posterior spines were $72\,\mu$ long. When these dimensions are compared with those of the largest adults from the same

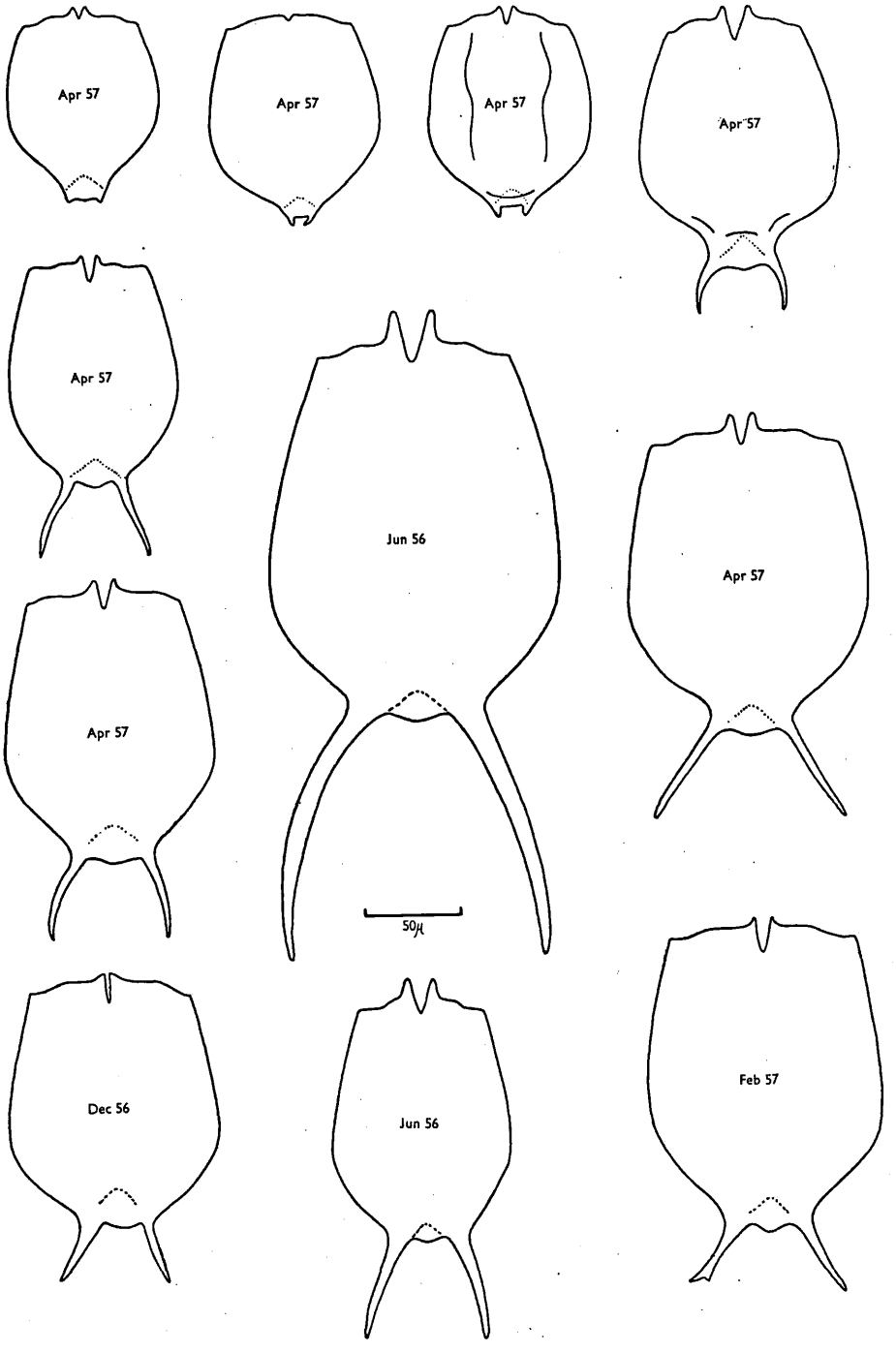

Fig. 6.—Variation in the form of *Brachionus caudatus*. All the specimens were taken from Fesafari and all are drawn to the same scale.

locality (lorica length 144 μ, posterior spine length 170 μ) they indicate that the posterior spine grows allometrically ; the lorica does not double its hatching length, while the posterior spines become more than twice as long as they were at hatching.

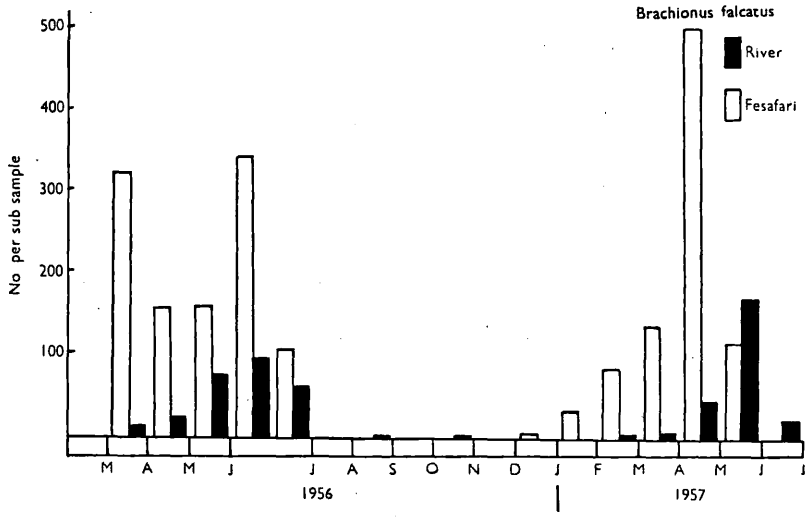

Fig. 7.—Seasonal occurrence of *Brachionus falcatus*.

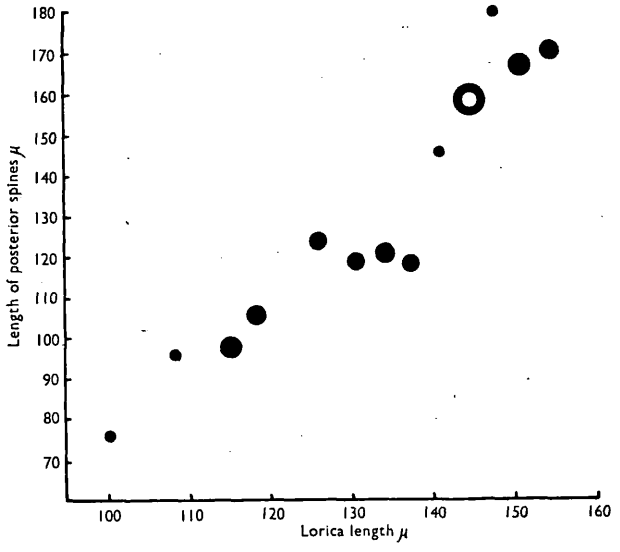

Fig. 8.—Relation between lorica length and posterior spine length in a sample of fifty *Brachionus falcatus* taken from Fesafari, 7th June 1956. The diameter of each dot is proportional to the number of animals on which it is based ; the largest represents fifteen and the smallest a single specimen.

Brachionus quadridentatus Hermann

A single specimen was found in the sample from Fesafari on September 19th, 1956. It was a remarkably fine specimen (Fig. 3 b) with long diverging anterior median spines with the middle thirds of their inner borders serrated. Specimens with less well developed spines were found in the main channel irregularly throughout the year. This is a cosmopolitan species regarded as a summer form in temperate climates.

Dipleuchlanis propatula (Gosse)

Small numbers of this species were found in the river in August, and it was somewhat more numerous (7 per cent of the total rotifers) in Fesafari in October.

Diplois daviesiae Gosse

No specimens were found in the main channel of the river, but there was a small outburst in Fesafari in September and October, when it formed up to 12 per cent of the total rotifers. It does not seem to have been recorded previously from West Africa.

Epiphanes macroura Barrois & Daday

Specimens from the Sokoto agree with the figures given by Beauchamp (1932) of East African specimens under the name *Notops mollis* Hempel, which is a synonym of *E. macroura.*

This species was present in small numbers in the river in June 1956 and May 1957. It was not found in Fesafari.

Euchlanis dilatata Ehrenberg

A single specimen was found in the sample from Fesafari on November 10th, 1956. This species has been recorded from Gambia (Berzins, 1957) and from the Belgian Congo (Evens, 1959).

Euchlanis triquetra Ehrenberg

A single specimen was found in the sample from the river on August 14th, 1956. This species was recorded as fairly common in the swamps south of Bolgatanga, Ghana, by Russell (1956) and a single specimen has been recorded from Gambia Berzins, 1957). The record by Russell is open to some doubt because he mentions a ventral plate, which is absent in the true *triquetra* Ehrenberg, but present in *E. incisa* Carlin which has been confused in the past with *E. triquetra.*

Keratella tropica (Apstein) (Fig. 3 c)

The synonymy of this species has recently been given in detail by Berzins (1955). It has frequently been regarded as a variety of *K. valga* (Ehrenberg), but it can be distinguished from that species by the sculpturing of the lorica. *Keratella valga* has three hexagons on the dorsal surface ; the last hexagon has the posterior border of the lorica as one of its sides. In *K. tropica* the dorsal surface has three hexagons and a small four-sided figure between the posterior border of the lorica and the last hexagon (Fig. 3 c). The two species differ

in their geographical distribution ; *K. valga* is predominantly found in Europe, with some records from temperate Asia ; *K. tropica* is found in the South-West of North America, South America, Africa, from Egypt to the Cape, but it has not previously been found in West Africa. There are also records from Madagascar, Ceylon, India, Indonesia, New Zealand, China, Japan, and the southern parts of the Soviet Union around the Caspian Sea.

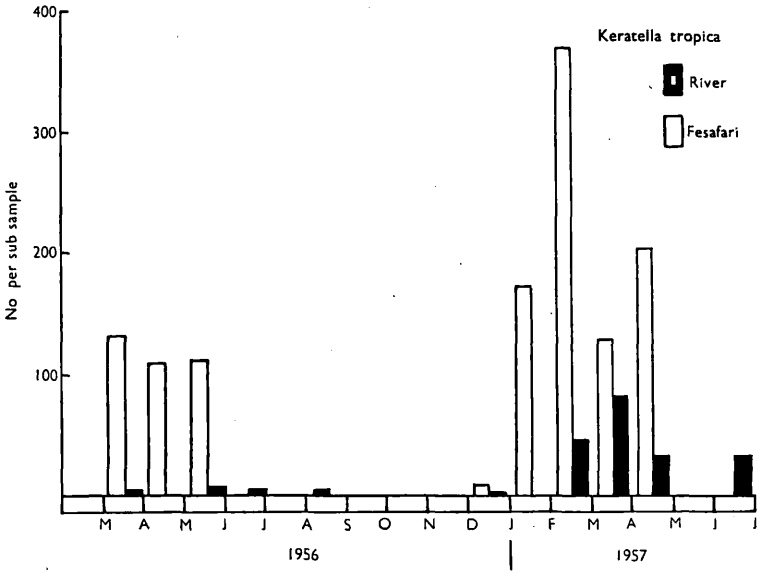

Fig. 9.—Seasonal occurrence of *Keratella tropica*.

The seasonal occurrence of this species is shown in Fig. 9. The numbers are generally much higher in Fesafari than in the main channel, but while the species disappears from the pool by the end of June it can still be found in the river as late as August.

Macrochaetus collinsi (Gosse)

This is a species characteristic of the flood waters. It appeared in the river in August and persisted until December, forming up to 8 per cent of the total rotifers. The numbers appearing in Fesafari were much smaller, and the seasonal occurrence was limited to October and November.

Mytilina ventralis (Ehrenberg)

Small numbers of this species were found when the river was in flood. The duration of occurrence in the river extended from August to December, but only from September to October in Fesafari.

Two distinct morphological forms were found : one (Fig. 3 i) corresponds with the form *brevispina* Ehrenberg, the other is similar to the var. *macracantha* (Gosse) but differs in that the median posterior part of the lorica is not extended into a spine (Figs. 3 g & h).

Notholca acuminata (Ehrenberg)

Three specimens were found in the sample from the river on May 5th, 1956. I have not seen any previous record of this species from Africa.

Platyias patulus (O. F. Muller)

This species appeared regularly in small numbers in the river from March to May in 1955, 1956 and 1957. It was also found in August 1956 and June 1957. In Fesafari it was found very sparsely in September and December 1956, presumably being washed into the pool by the flooding river.

Platyias quadricornis (Ehrenberg)

The occurrence of this species in the river paralleled that of *P. patulus* to a marked extent, the only difference of any note was the occurrence of *P. quadricornis* in the main channel in December 1956. In Fesafari this species was only found in small numbers from September to November 1956 ; again like *P. patulus* it was probably washed into the pool by the flooding river.

Both species of *Platyias* may be regarded as only partly planktonic, because they spend much of their time among water plants. Both species have widespread distributions and may be cosmopolitan.

Trichotria tetractis (Ehrenberg)

A single specimen was found in the sample from the river on August 14th, 1956.

Family LECANIDAE

Only one genus of this family is represented in the samples from the Sokoto, but this genus, *Lecane*, is the most abundant of all the rotifers during the wet season. The numbers per unit volume of water are low, but over half the rotifers found in the plankton during the wet season belong to this genus.

TABLE 3

Genus *Lecane*. Relative numbers of each species, as a percentage of the total number of rotifers. The first figures under each date refer to the main channel of the river and the figures in brackets refer to Fesafari. Only data for 1956 are given, because these include all the occurrences of the species apart from one record of *L. bulla* which formed 1 per cent of the total rotifers in the river in June 1957 (see Fig. 11).

	May	Jun	Aug	Sep	Oct	Nov	Dec
Lecane bulla	– –	– –	1 –	10 (51)	26 (23)	24 (27)	12 –
Lecane curvicornis	– –	– –	– –	– (3)	– (9)	4 –	2 –
Lecane hornemanni	– –	– –	– –	– –	– –	– (1)	– –
Lecane leontina	– –	– –	12 –	10 (8)	12 (11)	16 –	2 –
Lecane luna	– –	– –	17 –	4 (3)	18 (11)	– –	– –
Lecane quadridentata	– –	– –	– –	– –	– –	2 –	4 –
Lecane ungulata	1 –	1 –	2 –	– (2)	– (4)	6 –	– –

Lecane bulla (Gosse) (Fig. 10 d)

The appearance of this species in the plankton is associated with the floods which wash out rotifers from the bordering backwaters of the river. This is the most abundant of all the rotifers when the river level rises. Jennings (1900) noted that this was probably the most abundant species of its genus

in richly vegetated backwaters. This is a cosmopolitan species, but the only previous record of its occurrence in West Africa is that of Berzins (1957) from Gambia.

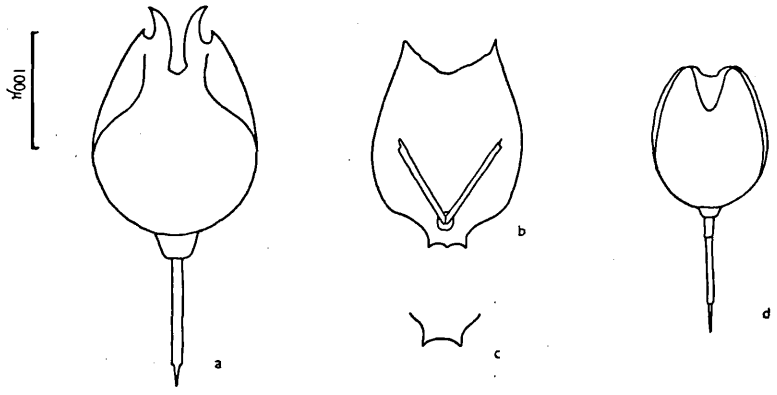

Fig. 10.—*Lecane* spp. from the River Sokoto, December 1956. a, *L. quadridentata*. b and c, *L. leontina*. d, *L. bulla*. All are drawn to the same scale.

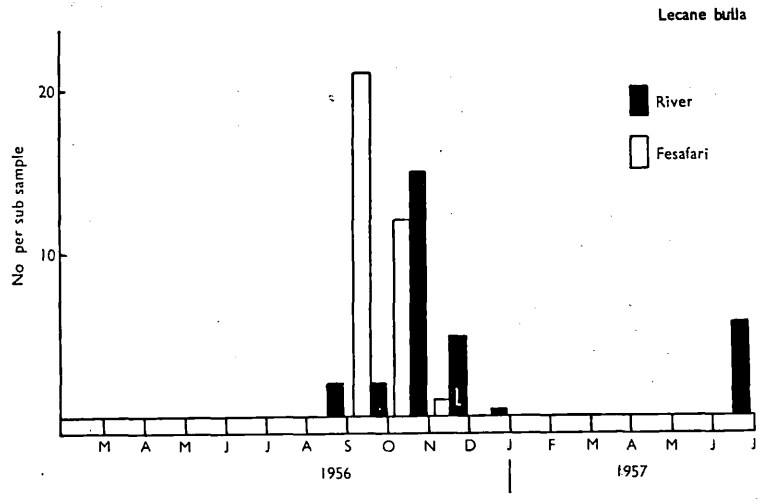

Fig. 11.—Seasonal occurrence of *Lecane bulla*.

The seasonal occurrence of *L. bulla* is shown in Fig. 11 ; it forms a marked contrast with the other seasonal distributions which have been figured.

Lecane curvicornis (Murray)

The specimens from the Sokoto agree in detail with the figures and description given by Hauer (1937/8) of a specimen from Sumatra. The species has also been found in North, Central and South America, Central Africa and Europe. Berzins (1957) records it from Gambia and Russell (1956) records it from Ghana.

In Fesafari this species was found in September and October 1956, and formed up to 9 per cent of the total rotifer population. It appeared somewhat later in the main river channel, being found only in November and December 1956, and never forming more than about 4 per cent of the total rotifers.

Lecane hornemanni (Ehrenberg)

Only a single specimen was found. This occurred in the sample taken from Fesafari on November 10th, 1956. This species does not seem to have been recorded previously from Africa.

Hauer (1937/8) gives the lorica length as ranging from 84–110 μ, and the toe length as 30–35 μ. The specimen from Fesafari had a lorica length of 101 μ, and the toes were 36 μ long.

Lecane leontina (Turner)

This is a cosmotropical species which has been recorded in West Africa from Ghana (Russell, 1956) and Gambia (Berzins, 1957). The Sokoto specimens included forms with a small median posterior spine (Fig. 10 b).

This species was found in the river from August to December, forming up to 16 per cent of the total rotifers. In Fesafari the seasonal occurrence was more restricted ; September and October were the only months in which it was found.

Lecane luna (O. F. Müller)

A cosmopolitan species which has been recorded from Ghana (Russell, 1956) and Gambia (Berzins, 1957).

This species was found in the river (August) before it was found in Fesafari (September) and it persisted in both localities until October, forming up to 18 per cent of the total rotifers in the river.

Lecane quadridentata (Ehrenberg) (Fig. 10 a)

This species has not previously been recorded in West Africa, and it is not included in the list of Belgian Congo species by Evens (1949). It was found sparingly in samples from the river in November and December, but was not found in samples from Fesafari.

Lecane ungulata (Gosse)

This cosmopolitan species occurred irregularly in samples from the river, occasional specimens being found in May, June, August, and somewhat higher numbers in November. In Fesafari the seasonal occurrence was restricted to September and October.

Family NOTOMMATIDAE

Scaridium longicaudum (O. F. Muller)

This widespread species usually lives among plants. It was found in small numbers in the river in December 1956, but was not found in the samples from Fesafari.

Family TRICHOCERCIDAE

Trichocerca bicristata (Gosse)

This species is generally found among plants and detritus, but it has also been found as a regular member of the plankton, for instance by Kofoid (1908) in the River Illinois. It has not previously been recorded in West Africa.

In the River Sokoto it was present from September to December, and formed up to 14 per cent of the total rotifers. It was less abundant in Fesafari and the duration of occurrence did not extend beyond November.

Trichocerca longiseta (Schrank)

A single specimen was found in the sample taken from Fesafari on April 6th, 1954. This species was recorded as rare in the swamps near Bolgatanga, Ghana, by Russell (1956), but was not recorded from Gambia by Berzins (1957).

Family ASPLANCHNIDAE

Asplanchna brightwelli Gosse

The seasonal occurrence of this species is shown in Fig. 12. The species is most abundant from April to June, with sporadic appearances at other times. The relative number in relation to other rotifers (i.e. as a percentage of the total rotifers) is much higher in the river than in Fesafari, and in 1957 it failed to appear in Fesafari. This may be a factor of some importance for the other rotifers because members of the genus *Asplanchna* are well known as predators of rotifers. In the sample taken from Fesafari on June 7th, 1956, several *A. brightwelli* were found with *Anuraeopsis navicula* in their stomachs.

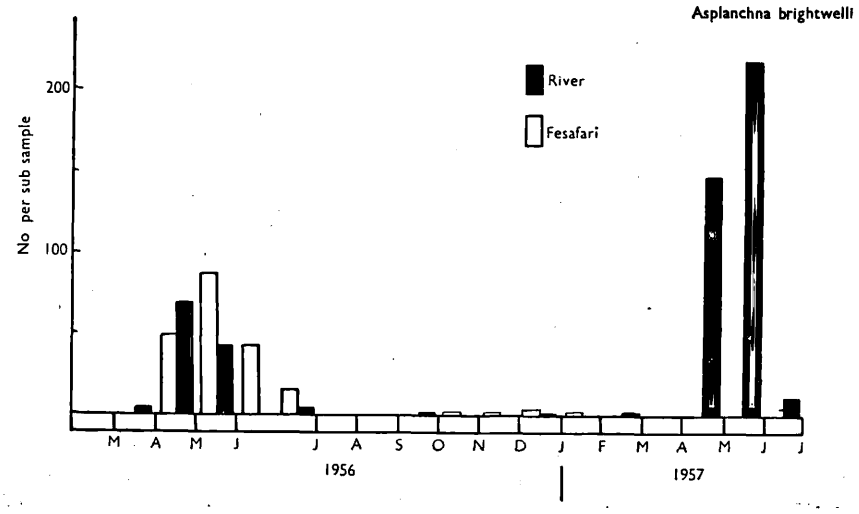

Fig. 12.—Seasonal occurrence of *Asplanchna brightwelli.*

Family SYNCHAETIDAE

Synchaeta sp.

There was an outburst of an unidentifiable *Synchaeta* species in Fesafari in December 1956. No specimens were found in the river, but in Fesafari this rotifer formed over 70 per cent of the total rotifers in the December sample. A few specimens were still present in the January sample, but none were found at any other time of year.

Polyarthra sp.

A single specimen of a *Polyarthra* species was found in the December 1956 sample from Fesafari. The body was about 100 μ long and 79 μ wide. The lateral spines were 108 μ long and 13 μ wide; ventral spines, about 40 μ long, were present. The specimen is clearly allied to *P. dolichoptera* (Idelson) as redescribed by Carlin (1943), but I have hesitated to asign it definitely to this species because it was not particularly well preserved.

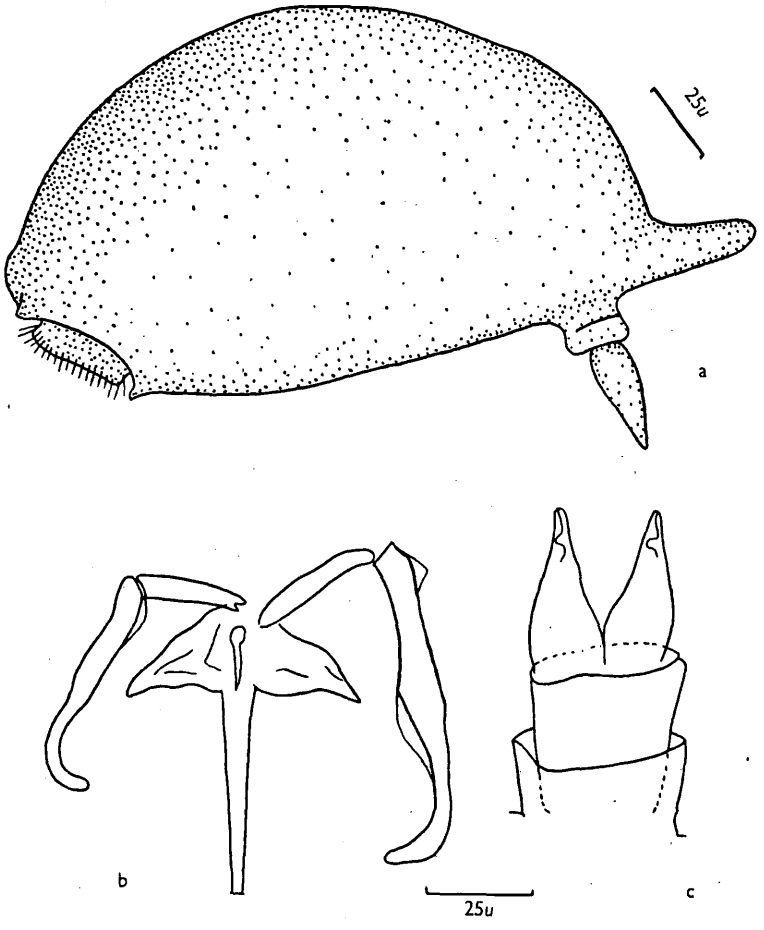

Fig. 13.—*Pseudoploesoma* sp. a, lateral view of uncleared specimen. b, ventral view of jaws cleared in sodium hydroxide. c, foot of cleared specimen, slightly squashed.

Pseudoploesoma sp. (Fig. 13)

A single specimen was found in the sample from Fesafari on November 10th, 1956. The body was 216 μ long, and the toes 36 μ long. These are close to the dimensions given for *P. formosum* Myers, but I have not assigned the specimen to this species because no fossettes were found on the lorica. Since *Pseudoploesoma* is known only from North America it was decided to examine the jaws of the Sokoto specimen to compare with the original description. The specimen was cautiously cleared with a solution of sodium hydroxide. This gave further details of the structure of the foot (Fig. 13 c) and showed that the jaws were virgate in form, agreeing in general with those figured by Myers (1934), but showing some differences in detail (Fig. 13 b). Treatment with sodium hydroxide also revealed the parabuccal processes, so that assignment to the genus *Pseudoploesoma* is justified. The lack of fossettes on the lorica, and the detailed differences in the jaws indicate that this specimen is distinct from *P. formosum*, but I do not feel justified in basing a new species on a single specimen which required drastic treatment to reveal its generic characters.

Sub-order *FLOSCULARIACEA*

Family TESTUDINELLIDAE

Testudinella patina

This cosmopolitan species occurred somewhat erratically in the samples, being most frequently found in the river during the wet season, but occurring in small numbers throughout the year. It was very scarce in Fesafari ; single specimens were found in January, April and October.

Filinia longiseta (Ehrenberg)

Small numbers were found in the river between May and September 1956, the only occurrence in 1957 was in April when it formed about 6 per cent of the total rotifers. Only single specimens were found in Fesafari in March and June 1956. This species is known from river plankton in Europe, North America and Manchuria.

Filinia sp.

Single specimens were found in Fesafari on the 7th and 25th June, 1956. The specimens had oval bodies, 108 μ long ; the terminal spine was 288 μ long, and the lateral spines varied between 268 and 342 μ. They run down to *F. maior* Colditz in Voigt (1957), but the spines are much shorter than the dimensions given by that author and by Carlin (1943). A further cause for doubt is the occurrence of *maior* as a winter form in Europe, whereas the specimens from the Sokoto were collected during the hottest season.

Tetramastix opoliensis Zacharias

The seasonal occurrence of this species is shown in Fig. 14. It is a summer form in Europe, and is widespread in the tropics. There are several records of its occurrence in flowing waters.

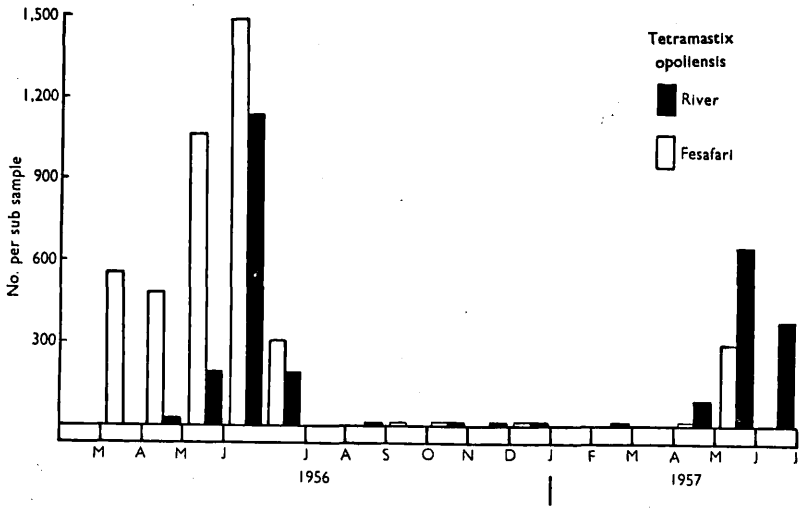

Fig. 14.—Seasonal occurrence of *Tetramastix opoliensis*.

Sub-order *COLLOTHECACEA*

Family COLLOTHECIDAE

Collotheca pelagica (Rousselet)

This is a planktonic representative of a group which is mainly sessile. Only a single specimen was found in the river in November 1956.

SEASONAL VARIATION IN NUMBER OF SPECIES

The total number of species observed in this study is forty-one ; samples from the river yielded thirty-one species, twenty of which were also found in the pool Fesafari. The samples from Fesafari yielded thirty species, of which ten were not found in the main channel. The two stations thus had roughly the same number of species and two-thirds of these they had in common.

Many of the species which were not shared by the two stations were rare, and were found only once or twice in the samples, but some showed a well-marked development of their population at one station while remaining absent from the other. The most striking example is the *Synchaeta* species which formed over 70 per cent of the total rotifers in Fesafari in December 1956 and yet was not found in the December sample from the river.

When the numbers of species in each monthly sample are considered it is seen that there is a marked increase when the river is in flood (*cf.* Table 4 and Fig. 1). This increase in the number of species occurs in spite of a great decrease in the total number of specimens (*cf.* Fig. 2). This is an unusual state of affairs. The general rule is that when conditions are favourable for animals then the number of species is high ; when conditions are unfavourable or extreme the number of species falls although the total number of individuals may be very large (*cf.* Hesse, Allee & Schmidt, 1937, p. 30). Conditions in the River Sokoto during the floods are unfavourable for plankton production

TABLE 4

Seasonal variation in the number of species present in a sample of 100 individuals.

Month	Number of species in main channel	Number of species in Fesafari	Total number of species in both samples
March	10	6	12
April	11	7	13
May	12	6	12
June	10	7	11
July	no sample	no sample	—
August	17	no sample	—
September	13	13	18
October	9	16	17
November	15	11	20
December	16	9	19
January	no sample	6	—
February	7	3	7
March	8	4	8
April	11	5	11
May	8	5	9
June	11	no sample	—

(Holden & Green, 1960), and yet we have an increase in the number of rotifer species. This increase is due to species which normally dwell among the vegetation of the bordering swamps being washed into the river. Many of the species occurring in the plankton samples during the floods are not true plankton species.

A consistent feature of the occurrence of the flood water species is the shorter period in which they may be found in Fesafari compared with the main channel of the river. This is due to the isolation of the pool causing a lag before the rising river reaches it, washing the vegetation dwellers into the plankton of the pool. The earlier reduction in the rate of flow through the pool allows the vegetation dwelling species to settle down again earlier than they do in the river.

The effect of rate of flow on the number of species is seen when the numbers of species at any one time in the river and in Fesafari are compared (Table 4). The number of species at the two stations is similar during the floods, but once the water level falls a consistently higher number of species is found in the river. Although the number of species is generally higher in the river at any one time, the number of species found in the two stations during the whole period of sampling is roughly the same.

Seasonal changes in the composition of the rotifer population are further emphasized when the relative importance of each species is considered. Two quite distinct assemblages can be distinguished. The flood assemblage is dominated by the *Lecane* species (Table 3 and Fig. 11), with additional regularly occurring species such as *Macrochaetus collinsi* and *Trichocerca bicristata*. At other times the brachionids are the dominant group (Tables 1 and 2), with *Tetramastix opoliensis* (Fig. 14) as an important auxiliary.

CYCLOMORPHOSIS

Seasonal changes in the body shape of successive generations have been described in a number of temperate zone rotifers (Lauterborn, 1900, 1903 ; Krätzschmar, 1908, 1913 ; Wesenberg-Lund, 1908, 1930 ; Hartmann, 1918 ; Carlin, 1943 ; Ruttner-Kolisko, 1949 ; Gallagher, 1957), but there is as yet no convincing explanation of the phenomenon. Indeed, some of the accounts are inadequate even from a descriptive point of view. When the cyclomorphosis of tropical rotifers is considered there does not seem to be any detailed information available.

Two of the species from the Sokoto showed marked seasonal variation in the length of their posterior spines. A detailed numerical study has been made of this variation to find in what respects the tropical species resemble the temperate zone species and to see if they throw any light on the mechanisms underlying cyclomorphosis.

Keratella tropica

This species was studied by measuring 50 individuals from each sample in which it was present. The points used in making the measurements are shown in Fig. 15 b.

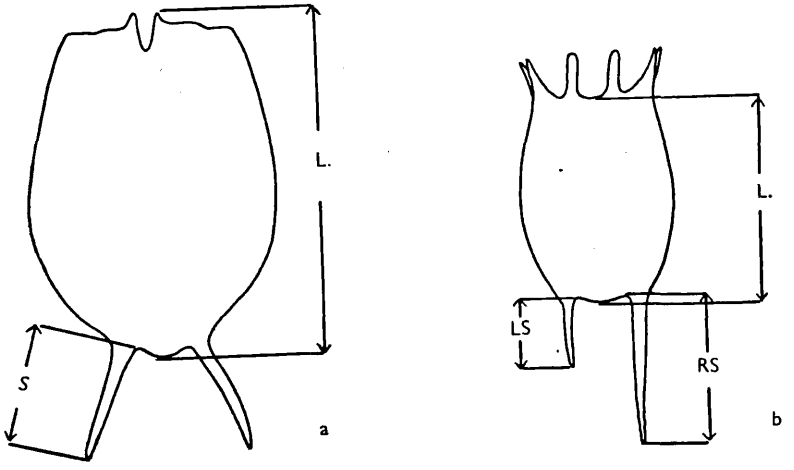

Fig. 15.—Diagram to show the measurements made on : a, *Brachionus caudatus* ; b, *Keratella tropica*. L—length of lorica ; LS—left posterior spine ; RS—right posterior spine ; S—posterior spine.

The data for 1954 extend from February 24th to April 6th ; no sample was available after the latter date. Samples were taken at weekly intervals during this period, so that they give a more detailed picture of part of the cycle which was studied at monthly intervals in later years. Fig. 16 shows that there was a steady increase in the length of the left posterior spine throughout this period, with only small changes in the length of the right posterior spine.

In a single sample taken in January 1955 none of the specimens had left posterior spines.

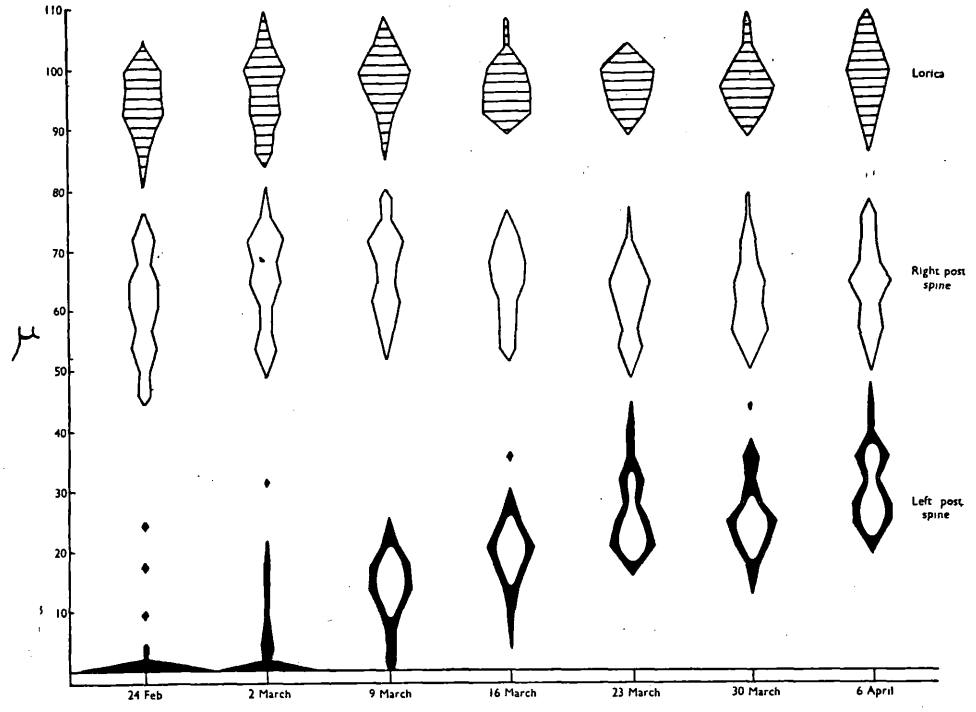

Fig. 16.—*Keratella tropica* : frequency distribution showing variation in dimensions in 1954.

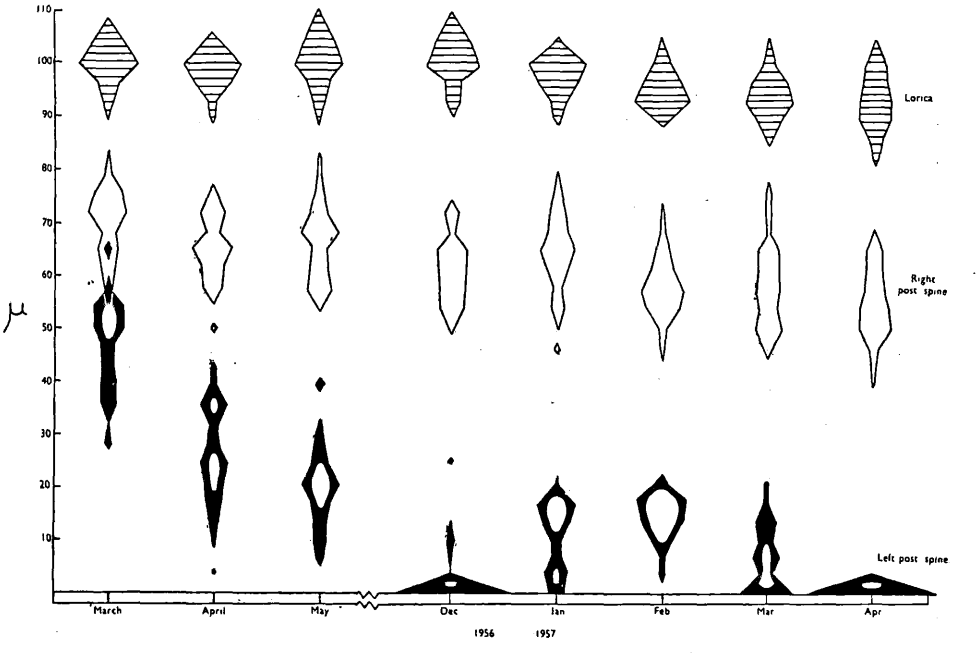

Fig. 17.—*Keratella tropica* : frequency distribution showing variation in dimensions during 1956 and 1957.

For 1956 samples taken at approximately monthly intervals are available from March to December. The species was present in the first sample, and had long left posterior spines (Fig .17). The length of the left spine decreased during April and May, and the species had disappeared from the samples by the end of June.

A reappearance, with a very short or no left posterior spine, was made in December. During the early part of 1957 a complete cycle was observed (Fig. 17). The length of the left spine increased until February then decreased to a very short length in April, and the species was absent from the May sample. The early decrease in length of the left spine and disappearance of the species in 1957 compared with other years is correlated with the rains beginning very early in 1957 ; the river in June 1957 was three times as high as in June 1956.

From the data presented above we may conclude that the normal cyclomorphosis of *Keratella tropica* in Fesafari starts with the appearance of the species as the floods subside in December. At this stage there is little or no left posterior spine. The left spine then increases in length and this is followed by a decrease, the detailed timing of which varies in different years. During the whole cycle the right posterior spine shows only small changes.

When correlations between the length of the lorica and the lengths of the spines in each sample are calculated some remarkable features are found (Table 5). The length of the right posterior spine is always positively correlated with the length of the lorica, but the left spine often shows no correlation with the length of the lorica, and in one sample a negative correlation was found. In yet other samples significant positive correlations were found.

The explanation of this bewildering array of correlation coefficients lies in the nature of the samples and the age composition of the population at the

TABLE 5

Seasonal variation in dimensions of *Keratella tropica*.

Date	Mean l. of lorica	Mean l. of left post. spine	Mean l. of right post. spine	Correlation (r) between lorica and left spine	Correlation (r) between lorica and right spine
	μ	μ	μ		
24 Feb 1954	94·74	1·18	60·74	0·06*	0·45
2 Mar 1954	95·88	3·12	65·30	0·31	0·68
9 Mar 1954	98·24	13·6	66·86	0·13*	0·66
16 Mar 1954	96·44	20·3	65·06	0·10*	0·65
23 Mar 1954	97·06	25·8	62·08	0·46	0·74
30 Mar 1954	97·20	25·5	62·78	0·26*	0·66
6 Apr 1954	99·02	30·5	64·48	0·12*	0·83
6 Jan 1955	95·58	0·0	54·46	0·0	0·28
23 Mar 1956	100·64	47·1	69·86	0·89	0·50
18 Apr 1956	99·34	26·46	65·08	0·36	0·61
7 May 1956	100·50	20·36	65·80	0·77	0·63
8 Dec 1956	100·90	1·73	61·17	—0·23*	0'33
8 Jan 1957	97·98	10·98	64·44	—0·30	0·51
3 Feb 1957	95·18	15·24	58·02	0·55	0·47
7 Mar 1957	94·44	6·02	58·18	0·11*	0·67
5 Apr 1957	93·60	0·28	54·88	0·15*	0·69

* Values not significantly different from 0 (P=0·05).

time of sampling. Once a rotifer has become fully grown it cannot, save by accident and mutilation, alter the length of its spines, so that the important period during which spine length is determined lies early in the animal's life, perhaps even as far back as the period of maturation of the oocyte in the maternal ovary. At any one time the population will contain old individuals which have been mature for a week or two and whose spines were determined under different conditions from those of younger individuals in the same sample. When the population as a whole is showing rapid changes in the length of the left spine then any correlation between the length of the left spine and the length of the lorica can be expected to be small or non-existent. When the population is not showing rapid changes in the length of the left spine there is a chance for the population to become more homogeneous, and a positive correlation can develop. This is of course based on the assumption that in constant conditions the left spine would be positively correlated with the length of the lorica.

An examination of Table 5 confirms that breakdown of correlation between the length of the left spine and the length of the lorica is associated with periods of rapid change in the length of the left spine.

The lack of correlation on 24th February, 1954, was caused by most of the specimens lacking a left spine. As small left spines appeared a low positive correlation developed (2nd March, 1954), but this broke down when the left spine increased in length more rapidly (9th March, 1954). A positive correlation reappeared when the rate of change of the left spine began to slow down (23rd March, 1954). The breakdown of correlation on 30th March, 1954, preceded a further increase in the length of the left spine (6th April, 1954).

The data for 1956 and 1957 are at monthly intervals, but the same process of interpretation seems to be applicable. In March 1956 the population was probably at its peak length of left spine ; the length fell in the next sample, so that the spine had probably finshed increasing in length and a high positive correlation might be expected. The drop in correlation in the next sample (18th April, 1956) was clearly due to the change in length of the left spine, and the increase in correlation in the May sample can be explained by the much smaller decrease in length of this spine.

The negative correlation in January 1957 is just significant at the level $P = 0{\cdot}05$. It is difficult to explain, but it may be due to chance (1 in 20 of such samples could give negative correlations of this value in the absence of true correlation in the population) or it may be due to a rapid increase in length of the left spine so that smaller, younger animals would have relatively longer spines than older, larger animals which matured when conditions stimulating growth of the left spine were not so favourable. The next appearance of a positive correlation coincides with the peak of left spine length for the year (3rd February, 1957), and is followed by a breakdown of correlation as the left spine decreases.

It is clear from the data analysed above that we cannot hope to glean much about the relative growth of the left spine from the analysis of individual samples. The right posterior spine presents better opportunities for analysis. Fig. 18 shows the relationship between right spine length and lorica length in

the samples in which the right spine was at its longest (23rd March, 1956) and shortest (5th April, 1957)—actually the shortest mean right spine length was recorded on 1st January, 1955, but in this sample the correlation with lorica

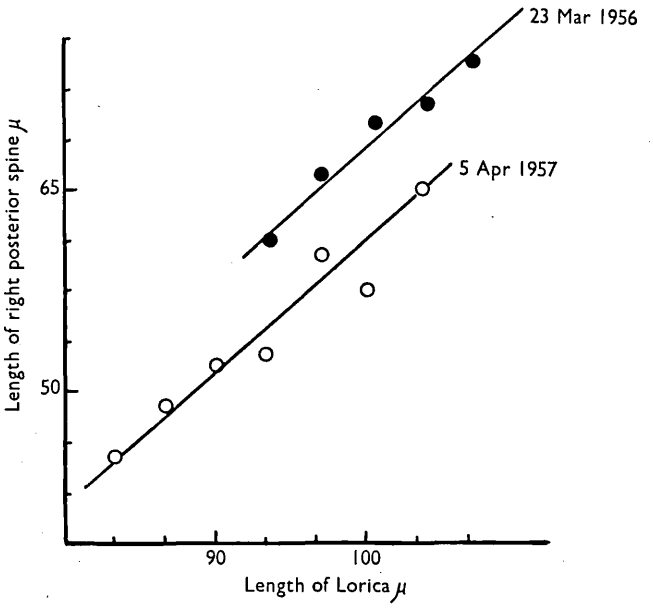

Fig. 18.—*Keratella tropica* : relation between length of lorica and length of right posterior spine. The points plotted are means of a varying number of animals (see frequency diagrams, Fig. 17).

length was unaccountably low so that the sample was not so favourable for analysis. The sample used for analysis does not differ significantly in its mean length of right spine from the lowest value, and if the data for 1st January, 1955, were plotted on Fig. 18 the points would lie around the lower line, but would be more scattered than those actually plotted. An interesting feature of this figure is that the two lines do not differ in slope, a fact which has been confirmed a comparison of the regression coefficients. The two lines do differ significantly ($P < 0{\cdot}0001$) in their heights and points of intercept of the axis. All the other samples have regression lines which lie between the two shown in Fig. 18.

Brachionus caudatus

The variation in form of this species was studied by measuring 100 specimens from each sample in which sufficient individuals were present ; only 30 were measured from the December 1956 sample. Only one of the posterior spines of each individual was measured because in most the spines were of equal length. If one spine was noticeably shorter than the other then the longer spine was measured.

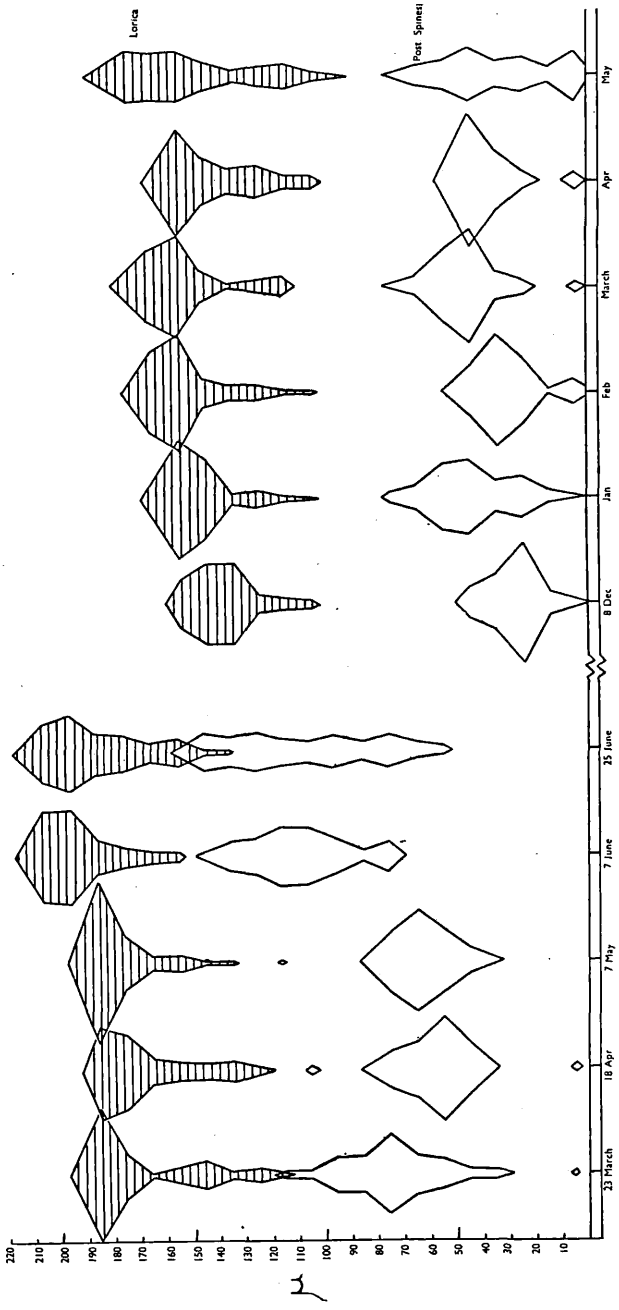

Fig. 19.—*Brachionus caudatus* : frequency distribution showing variation in dimensions during 1956 and 1957.

Fig. 19 shows that the length of the spines can vary enormously, but one never finds the majority of the population without spines ; individuals with very short or no spines are always in a distinct minority. Seasonal variation in spine length does not show as clear a cycle as that found with the left

posterior spine of *Keratella tropica*. There is a clear correlation between the length of the lorica and the length of the spines. The seasonal data show this fairly clearly, the only obvious exception being the February 1957 sample

TABLE 6

Seasonal variation in dimensions of *Brachionus caudatus*.

Date	Mean length of lorica μ	Mean length of posterior spines μ	Correlation (r) between lorica and posterior spines
23 Mar 1956	171·9	72·5	0·89
18 Apr 1956	167·5	56·7	0·70
7 May 1956	179·3	63·2	0·55
7 June 1956	194·1	109·0	0·69
25 June 1956	185·9	109·1	0·86
8 Dec 1956	140·0	28·3	0·48
8 Jan 1957	149·3	44·2	0·63
3 Feb 1957	154·2	31·8	0·47
7 Mar 1957	154·1	45·6	0·69
5 Apr 1957	144·1	39·8	0·65
17 May 1957	153·5	38·7	0·56

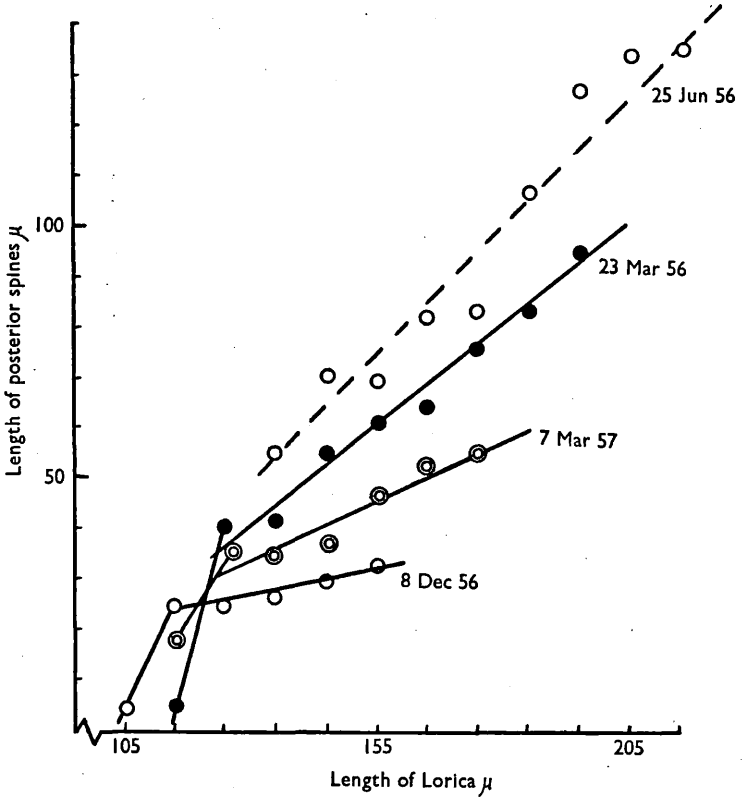

Fig. 20.—*Brachionus caudatus* : relation between length of lorica and lengths of posterior spines (see p. 519 for explanation).

when the mean lorica length increased slightly while the mean spine length decreased. Within each sample of 100 specimens the correlation between spine length and lorica length is generally strong as shown in Table 6.

When the relationship between spine length and lorica length is analysed it is found that the relationship varies with the mean length of the lorica. When the mean lorica length of a sample is high the spines are longer than when the mean lorica length is low ; the slope of the relationship is altered (Fig. 20) so that the smaller animals from a sample with a high mean lorica length have longer spines than animals of the same lorica length from samples with a low mean lorica length. At high lorical lengths the relationship is approximately linear, with the lines of all the samples converging back towards a point at which the lorica length would be about 105 μ and the spine length about 20 μ. This does not, however, happen. The lowermost points of three of the lines in Fig. 20 are much lower than one would expect from the trends of the other points. This may indicate that a critical size must be reached before the spines begin to grow rapidly ; all the individuals with no posterior spines had loricas less than 115 μ long. Once the critical size has been passed the length of the posterior spines varies with the mean size of the animals in the sample.

In Fig. 20 the lines have been fitted to the data by eye, but regression coefficients (b yx) have been calculated and compared ; these indicate that the slopes of the lines differ significantly at a level $P < 0{\cdot}001$.

DISCUSSION

The cyclomorphosis of *Keratella tropica* shows a distinct addition to the length of the left posterior spine in the early part of the season and a decrease in the length of this spine at the end of the season. This is similar to the cyclomorphosis of the European species *Keratella valga* as described by Hartmann (1918), whose results have been questioned by Ahlstrom (1943). In view of the results presented here concerning *K. tropica* there seems no valid reason for questioning the accuracy of Hartmann's observations.

When *Keratella tropica* and *Brachionus caudatus* are compared both similarities and differences are found. The two species are similar in that both had much longer spines in 1956 than in 1957. Clearly the environment can influence the growth of spines in both species in a similar manner. But the differences are great, particularly in relation to the changes in length of the left posterior spine of *K. tropica*. It may be that the right spine of this species is peculiar in that it is fairly constant in length and does not show the large variations in length which are found in the left spine and in both the posterior spines of *B. caudatus*. The two species also differ in their relationship between spine length and seasonal occurrence. Although both had longer spines in 1956, *K. tropica* decreased the length of its left posterior spine and disappeared from the plankton by the end of June, while *B. caudatus* persisted throughout June and increased the lengths of its posterior spines.

The problems of spine length in *Keratella tropica* are more complex than in *Brachionus caudatus*. The large variation in the left spine of *K. tropica* while the right spine remains fairly constant in length can only mean that

there must be some advantage in retaining the right spine. It also poses a problem in relation to the causal mechanism of cyclomorphosis. The environment probably controls the variations of the left spine, but internal factors must surely be responsible for the retention of the right spine. Fig. 18 shows that the length of the right spine varies with the length of the lorica ; this can be interpreted as showing that a larger body produces more of the internal factor influencing spine growth. The difference between the right and the left spine implies unequal distribution of this factor. The data concerning *B. caudatus* may also be related to an internal factor influencing spine growth. Here too the spines vary with the length of the lorica and seem to be influenced by the growth rate of the rotifer. The greater spine lengths achieved by animals which are growing more rapidly may be attributed to a more rapid production of the internal factor controlling spine growth.

It seems reasonable to assume that a high rate of growth will occur when the rotifer is well fed. If this is so then it is difficult to reconcile the relation between spine length and lorica length with the results of Büchner, Mulzer & Rauh (1957) who maintain that poor feeding enhances spine growth in *Keratella cochlearis* and *Brachionus calyciflorus* (= *pala*). They did not, however, actually measure the spines of the animals they studied, and it is uncertain whether their control of feeding was adequate to eliminate chance results due to bacteria, which might form suitable food for rotifers.

Among the possible factors which may control the growth of rotifers both the amount and the nature of the food available should be considered. It is known for instance that some species of algae are better than others of the same genus as rotifer food (Pourriot, 1957). Oxygen, pH and temperature may also be involved. Dissolved oxygen is known to be a factor in the complex controlling the growth of other planktonic animals such as *Daphnia* (Green, 1956). Hydrogen ion concentration is well known to be of importance in governing the distribution of rotifer species ; it seems likely that there is an optimum pH for the growth of particular species. An increase in temperature would accelerate the rate of growth, but it may also accelerate the onset of maturity so that the rotifer may become mature at a smaller body size than if it grew more slowly at a lower temperature. It would be possible for the effect of an increase in temperature on spine growth to vary according to whether it accelerated maturation more or less in relation to growth.

It is worth noting that the decrease in length of the left spine of *K. tropica*, from March to May 1956, was undergone while the temperature remained consistently between 26 and 29°C. The complete cycle of increase and decrease, from December 1956 to April 1957, happened when the temperature first of all fell from 21°C. in December to 16°C. in January and then rose steadily to 27°C. at the beginning of April. There is no obvious correlation between the cycle of *K. tropica* and the temperature of its environment.

It is clear from the data presented above that the factors controlling the variation in spine length in rotifers are likely to be complex, and that simple theories, such as those of Wesenberg-Lund (1900, 1908) and Ostwald (1902) cannot be applied. There is no simple relation between spine length and temperature such as would be demanded by these theories. Investigations

in Europe have shown that the posterior spines of *Keratella quadrata* decrease in length during the warmer months (*cf.* Carlin, 1943), so that in this species at least there is no question of increased spine length compensating for a decrease in the viscosity of water as temperature rises.

Gallagher (1957) claims to have established a positive correlation between temperature and posterior spine length in a North American population of *Keratella cochlearis*. This claim deserves scrutiny because it is in direct opposition to the classical work of Lauterborn (1903), who found a decrease in the length of the posterior spine of this species during the summer months in Germany. Gallagher's correlation was calculated between mean air temperature of the twenty-four hours in which his collections were made and the mean spine length of a varying number of animals. The use of mean spine length in this way is not a valid statistical procedure, since it automatically removes much of the variance which a correlation coefficient is intended to estimate. Even if the use of mean spine length was acceptable the significance of some of the means given by Gallagher is open to question. The range of mean spine length which he gives is small (22–36 μ), and the lowest of these is based on but four individuals. The standard error of this mean is given as 8·51, which, unless it is a typographical error, is so great as to exclude this particular mean from having any significance in relation to the rest of his data. There are significant differences between some of his mean values, perhaps the most striking of these is the mean spine length of 32 μ when the mean air temperature was 7·2°C. which is significantly higher than the mean spine length of 23 μ which was recorded when the mean air temperature was 9·4°C. Discrepancies such as these are sufficient to invalidate Gallagher's conclusion that there is a positive correlation between air temperature and spine length.

As rotifers live in water, and water temperatures are generally less variable than air temperatures, one might more justifiably seek a correlation between water temperature and spine length, but Gallagher admits that if the correlation is calculated between water temperature and spine length then there is no significant correlation at a level of $P = 0{\cdot}05$.

It would also seem to be desirable to consider spine length in relation to the size of the animal bearing the spine ; Gallagher provides no data of this type.

The most detailed and thorough work on the cyclomorphosis of rotifers is that of Carlin (1943) who showed that *Kellicottia longispina*, *Keratella quadrata* and *Keratella stipitata* (= *cochlearis*) decrease the length of their spines when the water temperature is at its highest. Carlin reconciles these findings with ideas of flotation by considering the turbulence of the water. In the warmer weather the viscosity of the water will be lowered so that turbulence might be expected to be greater for a given amount of energy expended in producing a current (for instance by wind or inflow into a pond). An increase in turbulence would tend to keep the rotifers afloat, so that their need for long processes would be reduced. In winter the viscosity of the water will be higher and turbulence will be weaker, lending less support to the rotifers so that their spines need to be longer. This idea may be capable of wider application, but

it is a teleological explanation offering only a partial explanation of cyclomorphosis.

Any future approach to the problem of cyclomorphosis in rotifers must consider the length of the spines in relation to the body size of the rotifer. Margalef (1947) took a step in this direction when he used logarithmic scales to plot the spine lengths and lorica lengths of *Keratella quadrata*, as measured by various other authors, and found that the relationship could be expressed as a straight line. This indicates that the longer spines are a result of allometric growth. It would be of interest to plot similar data for *K. quadrata* on a seasonal basis to see if there is any parallel to the relationships described on p. 519 for *Brachionus caudatus*.

It may be unwise to seek a unifying explanation of variation in all the various protuberances in diverse families and species of rotifers. The specific line of approach seems to be the most profitable at present. Only when there is sufficient information available concerning the relationship between environmental factors and seasonal forms of a considerable range of species will it be worthwhile examining the possibility of a general explanation.

The two species described in this paper illustrate the difficulties that any general explanation will encounter. It will have to explain why two species in the same habitat, both having two posterior spines, differ in that one varies both its spines in a similar manner, while the other varies the left spine without much variation in the right spine. Two lines for future study may be suggested. First a study of the relationship between growth and the development of spines to determine the environmental complex of factors controlling the relationship. The second line of approach might deal with the physical effects of variation in length of spines on the locomotion of rotifers, particularly in relation to the turbulence of water. This might throw some light on the significance of cyclomorphosis as an ecological and evolutionary phenomenon

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ZOOPLANKTON OF THE RIVER SOKOTO
THE FRESHWATER MEDUSA *LIMNOCNIDA*

BY

J. GREEN

Bedford College, University of London

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(With 3 figures in the text)

The freshwater medusa *Limnocnida victoriae* was found from December 1956 to April 1957 in a pool which becomes separated from the main channel of the River Sokoto during the dry season. The medusae are described and the importance of the number and size of statocysts as a systematic character within the genus is discussed. A map showing the distribution of *Limnocnida* in Africa is given.

CONTENTS

INTRODUCTION

The present paper is concerned with a medusa which appeared in the pool Fesafari which becomes separated from the main channel of the river during the dry season. A general account of the hydrology of this North Nigerian river and of the main fluctuations in the total plankton is given by Holden & Green (1960).

The history of the discovery of *Limnocnida* has been told many times (e.g. Günther, 1893 ; Moore, 1899 ; Brown, 1908 ; Schouteden, 1939 ; Kramp, 1954) and recently a detailed monograph of the genus has been given by Bouillon (1956–7). The last author considers that there are three African species in the genus, while other authors (Leloup, 1951 ; Kramp, 1954) have been inclined to the view that all the African forms belong to one species. The specimens from Fesafari have been examined to see if they support either of these opposing views.

TEMPORAL OCCURRENCE AND SIZE

Although the area was visited and plankton samples were taken by Mr Holden at intervals throughout the various years there were no signs of the medusa in 1954, 1955 or 1956. The first large specimens were found in February 1957, but a careful search of plankton samples taken in January of that year and December 1956 revealed the presence of very small medusae. The numbers of medusae found in a standard plankton tow over a distance of

300 metres using a net with a mouth diameter of 30·5 cm. were as follows : December, 2 ; January, 7 ; February, 35 ; March, 21 ; April, 7. There was a clear peak of abundance in February, followed by a gradual decline. Fig. 1 shows that production of small medusae persisted into March. No small specimens were found in April, and by May all the medusae had disappeared.

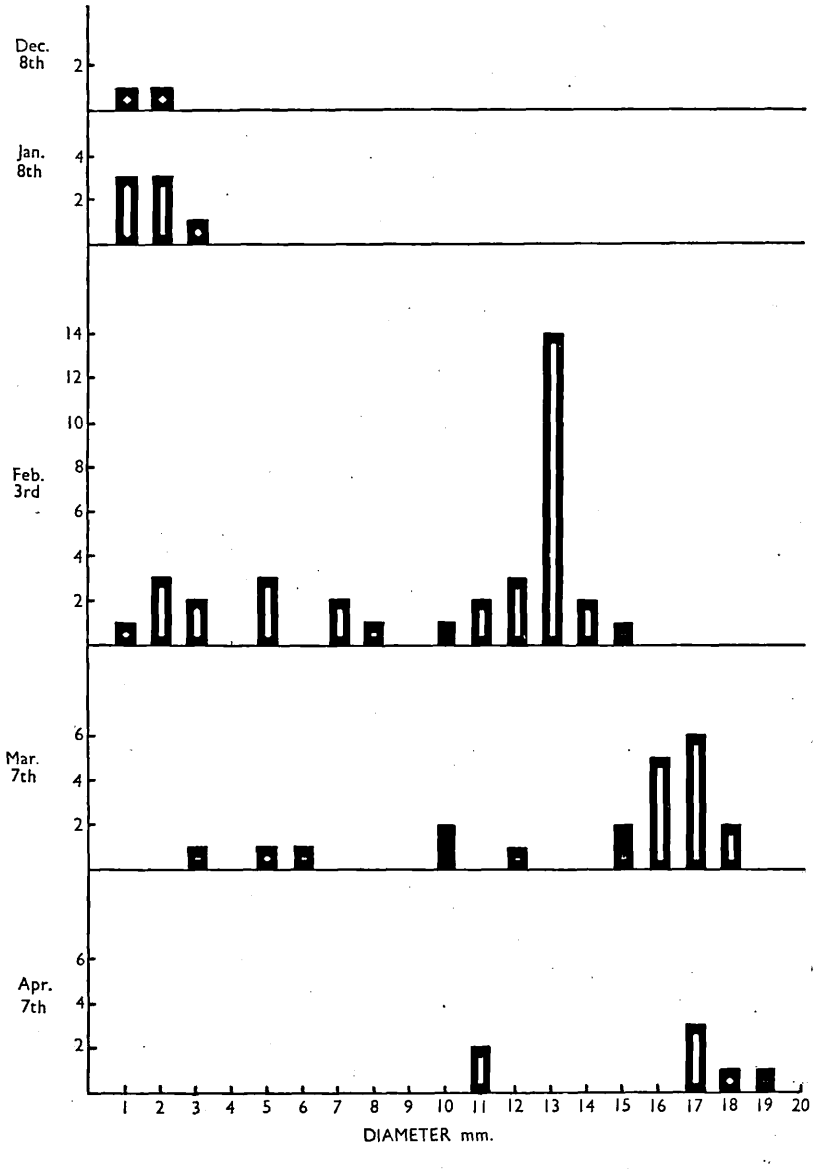

Fig. 1.—Diagram showing the frequencies of size groups of medusae of *Limnocnida* during the period of occurrence in the Sokoto river system.

Some of the larger specimens collected in February had well-developed gonads and so were presumably capable of breeding. The largest specimens

from Fesafari are distinctly smaller than those recorded from elsewhere. Diameters up to 25 mm. have been recorded from other parts of Africa, but the largest specimens from Fesafari only reach a diameter of 19 mm. There were no signs of asexual reproduction by budding around the edge of the manubrium, as is found in specimens from Lake Tanganyika.

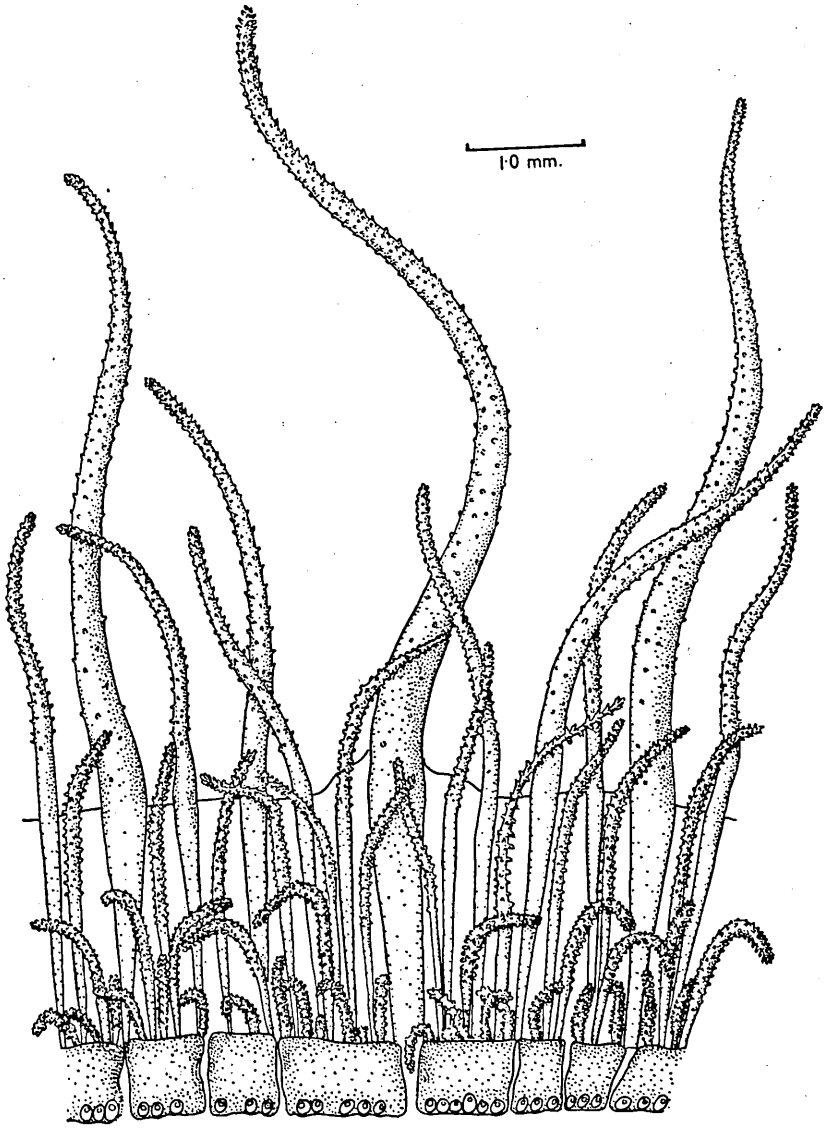

Fig. 2.—Part of the margin of a medusa of *Limnocnida* from the Sokoto river system. (Scale 1·0 mm).

The observed occurrence in Fesafari contrasts with that known in other areas. Medusae have been found in all months in Lake Tanganyika (see table on p. 267 in Bouillon, 1956–7), and in Lake Mohasi sexually reproducing individuals are found from mid-April to September (Bouillon, 1954).

DESCRIPTION OF THE MEDUSA

The characters which have been used to separate species of *Limnocnida* have been mainly the number and size of the tentacles and the marginal statocysts which lie in a row at the inner margin of the ring of nematocysts.

In the specimens from Fesafari there are about 400 tentacles on an individual with a diameter of 15 mm. The tentacles are arranged in a fairly definite series (Fig. 2). The largest tentacles are about 1 cm. in length. The number of tentacles increases as the animal grows. Bouillon (1955) notes that when freshly budded from the hydroid the medusa has a diameter of 0·4 mm. and has eight tentacles. The smallest specimen found in the plankton from Fesafari had a diameter of 0·6 mm. and had eight tentacles. A specimen with a diameter of 1·0 mm. had eight tentacles and the rudiments of eight more, while a specimen with a diameter of 1·4 mm. had eight long tentacles, eight shorter tentacles and the rudiments of 16 more ; none of these small specimens had statocysts. A specimen with a diameter of 2·5 mm. had a total of 32 tentacles, with the rudiments of 32 more, and 16 statocysts were present. Edney (1939) noted that the first eight statocysts appeared in Rhodesian specimens when the diameter was about 2·0 mm.

The size of the statocysts has been thought to be of systematic importance. Most authors give a single diameter, as if the statocysts were always spherical, but in the specimens from Fesafari most of the statocysts are egg shaped. The range of size of statocysts from a 13 mm. diameter specimen can be judged from the following series of measurements made on 10 successive neighbouring statocysts (measurements in μ) :

130×104, 91×65, 130×117, 78×78, 104×91, 65×65, 130×130, 104×91, 95×78, and 104×91.

The average size is thus somewhat larger than that found in specimens from Lake Tanganyika, where Bouillon (1956–7) found an average diameter of 75 μ, and Arnold & Boulenger (1915) give a range of diameters from 70–90 μ. However, Bouillon has pointed out that some statocysts in Tanganyika specimens reach a diameter of 142 μ. It would seem that not much importance can be attached to the size of the statocysts.

The number of statocysts in relation to the number of tentacles might be of systematic importance. In specimens from Fesafari the ratio is about 0·6.

Bouillon (1956–7) divides the African species of *Limnocnida* as follows :

L. tanganyicae Günther : 300 tentacles, with maximum length of 2·0 cm., ratio of statocysts to tentacles 0·80, medusae produce buds.

L. victoriae Günther : 400 tentacles, with maximum length of 1·6 cm., ratio of statocysts to tentacles 0·93, medusae do not bud.

L. congoensis Bouillon : 800 tentacles, with maximum length of 1·2 cm., ratio of statocysts to tentacles 0·48, medusae do not bud.

On the basis of the number of tentacles and the lack of budding the medusae from Fesafari clearly belong to *L. victoriae*. The tentacles do not, however,

reach a length greater than 1·0 cm., though this may be due to shrinkage on fixation. The ratio of the number of statocysts to the number of tentacles is distinctly low. This may be due to some extent to the generally small size of the Sokoto specimens. Browne (1908), in his study of two specimens from the Niger delta, noted that the larger specimen (18 mm. diam.) had an almost continuous row of statocysts all touching each other, while the smaller specimen (11 mm. diam.) had a much less crowded row with small gaps.

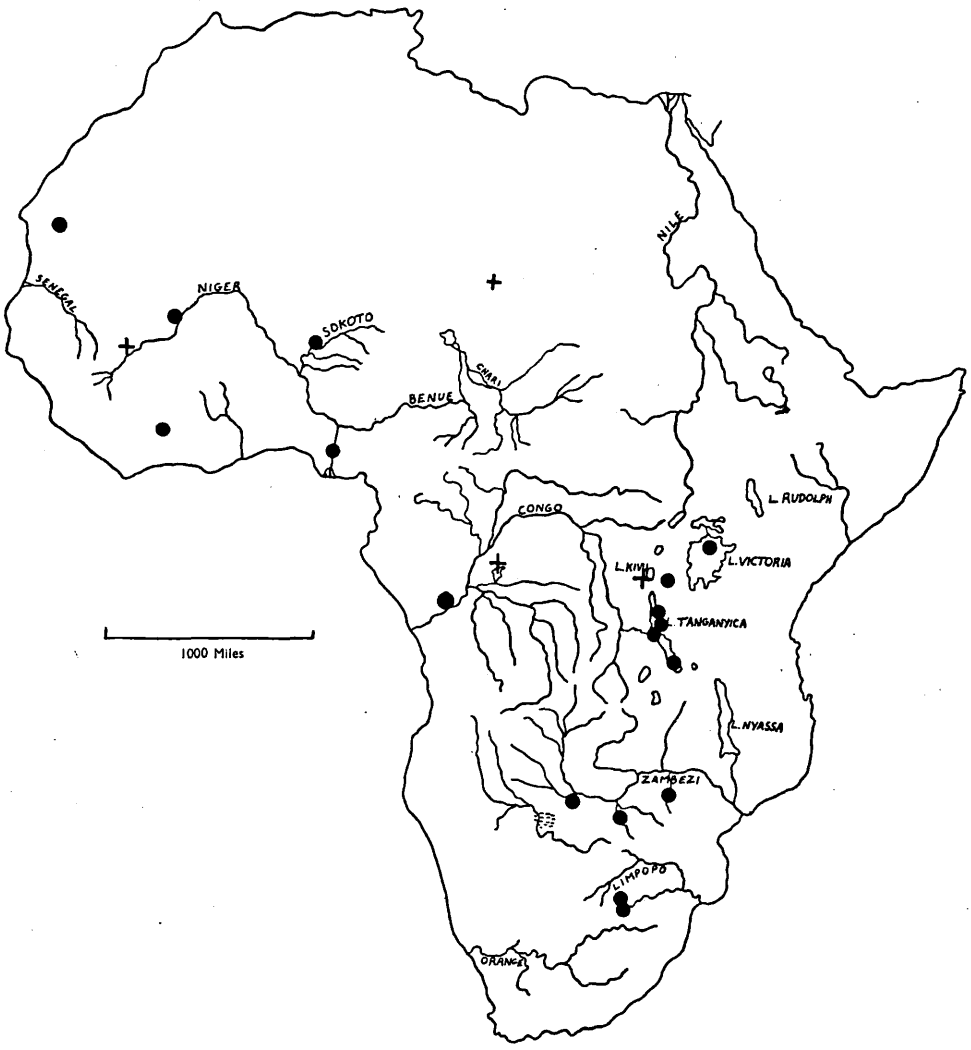

Fig. 3.—The distribution of *Limnocnida* in Africa. Dots indicate localities where medusae have been collected. Crosses indicate places where medusae have been observed but not collected.

The specimens from Fesafari do not invalidate the specific categories set up by Bouillon, but they do indicate that the ratio of statocysts to tentacles is

not a very reliable character, and certainly not one of sufficient accuracy to warrant quoting to the second decimal place.

DISTRIBUTION OF *LIMNOCNIDA* IN AFRICA

On Bouillon's map (1956–7, p. 469) only a single record from West Africa is shown ; this is the record by Daget (1950) from the upper reaches of the Niger. In fact there are at least three other authentic records of *Limnocnida* from West Africa. Browne (1908) gave a detailed description of two specimens collected by J. S. Budgett from the Niger delta, and Dekeyser (1955) gives details of the other occurrences. A revised map of the distribution of *Limnocnida* in Africa is given in Fig. 3. Of the three species recognized by Bouillon (1956–7), *L. tanganyicae* is found only in Lake Tanganyika, *L. congoensis* is found in the River Congo, and *L. victoriae* occurs in all the other localities.

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(*Reprinted from Nature, Vol.* 189, *No.* 4760, *pp.* 227–228, *January* 21, 1961)

Biliverdin in the Eyes of *Polyphemus pediculus* (L.) (Crustacea, Cladocera)

THE large median compound eye of the Cladocera begins its development as two separate rudiments which enlarge gradually and fuse in the mid-line. In the majority of Cladocera these rudiments are red in colour and gradually darken so that they are black just before fusion. In embryos of *Polyphemus pediculus* the eye rudiments are green in colour, and when treated with yellow nitric acid they give a positive Gmelin reaction, indicating the presence of a bile pigment.

A large sample of female *Polyphemus*, containing numerous embryos, was first extracted with acetone to remove carotenoids and then extracted with acid methanol (5 per cent hydrochloric acid), giving a deep olive-green solution. When this was diluted with water and shaken with chloroform the chloroform phase became blue in colour while the acid methanol phase became orange-brown. The chloroform phase was washed with dilute sodium bicarbonate, then with water and evaporated to dryness. When the pigment was redissolved in acid methanol the absorption spectrum showed a sharp peak at 374 mμ and a lower, more rounded, peak at 685 mμ. These are close to the absorption maxima of biliverdin hydrochloride ; the small differences can be attributed to the presence of other substances in the extract.

The identity of the green pigment as biliverdin was confirmed by the addition of zinc acetate and a little iodine to a methanol solution in the presence of a trace of ammonia. The result was a green solution with a brilliant pink fluorescence in ultra-violet light, and a sharp absorption band at 635 mμ showing that the pigment had proto not meso side-chains.

The presence of biliverdin in the eyes of adults was demonstrated by selecting females without embryos and extracting with methanol. The addition of zinc acetate and iodine produced the same results as with the larger sample.

It has not been possible to demonstrate a Gmelin reaction in the eyes of adult *Polyphemus* because of the presence of large amounts of a dark masking pigment. This pigment is also extracted with acid methanol, but remains in the methanol phase when

it is diluted and shaken with chloroform. Further studies are being made of this and similar pigments in other Cladocera; meanwhile, it is possible to say that they are similar in many respects to ommochromes, being insoluble in most organic solvents, but easily soluble in acids, and showing reversible changes in colour according to whether they are oxidized or reduced.

Biliverdin is rare in the Crustacea; it has been found only in the digestive gland of the crayfish *Cambarus*[1], and in the roots of certain parasitic cirripedes[2]. The present record of biliverdin in *Polyphemus* adds another crustacean order to the few known to include bile pigments in their metabolism[3]. It is interesting in this connexion to note that, although searched for, no bile pigments have been found in the cladoceran *Daphnia*[4].

J. GREEN

Zoology Department, Bedford College,
Regent's Park, London, N.W.1.

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A NEW SPECIES OF *SABELLACHERES* (CRUSTACEA : COPEPODA) PARASITIC ON THE FAN WORM *EUDISTYLIA POLYMORPHA* (JOHNSON)

BY

J. GREEN

Bedford College, University of London

[Accepted 8th November, 1960]

(With 14 figures in the text)

The female and nauplius of *Sabellacheres dalesi* sp. n. are described and figured. A key to the genera and species of the Gastrodelphyidae is given.

CONTENTS

INTRODUCTION

The remarkable copepod described below was discovered by Dr R. P. Dales during his researches into the physiology of fan worms. It gives me great pleasure to name the species after him.

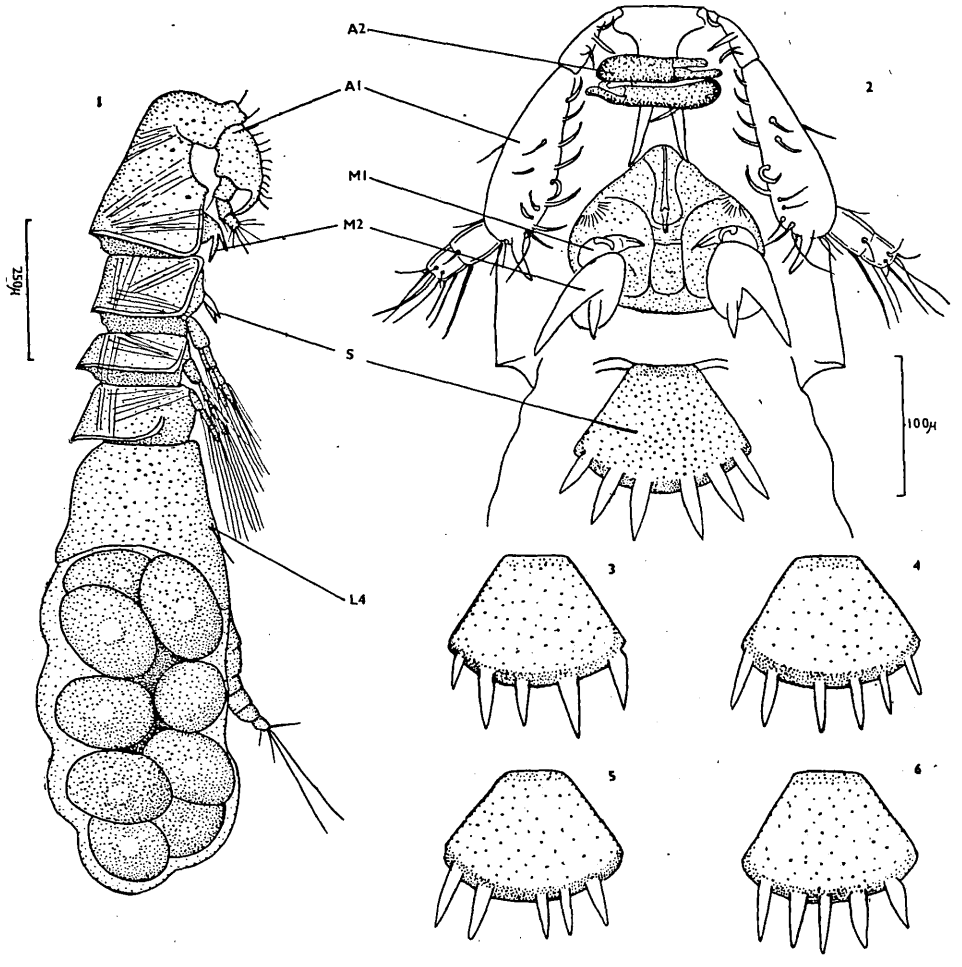
SABELLACHERES DALESI sp. n.

Female. The general form of the body is shown in Fig. 1. The total length including the brood sac is about 1·4 mm. There is a distinct cephalon bearing a pair of large antennules antero-laterally. The rostrum forms the antero-ventral part of the cephalon and can be separated as a distinct sclerite prolonged posteriorly in the form of two large spines (Fig. 9).

The antennule consists of five podomeres, the two basal ones being enlarged and flattened. Some of the setae on these basal podomeres have the outer layer of the podomere cuticle continued over the setal surfaces, so that a curious flanged appearance results when viewed by transmitted light. The second podomere is prolonged in the form of a large spine distally (Fig. 8). Thickenings of the integument near the base of this spine give the appearance of a line across the base, but the spine is not articulated. When viewed from the ventral surface the enlarged second podomere obscures the third podomere (Fig. 2), but this is easily seen in lateral view (Figs. 1 & 8). The three distal podomeres are cylindrical in form.

The antennae lie crosswise between the bases of the antennules (Fig. 2) ; this is clearly an adaptation to grip the filaments of the host crown. There

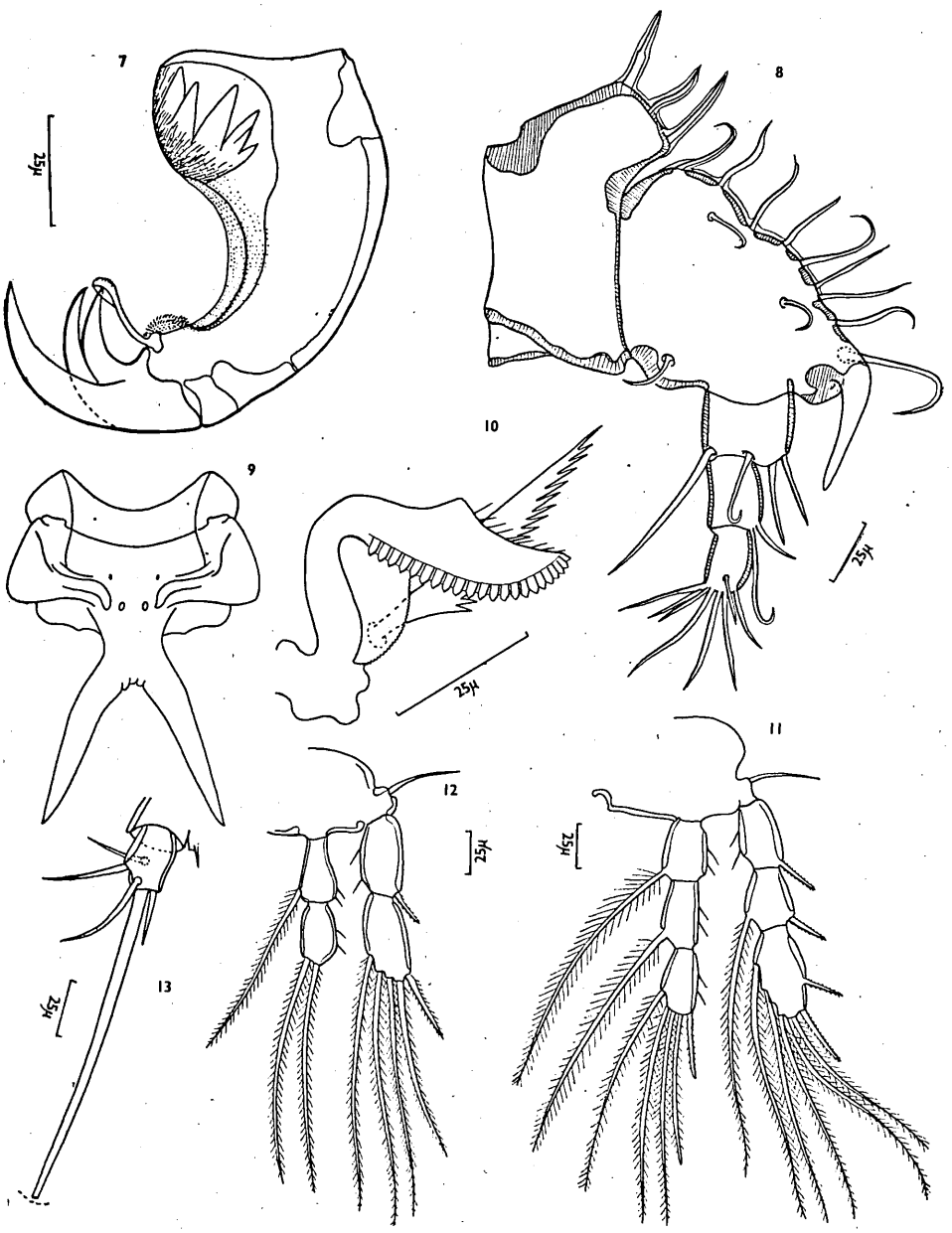
are four podomeres forming each antenna ; the last one bears a plate with a strongly serrate edge near the base and three heavy claws at the tip. A curious spoon-shaped seta arises near the base of the most proximal of the three claws and caps its tip (Fig. 7). At the base of the spoon-shaped seta is a pad bearing minute spinules.



Figs. 1–6—*Sabellacheres dalesi* sp. n. 1, lateral view of female. 2, ventral view of the anterior end of female. A1—antennule, A2—antenna, M1—first maxilla, M2—second maxilla, S—sternal plate, L4—fourth leg. 3–6, variations in the spines on the sternal plate.

The mandible (Fig. 10) has its main body bent into the form of an S. The basal portion is expanded into a thin flange with a very finely milled edge. A large spine arises from the basal portion and projects diagonally forwards towards the mouth. The medial edge of this spine bears about 18 large serrations. The main body of the mandible continues obliquely in a posterior direction in the form of a tongue-shaped process bearing about twenty-two distinct teeth along one border and a graduated series of small hairs along

the other. There is a large triangular projection near the middle of the border bearing the hairs. In Fig. 10 the toothed process is shown as being flat, but when in its normal state inside the mouth the distal tip curves ventrally.



Figs. 7–13—*Sabellacheres dalesi* sp. n. 7, apex of antenna. 8, left antennule, medial view. 9, rostrum separated from the rest of the exoskeleton. 10, left mandible, dorsal view, somewhat flattened. 11, first swimming leg. 12, third swimming leg. 13, right caudal ramus, ventral view.

The first maxilla bears two medially directed processes and a longer ventrally directed spine.

The second maxilla is larger than the first and is prolonged in the form of two posteriorly directed spines, the lateral one being much larger than the medial one.

No maxilliped was found.

Behind the cephalon are three apparent segments, the first of which bears two distinct structures. The anterior structure takes the form of a plate bearing five or six stout spines on the posterior border. There is some variation in the size of these spines, and they do not always form an even series (Figs. 3–6). The homology of this plate is uncertain, but there are distinct sternal articulations at the antero-lateral corners, and large muscles run to various parts of the plate. In Figs. 1 and 2 the noncommital label ' sternal plate ' has been applied. In List's (1890) descriptions of *Gastrodelphys clausi* and *G. myxicolae* the term ' Bauchwirbelkörper ' is applied to this structure.

The second structure on the first apparent free segment is the first pair of swimming legs (Fig. 11). The second and third apparent segments each bear a pair of swimming legs. The second swimming legs are similar in all respects to the first except that the terminal podomere of the exopodite bears an extra plumose seta. The third swimming legs have only two podomeres in both the exopodite and the endopodite (Fig. 12). The last or 4th leg is represented by a small papilla bearing a single seta.

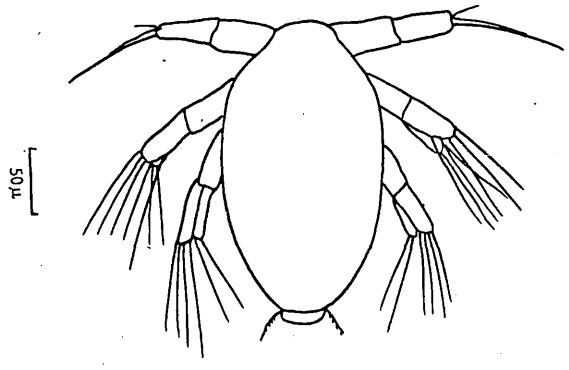


Fig. 14—*Sabellacheres dalesi* sp. n. Newly hatched nauplius, dorsal view.

The brood sac projects dorsally beyond the tip of the abdomen, which is small and consists of four segments. Each caudal ramus (Fig. 13) bears five setae, one of which reaches a length of 280 μ while the others are all much shorter.

Male. Unknown.

Nauplius. The newly hatched nauplius is shown in Fig. 14. The total length of the body, excluding setae is about 220 μ. The antennules are uniramous with one long and one short terminal seta. The antennae are biramous with five setae on the exopodite and two setae on the endopodite ; the mandibles are similar in structure but have only four setae on the exopodite. The furcal setae are feathered along their medial borders.

Occurrence. Five females were found by Dr R. P. Dales on the branchial crowns of *Eudistylia polymorpha* Johnson (Polychaeta, Sabellidae) which had been sent to him from a collecting station three miles North of Point Dume at the western end of Zuma Beach, near Los Angeles, California. The worms were collected from between rocks in water varying in depth from 25 to 50 feet, in mid August 1959.

Dr Dales noted that the females were capable of active swimming and could detach themselves from the host crown, swim around for a short while, and then reattach themselves.

DISCUSSION

The species described above is similar in many respects to *Vermiclavella elongata* described by Markewitsch (1940) from *Myxicola*, but it is a much smaller species (total length 1·4 mm. compared with 4·6 mm. in *V. elongata*), and both the rami of the third swimming leg have only two podomeres compared with three in *V. elongata.*

The two species of *Gastrodelphys* described by List (1890) have the swimming legs greatly reduced and are presumably incapable of swimming. It seems probable that here we have a series of stages showing reduction of locomotory activity associated with the adoption of a parasitic mode of life. If we regard all the forms as belonging to one genus some problems of nomenclature become apparent. *Vermiclavella* Markewitsch 1940 would become a synonym of *Gastrodelphys* Graeffe 1883. The problem is complicated by the fact that Sars (1862) gave a brief description of a copepod parasitic on *Myxicola* and gave it the name *Sabellacheres gracilis.* A comparison of Sars's description with the figures given by Markewitsch (1940) indicates that they were looking at the same species. If all the species are congeneric the correct name would be *Sabellacheres* Sars. It seems preferable at the moment to retain *Gastrodelphys* for those species with greatly reduced swimming legs, so that the hitherto described species of the family Gastrodelphyidae can be separated as in the following key.

FAMILY GASTRODELPHYIDAE LIST 1890

The diagnosis of this family will have to be extended from that given by List to include forms with well-developed swimming legs. Antennules with five podomeres, the first two of which are flattened and enlarged. Antennae with four podomeres the last of which bears three large claws. First apparent thoracic segment bearing a sternal plate with posteriorly directed spines. Brood sac dorsal, projecting above the abdomen. Parasitic on annelids.

1 (4). Three pairs of swimming legs well developed, with at least two podomeres in both rami ..... *Sabellacheres* Sars 1862.

2 (3). Third swimming legs widely separated from the first two pairs and with three podomeres in both rami. Large species, up to 5 mm. long, parasitic on *Myxicola* ..... *S. gracilis* Sars 1862 (syn. *Vermiclavella elongata* Markewitsch 1940).

3 (2). Third swimming legs not widely separated from first two pairs and with only two podomeres in exopod and endopod. Length, including egg sac, 1·4 mm. Parasitic on *Eudistylia.* *S. dalesi* sp. n.

4 (1) Swimming legs reduced. Endopods of single podomere or absent ..... *Gastrodelphys* Graeffe 1883.

5 (6). Endopods present on first two pairs of legs. Abdomen with three segments. Parasitic on *Bispira volutacornis* ..... *G. clausi* Graeffe 1883.

6 (5). Endopods absent. Abdomen with one segment. On *Myxicola* ..... *G. myxicolae* List 1890.

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ZOOPLANKTON OF THE RIVER SOKOTO. THE CRUSTACEA

BY

J. GREEN

Bedford College, University of London

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(With 95 figures in the text)

Fifty-one species of Crustacea are recorded from plankton samples taken from the Sokoto river system. The following new species are described: *Alona holdeni, Microcyclops pseudo-anceps, Cyclops gauthieri, Physocypria minicapensis.* An account is given of the seasonal occurrence of each species in the main channel of the river and in a pool which becomes isolated from the river during the dry season. Two assemblages of Crustacea are distinguished; in the dry season *Diaphanosoma excisum, Moina dubia,* two species of *Thermocyclops* and *Tropodiaptomus laurentii* form the bulk of the plankton, but in the wet season, when the river is in flood the dominant crustaceans are members of the cladoceran genus *Alona.* The specific composition of the crustacean plankton of the Sokoto is compared with that of the Illinois and the Nile; in all three rivers there are approximately three times as many species of Cladocera as of cyclopoid copepods.

CONTENTS

INTRODUCTION

A general description of the hydrology of the River Sokoto and an account of the fluctuations of the major groups of planktonic organisms has been given by Holden & Green (1960). The dominating feature of the hydrology is the flooding of the river during the single wet season (see Fig. 1). During the floods

the pH of the river water is lowered by acid water washed out of swamps bordering the river. The alkalinity of the river water is high so that even during the floods, when rainwater causes dilution, the pH rarely falls below 7. The temperature of the river is normally high, in the region of 25°C or over, but during December and January the harmattan, a cold wind from Sahara, blows southwards and cools the river below 20°C.

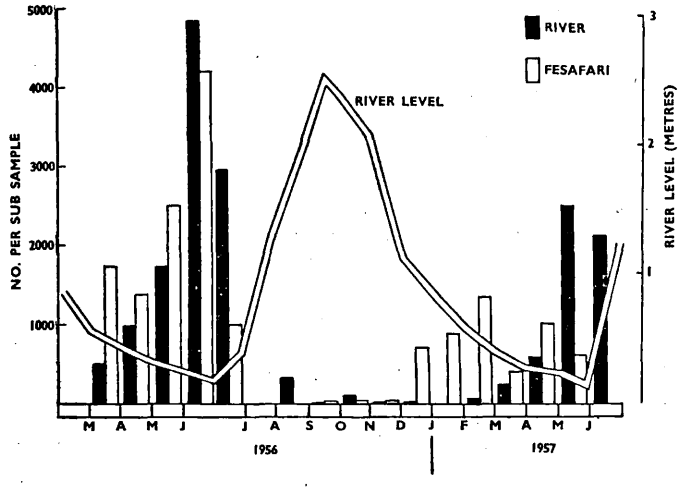


Fig. 1—Seasonal variation in total Crustacea in relation to the flood cycle of the river. Notice in this figure that two samples were taken in June 1956, but no sample was taken in July ; this also applies to the other diagrams showing seasonal occurrence. Each sub sample represents the number of individuals in approximately one third of a cubic metre.

The present paper deals with the detailed systematics of the Crustacea, and gives an account of their seasonal occurrence in the plankton. During the dry season the level of the river falls and a pool called Fesafari becomes isolated from the main channel. The abundance and seasonal occurrence of the planktonic species in this pool have been studied for comparison with the main river channel.

METHODS

The plankton was collected by Mr M. J. Holden using a fine meshed plankton net which was towed for a fixed distance just below the surface of the water. Details of the main sampling stations in the main channel and in the pool Fesafari are shown on a map in Holden & Green (1960).

The samples were preserved in 5 per cent formalin. The volume of each sample was made up to 30 ml., and after thorough mixing sub samples of 0·5 ml. were taken with a wide mouthed graduated pipette and ejected into a petri dish containing a thin film of water. The water in the petri dish was gently agitated so that the animals were evenly distributed. All the animals in the sub sample were then counted under a binocular microscope with a magnification of 30 diameters and assigned to major groups such as Cladocera, Cyclopoida, Calanoida etc. This gave a measure of the actual abundance of each group. The relative abundance of each species within each of the major

groups was estimated by another series of samples in which 100 specimens of each group were identified to specific level. In some of the samples taken during the floods animals were so scarce that 100 specimens of some of the groups could not be found and the relative abundance had to be estimated from lower numbers. The figures showing seasonal abundance were drawn using data calculated from the relative abundance of each species and the actual abundance of its group as estimated by the first series of samples.

The Cladocera were identified and counted after they had been mixed with a little lactic acid containing lignin pink. Preparations made in this way were examined on a glass slide with lines ruled at intervals just a little under the field diameter of a monocular microscope with a 16 mm. objective. A parfocal higher powered objective was used to identify specimens which were not recognisable under the lower power. This slide was moved with a mechanical stage and each row was systematically searched for Cladocera until 100 had been identified.

The Cyclopoida and Calanoida were picked out of the sub samples in the petri dish and laid in rows in a film of lactic acid on a glass slide. In this way a row of about ten specimens could be examined in detail and manipulated or even dissected under the microscope. This method is tedious, but to ensure accurate identification it is essential that cyclopoids should be examined in such a way that they can be dissected to reveal their diagnostic characters. Because of the labour involved only fifty cyclopoids from each sample were identified to specific level.

The few ostracods in the samples were picked out and dissected in polyvinyl-lactophenol containing lignin pink, and permanent preparations were made.

SYSTEMATIC SURVEY AND SEASONAL OCCURRENCE

Sub-class **BRANCHIOPODA**

Order *CLADOCERA*

Family **Sididae**

Pseudosida szalayi Daday

Several specimens of this species were found in the sample taken from the river in October 1956, and single specimens were found in the September and November samples of the same year. This is not a true planktonic species ; its presence in the plankton being due to flood water washing it from its normal habitat among vegetation.

This species can be distinguished from the closely allied *P. bidentata* Herrick by the median projection near the end of the post abdomen. Stingelin (1904) and Rzòska (1952) show this process as reaching beyond the second basal spine of the post abdominal claw, but in my specimens the process was much shorter and barely reached beyond the first basal spine (Fig. 2). Rzòska noted ten or eleven groups of spinules along the side of the post abdomen, but the Sokoto specimens had thirteen or fourteen groups. This difference is probably due to a difference in size ; Rzòska's specimens only reached a length

of about 1·5 mm., while those from the Sokoto ranged up to 2 mm. Birge (1910), in his detailed re-examination of *P. bidentata* found up to fifteen groups of spinules on the post abdomens of specimens reaching a length of 2 mm.

There are records of *P. szalayi* from localities in Tanganyika (Harding, 1957b) and Nysaland (Daday, 1910), and Rahm (1956) has recorded it from the Ivory Coast ; outside Africa it is known from Ceylon, India, Siam and Sumatra.

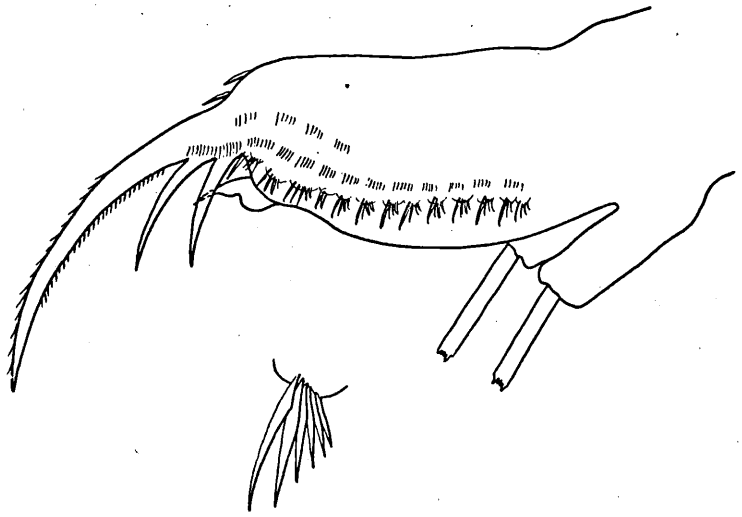


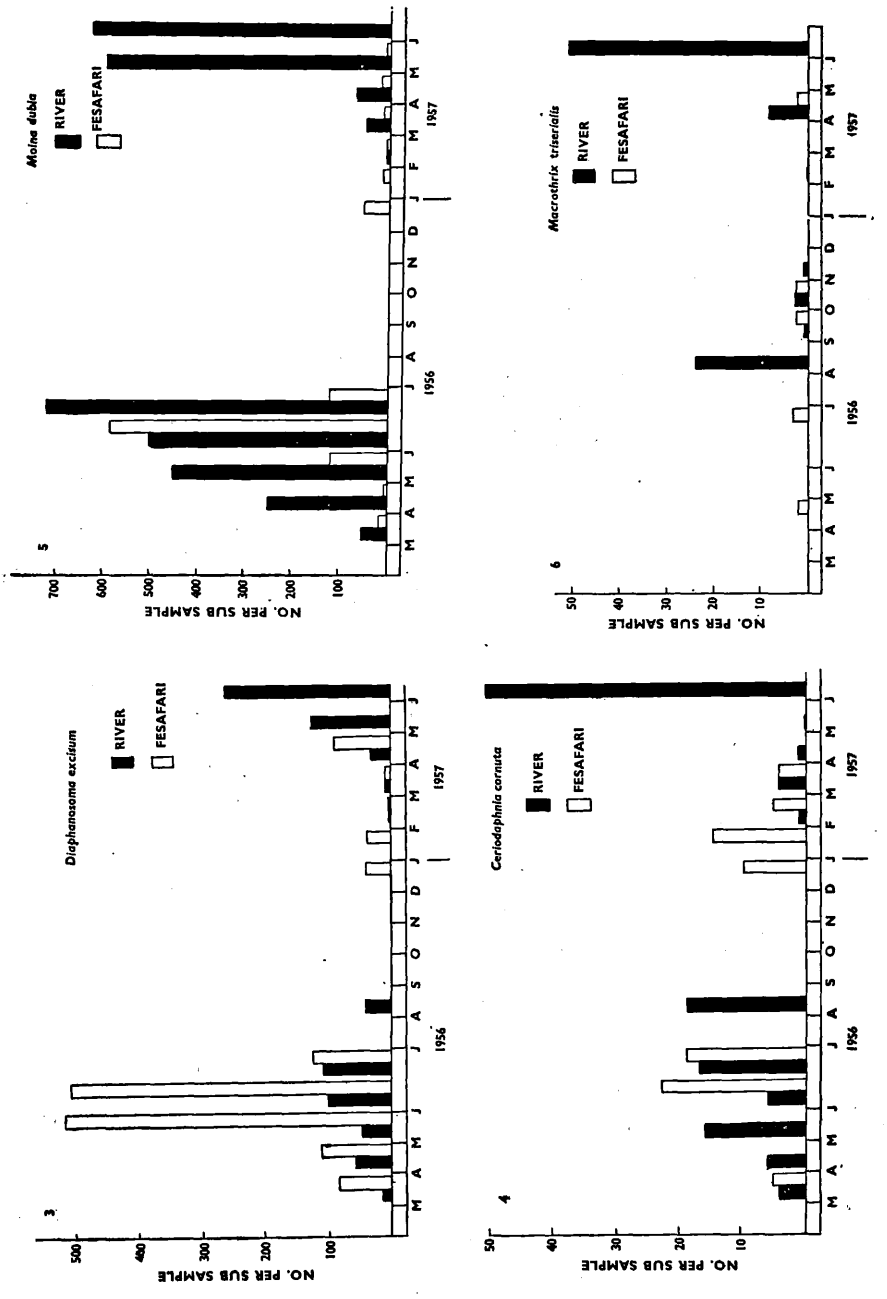
Fig. 2—*Pseudosida szalayi*, post abdomen and detail of a lateral fascicle.

Diaphanosoma excisum Sars

The seasonal occurrence of *D. excisum* is shown in Fig. 3. This was the dominant cladoceran in Fesafari in 1956, sometimes forming over 90 per cent of the total Cladocera, but it was much less abundant in 1957. In contrast the river developed a larger population in 1957 than in 1956.

The specimens from the Sokoto agree with Sars's (1885) original description, except that the infolding or duplicature of the ventral margin of the carapace is broader than shown in his figures, and in some of the specimens the antennae are long enough to reach back as far as the posterior margin of the carapace. The Sokoto specimens also have fine hairs on the post abdomen, a feature noted by Jenkin (1934) in material from the Rift Valley Lakes in Kenya, and to which she gave the name var. *Stingelini*. This variety is also characterised by a wider duplicature of the carapace than the type. The number of teeth on the posterior ventral corner of the carapace is variable, sometimes differing on the two sides of one specimen, for instance : a specimen from Fesafari in June 1956 had five teeth on the left valve and two teeth on the right valve ; such variation has also been noted in East African material (Jenkin 1934).

This species is widely distributed in tropical and sub tropical parts of Australia, Asia and Africa, and has previously been recorded in West Africa from the High Volta, the Ivory Coast and the region of Lake Chad.



Figs. 3–6—Seasonal occurrence. 3. *Diaphanosoma excisum*. 4. *Ceriodaphnia cornuta*. 5. *Moina dubia*. 6. *Macrothrix triserialis*.

Family **Daphniidae**

Ceriodaphnia cornuta Sars

The seasonal occurrence of this species is shown in Fig. 4. Its abundance does not seem to be so immediately influenced by the flood water as that of the two dominant cladoceran species: *Diaphanosoma excisum* and *Moina dubia*, although when the flood reaches its maximum (September in 1956) *C. cornuta* disappears from the samples and does not reappear until December in Fesafari, and even later in the river.

Most of the specimens could be classified as *C. rigaudi* Richard, which was reinstated as a separate species by Jenkin (1934), but is now generally regarded as a form of *cornuta* (Cf. Gauthier, 1951 ; Rzòska, 1956 ; Harding, 1957b). This species is widespread through the tropical and sub-tropical regions of the world.

Simocephalus serrulatus (Koch)

A few specimens were found in the sample taken from Fesafari in September 1956, and a single specimen in the sample from the river in March 1956.

The specimens from Fesafari agree with Sars's (1895, 1916) figures of *S. capensis* (a synonym of *serrulatus*) except that the front of the head is not so sharply angled. This more rounded front to the head is similar to the var. *rotundifrons* described by Brehm (1933 a) from the High Volta. The specimen from the river had the more sharply angled head characteristic of the type form.

This species is probably cosmopolitan.

Moina dubia De Guerne & Richard

This is the dominant cladoceran in the river during the dry season, often forming 80 per cent or more of the total Cladocera, and in Fesafari it becomes co-dominant with *Diaphanosoma excisum* in 1956, but failed to develop a large population in 1957 (Fig. 5).

Several African species of *Moina* have been examined and figured in great detail by Gauthier (1954), the specimens from the Sokoto agree with his diagnosis of *M. dubia*.

Family **Bosminidae**

Bosminopsis deitersi Richard

A single specimen of this widespread species was found in the sample taken from Fesafari in March 1956. A single egg was present in its brood pouch.

Family **Macrothricidae**

Macrothrix triserialis Brady

The seasonal occurrence of this species is shown in Fig. 6. It is always sparse in Fesafari, but the river samples show a maximum at the beginning of the floods, the peak being earlier in 1957 than in 1956. This is not a true planktonic species, but generally lives among vegetation and near the bottom.

Diagnostic characters of the species are shown in Figs. 7–10. The two spines at the apex of the antennule are particularly well developed in these Sokoto specimens.

This species, which includes *M. chevreuxi* De Guerne & Richard, seems to be generally distributed in the tropics, and extends into S. Africa and New South Wales (see Gauthier (1939) for details).

Macrothrix goeldii Richard

This species was much less abundant than *M. triserialis*, and was only found in small numbers in the sample taken from the river in August 1956.

Diagnostic characters of the species are shown in figs. 11–14. The form of the antennule in *Macrothrix* species can look very different according to the precise angle from which it is viewed, so that I have given two views for the species found in the Sokoto.

This species is known from Chile, India and West Africa.

Grimaldina brazzai Richard

A single specimen of this characteristic form was found in the sample taken from the river in August 1956.

The distribution of this species extends from tropical Africa to New Guinea in the East, and to Brazil and the Southern U.S.A. in the West.

Ilyocryptus spinifer Herrick

This species was found in small numbers in the samples from the river at irregular intervals throughout the year. No specimens were found in the samples from Fesafari. This is a bottom dwelling species whose occurrence in the plankton is accidental ; its occurence only in the samples from the river indicates that considerable water movement is necessary to swirl this animal up into the plankton. *Ilyocryptus halyi* Brady and *I. longiremis* Sars are synonyms of this cosmotropical species.

Family **Chydoridae**

Members of this family are to a large extent bottom and weed dwellers, but many of them can swim actively and are regularly recorded in plankton samples, particularly when filamentous blue-green algae are present in the plankton. A good example of this is given by Berg & Nygaard (1929) in their study of the plankton of Frederiksborg Castle Lake, where the occurrence of *Chydorus sphaericus* in the plankton is associated with an abundance of Cyanophyceae.

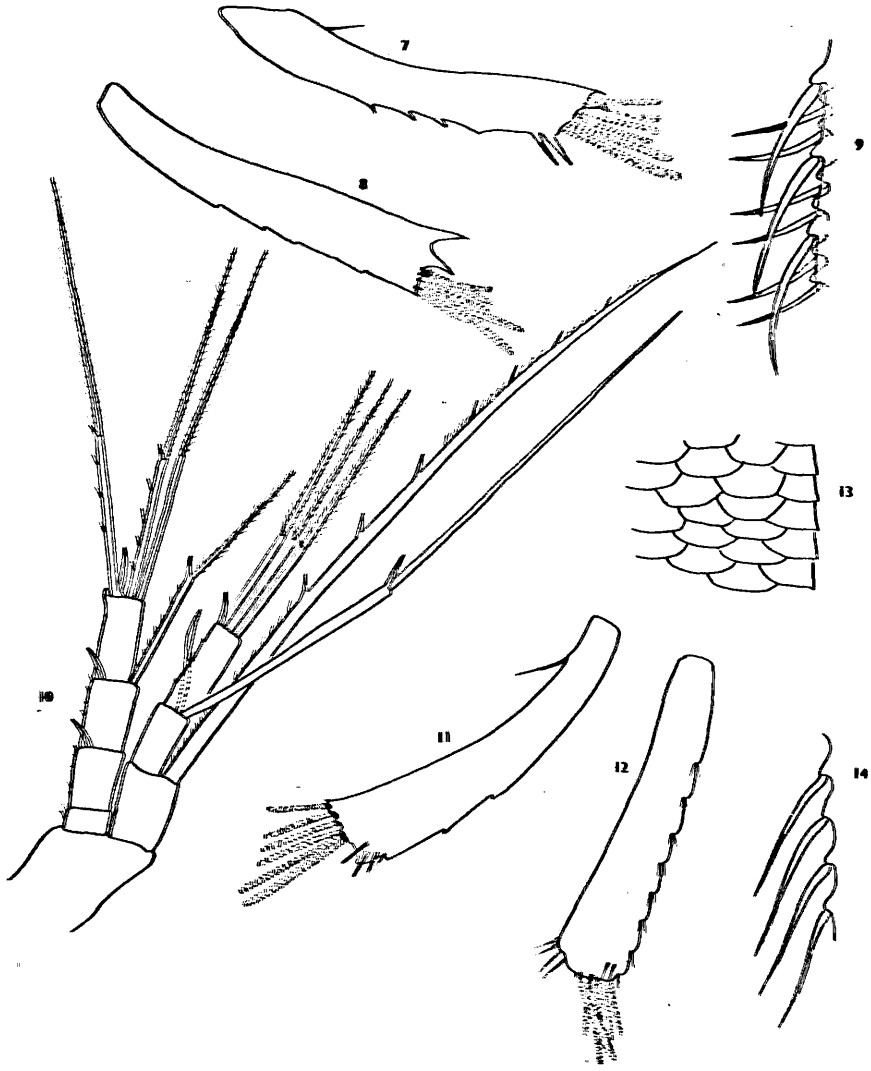
Alona affinis Leydig

A few typical specimens of this widespread species were found in the samples taken from the river in September and October 1956.

Alona diaphana King

This species was found in the river from August to December 1956 but in Fesafari its occurrence in the plankton was more restricted, and it was only

found in the samples taken in the period from September to November. In both the pool and the river it was most numerous in October.



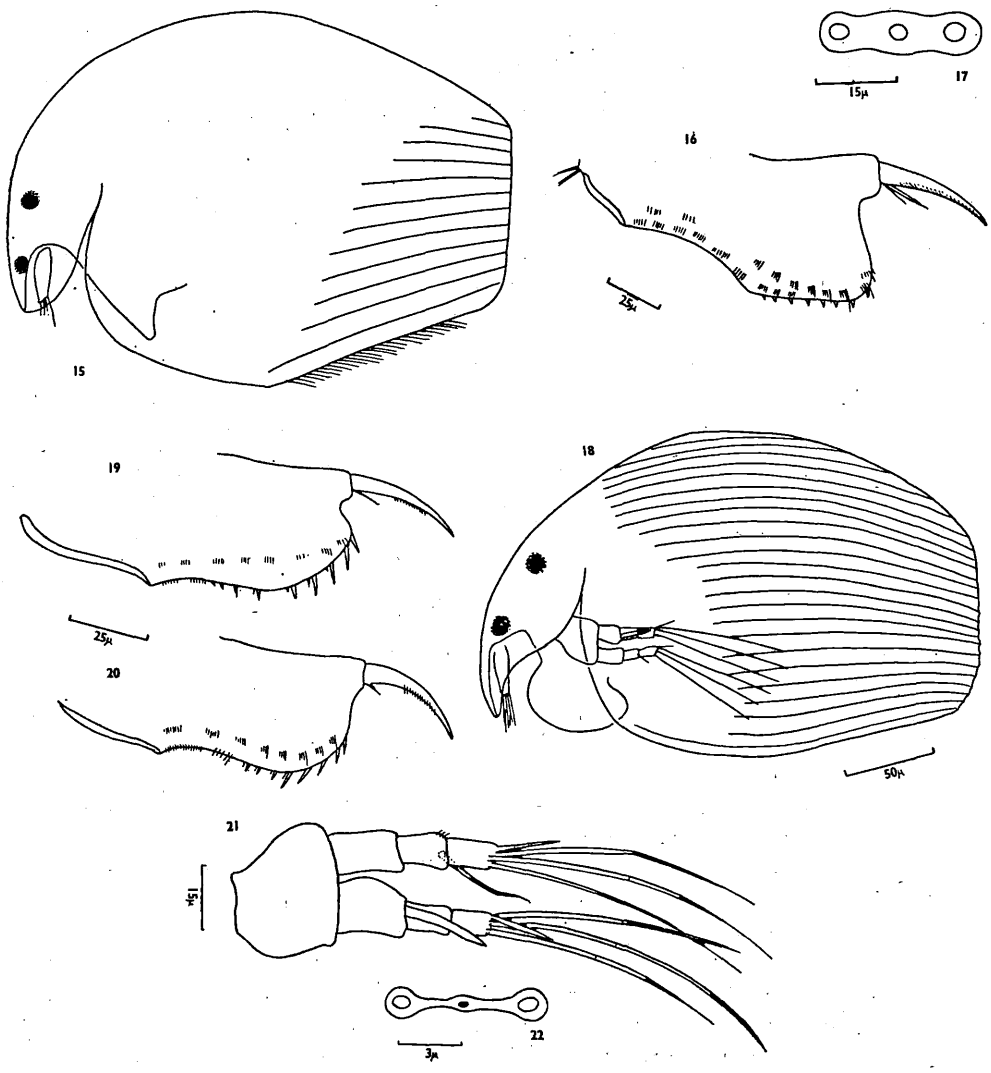
Figs. 7–14.—*Macrothrix* species. 7–10, *M. triserialis*: 7 & 8, different views of one antennule; 9, part of the anterior border of the carapace; 10, antenna. 11–14, *M. goeldii*; 11 & 12, different views of one antennule; 13, part of the dorsal margin of the carapace; 14, part of the ventral margin of the carapace.

The specimens had lateral fascicles on the post abdomen, so that they would be identifiable as *A. davidi* Richard if this was regarded as a separate species, but I am following Brehm (1933), Harding (1955, 1957b) and Fryer (1957a) in regarding it as a synonym of *A. diaphana*.

Alona eximia Kiser

This species was found in the samples from Fesafari from June to November 1956, and in the main channel of the river from September to November 1956, and a few specimens in February 1957.

I have asigned these specimens to *A. eximia* largely on the structure of the post abdomen (Fig. 16) which agrees well with the figure given by Kiser (1948). There are some small differences in the arrangement of the lateral fascicles, but I do not regard these as important, since the more anterior groups, which Kiser



Figs. 15–22.—*Alona* species. 15–17, *A. eximia* : 15, lateral view of head and carapace ; 16, lateral view of post abdomen ; 17, dorsal pores. 18–22, *A. holdeni* : 18, lateral view of head and carapace ; 19 & 20, lateral views of post abdomens ; 21, antenna, lateral view ; 22, dorsal pores.

does not figure, are difficult to see without using an oil immerison objective and critical lighting.

The rostrum is short and blunt. The antennules do not quite reach the tip of the rostrum, but the antennulary setae extend a short distance beyond it ; one of these setae is much longer than the others. The labral keel is a little different in shape from that figured by Kiser, who shows it projecting somewhat more anteriorly.

The ventral part of the carapace is clearly striated, but the striae fade away dorsally, particularly in the larger egg bearing females. The striae are somewhat closer together than in Kiser's figures.

On one specimen I was able to see the form of the dorsal pores (Fig. 17), which are quite different from the dorsal pores of *A. intermedia* Sars (see Frey 1959).

The only previous record of *A. eximia* seems to be the original description from the Pearl River, China.

ALONA HOLDENI sp. n.

The main diagnostic features of this species are shown in Fig. 18–22. The total length of the adult female ranges up to 0·3 mm., but most specimens are about 0·25 mm. long. The rostrum is moderate in length with the antennules not reaching its tip, but with the antennulary setae projecting a little beyond the tip. The eye is only a little larger than the ocellus. The labral keel is smoothly ovoid, without any teeth. The carapace is strongly arched dorsally and only slightly bent along the ventral edge. The posterior ventral corner of the carapace does not bear any teeth. The valves of the carapace are clearly striated with between twenty-seven and thirty striae.

The antenna bears a striking character in the form of a large spine like seta on one of its branches (Fig. 21). Other species of *Alona* that I have examined have a small seta in this position and it does not reach beyond the apex of the second podomere. I have not been able to examine all the described species of *Alona*, and since most authors do not describe the antennae of their specimens it is at present unknown if this character is unique to *A. holdeni*.

The post abdomen shows some variation in shape (Figs. 19 & 20), most of the specimens looking like Fig. 20. Lateral fascicles are present, and the distal ones overlap the margin of the post abdomen. There is also some variation in the length of the basal spine on the claw, sometimes it is almost a quarter the length of the claw, whilst more often it is about one sixth the length of the claw.

Dorsal pores were found in one specimen, only the median row was seen ; this consisted of three pores, the middle one of which was the smallest.

No males were found.

There was a small outburst of this species in Fesafari in May and June 1956, and single specimens were found in Fesafari in January 1957, and in the main channel of the river in April 1957. The relative abundance of this species in Fesafari in May, before the river was in flood, indicates that this may be a genuine member of the plankton and not merely a form swept into the plankton as are most other species of the genus.

Alona pulchella King

This cosmotropical species was most numerous during the period when the river was in flood, but small numbers were also found in the river in May 1956, February and March 1957. In Fesafari it was only found in the samples taken in September and October 1956.

In identifying and counting this species I have been guided mainly by the structure of the post abdomen (Fig. 24) and have not distinguished the form *camboueí* De Guerne & Richard, which is at most a variety of *A. pulchella.*

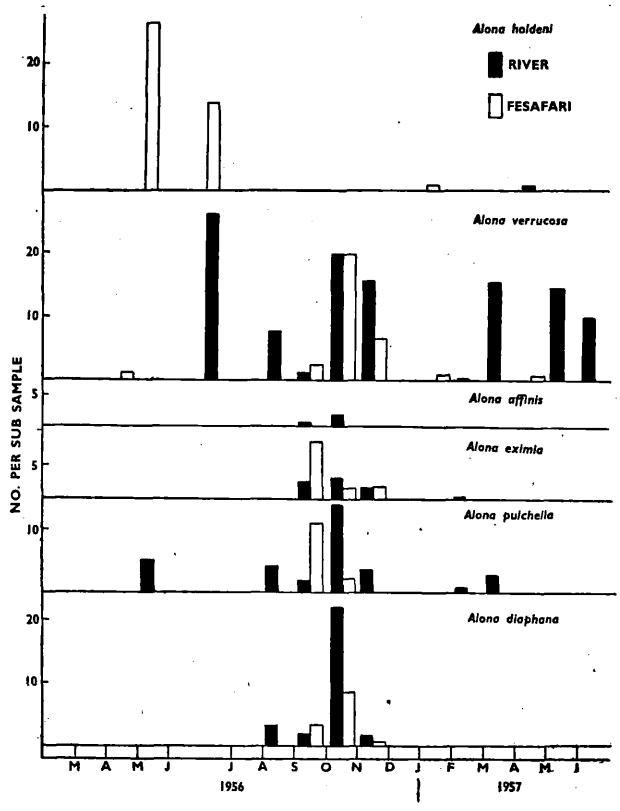


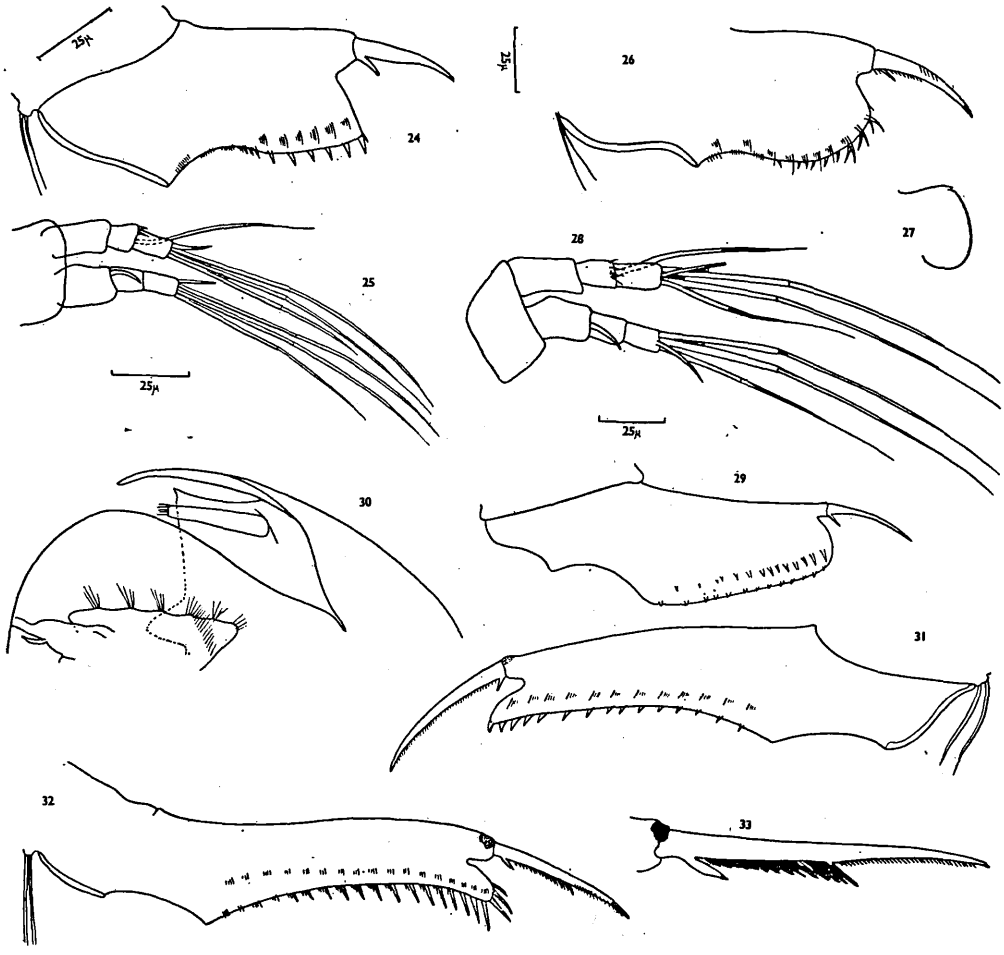
Fig. 23—Seasonal occurence of *Alona* species.

Alona verrucosa Sars

The seasonal occurrence of this species in the plankton was somewhat erratic (Fig. 23), both in the main channel and Fesafari. It may be that this is a more active swimmer than most other members of the genus so that it is more frequently caught in the plankton, even when the river is not flooding.

The nomenclature of this form has recently been discussed in detail by Johnson (1956). The labrum of the Sokoto specimens bore an antero-ventral notch, and the lateral fascicles of the post abdomen had long distal bristles reaching beyond the margin of the post abdomen (Fig. 26). The characteristic " verrucae " were not always present on the carapace.

This species does not seem to have been recorded previously from West Africa, though it is known from East Africa, South America and South-east Asia.



Figs. 24–33.—Chydorids from the River Sokoto. 24, *Alona pulchella*, lateral view of post abdomen. 25, *A. pulchella*, lateral view of right antenna. 26, *Alona verrucosa*, lateral view of post abdomen. 27, *A. verrucosa*, labral keel. 28, *A. verrucosa*, lateral view of right antenna. 29, *Alonella globulosa*, lateral view of post abdomen. 30, *Kurzia longirostris*, lateral view of head and anterior part of carapace ; the eye has bleached in the preservative. 31, *K. longirostris*, lateral view of post abdomen. 32, *Euryalona occidentalis*, lateral view of post abdomen. 33, *E. occidentalis*, enlargement of claw.

Alonella excisa (Fischer)

A single specimen of this cosmopolitan species was found in the sample taken from the river in November 1956.

Alonella globulosa Daday

In both the river and Fesafari this species was found in the plankton from September to November 1956.

The post abdomen of this species is shown in Fig. 29. The Sokoto specimens would be assigned to *A. sculpta* Sars, if this was regarded as a separate species.

Kurzia longirostris (Daday)

A single specimen was found in the sample taken from the river in August 1956. The post abdomen of this specimen is shown in Fig. 31 ; lateral fascicles were clearly visible under a 4 mm. objective when the specimen was mounted in polyvinyl-lactophenol and stained with lignin pink. A characteristic feature is found in the shape of the labral keel, which projects ventrally to form an almost right angled corner. The base of the first limb bears a large flange which projects dorsally alongside the posterior part of the labrum (Fig. 30). Confusion of this part of the first limb with part of the labrum might account for the peculiarly shaped labrum figured by Daday (1910), under the name *Pseudalona longirostris*, for a specimen from the Nyasa region.

This is a widespread species in the tropics ; there are records from South America, New Guinea, East Indies, Ceylon as well as East and West Africa.

Euryalona occidentalis Sars

A single specimen was found in the sample taken from the river in October 1956. The post abdomen of this specimen is shown in Fig. 32. The claw (Fig. 33) agrees in detail with Harding's (1957 a) figure, and clearly distinguishes the specimen from *E. colletti* Sars, a South African form. The labrum also carried the peculiar lateral creases noted by Harding.

There is still some doubt whether or not this species is a synonym of *E. orientalis* (Daday). Rzòska (1952) identified specimens from the White Nile as *E. orientalis*, and his figure of the post abdomen agrees fairly well with mine, but Daday's figure of the post abdomen is shorter and broader than mine and Rzòska's. Daday (1910) gives a figure of the labrum of a specimen of *E. orientalis* from East Africa which differs in shape from Harding's figure of *E. occidentalis* and does not show the lateral creases.

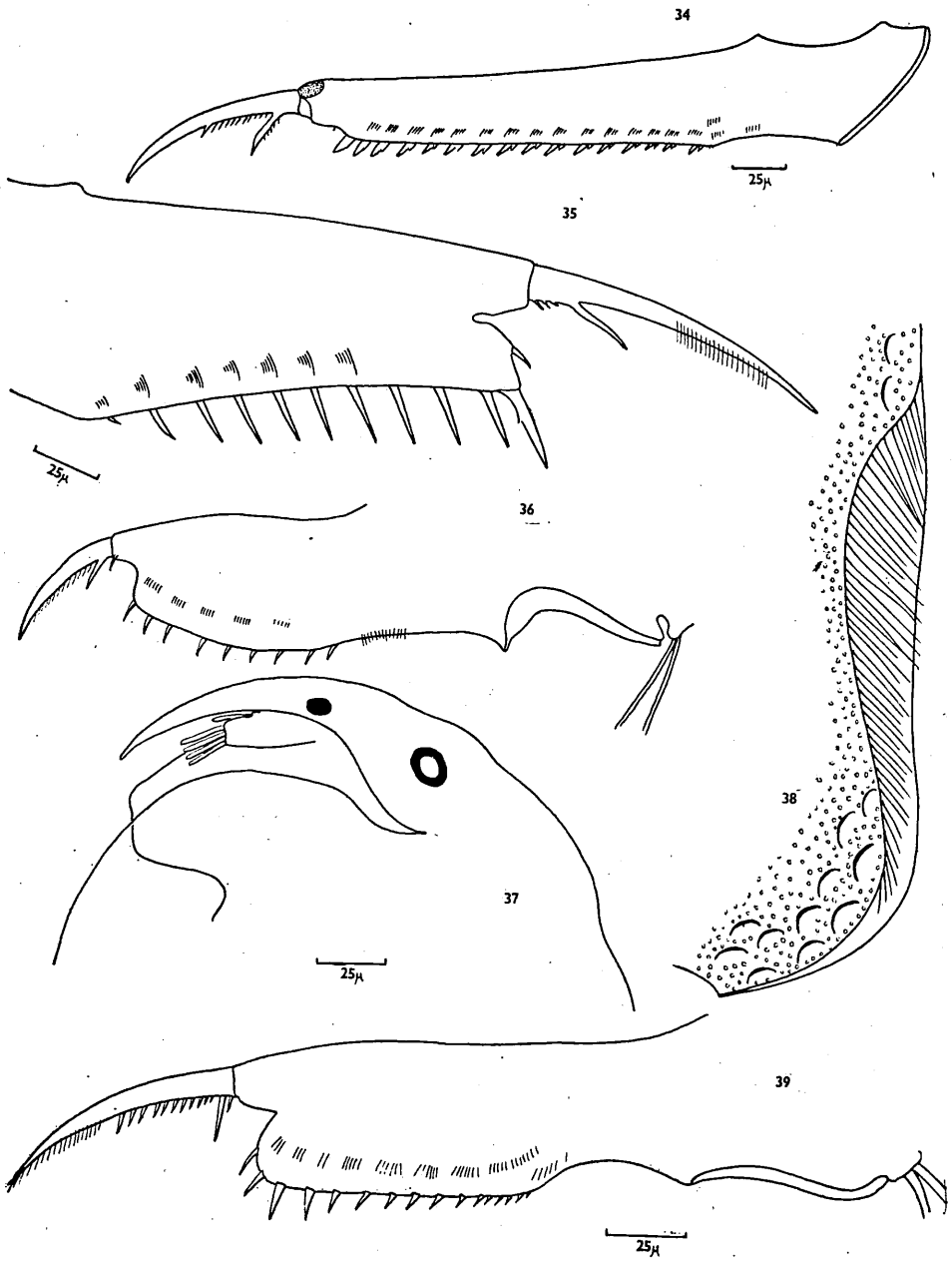
Apart from various records from Africa this species has been recorded from South America.

Camptocercus rectirostris Schödler

This species was only found in the sample taken from the river in October 1956, when it formed about 5 per cent of the total Cladocera, and in November 1956, when a single specimen was found.

The post abdomen (Fig. 34) agrees with typical European specimens, but the carapace lacks any post ventral teeth, which is typical of tropical specimens in contrast to European specimens which usually have two or three such teeth.

This species seems to be cosmopolitan. Harding (1957 b) has suggested that the tropical and southern hemisphere forms lacking post ventral teeth might be separated as a distinct sub species under the name *australis* Sars.



Figs. 34–39—Chydorids from the River Sokoto. 34, *Camptocercus rectirostris*, lateral view of post abdomen. 35, *Oxyurella singalensis*, lateral view of apex of post abdomen. 36, *Chydorus eurynotus*, lateral view of post abdomen. 37, *C. eurynotus*, lateral view of head. 38, *C. eurynotus*, posterior ventral margin of the right valve of the carapace. 39, *Chydorus globosus*, lateral view of post abdomen.

Oxyurella singalensis (Daday)

Specimens of this species were found in the river in October and December 1956, and in Fesafari in January 1957.

The apex of the post abdomen of a specimen from the river is shown in Fig. 35. This agrees fairly well with Daday's (1910) figure of *Alonopsis singalensis*, which Rzòska (1952) places in the genus *Oxyurella*. The Sokoto specimens also agree with Rzòska's description of specimens from the White Nile ; a minor difference is found in the presence of three minute teeth at the base of the claw instead of two as shown by Rzòska. Fryer (1957a) has collected specimens from one of Daday's Localities and confirms that they agree with the Nile specimens described by Rzòska. A very similar figure of the post abdomen is given by Harding (1957b) but under the name *Oxyurella tenuicaudis* Sars. It is highly probable that this was in fact a specimen of *O. singalensis*, which can be distinguished from *O. tenuicaudis* by the three distal spines on the post abdomen forming part of a series which gradually increases in length, instead of being abruptly larger than the more proximal spines, as in *O. tenuicaudis*. Harding also figures the post abdomen of a specimen which is the true *O. tenuicaudis*, and which agreed with European specimens with which it was compared.

Rzòska (1952) and Fryer (1957a) have already suggested that *Alona gauthieri* Brehm (1933 a) is a synonym of *O. singalensis*. Brehm's figure shows a post abdomen which agrees with fig. 35, and shows three small teeth at the base of the claw. A difference is found in that Brehm shows the lateral fascicles as consisting of a single setule, but it is uncertain whether this small difference is real or observational ; the posterior members of a fascicle are often extremely difficult to see.

Brehm (1953) has also described *Oxyurella lindbergi* from India, and Rahm (1956) records it from the Ivory Coast, but it seems very doubtful if it differs sufficiently from *O. singalensis* to be regarded as a separate species.

Acroperus harpae Baird

This species was scarce in the samples, only a few specimens were taken from Fesafari in October and November 1956, and a single specimen was taken from the river in November 1956.

The specimens from Fesafari had straight dorsal margins, and would be referable to *A. angustatus* Sars, if this was regarded as a good species ; the specimen from the river was a typical female of *A. harpae*.

This is mainly a species of the temperate and arctic Northern Hemisphere, but it has also been recorded from the Sunda region and from Africa (French West Africa and near Lake Tanganyika).

Leydigia ciliata Gauthier

Single specimens agreeing with Harding's (1955) redefinition of this species were found in the samples taken from the river in August and October 1956. None was found in the samples from Fesafari. This is a mud dwelling form which is only accidentally swept into the plankton.

Previous records of this species are known from West Africa and South America.

Chydorus eurynotus Sars

This bottom dwelling species only appeared in the samples during the floods, from September to November 1956 in both the river and Fesafari, forming up to 5 per cent of the total Cladocera.

The post abdomen is shown in Fig. 36. This agrees with Brehm's (1933 a) figure of the post abdomen of *C. kallipygos* except that Brehm omits the small lateral fascicles, but I agree with Gauthier (1939) that Brehm's species is a synonym of *C. eurynotus*. Harding (1955) maintains the two as separate species, and his figures of the post abdomens show certain differences. Nevertheless one of Gauthier's figures (11D) is intermediate between the two figures given by Harding. The labral keel of *C. eurynotus* is variable in form ; Harding (1957 b) figures a whole series, some of which are rounded while others are flattened at the end, as in Fig. 37. The keel may still be useful in separating *C. eurynotus* from other species by the fact that it never seems to be prolonged into a point of the type found in the closely allied *C. sphaericus* (O. F. Müller) and *C. herrmanni* Brehm.

The sculpturing of the carapace also seems to be variable. Gauthier (1939) shows a reticulated pattern of wavy lines, but Harding (1957b) says that these are sometimes absent. The specimens from the Sokoto did not have a reticulated, pattern, but were finely punctured all over, with a few crescentic depressions near the post ventral border (Fig. 38).

Chydorus eurynotus is known from South America, East and West Africa.

Chydorus barroisi Richard.

This species is swept into plankton samples by the floods, forming up to 20 per cent of the total Cladocera in Fesafari in October 1956, but only up to 8 per cent in the main channel of the river. A few specimens were also found in the samples taken from Fesafari in January and March 1957.

This species is widespread in Africa, and has been recorded from North and South America, South Asia, Palestine and New Zealand.

Chydorus globosus Baird

A single specimen of this well-known species was found in the sample taken from Fesafari in December 1956. The post abdomen (Fig. 39) shows the long and short spines at the base of the claw quite clearly, and there is a series of groups of lateral fascicles as described by Birge (1918) for North American specimens. It seems probable that Lilljeborg (1900) overlooked these fascicles in his description of European specimens ; they are certainly present in the British specimens of this species that I have examined.

Pleuroxus aduncus (Jurine)

A single specimen of this cosmopolitan species was found in the sample taken from the river in September 1956, and single specimens were taken from Fesafari in September 1956 and February 1957.

The synonymy of this form has recently been given in detail by Harding (1955).

Pleuroxus laevis Sars.

Single specimens of this species were found in the samples taken from the river in October and November 1956, and from Fesafari in December 1956.

The post abdomen of one of these specimens is shown in Fig. 41, and a lateral view of the head and carapace in Fig 40. In the latter figure the labral keel is somewhat unusual in shape ; this I believe to be due to contraction of the animal when it died.

I suspect that *Pleuroxus incertus* Brehm (1933 a) belongs to this species ; his figures show that the post abdomen is very similar to that of *P. laevis*, and show the post-ventral corner of the carapace as having a single tooth.

This species is widespread in temperate and subtropical regions, but does not seem to have been recorded from the Americas.

Graptoleberis testudinaria (Fischer)

In Fesafari this species was only found in October, but in the main channel it was found from September to November, always in small numbers.

This species is probably cosmopolitan, being common in Europe as well as in the tropics, and extending as far North as Greenland.

Order *CONCHOSTRACA*

Family **Cyclestheriidae**

Cyclestheria hislopi (Baird)

Two very small specimens of this cosmotropical species were found in the sample taken from the river in October 1956.

The carapaces of these specimens were only 1·04 mm. long. The head bore a relatively large dorsal organ (Fig. 42), and the post abdomen (Fig. 43) had not yet developed its full complement of spines and spinules, so that it appeared a little different to figures given by various authors of the adult post abdomen (cf. Daday, 1926). A comparison with young and adult specimens from other localities in the collection at the British Museum (Natural History) showed that these differences are entirely due to the juvenile state of the specimens.

Sub-class **COPEPODA**

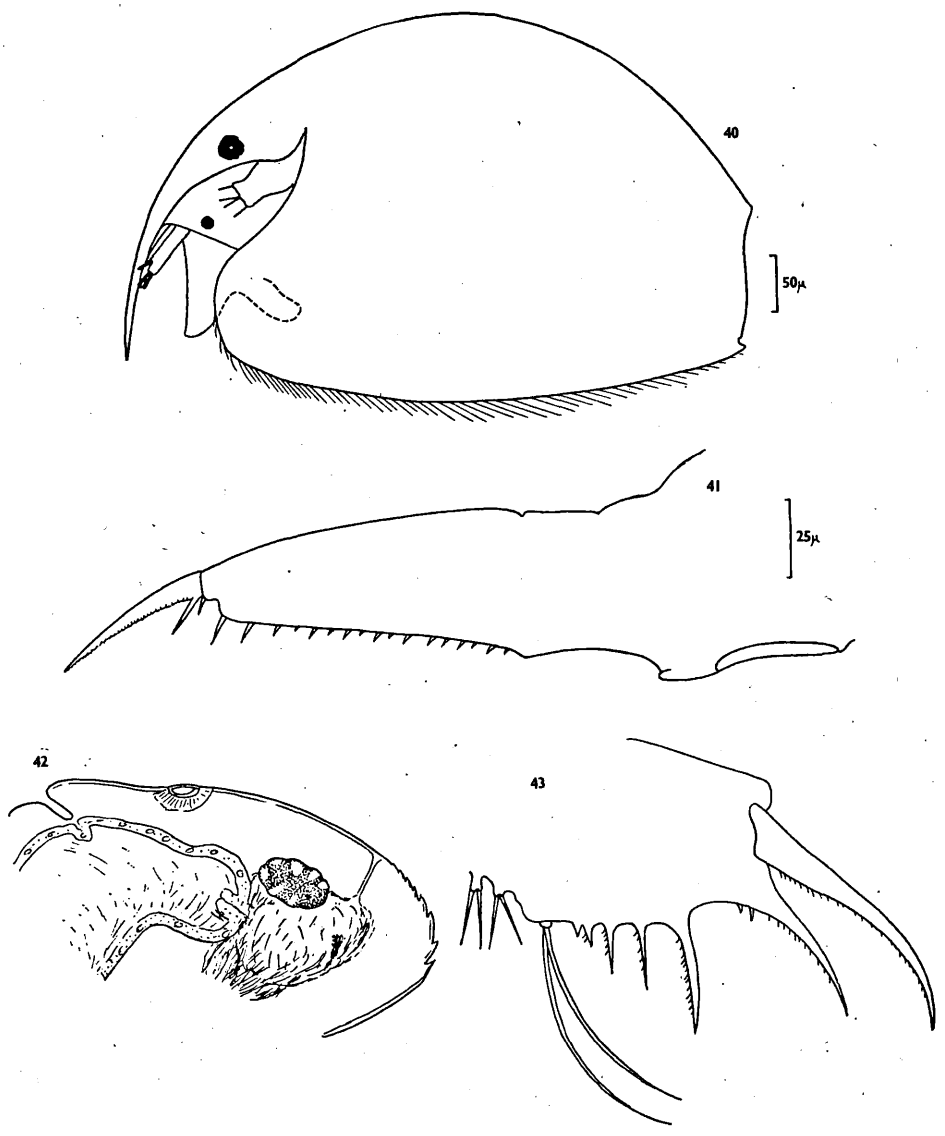
Order *CALANOIDA*

Family **Diaptomidae**

Tropodiaptomus laurentii Gauthier

This species has only recently been described from Senegal by Gauthier (1951). Only a single specimen of another calanoid copepod was found, so that *T. laurentii* was practically the only calanoid present. This fortunate chance has made it possible to study the biology of this species in more detail than the cyclopoids, whose nauplii were not separable from each other. The biology of *T. laurentii* was studied using the samples from Fesafari because these would show less variation than the changing population swept down by the river.

In the account which follows the lengths of nauplii have been measured from the anterior margin of the head to the ends of the rudimentary caudal rami, and the lengths of the copepodid stages have been measured from the anterior margin to the junction of the metasome and urosome ; this is a more reliable measure of length than total length because many of the specimens had the urosome curved and at an angle to the metasome so that errors of measurement would be likely to occur. Metasome length has been found to be a convenient



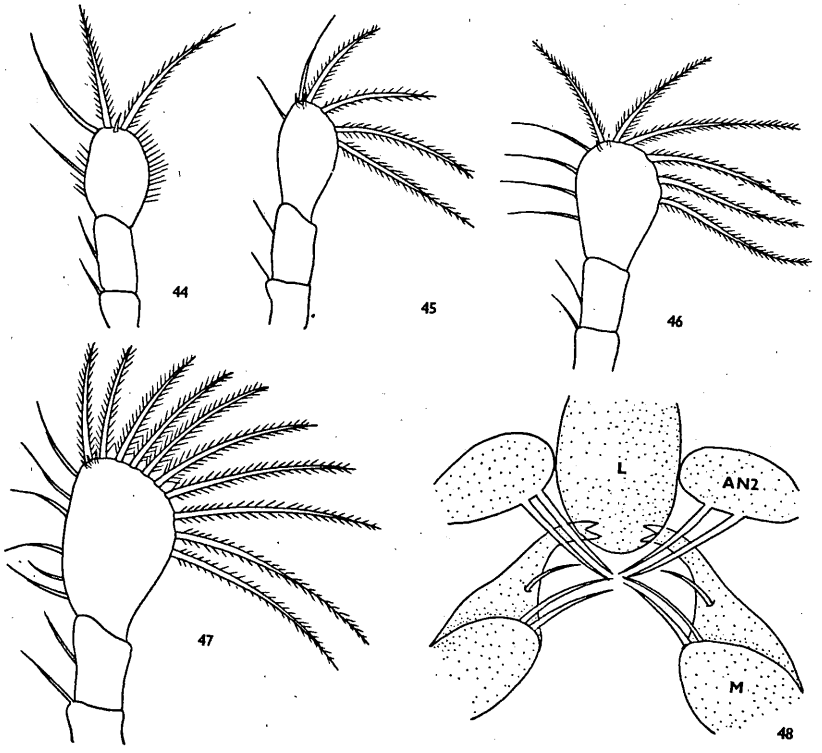
Figs. 40–41—*Pleuroxus laevis*. 40, lateral view of head and carapace. 41, lateral view of post abdomen.

Figs. 42–43—*Cyclestheria hislopi*, juvenile specimen. 42, lateral view of head. 43, lateral view of post abdomen.

measure of size by other workers on calanoid copepods (e.g. Comita & Anderson, 1959, Digby, 1950 ; Marshall, 1949, Tonolli, 1949).

It has been possible to recognise six naupliar stages in the material ; these may be characterized as follows.

Stage 1. Length 150 to 180μ. Three large setae on antennule, 1 small seta on inner border, and numerous fine hairs on the outer border (Fig. 44).
Stage 2. Length 198 to 220μ. Four or five large setae on antennule, one seta on inner border.
Stage 3. Length 225 to 250μ. Six large setae on antennule, four setae on inner border.
Stage 4. Length 240 to 320μ. Eight large setae on antennule, four setae on inner border.
Stage 5. Length 330 to 340μ. Nine large setae on antennule, four or five setae on inner border.
Stage 6. Length 352 to 370μ. Ten or eleven large setae on antennule, six setae on inner border (Fig. 47).



Figs. 44–48—Nauplii of *Tropodiaptomus laurentii*. 44, antennule of 1st stage. 45, antennule of 2nd stage. 46, antennule of 3rd stage. 47, antennule of 6th stage. 48, Ventral view of the appendages near the mouth of 5th stage nauplius. L, labrum ; An 2, antenna ; M, mandible.

Six copepodid stages were also recognised, and can be distinguished as follows :—

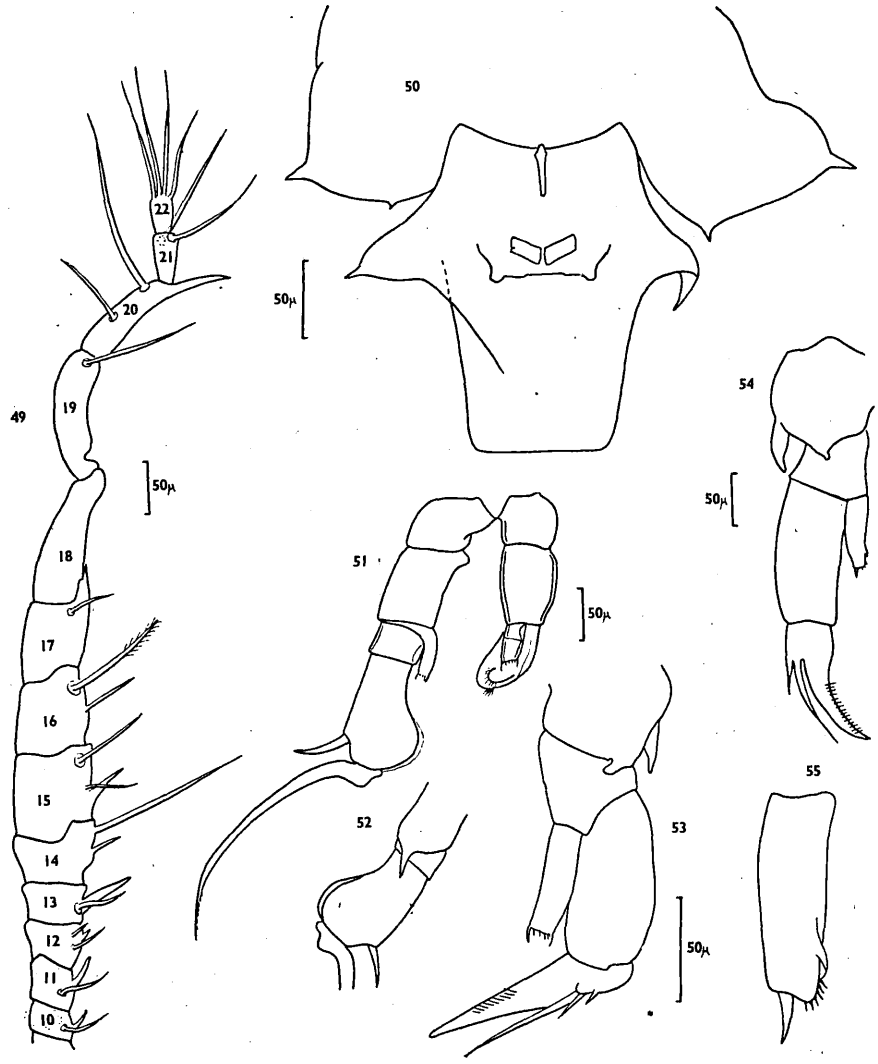
Copepodod I. Length 340 to 380μ. Two pairs of swimming legs ; four free segments in metasome ; urosome of one segment ; caudal rami each with two large setae.
Copepodid II. Length 400 to 500μ. Three pairs of swimming legs ; five free segments in metasome (this also applies to all the later stages) : urosome of one segment ; caudal rami each with four large and two small setae.
Copepodid III. Length 490 to 560μ. Four pairs of swimming legs ; five large setae and one small seta on each ramus (applies to all later stages) ; urosome with two segments.

Copepodid IV. Length 580 to 710μ. Five pairs of legs, the right fifth leg of the male is slightly longer than the left ; urosome of three segments.

Copepodid V. Length 650 to 900μ. Females larger than males. Five pairs of legs, right fifth legs of males distinctly longer than left ; urosome of four segments in male, three segments in female.

Copepodid VI. Adult male, length 780 to 910μ. Five pairs of legs, fifth legs fully developed (Figs. 51 and 52) ; urosome of five segments ; right antennule modified and swollen (Fig. 49).

Adult female, length 920μ—1·06 mm. Five pairs of legs, fifth pair fully developed (Fig. 53) ; urosome of four segments.



Figs. 49–55—Diaptomids from the River Sokoto. 49–53, *Tropodiaptomus laurentii*. 49, right antennule of male. 50, genital segment of female, ventral view. 51, fifth legs of adult male, anterior view. 52, apex of right fifth leg of adult male, posterior view. 53, fifth leg of adult female, anterior view. 54–55, *Diaptomus* sp. female. 54, fifth leg, anterior view. 55, endopod of fifth leg.

The two years for which sufficient data are available show great differences (Table 1). In 1956 the population was well established by March, but still only 6 per cent of the females were carrying spermatophores. As the season progressed the ratio of males to females increased greatly, and the percentage of females with spermatophores increased correspondingly. The sex ratio became almost unity at the end of June, just before the floods dispersed the population to such an extent that no specimens were found in the samples until December.

There were no nauplii or young copepodids in the December 1956 sample, and none of the females carried spermatophores. Nauplii were found in the January 1957 sample, and as the season progressed their numbers increased, but only slightly. The season ended earlier than in the previous year and the population only reached a small fraction of the size it was in 1956. As in the

Table 1—*Tropodiaptomus laurentii* : numbers per sub sample from Fesafari.

Date	No. of nauplii	No. of copepodid stages I–IV.	No. of copepodid stage V.	No. males	No. females	sex ratio
23 March 56	30	52	66	46	34 (6)*	1·33
18 April 56	33	25	28	70	19 (28)	3·66
7 May 56	154	95	72	108	23 (40)	4·80
7 June 56	640	567	220	275	128 (44)	2·14
25 June 56	105	43	43	94	105 (59)	0·88
8 Dec. 56	0	0	4·5	2·8	2·8 (0)	1·0
8 Jan. 57	1·3†	1·5	0·5	0·8	1·0 (25)	0·8
3 Feb. 57	1·4	1·6	3·1	1·6	1·7 (20)	0·94
7 March 57	2·7	0·8	1·3	1·5	0·3 (25)	5·0
5 April 57	4·1	19·7	9·2	0·4	1·0 (0)	0·4
17 May 57	0·1	0·1	0·6	0·7	0·6 (0)	1·15

*Figures in brackets following the number of females give the percentage carrying spermatophores.

†Fractional numbers are due to the low numbers of copepods necessitating the examination of several sub samples in order to obtain enough specimens to estimate their abundance.

previous year the ratio of males to females increased early in the season and fell at the end. The number of females with spermatophores did not reach the level of the previous year and fell away at the end so that no females were found with spermatophores in the last two samples. The failure of the population in 1957 is reflected in the low percentage of females with spermatophores and in the small numbers of nauplii.

The data available seem to indicate that in 1957 in Fesafari there was only one generation of *T. laurentii*. The population appears to have passed through the floods in the form of 5th stage copepodids and non reproducing males and females. The males appear to become mature before the females ; this would account for the great increase in the sex ratio which was found between March and May in 1956 and the high ratio in March 1957. The later fall in the sex ratio is probably due to the maturation of females, but it may also be partly due to the earlier death of some of the older males. In 1956 there might have been more than one generation in the season, but more detailed sampling would be required to establish this point.

Diaptomus sp. (sens. lat.)

A single female diaptomid, differing from the females of *Tropodiaptomus laurentii*, was found in the sample taken from Fesafari in December 1956. All the males in the sample belong to *T. laurentii*, the females of which differ from the present specimen in various characters, particularly in the structure of the 5th leg (Figs. 54–55).

In the absence of males it is not possible to place this female in one of the genera created by Kiefer (1932 b), because the diagnostic characters of these genera are almost entirely based on males.

Order *CYCLOPOIDA*

Family **Cyclopidae**

Macrocyclops albidus (Jurine) *oligolasius* Kiefer

This form was only found in the sample taken from the river in December 1956. Kiefer (1934 a) remarks that the tropical forms of this species have relatively shorter caudal rami than palaearctic specimens. The Sokoto specimens are in agreement with this ; the rami are not quite twice as long as wide (one specimen had rami 57μ long and 32μ wide), whereas in palaearctic specimens the length is usually much more than twice the width. The subspecies *oligolasius* has previously been recorded from West Africa by Kiefer (1934 b).

Eucyclops serrulatus (Fischer)

This cosmopolitan species was not found in Fesafari, but occurred in small numbers in the samples taken from the river in March, both in 1956 and 1957, and in December 1956.

Eucyclops gibsoni (Brady)

Single specimens of this widely distributed African species were found in the samples taken from the river in March 1956, March 1957 and June 1957. It was not found in the samples from Fesafari.

The specimens were distinctly small, one female, carrying five eggs in each egg sac, had a total length, excluding furcal setae, of 600μ ; the caudal rami were 98μ long and 14μ wide, giving a ratio of 7 : 1.

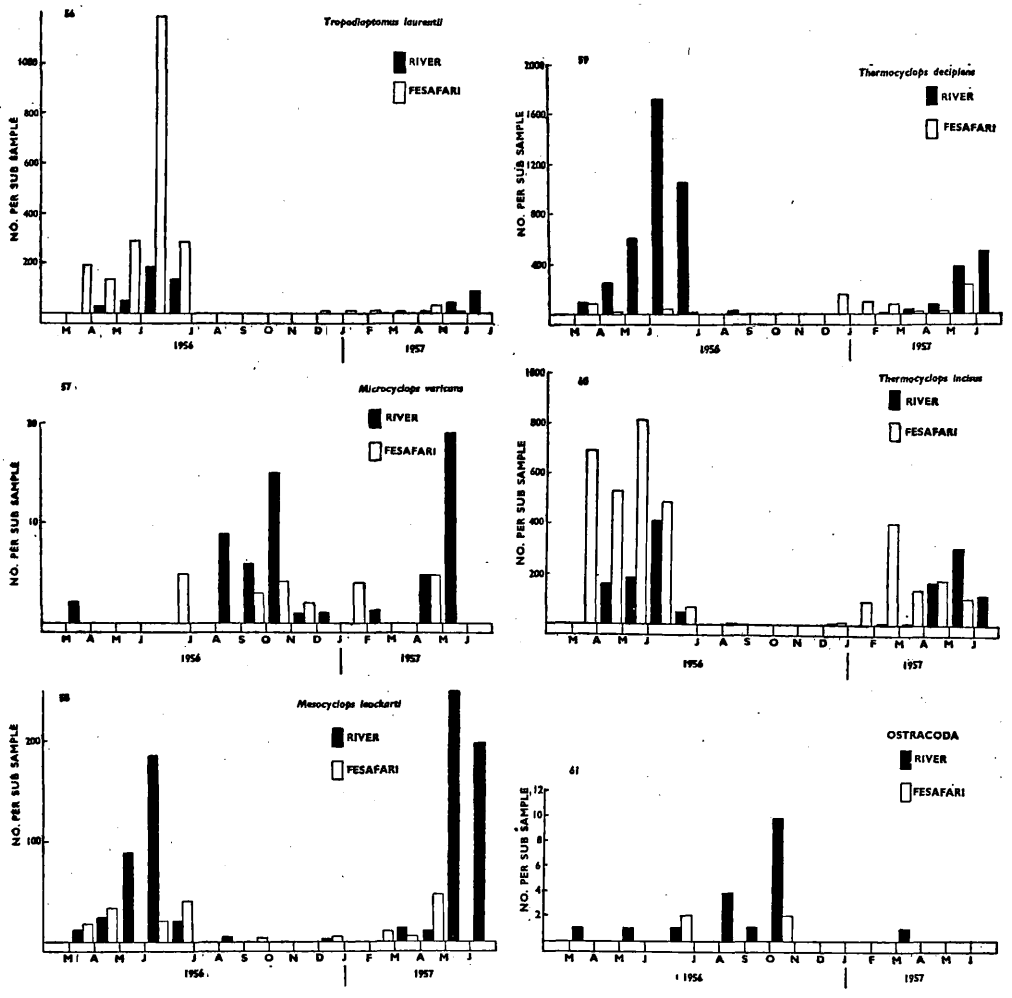
Ectocyclops compactus (Sars)

Single specimens were found in samples from the river in December 1956 and February 1957, and from Fesafari in November 1956. The antennules were short and did not reach the posterior border of the cephalothorax ; each antennule was formed by eleven podomeres, agreeing with Fryer's (1955) re-assessment of this species.

Ectocyclops phaleratus (Koch) sens. lat.

A single specimen of an *Ectocyclops* species was found in the sample taken from the river in December 1956. This specimen seems to belong to the *phaleratus* complex as reviewed by Fryer (1955). Some features of the specimen

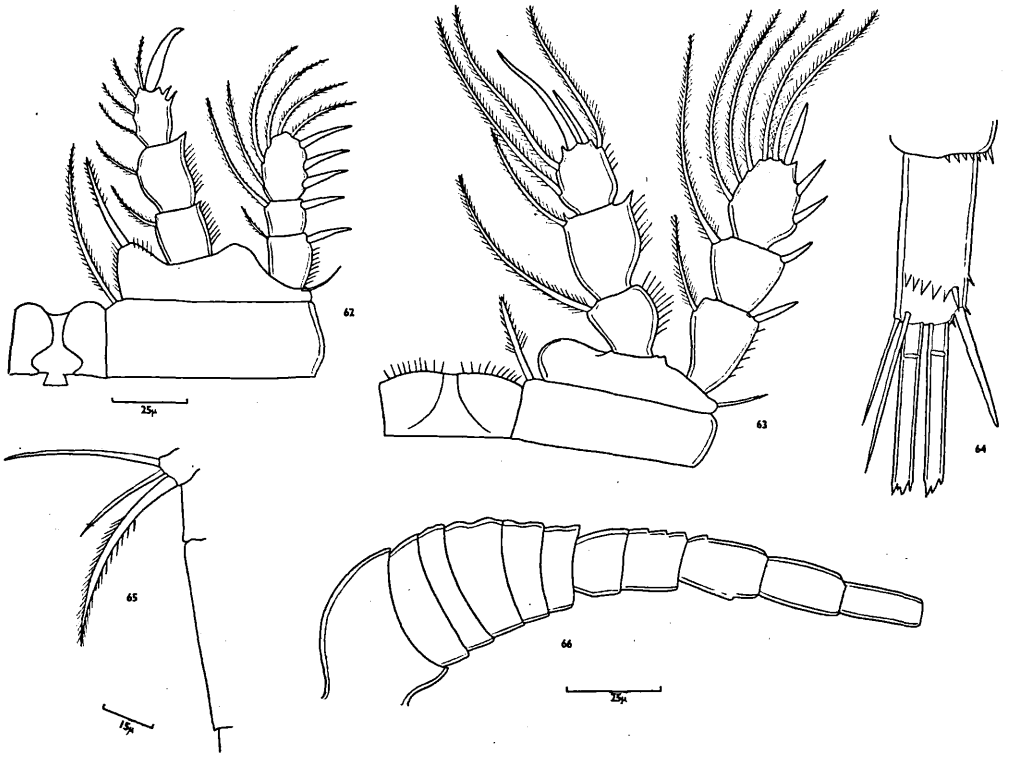
are shown in Figs. 62–66. There are some resemblances to *E. phaleratus ilariensis* Onabamiro (1952), for instance the two outer setae of leg 5 are smooth and shorter than the innermost seta, and the outer terminal seta of the caudal ramus is stout while the inner seta is very thin. However, each caudal ramus



Figs. 56–61—Seasonal occurrence in the plankton. 56, copepodid stages of *Tropodiaptomus laurentii*. 57, copepodid stages of *Microcyclops varicans*. 58, copepodid stages of *Mesocyclops leuckarti*. 59, copepodid stages of *Thermocyclops decipiens*. 60, copepodid stages of *Thermocyclops incisus*. 61, ostracods.

is a little over twice as long as wide, measuring 57μ in length and 25μ in width ; the uniting lamella of leg 4 has numerous fine setae and the basis of the leg does not have an inner spine. A further difference from *E. p. ilariensis* is found in the relative lengths of the spines at the apex of endopod 3 of leg 4. In the present specimen the inner spine is twice as long as the outer spine, but

in *E. p. ilariensis* the inner spine is over three times as long as the outer one. Each antennule is composed of eleven podomeres (Fig. 66).

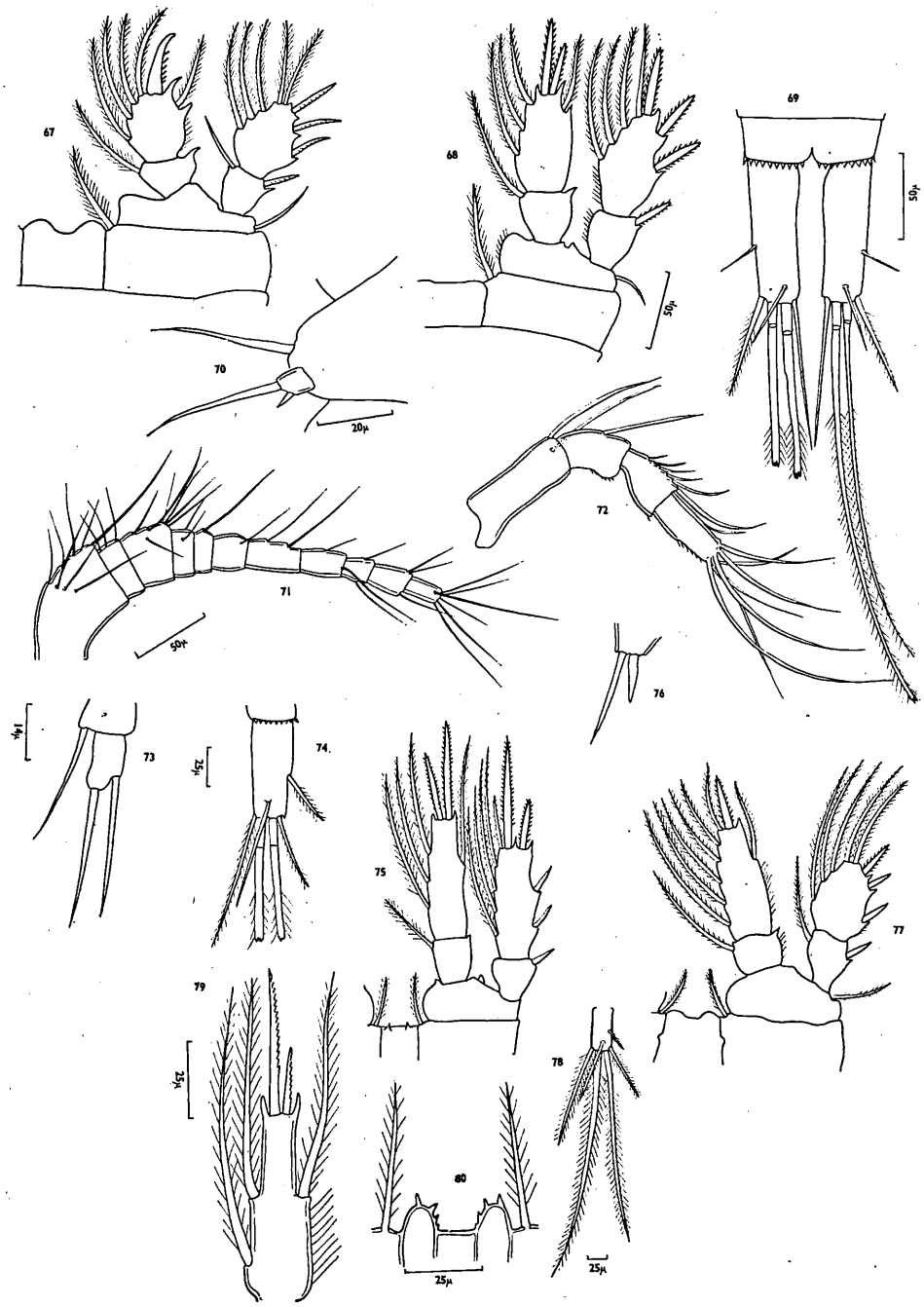


Figs. 62–66—*Ectocyclops phaleratus*. 62, leg 1. 63, leg 4. 64, caudal ramus. 65, leg 5. 66, antennule with all setae omitted. Figs. 62–64 are all to the same scale.

MICROCYCLOPS PSEUDO-ANCEPS sp. n.

Female. Total length, excluding furcal setae, 0.65 mm. The front of the head is rounded or slightly flattened. The genital segment is as long or a little longer than the last three segments. The receptaculum has distinct lateral arms. The caudal rami are about 82μ long and 25μ wide, with the innermost seta (length 93μ) longer than the outermost seta (length 65μ), and the dorsal seta (length 40μ) shorter than both these. The lateral seta is inserted about two fifths of the length of the ramus from the apex. The borders of the rami are smooth, but the last segment bears a series of small spinules around the base of each ramus (Fig. 69).

The antennules are short, not reaching beyond the posterior border of the cephalothorax ; each consists of eleven podomeres (Fig. 71). Each antenna has four podomeres (Fig. 72). The mouthparts are normal for the genus ; the claw of the maxilla bears four distinct teeth.



Figs. 67–80—Cyclopoids from the River Sokoto. 67–72, *Microcyclops pseudo-anceps* ; 67, leg 1. 68, leg 4. 69, caudal rami. 70, leg 5. 71, left antennule, ventral view. 72, left antenna, ventral view. Figs. 67, 68, 71 & 72 are all to the same scale.

73–77, *Cyclops* (*Diacyclops*) *gauthieri*. 73, leg 5. 74, caudal ramus. 75, leg 4. 76, leg 6. 77, leg 1. Figs. 74, 75 & 77 are to the same scale.

78–80, *Thermocyclops decipiens*, 78, caudal ramus. 79, leg 4, endopod 3. 80, connecting plate of legs 4.

The endopodites and exopodites of legs 1 to 4 all consist of two podomeres. The inner borders of the second basipodites of all these legs lack setae or spines. The spine formula is 3 4 4 3. The inner apical spine of leg 4 endopod 2 is one and a third times as long as the outer apical spine. The fifth leg consists of a single short podomere with a long terminal seta and a small but distinct inner spine.

The male is unknown.

This species runs down to *M. anceps* Sars in Lindberg's (1957) key to the species of *Microcyclops*, but it can be distinguished from that species by the presence of only eleven podomeres in the antennule ; the fifth leg is also somewhat shorter than in *anceps* as figured by Lowndes (1934) in his detailed redescription. A further difference from *anceps* is the absence of conspicuous setae on the face of the second podomere of the endopods of legs 2 to 4.

This species was only found in small numbers in the river in November and December 1956.

Microcyclops varicans (Sars)

The seasonal occurrence of this cosmopolitan species is known in Fig. 57. It occurred irregularly throughout the year, and reached its greatest abundance in the river in October 1956 and May 1957. Even at its most abundant it did not exceed twenty individuals per sub sample, so that it cannot be regarded as a serious competitor with the *Thermocyclops* species which occur in much greater numbers.

CYCLOPS (DIACYCLOPS) GAUTHIERI sp. n.

This species was described by Gauthier (1951) but he did not give it a name. The main diagnostic characters of the species are as follows. The antennules have only ten podomeres. The endopodites and exopodites of legs 1 to 4 are all formed of two podomeres. The terminal podomere of the endopodite of leg 4 is a little over three times as long as wide (Fig. 75) and the outer terminal spine is one and a half times the length of the inner terminal spine. There is some discrepancy between Gauthier's description of this podomere and my specimens, but since he had only a single specimen I think it is possible that the exopod may have overlaid the endopod with some confusion of the armature of the two rami. Leg 5 has two podomeres, the first bearing a long seta and the second bearing two long setae. Leg 6 bears a long outer seta and a shorter stouter inner seta.

The caudal ramus is shown in Fig. 74. The margins are smooth but the base is surrounded by a row of short, stout spinules. The posterior borders of the abdominal segments are also spinulated.

The correct genus for this species is difficult to assess ; the fifth leg is similar to that of *Mesocyclops*, but the reduction of the antennules to ten podomeres and the swimming legs to two podomeres in each ramus is more like the sub genus *Diacyclops* or even *Microcyclops*. The structure of the fifth leg prevents it from being placed in *Microcyclops*, so I have provisionally placed it in the sub genus *Diacyclops*.

This species was found in small numbers at irregular intervals throughout the year, both in the river and in Fesafari.

The only previous record is the unnamed description of a specimen from Senegal by Gauthier (1951).

Mesocyclops leuckarti (Claus)

This well-known and widespread species was much more abundant in the river than in Fesafari, reaching its maximum in June in 1956 and in May in 1957 (Fig. 58). In contrast to the most abundant cyclopoids, *Thermocyclops incisus* and *T. decipiens*, the numbers in 1957 were somewhat greater than in 1956. This is a larger, more robust, species than the *Thermocyclops*, and may well have different ecological requirements which were better suited in 1957 than in 1956. Fryer (1957b) has shown that *M. leuckarti* is a carnivorous species, while the food preferences of the two *Thermocyclops* species are as yet unknown.

The caudal rami of the Sokoto specimens are mostly a little less than three times as long as wide, so that they could be placed in the sub species *aequatorialis* of Kiefer (1929 b).

Thermocyclops decipiens Kiefer

This form is very closely allied to *T. hyalinus* Rehberg, and Gurney (1933) does not regard the two as separable. I have maintained *T. decipiens* as a separate form on the basis of the shape of the receptaculum, which agreed with the original figure given by Kiefer (1929 a). Some other features of the species are shown in figs. 78–80.

The seasonal occurrence of *T. decipiens* is shown in Fig. 59. It was much more abundant in the river than in Fesafari and reached its maximum early in June ; thereafter the population declined rapidly and the species was not found in most of the samples taken during the flood period of the river.

Thermocyclops incisus Kiefer

This species, which was originally described from West Africa (Kiefer, 1932 a), is very closely allied to *T. emini* (Mrazek), but the caudal rami are relatively shorter, being barely three times as long as wide, while in *T. emini* they are generally over three times, occasionally four times, as long as wide. The dorsal seta on each ramus is slightly shorter than the innermost seta.

The seasonal occurrence of *T. incisus* is shown in Fig. 60. The population increased more rapidly in Fesafari than in the river and reached a greater density.

The occurrence of this species contrasts in several respects with the occurrence of *T. decipiens*. The later species was abundant in the river, but scarce in Fesafari, while *T. incisus* was abundant in Fesafari, but also fairly abundant in the river. The earlier reduction of rate of flow through Fesafari enabled the population of *T. incisus* to increase more rapidly in the pool, probably at the expense of *T. decipiens* which thrives better in the main channel of the river.

Order *LERNAEOIDA*

Family **Lernaeidae**

Lernaea sp.

Copepodid stages of a *Lernaea* species were found in the samples at irregular intervals throughout the year. The adults are parasitic on fish, and the characters used to separate species are mostly to be found only in the adults, so that any further identification of the copepodids is not practicable.

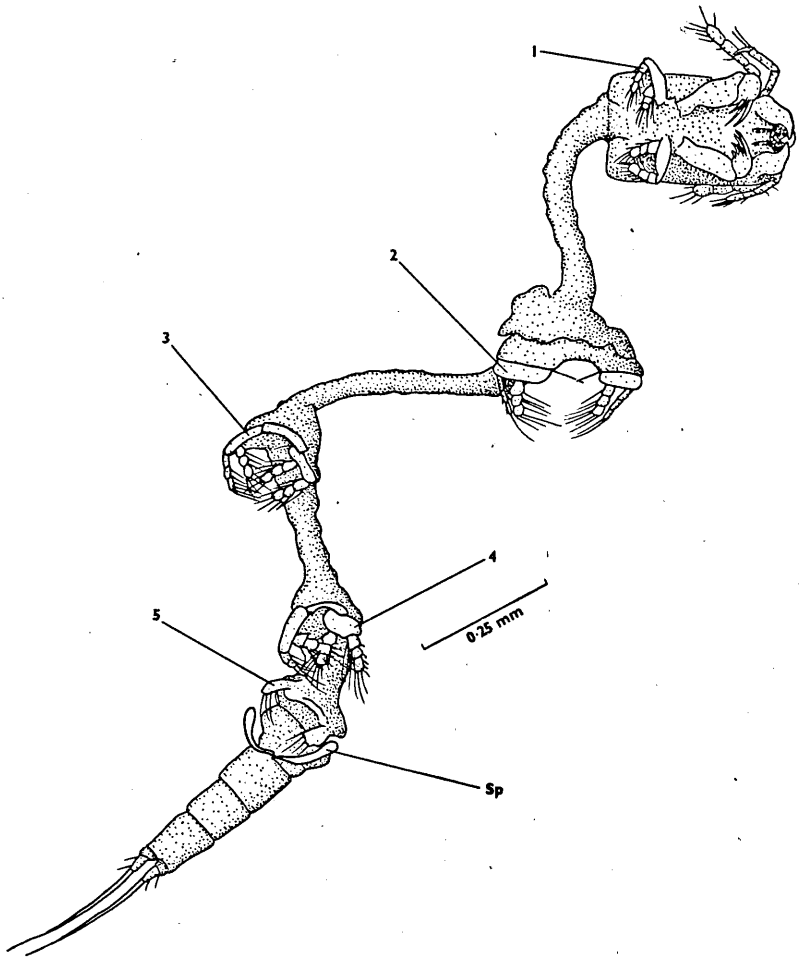


Fig. 81—*Lernaea* sp., early stage in metamorphosis ; legs 1–5 have been numbered. Sp–spermatophore.

An interesting stage was found in the sample taken from the river in June 1956 (Fig. 81). This is an early stage in metamorphosis which must have been dislodged from a fish. The body has elongated, separating the swimming legs, but it has not begun to swell into the characteristic adult form. The head still retains its larval characteristics without any development of the antler-like processes found in the adult.

Order *BRANCHIURA*

Family **Argulidae**

Argulus sp.

A single specimen of a larva of an *Argulus* species was found in the sample taken from Fesafari in April 1957. Because records of argulids from West Africa are scarce I have illustrated this larva even though it is not yet possible to assign it to a species. The presence of only two sharp spines on the basipodite of the maxilla, and the form of the respiratory areas (Fig. 82) are features which may prove to be of importance in identifying this larva.

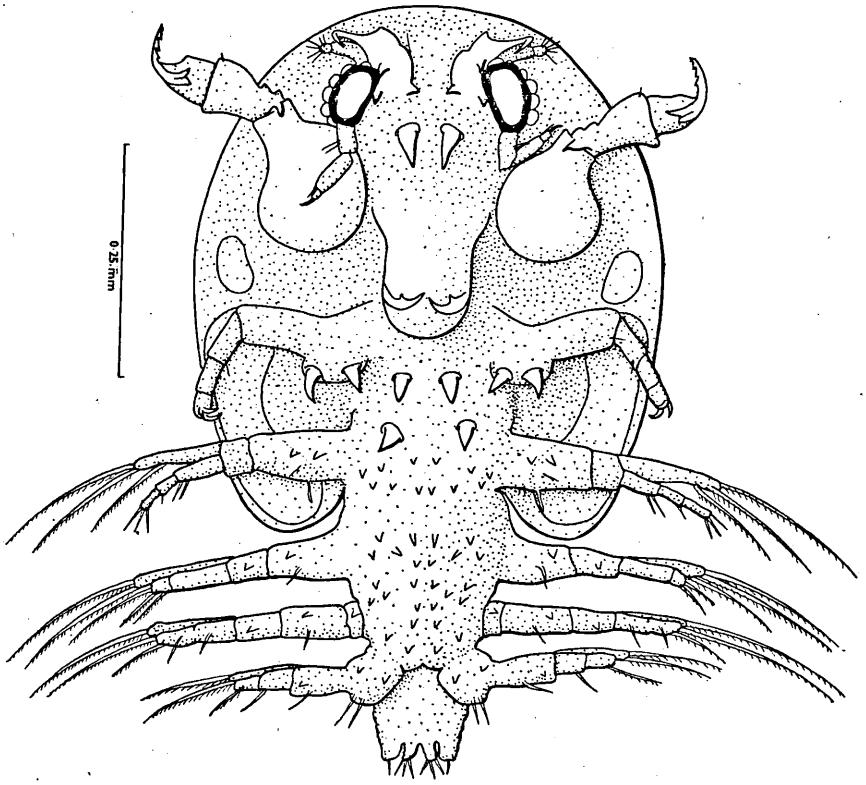


Fig. 82—Larva of *Argulus* sp.

Sub-class **OSTRACODA**

Ostracods were only found in small numbers in the samples, mainly in the river during the flood period (Fig. 61). Many of the specimens were immature and so could not be determined to specific level.

Family **Cypridae**

Cypretta globula Sars

This species was originally described from Australia by Sars (1889) as a member of the genus *Cypridopsis*, and was later transferred to *Cypretta* when recorded from South Africa (Sars, 1924). Two species of *Cypretta* have been recorded from the region of Lake Chad by Gauthier (1939) who gives diagnostic characters which clearly separate them from *C. globula*.

This species was responsible for the highest number of ostracods recorded in the river, during October 1956. A few specimens were also found in the sample taken from the river in August 1956.

Cypridopsis sp.

Klie (1935) has remarked that Africa is the Dorado of *Cypridopsis*, there being about sixty species in the genus and about two thirds of these are African. In view of this and the fact that the descriptions of many of the species described as new by Sars (1924) were concerned only with the shell I have not attempted to identify to specific level the two specimens which clearly belonged to this genus and were found in the sample taken from the river in August 1956.

PHYSOCYPRIA MINICAPENSIS sp. n.

Single specimens of a *Physocypria* species were found in the samples from the river and from Fesafari in October 1956. Both specimens were males.

The length of the shell is about 0·43 mm. The valves have fine but distinct longitudinal striae which branch and anastomose. The left valve overlaps the right valve anteriorly. The anterior margin of the right valve bears small but distinct tubercles. When viewed from above the width of the shell is less than one third of the length. The limbs are similar in most respects to the limbs of *P. capensis* (Sars). The palp of the right maxilliped is larger than the left and the proximal podomere is produced into a sharp spine. The caudal furca is shown in Fig. 90 ; the anterior claw is 54μ long and the posterior claw 36μ long. The seta on the posterior border is very inconspicuous.

Each Zenker's organ has seven wreaths of chitinous spines (Fig. 86), and the proximal end of the tube is expanded and bladder like. The copulatory appendage has one lamella broadly triangular while the other is much narrower (Fig. 87).

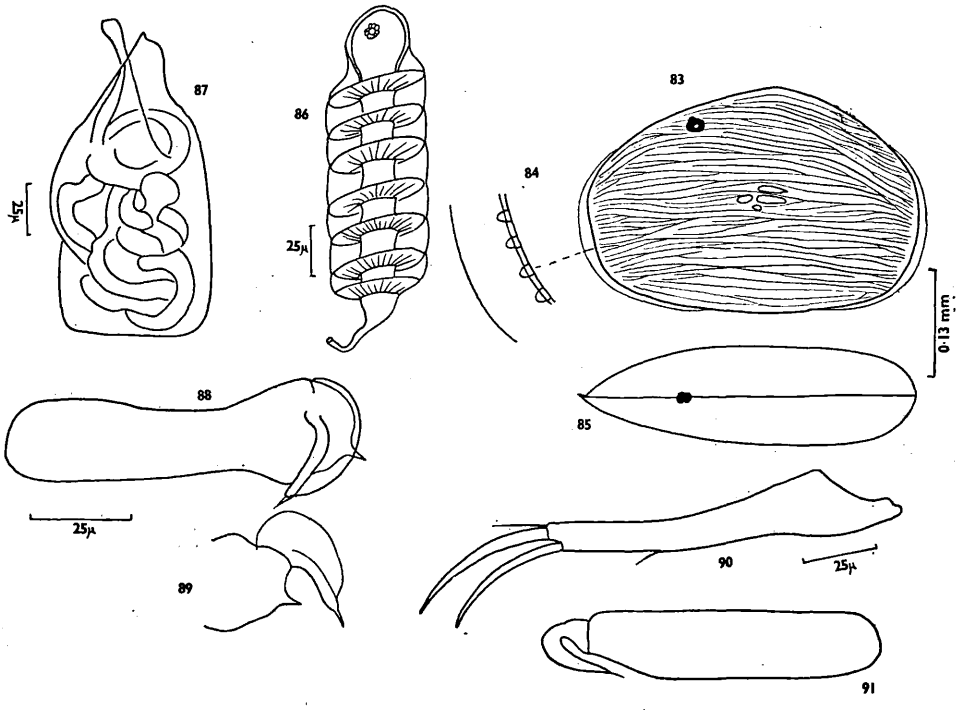
This species is closely allied to *P. capensis*, but can be distinguished from that species by the following characters. The shell is smaller and the striations are more distinct. The palps of the maxillipeds, particularly the right one have a different shape (cf. Sars, 1924, plate X). The posterior border of each furcal ramus bears only a small inconspicuous seta instead of a long and distinct one. Finally, the copulatory appendage has one lamella much more broadly triangular than the other.

The female is unknown.

Family **Darwinulidae**

Darwinula sp.

A single specimen belonging to this genus was found in the sample taken from Fesafari in June 1956. The specimen was somewhat damaged and specific identification was not possible.



Figs. 83–91—*Physocypria minicapensis*, male. 83, lateral view of shell. 84, anterior margin of right valve. 85, dorsal view of shell. 86, Zenker's organ. 87, copulatory appendage. 88, palp of right maxilliped. 89, apex of palp of right maxilliped in open position. 90, furcal ramus. 91, palp of left maxilliped, to same scale as 88.

Sub-class **MALACOSTRACA**

Order *DECAPODA*

Family **Atyidae**

Caridina africana Kingsley

This small shrimp was only found in one of the samples, taken from Fesafari in January 1955. The rostrum reaches the end of the peduncle of the antennule, but is not quite as long as the antennal scale. The dactylopodite of leg 3 is about a quarter of the length of the propodite, and the dactylopodite of leg 5 is a little over a quarter the length of the corresponding propodite.

This species is widespread in Africa and numerous varieties have been described, but they are not always clearly separable and intermediates are frequent.

SEASONAL VARIATION IN NUMBER OF SPECIES

The total number of crustacean species observed in this study is fifty-one ; samples from the river yielded forty-five species, twenty-seven of which were also found in the pool Fesafari. The samples from Fesafari yielded thirty-three species, of which six were not found in the main channel of the river. Most of these species which were not shared by the river and the pool were distinctly rare ; several of the species recorded from the river were only found as single specimens.

In considering the number of species in each monthly sample one finds a marked contrast between the Cladocera and the Cyclopoida (Tables 2 & 3). The Cladocera show an increase in number of species when the river is in flood. This increase occurs in spite of a great decrease in the total number of individuals. The Cyclopoida on the other hand show a decrease in the number of species during most of the time that the river is in flood. Remarkably enough the greatest number of cyclopoid species is found in December when the river level is dropping rapidly (cf. Fig. 1). An explanation of this might be found in the lowered temperature at this time of year, for this is the time of the harmattan, the cold wind which blows southwards from the Sahara. The lowered temperature might in part be responsible for the occurrence of cyclopoid species which are widespread in temperate regions, such as *Macrocyclops albidus*, *Eucyclops serrulatus* and *Ectocyclops phaleratus*.

Table 2—Seasonal variation in the number of species of Cladocera present in each sub sample.

Month	Number of species in main channel	Number of species in Fesafari	Total number of species in both samples
March 56	5	4	6
April 56	4	4	6
May 56	4	4	5
June 56 (early)	3	5	5
June 56 (late)	7	6	9
July 56	no sample	no sample	—
August 56	11	no sample	—
September 56	14	10	16
October 56	16	9	17
November 56	15	8	16
December 56	2	5	7
January 57	no sample	7	—
February 57	8	4	9
March 57	5	4	6
April 57	5	4	7
May 57	5	2	5
June 57	5	no sample	—

Among the Cladocera several of the flood water species are found in the plankton for a shorter period in the pool Fesafari than in the main channel of the river. This is particularly well shown by *Alona diaphana*, *A. pulchella* and *Graptoleberis testudinaria*. As the river rises it sweeps these bottom dwelling species up into the plankton, and continues to keep them afloat when the level falls and Fesafari becomes isolated so that the individuals in the pool can settle down earlier.

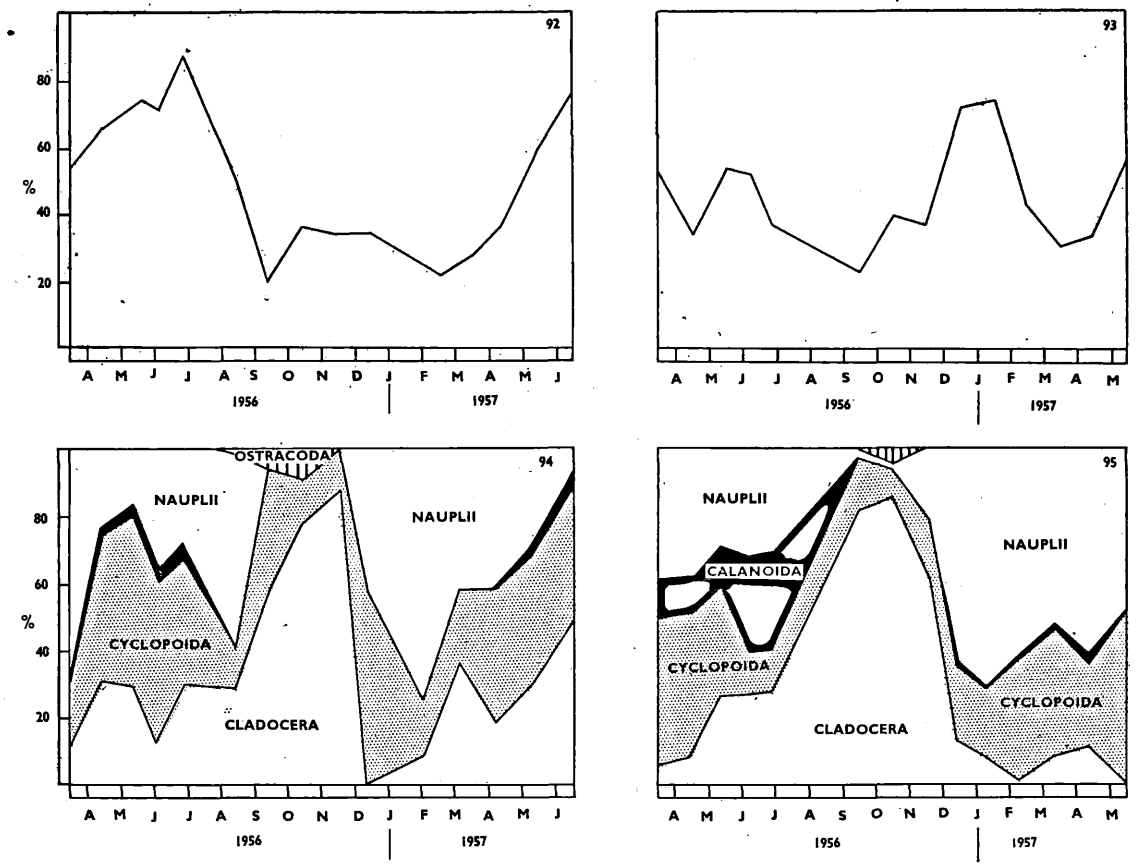
Table 3—Seasonal variation in the number of species of cyclopoids present in a sample of fifty individuals.

Month	Number of species in main channel	Number of species in Fesafari	Total number of species in both samples.
March 56	5	3	6
April 56	3	4	4
May 56	4	2	4
June 56	3	5	5
July 56	no sample	no sample	—
August 56	4	no sample	—
September 56	1	2	2
October 56	1	1	1
November 56	2	2	3
December 56	8	3	9
January 57	no sample	3	–
February 57	4	4	6
March 57	6	3	6
April 57	4	4	4
May 57	4	2	4
June 57	4	no sample	–

The effect of rate of flow in increasing the number of cladoceran species taken in the plankton samples is only shown during the floods ; during the dry season the numbers of species in the main channel and in Fesafari are very similar. This is in contrast with the effect found in the Rotifera (Green, 1960), which showed a greater number of species in the main channel throughout the dry season. This difference between Cladocera and the Rotifera is probably a reflection of the larger size and superior locomotory powers of the Cladocera which enable them to maintain themselves in the correct habitat in the face of moderate water movements.

DISCUSSION

The seasonal occurrences shown in the figures can be divided into two main types. The first shows an increase in number of individuals during the dry season when the river level is low, and shows a rapid decrease as the level of the river rises ; it is to this group that all the truly planktonic Crustacea belong. The second type shows an increase in the number of individuals found in the samples as the river level rises and a decrease as the river falls. The numbers of individuals involved is much smaller than in the case of the truly planktonic species. A possible third type shows no definite cycle, but erratic occurrences of small numbers at irregular intervals ; this group cannot be clearly separated from the second group because the irregular occurrences tend to be more frequent during the floods. These last types of seasonal occurrence involve animals which are not strictly planktonic, and whose occurrence in the plankton depends mainly on water movements. Another possible factor would be the build up of a bottom or weed dwelling population followed by active movement into the plankton for a short period when the population became dense.



Figs. 92–95—Seasonal variation in the relative importance of Crustacea in the zooplankton of the River Sokoto and the pool Fesafari. 92, Crustacea as percentage of the total zooplankton in the river. 93, Crustacea as percentage of the total zooplankton in Fesafari. 94, Composition of the crustacean zooplankton in the river ; each group is plotted as a percentage of the total number of Crustacea. 95, Composition of the crustacean zooplankton in Fesafari.

The different types of seasonal occurrence have the result that two distinct plankton assemblages can be distinguished. During the dry season two cladocerans, *Diaphanosoma excisum* and *Moina dubia*, two cyclopoids, *Thermocyclops incisus* and *T. decipiens*, and, in 1956, the calanoid *Tropodiaptomus laurentii*, form the main bulk of the plankton. As the river level rises these species disappear and are replaced by much smaller numbers of flood water species. The flood assemblage is dominated by the *Alona* species, which sometimes form over eighty per cent of all the Cladocera between September and November. Among the cyclopoids *Microcyclops varicans* is the main species found in the flood water, indeed in some of the samples it is the only cyclopoid, but its occurrence is not restricted to the flood periods ; small numbers were found at irregular intervals throughout the year.

The Crustacea form a very high proportion of the total zooplankton (Figs. 92 & 93), but they form a higher proportion of the plankton in the river in June than at any other time, while in the pool the proportion of Crustacea is highest in December and January. It must be remembered that we are here considering the Crustacea as a proportion of the total zooplankton, and the absolute numbers shown in Fig. 1 should be borne in mind throughout this comparison.

The relative numerical importance of the various groups of Crustacea during the whole year is shown in Figs. 94 & 95, where each group is plotted as a percentage of the total Crustacea. A remarkable feature brought out by these figures is the overwhelming dominance of the Cladocera during the floods. The absolute numbers of Crustacea fall to a very low level during the floods, but of those that are found the great majority are cladocerans.

The effect of rate of flow on the occurrence of ostracods in the plankton is also shown in Figs. 94 and 95. The less turbulent conditions in the pool are reflected in the lesser relative abundance of ostracods and in their shorter duration of occurrence.

When the planktonic Crustacea of the River Sokoto are compared with those found in other rivers there are many similarities, but also differences. Table 8 compares the Sokoto with the Illinois (from data given by Kofoid, 1908) and the Nile (from data given by Brook and Rzòska, 1954, and Rzòska, 1957). A striking feature of this comparison is the similarity of the cladoceran populations in the three rivers. The most important species in the Illinois differ from those in the Sokoto and the Nile, but belong to the same genera. The similarity is less marked in the cyclopoids, though it should be noted that *Mesocyclops edax* is closely related to *M. leuckarti*, and Kiefer (1929) regarded it as a sub species of *M. leuckarti*. The calanoids show the widest divergence, a feature which is in agreement with the fact that freshwater calanoids are generally much more restricted geographically, both in species and genera, than the cyclopoids and Cladocera.

A further point brought out by this comparison is that the number of species in each of the three groups is of the same order in the three rivers. The figures for Cladocera and Cyclopoida in the Nile are at present too low, but even in their present state they indicate that in each river the number of species of Cladocera is about three times as high as the number of cyclopoid species. In each river there is only one really abundant calanoid ; the higher total of

Table 4—Comparison of the planktonic Crustacea of the Rivers Sokoto, Illinois and Nile.

CLADOCERA	Sokoto	Illinois	Nile
Total number of species (including adventitious spp.)	30	25	18*
Dominant planktonic species.	*Moina dubia*	*M. micrura*	*M. dubia*
	Diaphanosoma excisum	*D. brachyurum*	*D. excisum*
	Ceriodaphnia cornuta	*C. scitula*	*C. cornuta*
	—	*Daphnia cucullata*	*D. lumholtzi*
CYCLOPOIDA			
Total number of species	11	8	4*
Dominant planktonic species	*Thermocyclops incisus*	—	*T. neglectus*
	Thermocyclops decipiens	—	—
	Mesocyclops leuckarti	*M. edax*	*M. leuckarti*
	—	*Cyclops bicuspidatus*	—
CALANOIDA			
Total number of species	2	3	5
Dominant planktonic species	*Tropodiaptomus laurentii*	—	—
	—	*Diaptomus siciloides*	—
	—	—	*Thermodiaptomus galebi*

*The numbers of species in the Nile are probably too low because full lists have not yet been given. The numbers in this Table represent the minimum deduced from the list of genera given by Brook & Rzòska (1954).

calanoid species in the Nile is due to a detailed examination by Rzòska (1957) over a great geographical distance including both the White and Blue Nile.

Certain of the genera and species in Table 4 occur in river plankton in other parts of the world ; *Diaphanosoma brachyurum* for instance has been found in the Sungari River, Manchuria (Ueno, 1936), in the Elbe at Dresden (Schorler, 1900), in the Danube at Vienna (Steuer, 1901) and in the Volga (Behning, 1938). It is however by no means restricted to rivers and forms an important constituent of lake plankton in various parts of the Northern Hemisphere ; to take only a few of many examples, it occurs in Lake Mendota (Birge, 1897), Frederiksborg Castle Lake, Denmark (Berg & Nygaard, 1929), and in the Lago Maggiore (Pirocchi, 1947). This introduces the question as to whether any of the Crustacea can be regarded as belonging only to the river plankton, or potamoplankton in the terminology of Zacharias (1898). In fact there does not seem to be a single species which is not also found in ponds or lakes. Zimmer (1898) noted this over sixty years ago, and no definite examples have been found since. Nevertheless some species thrive better than others in rivers ; we have already seen (p. 441) how the two species of *Thermocyclops* in the Sokoto differ in that one is more abundant in the river while the other is more abundant in the pool Fesafari.

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Bile Pigment in *Eucypris virens* (Jurine)

Eucypris virens (Crustacea : Ostracoda) is a common ostracod with green patches on the valves of its carapace. When treated with yellow nitric acid these green patches become purple, then red, orange and finally yellow. This sequence is typical of the later stages of the Gmelin reaction, indicating the presence of a bile pigment.

Several hundred *E. virens* were soaked overnight in acid methanol (5 per cent hydrochloric acid), to give a clear blue-green solution with an absorption maximum at 655 mμ. The pigment passed readily into chloroform after dilution of the acid methanol with water. In dry chloroform absorption maxima were found at 362 and 660 mμ. The chloroform solution was then evaporated and the pigment taken into methanol. The solution in methanol did not fluoresce in ultra-violet light, nor did it fluoresce on the addition of zinc acetate, but when a trace of iodine was added a brilliant pink fluorescence appeared. This fluorescent solution, stabilized with a trace of ammonia, showed a strong absorption maximum at 625 mμ. This indicates that the pigment was mesobiliverdin, or a pigment very similar to it.

A pigment with similar properties has been obtained by cold acid hydrolysis of *C*-phycocyanin[1]. This raises the possibility that the original pigment in the ostracod was not a bilatriene of the mesobiliverdin type, but a biladiene similar to those occurring in the blue-green algae on which this ostracod feeds. This possibility was tested by taking fresh *E. virens* which had been passed through several changes of clean water to allow them to empty their intestines, and homogenizing them in 2 per cent hydrochloric acid in methanol. The blue-green solution was cleared by centrifuging, then diluted with water and the pigment taken, not very easily, into chloroform. The chloroform solution was washed with dilute sodium bicarbonate, and a small volume of methanol was added. This solution showed a pink fluorescence in ultra-violet light, and the fluorescence was greatly intensified by the addition of zinc acetate. Addition of iodine increased the fluorescence slightly, but then turned it to a pale green. The pink fluorescence was completely quenched by the addition of mineral acids, thus distinguishing it from the fluorescence of por-

phyrins, and indicating that the pigment was a biladiene of the *a*, *b* type.

It has previously been shown[2] that another ostracod, *Heterocypris incongruens* (Ramdohr), accumulates biladienes, derived from blue-green algae, in the form of small granules in the wall of its gut, but not outside the gut in the valves of the carapace. *Eucypris virens* also accumulates biladienes in the wall of its gut, but in addition transfers some to the valves of its carapace. The mesobiliverdin obtained from the first extract was due to the hydrolysis of the biladiene in the strong acid; when the pigment was extracted in weaker acid and examined immediately to minimize hydrolysis the indications were that in the ostracod the pigment was present as a biladiene.

It is probable that the green colour in the carapace valves of other ostracods is due to bile pigments: the patches on the shell of *Cypridopsis aculeata* (O. G. Costa) give a positive Gmelin reaction.

J. GREEN

Department of Zoology,
Bedford College,
London.

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SEASONAL POLYMORPHISM IN *SCAPHOLEBERIS MUCRONATA* (O. F. MÜLLER) (CRUSTACEA: CLADOCERA)

By J. GREEN

Bedford College, University of London

The following account gives a picture of the seasonal variation in form of *Scapholeberis mucronata* (O. F. Müller) in one particular habitat. This cladoceran has, at least when young, clearly defined spines on the head and at the posterior ventral extremity of the carapace (Fig. 1). For the sake of convenience these will be referred to as the head spine and the tail spine. Earlier observations on seasonal variation in the length of these spines have been made by Gruber (1913), and isolated samples have been measured by Rammner (1927), but these authors examined relatively few samples and missed several basic features of the seasonal cycle. Gruber's main conclusion was that the head spine decreased during the life of an individual, and decreased with successive generations as the season progressed. As will be seen from the following account the first part of this conclusion is not correct at all seasons, and the second is a misinterpretation of the nature of the seasonal cycle.

METHODS

Each sample was collected from Hampton Court Long Water and examined on the same day. All observations and measurements were made on living animals, so that artifacts due to fixation and preservation were avoided. The samples were collected with a fine-meshed bolting silk hand net. The mesh of this net (180/in., 7·2/~~cm~~) was fine enough to retain all stages of *Scapholeberis*. No attempt was made to estimate the population in terms of number per unit area or volume. Simple inspection of the habitat was sufficient to show that figures of population density obtained by the application of any of the usual random sampling methods would be quite inadequate unless applied on a vast and unrealistic scale. The animals often occurred in small distinct swarms, so that one sample might contain several thousand individuals, while another taken only a few decimetres away would contain less than fifty. The density of the population was estimated and recorded under the following categories:

1. One to twenty specimens collected in 10 min.
2. Twenty to one hundred specimens collected in 10 min.
3. Small numbers visible in the water without the use of the hand net, several hundred collected in 10 min.
4. Small swarms visible in the water, several thousands collected in less than 10 min.
5. Dense swarms forming a band several decimetres wide along the edge of the water; possible to collect over a million if required.

This scale is sufficient to give a general picture of the seasonal abundance of *Scapholeberis*, but is not sensitive enough to record small fluctuations of the population.

The composition of the population was estimated by taking random samples from the main sample with a wide pipette and ejecting them on to a glass plate, then draining off most of the water and examining under a binocular microscope. In this way a sample

of at least 100, more frequently about 300, individuals was examined. The categories into which specimens were grouped were as follows:

1. Females with parthenogenetic eggs or embryos in the brood pouch, or with large ovaries indicating that such eggs were about to be laid.
2. Females of mature size without eggs or large ovaries, but possessing the long abdominal process with which eggs are retained in the brood pouch. These are females which have become temporarily or permanently sterile.
3. Females with ephippia, or with the carapace showing signs of ephippium formation and the corresponding appearance of ephippial eggs in the ovary.
4. Immature females, smaller than the mature females and lacking the long abdominal process which retains eggs in the brood pouch.
5. Males, including immature specimens.

The lengths of the body and the spines were measured between the limits shown in Fig. 1. All measurements were made with the same calibrated eyepiece micrometer.

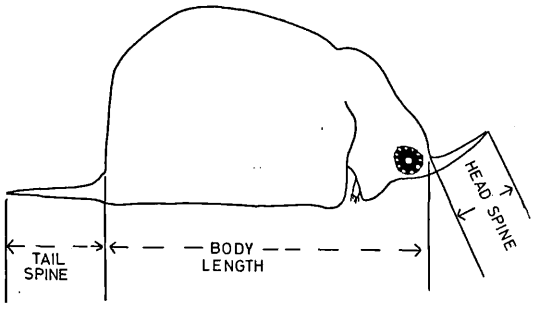

FIG. 1. Lateral outline of the head and carapace of an adult female of *Scapholeberis mucronata* to show the limits between which measurements were made.

Egg numbers were counted after dissecting the eggs or embryos from the brood pouch of each female under a low-powered binocular microscope. All the embryos in any one brood were found to be at the same stage of development, and each brood is retained in the pouch until the young are liberated as miniature immature adults. Estimates of egg production per female can thus be based on both eggs and developing embryos. For convenience the term 'egg number' is used irrespective of whether eggs or embryos were counted. The time interval between broods is only a few days, and so is much less than the interval between samples. This means that the egg number will reflect conditions of nutrition and temperature close to the time at which each sample was taken.

Water temperature was measured at 09.00 hours (GMT or BST according to season) on days that collections were made. A total-immersion thermometer was held about 10 cm below the surface of the water and the temperature read after 2 min with the thermometer completely immersed.

The chlorophyll content of the water was estimated by taking 100 ml and filtering through a Millipore DA filter with a pore diameter of 0·65 μ. This gives a high rate of filtering while retaining nannoplanktonic algae. The filter and the algae retained by it were homogenized with acetone in a Griffith type thimble homogenizer. The resulting homogenate was filtered through Whatman's filter paper which had been soaked in

acetone, and during filtration the filter funnel was covered to prevent evaporation. The volume of filtrate was made up to 5 ml and the absorption of light through a 1 cm path at a wavelength of 665 mμ was measured in a Unicam SP 500 spectrophotometer. Measurements of absorption were also made at 10 mμ intervals between 400 and 460 mμ to check on the possibility that different chlorophylls might be important at different times of year. This method of estimating chlorophyll gave consistently reproducible results with samples from the Long Water, and no signs of incomplete extraction were ever noted. The method may not be applicable to all freshwater phytoplankton; some attempts to estimate the chlorophyll content of samples from Regent's Park Lake failed because the acetone did not extract all the pigment.

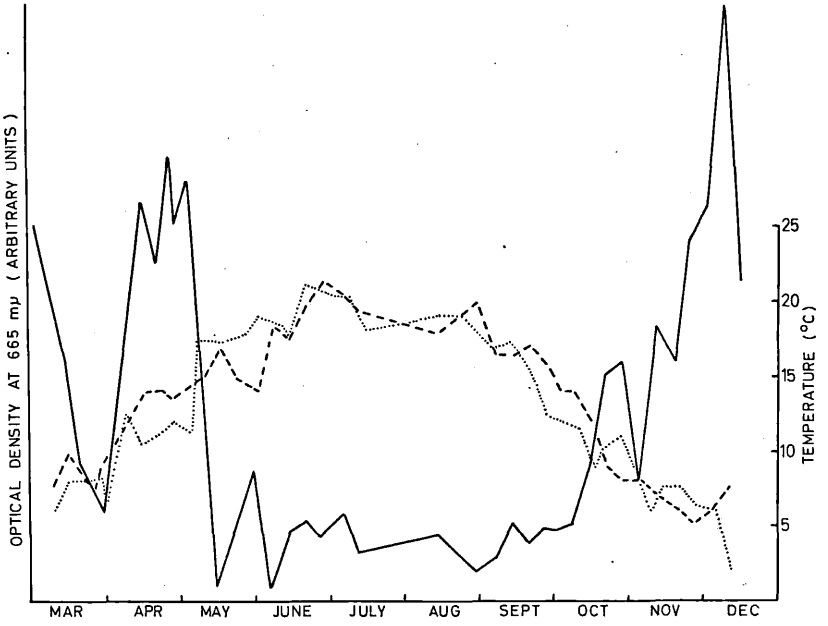

FIG. 2. Seasonal variation in temperature (. . . . . , 1960; - - - -, 1961) and chlorophyll content (———, 1961) of the water in Hampton Court Long Water.

THE ENVIRONMENT: TEMPERATURE AND FOOD

Hampton Court Long Water is rectangular in shape, about 1000 m long and 30 m wide. The greatest depth is about 2 m. There is a controlled overflow at the end away from Hampton Court Palace, so that the water level can be maintained fairly constant. This gives great stability to the margins of this body of water, while it retains a pond-like character. These conditions, coupled with the abundant vegetation along the margins and the patches of water lillies (*Nymphaea*) at intervals in deeper water, make the Long Water an ideal place for the development of considerable populations of Cladocera. The vegetation is important in providing sheltered areas of still water which are particularly favoured by *Scapholeberis mucronata*.

Temperature measurements are shown in Fig. 2. The 1960 spring was distinctly cooler than that of 1961, and the earlier appearance of *S. mucronata* in 1961 can be related to a period of warm weather, just before 14 March, which raised the temperature to 10° C

by that date. The water temperature did not reach 10° C until after the beginning of April in 1960.

The chlorophyll content of the water, which can be regarded as an estimate of the food available to Cladocera, was not measured during 1960, but in 1961 it showed a distinct peak during April, with a rapid fall at the beginning of May. During the summer there were small fluctuations, but the level was generally low. There was a remarkable increase in the amount of chlorophyll towards the end of the year (Fig. 2).

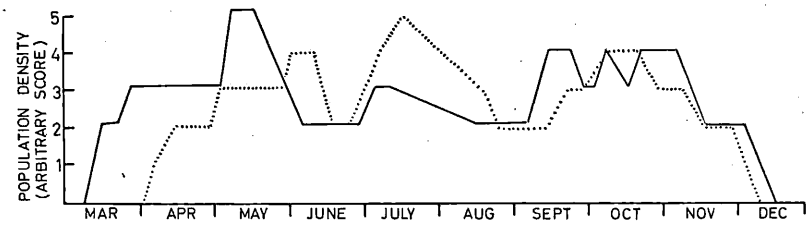

FIG. 3. *Scapholeberis mucronata*: seasonal variation in population density (. . . . . , 1960; ———, 1961). See text for explanation of the density scale.

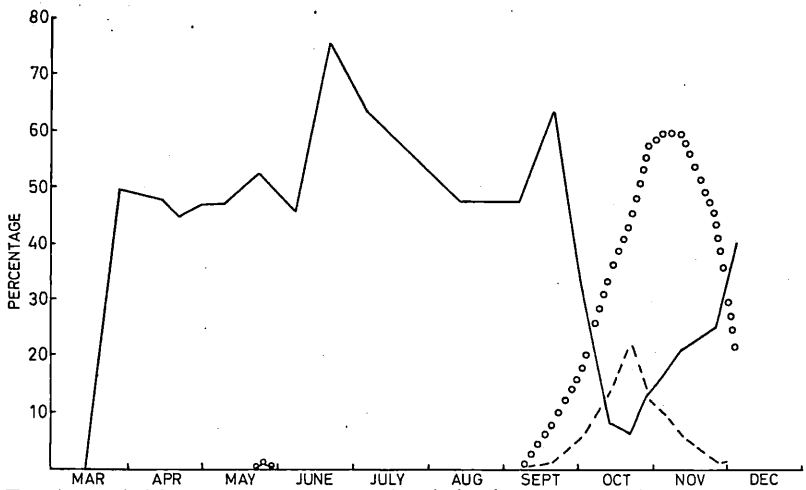

FIG. 4. *Scapholeberis mucronata*: seasonal variation in the composition of the population (———, females with eggs; OOOOO, ephippial females; — — —, males).

POPULATION DENSITY AND COMPOSITION

Estimates of population density made during the years are shown in Fig. 3. In each year there were indications of three main pulses; the first in April and May, followed by a somewhat reduced population in June. In both years a small number of ephippial females appeared as the first pulse was declining (Fig. 4). There were indications of an increased population in July, with lowered densities in August. From the middle of September the population was dense and increasing numbers of females were found with ephippia. The actual percentages were not recorded in 1960, but at the beginning of November 1961 about 60% of the whole population was made up by females with, or about to produce, ephippia.

Males were produced in small numbers at the end of May 1961, then disappeared from the samples until the middle of September, when they reappeared with the ephippial females. The highest percentage of males was recorded on 23 October, which is somewhat before the peak production of ephippia. Thereafter the percentage of males dropped rapidly.

The percentages of females with parthenogenetic eggs or embryos in the brood pouch showed several interesting features during 1961. The rapid rise at the end of March was due to the maturation of females hatching from ephippia. The two obvious peaks in Fig. 4 are related to periods when the egg number was particularly low (see Fig. 5). This resulted in the percentage of immature females being very low, so that the percentage of mature animals was higher than usual. The dramatic fall in percentage carrying eggs at the end of September is clearly related to the changeover to sexual reproduction. There was a gradual increase in the percentage carrying eggs during November, but the number of eggs per female was falling and the population diminished, so that on 11 December only a single specimen was found after an intensive search, and none was found after that date until the following spring.

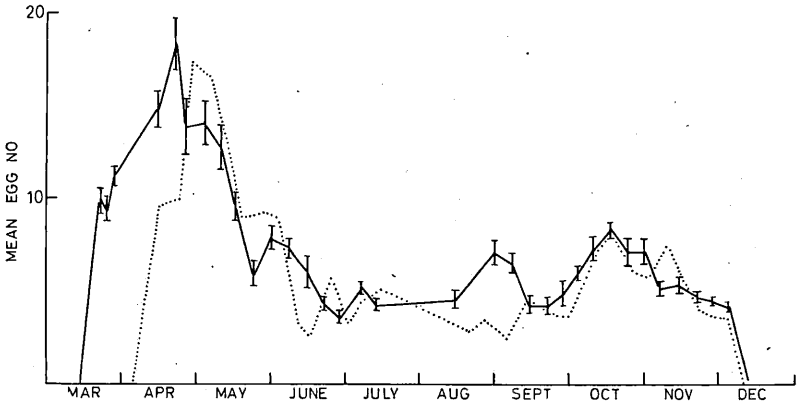

FIG. 5. *Scapholeberis mucronata*: seasonal variation in egg production (. . . . . , 1960; ———, 1961). Each point is based on twenty females with eggs; the short vertical lines indicate one standard error above and one below the mean.

SEASONAL VARIATION IN EGG PRODUCTION

In 1960 the first immature female was found on 7 April, and by 14 April the first eggs were present in the brood pouches of mature females. The mean egg number then rose rapidly to a maximum of 17·4 on 27 April, then began to fall until by 14 June there were only two or three eggs in the brood pouch of each female. The number remained low, with small fluctuations throughout the summer, but showed a distinct rise in October, and finally fell at the end of November.

The same sequence of events was repeated in 1961, except that the first females with eggs were found much earlier. All the females brought into the laboratory on 14 March were immature, but some were in their adolescent intermoult (*cf.* Green 1956), and laid their first eggs on the following day. The season thus started almost a month earlier than in 1960. The mean egg number rose to a maximum of 18·3 on 20 April then began to fall, but it did not fall as rapidly as in 1960. There was again a distinct rise in egg number during October, and the final fall came at the beginning of December.

The high egg production in April can be related to the large amount of food available at this time, as indicated by the chlorophyll estimates. The increased egg number in October can also be related to increasing amounts of chlorophyll, but it should be noted that the percentage of females carrying eggs was low in October. High density of population and decreasing temperatures, both of which are known to induce bisexual reproduction in Cladocera (Banta 1939; Berg 1934), were clearly influencing the population and causing the production of males and ephippial females, in spite of the increasing amounts of food which were becoming available.

VARIATION IN THE SIZE OF MATURE FEMALES

The description of the variation in mean egg number applies almost identically to the variations in length of the females carrying the eggs. The two fluctuate together. The longest females were found in spring. When they died they were replaced by smaller females, and it was not until October that the mean size increased significantly again (Fig. 6).

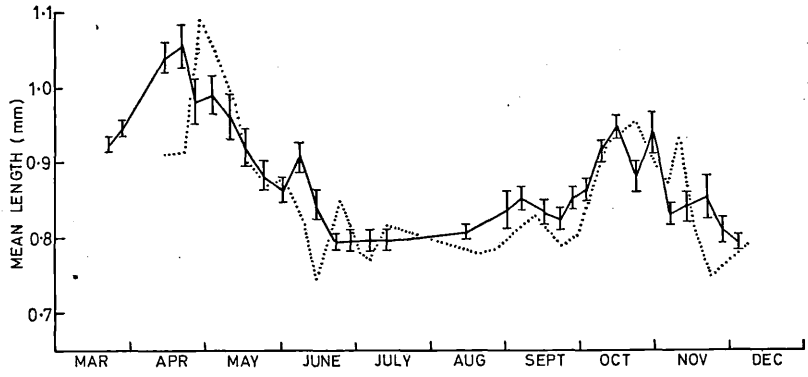

FIG. 6. *Scapholeberis mucronata*: seasonal variation in the body length of adult females (. . . . . , 1960; ———, 1961). The short vertical lines indicate one standard error above and one below the mean.

VARIATION IN THE HEAD SPINE

During 1961 two sets of samples were taken to study the variation in length of the head spine and the tail spine. The first set of samples was restricted to mature females with eggs; twenty specimens were measured in each sample, and altogether thirty-two such samples were measured. The second series was also restricted to females, but each sample of 100 was taken at random so that all ages and reproductive stages were included; fifteen such samples were taken.

The results of the first set of samples are shown in Fig. 7. In these mature females the head spines were long in March and April, then gradually diminished in length during the successive generations of the summer. There were indications of an increase in the length of the head spine of mature animals in November, but the lengths were never as great as they had been in spring. This general description applies whether one considers absolute length of the head spine, or the relative length as a percentage of the body length.

Fig. 8 shows the results of the second series of samples. In March and April the length of the head spine increased with increasing body length but as the season progressed

there was a gradual change in this relationship, so that by the middle of June the head spine decreased in length with increasing body length. This persisted throughout the summer, the reduction being greatest in September. Towards the end of the season the reduction was less marked, and in some individuals the head spines had clearly increased in length.

During the whole season there were only small variations in the head spine length of newly hatched individuals.

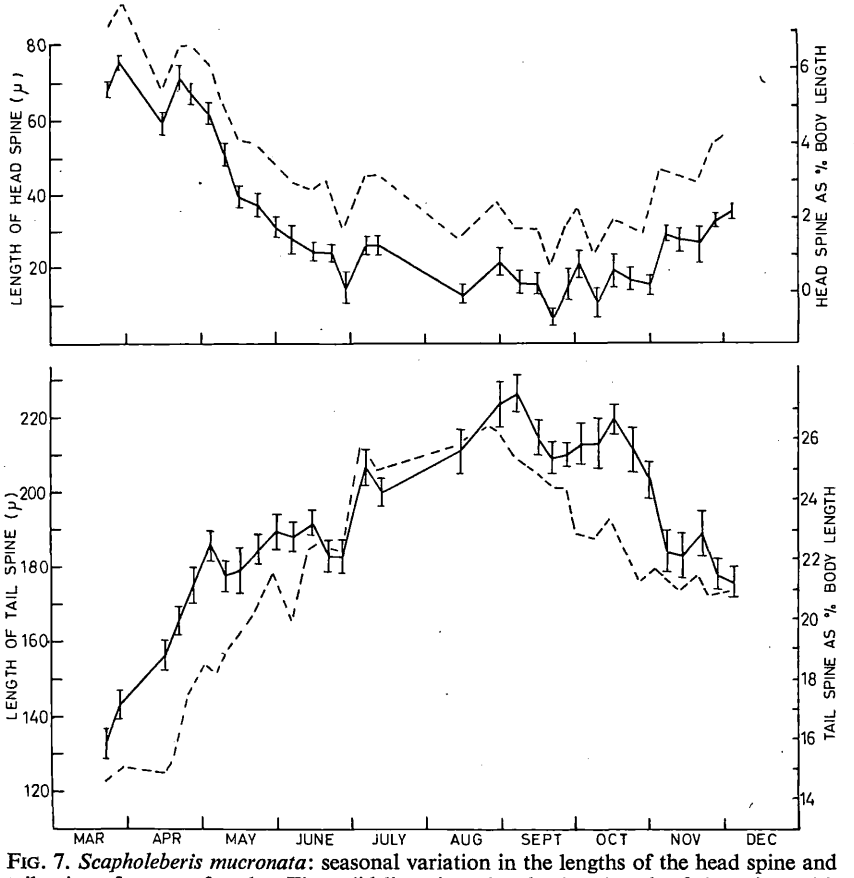

FIG. 7. *Scapholeberis mucronata*: seasonal variation in the lengths of the head spine and tail spine of mature females. The solid line gives the absolute length of the spine, with standard errors indicated above and below each point. The broken line gives the spine length as a percentage of the body length.

VARIATION IN THE TAIL SPINE

Variations in the length of the tail spine in mature females are shown in Fig. 7. It is clear that the mean length varies inversely with that of the head spine. The tail spines of mature females are shortest in spring, longest in summer, and in the autumn generations there is a reduction in length, but the spines do not become as short as those of the spring generations.

Fig. 9 shows that in all samples the tail spine increased in length with increasing body length, but it does so at different rates at different times of the year, the rate being low in spring and autumn, and high in summer.

The tail spines of very young females are distinctly longer in summer than in spring; this coupled with the high rate of relative growth during summer results in the very long spines of the mature females at the end of August and the beginning of September.

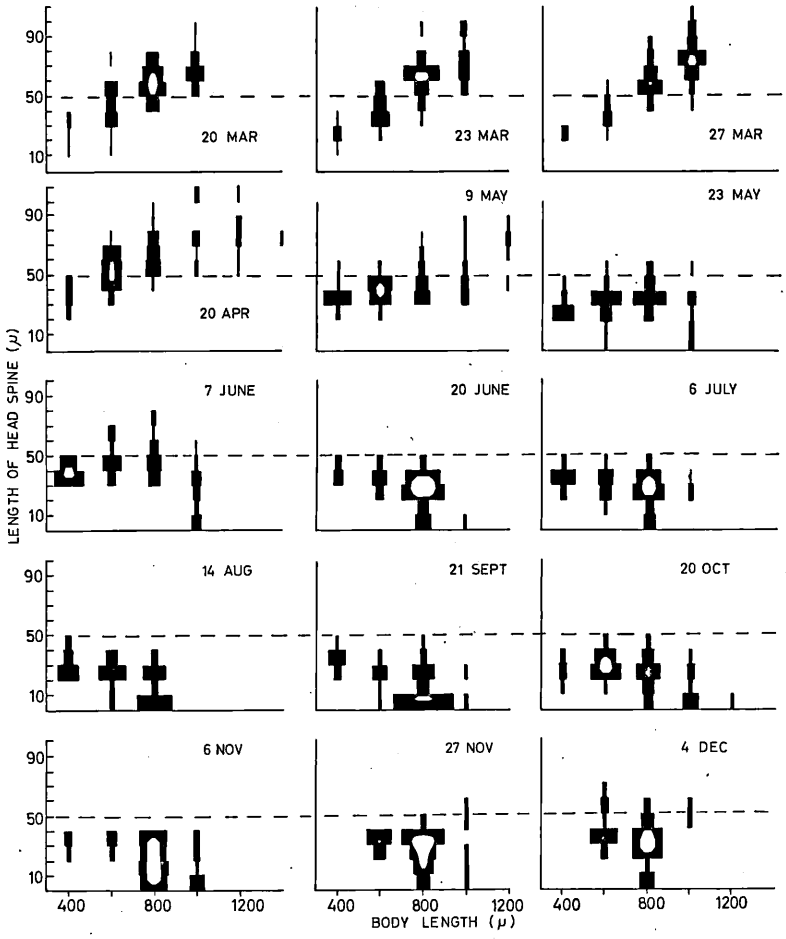

FIG. 8. *Scapholeberis mucronata*: seasonal variation in length of the head spine in relation to body length. The broken line at 50 μ is put in to aid comparison of the several rows.

EPHIPPIAL FEMALES

It is well known from researches on other members of the Daphniidae that any one female is capable of producing both parthenogenetic and ephippial eggs (*cf.* Berg 1934). It is of interest to discover if females which produce ephippia differ from those which continue to produce parthenogenetic eggs. Figs. 10 and 11 show that although the size-ranges overlap, the ephippial females of *Scapholeberis mucronata* are generally smaller than the normal parthenogenetic females, and further that the head and tail spines are shorter, at least in the upper part of the size-range.

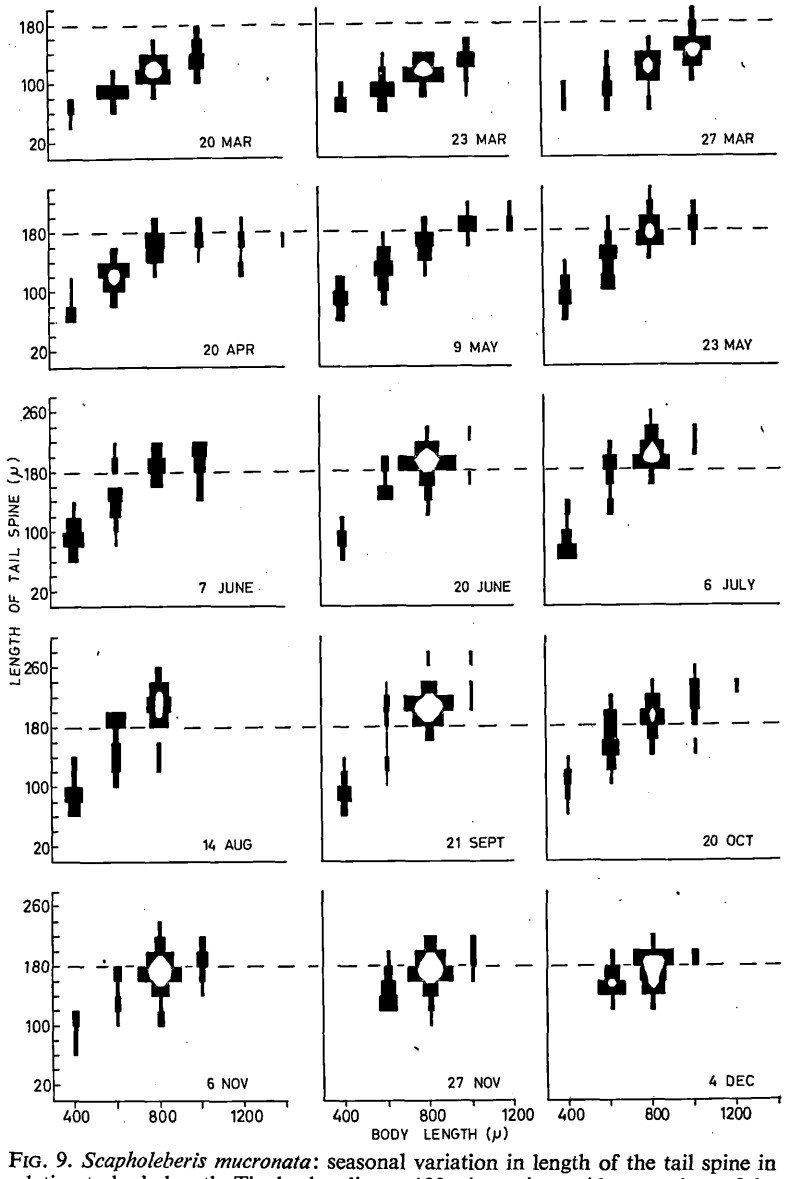

FIG. 9. *Scapholeberis mucronata*: seasonal variation in length of the tail spine in relation to body length. The broken line at 180 μ is put in to aid comparison of the several rows.

MALES

Most males of *Scapholeberis mucronata* lack head spines, but occasional specimens are found with very short head spines. For instance, out of fifty males measured on 20 Oct. 1961 only one had a head spine, and this was only 11 μ long.

Gruber (1913) stated that the males had longer tail spines than the females, and considered this as a compensation for the lack of a head spine. He did not present any detailed numerical data to support this statement. The measurement of fifty males on

20 Oct. 1961 revealed that small males had tail spines of about the same length as those of ephippial females, but they did not show any significant increase in length as the animal grew—at least over the range of body lengths from 500 to 800 μ. This lack of growth results in larger males having slightly shorter tail spines than ephippial females of the same body length (Table 1).

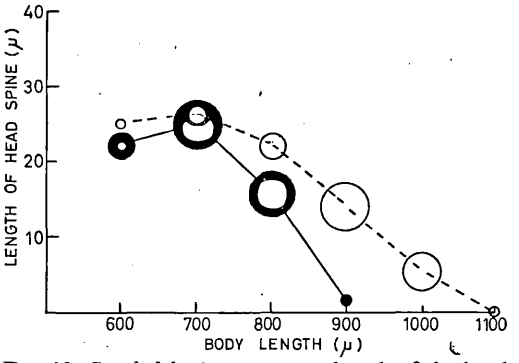

FIG. 10. *Scapholeberis mucronata*: length of the head spine of ephippial females (———●) compared with females carrying parthenogenetic eggs (- - - - ○). The size of each dot is proportional to the number of animals that it represents: the smallest representing a single individual while the largest represents twenty. Comparison made on sample collected on 20 Oct. 1961.

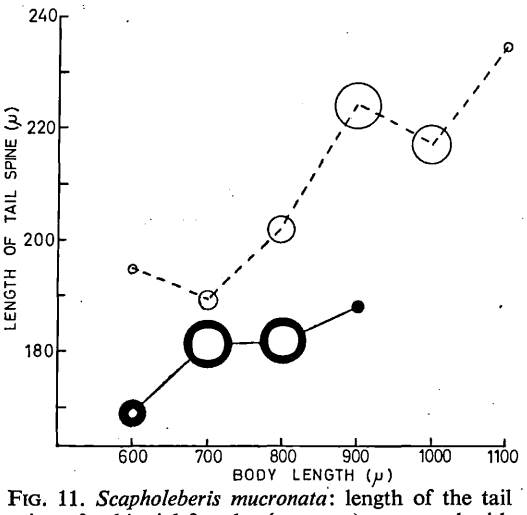

FIG. 11. *Scapholeberis mucronata*: length of the tail spine of ephippial females (———●) compared with females carrying parthenogenetic eggs (- - - - ○). The conventions of Fig. 10 are used.

NOTES ON OTHER POPULATIONS OF *SCAPHOLEBERIS MUCRONATA*

Table 2 gives a summary of measurements made on samples of twenty mature females from Slapton Ley, Devon, from two populations on Disko Island, Greenland (Lat. 69° N)

and from the mouth of the River Toce where it enters Lago Maggiore in Northern Italy. Some comparative samples from the Long Water are also entered in the table.

Slapton Ley is a substantial body of water; the Lower Ley, from which the samples were taken, is about 2·3 km long and 500 m wide. The two samples show remarkable agreement with each other, and do not differ significantly from comparable samples

Table 1. *Lengths of tail spines in males and females of* Scapholeberis mucronata (*fifty of each group measured*)

Body length (μ)	Length of tail spine (μ)		
	♂♂	Ephippial ♀♀	♀♀ with eggs
500–600	164·1 ± 5·3	–	–
600–700	166·7 ± 4·1	169·5 ± 4·1	195·0
700–800	167·5 ± 4·8	181·8 ± 3·8	189·3 ± 4·8
800–900	–	182·1 ± 4·0	202·8 ± 5·2
900–1000	–	183·3 ± 5·2	224·4 ± 5·2
1000–1100	–	–	217·8 ± 4·8
1100–1200	–	–	235·0

Table 2. *Comparison of mature females of* Scapholeberis mucronata *from different localities*

Locality and date	Water Temperature at 9 a.m. (° C)	Mean length (mm)	Mean length of head spine (μ)	Mean length of tail spine (μ)	Mean egg number
Slapton Ley					
8 Apr. 1961	12	1·06 ± 0·02	90·1 ± 2·7	170·7 ± 3·0	19·3 ± 1·6
2 May 1962	12	1·07 ± 0·03	87·6 ± 3·0	168·5 ± 5·0	19·6 ± 1·7
Long Water					
20 Apr. 1961	14	1·05 ± 0·02	71·6 ± 3·4	166·1 ± 3·6	18·3 ± 1·4
10 May 1962	16	1·12 ± 0·03	73·9 ± 2·5	176·2 ± 3·4	21·0 ± 1·3
21 Sept. 1961	17	0·83 ± 0·01	6·8 ± 2·1	209·4 ± 4·1	4·3 ± 0·4
Greenland					
Godhavn					
31 July 1961	8	0·95 ± 0·01	4·3 ± 1·2	89·1 ± 1·9	2·8 ± 0·2
Kangarssuk					
28 July 1961	11*	0·84 ± 0·02	0·4 ± 0·3	79·8 ± 2·3	1·9 ± 0·2
Lago Maggiore Fondo Toce					
9 Aug. 1962	25†	0·74 ± 0·02	31·2 ± 4·7	142·7 ± 13.0	7·4 ± 0·5

* Temperature measured at noon.
† Temperature measured at 3 p.m.

taken from the Long Water. This agreement between the two samples, in spite of the 1962 sample being taken almost a month later, can be attributed to the late spring of 1962 delaying the development of the population so that they were at comparable stages in the two years.

The agreement between the Long Water and Slapton Ley indicates that the cycle

described for the Long Water is probably applicable to other localities of a similar size in the same latitude and at the same altitude. Very much smaller water bodies, particularly if unstable near their margins, and large lakes may well exhibit different cycles, more so if they are situated in different latitudes.

The Greenland samples come from localities which are subjected to much lower temperatures than the British samples. The populations were found in pools under 50 m in diameter and not much over a metre in depth. Such pools on Disko are frozen from October to May (Røen 1962), and though the temperature in midsummer can reach 20° C this is exceptional. The air temperature at Godhavn in summer normally varies between +12° and −5° C, so that pool temperatures can show considerable fluctuations from day to day. As a rough approximation the average summer temperature of these pools is about 10° C below that of pools in southern England.

The significant features of specimens from Greenland are the very short head spines and the relatively short tail spines. These are emphasized by comparison with the sample taken from the Long Water on 21 September when the population showed the greatest reduction of the head spine. In the sample from the Kangarssuk Peninsula eighteen of the females were without head spines, and in the sample from Godhavn eleven were without head spines. The longest head spine on a mature female from Godhavn was only 14 μ, but an immature specimen had a head spine 25 μ long. Ten immature females from Godhavn (mean length 620 μ) had a mean head spine length of 17·5 μ and a mean tail spine length of 77·8 μ. Comparison of these figures with those of mature females in Table 2 indicates that the head spine diminishes rapidly in size as the animal grows, and the tail spine does not grow very much; the longest tail spine found in a mature female from Godhavn was 104 μ.

The mouth of the River Toce represents a habitat with a consistently higher temperature than the Long Water. The significant features of the specimens from this habitat are the small body length—smaller than any sample from the Long Water, and the moderate length of the tail spine when compared with specimens from the Long Water. The tail spine was much more variable than in any of the other samples; note the large standard error in Table 2. The mean length of the head spine in these Italian specimens is comparable to the mean length found in the Long Water in May or late November.

DISCUSSION

A provisional explanation of the seasonal cycle of *Scapholeberis mucronata* can be made in terms of differential influences of temperature and food. The cycle of the tail spine in the Long Water can be attributed mainly to the influence of temperature. The length of the tail spine being greatest when the temperature is highest. It does not show any sign of being influenced by food shortage. When food shortage becomes more severe than in the Long Water then the tail spine may be reduced. Gruber (1913) showed that starvation reduced the length of both the head spine and the tail spine. Now at a given temperature the mean egg number can be taken as a guide to the nutritional state of a cladoceran (Ingle, Wood & Banta 1937; Fox, Hardcastle & Dresel 1949; Green 1954, 1956), and on this basis the Greenland populations were in a much poorer state than was ever found in the Long Water. The summer temperature of the water in the pools in Greenland was roughly comparable with the Long Water at the beginning of spring, but the tail spines of the Greenland specimens were much shorter than the tail spines of even the

earliest specimens in the Long Water. The very short tail spines of the Greenland specimens can be attributed to the combined effects of low temperature and food shortage.

The tail spines of the Italian specimens were much more variable than those of the other samples, but the mean length was distinctly lower than that of the summer samples from the Long Water. This may indicate that high temperatures, above those found in the Long Water, begin to have a retarding effect on the growth of the tail spine. The reduction in length of the tail spine in the Italian specimens is not the result of food shortage since the mean egg number is well above that of Long Water specimens in September 1961. These Long Water specimens were only producing about four eggs each, yet their tail spines were longer than the Italian specimens which were producing seven eggs.

The head spine seems to be more susceptible to food shortage than the tail spine. In the Long Water the first significant drop in its length in mature females occurred at the beginning of May, when the chlorophyll began to fall. At the same time the larger random samples began to show that the rate of growth of the head spine was slowing down, and in some females it had decreased in length. By 20 June the head spine was decreasing in all the females as they grew in length. The absence of head spines from the majority of the adult females in the Greenland samples can be attributed to food shortage, taking the low egg production as a guide. The head spines of the Italian specimens are in agreement with those of specimens from the Long Water on 31 May 1961, when the egg number was identical with that of the Italian sample.

Gruber (1923) summarizing results from several different habitats reached the conclusion that there was a correspondence between the size of a habitat and the length of the head spine. Specimens from large lakes had long head spines while those from small pools were often without head spines. There is some element of truth in this idea. When I visited Lake Ohrid in August 1959 I noted that the specimens of *S. mucronata* from the main body of the lake had extraordinarily long head spines while those from small pools in Ohrid Marsh had much shorter head spines. Brehm & Zederbauer (1904) have also noted very long head spines in specimens from Lake Garda. Although this effect may apply when comparing very large with very small bodies of water, covering a range of several thousandfold, it can be overridden by other factors when two water bodies differ only slightly in size. Rammner (1927) compared samples from two pools, one of which was six times the area of the other; the smaller one having an area of approximately 200 m^2. The head spines of the animals from the smaller pool were distinctly longer at all stages. A possible criticism of Rammner's work is that the samples were taken about a month apart, that from the smaller pool being taken in August, while that from the larger pool was taken in July. In the normal course of events one would expect the later sample to have shorter head spines, but in Rammner's samples the later sample from the smaller pool had longer head spines so that the difference in habitat size was overridden by other factors.

It is not possible to relate the results from the Long Water to any general theory of cyclomorphosis, such as that of Wesenberg-Lund (1908, 1926). Such theories served a useful purpose in that they stimulated a great deal of research, but general application to a wide range of planktonic animals must now be abandoned. The Rotifera do not comply in any way with the flotation theory as formulated by Wesenberg-Lund (Carlin 1943; Ruttner-Kolisko 1949; Green 1960), and within the Cladocera an additional factor, turbulence, is of importance (Brooks 1947; Hrbáček 1959; Jacobs 1962). There is a definite effect of temperature on the development of head crests in *Daphnia* (Coker &

Addlestone 1938; Jacobs 1961), but the present results indicate that the head spine of *Scapholeberis* is much more susceptible to changes in nutrition than it is to changes in temperature. While, at the levels of nutrition found in the Long Water, the length of the tail spine varies in unison with the temperature. The explanation of the curious inverse relation between the head spine and tail spine of *S. mucronata* which has been offered in this discussion is of necessity tentative, but the data from this field study have provided a clear picture of the nature of the seasonal cycle and have enabled the formulation of hypotheses which can be tested experimentally.

ACKNOWLEDGMENTS

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SUMMARY

1. A population of *Scapholeberis mucronata* in Hampton Court Long Water has been studied over a period of 2 years to obtain a picture of the seasonal changes in form, particularly in size of head and tail spines shown by this species.

2. Maximum production of parthenogenetic eggs per female was found towards the end of April; egg production during summer was generally low and there was an increase in October. The mean length of adult females with parthenogenetic eggs followed a cycle similar to that of egg production.

3. There were two periods of sexual reproduction, the first being very slight and lasting only 2 weeks in May; the second was more prolonged, lasting from the middle of September to the end of November.

4. The length of the head spine in this population seemed to be influenced mainly by changes in nutrition, being longest in spring when food was abundant and showing a marked decrease in summer when food was scarcer.

5. The length of the tail spine was not readily related to changes in nutrition, but it increased with increasing temperature, showing a clear increase in summer, at a time when the head spine decreased.

6. Samples from Greenland and Italy have been measured to compare with the seasonal results from the Long Water. The Greenland samples emphasized the correlation between low nutritional level and reduction of the head spine, and the Italian sample indicates that temperatures higher than those found in the Long Water may cause some reduction in the length of the tail spine.

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ZOOPLANKTON OF THE RIVER SOKOTO, THE RHIZOPODA TESTACEA

BY

J. GREEN

Bedford College, University of London

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(With 33 figures in the text)

Thirty-two species of testaceous Rhizopoda have been identified from the plankton of the River Sokoto. Most of these species are adventitious in the plankton, and are most abundant in the wet season when the river is in flood. Four species : *Arcella vulgaris*, *Difflugia limnetica*, *D. lobostoma* and *Centropyxis aculeata* occur regularly in the plankton during the dry season, and show evidence of producing light shelled forms which adopt a planktonic mode of life.

CONTENTS

INTRODUCTION

This is the final paper in a series dealing with the zooplankton collected by Mr M. J. Holden during a tour of duty as a fisheries research officer in the North Nigerian province of Sokoto. A general description of the river and its plankton has been given by Holden & Green (1960). The present paper gives a detailed account of the systematics and seasonal occurrence of the testaceous rhizopods. The main object has been to discover which species are responsible for the production of light shelled planktonic forms.

The Testacea differ from the groups previously considered (cf Green, 1960a, 1960b & 1962) in that they are not primarily planktonic, and only achieve a relative importance during the flood period of the river (see Fig. 1.). When the floods subside a pool called Fesafari becomes separated from the main channel of the river; a series of samples was taken from this pool for comparison with the river.

METHODS

The plankton was collected by Mr Holden by towing a fine meshed plankton net for a fixed distance just below the surface of the water. Details of the two sampling stations are given by Holden & Green (1960). The samples were preserved in 5 per cent formalin; the resulting fixation was of a high standard;

many of the Testacea had clearly distinguishable nuclei and in some pseudopodia were preserved in a remarkably lifelike manner.

The volume of each sample was made up to 30 ml, and after thorough mixing sub-samples of 0·5 ml were taken with a wide mouthed graduated pipette and ejected into a petri dish containing a thin film of water. All the animals in the sub-sample were then counted under a binocular microscope with a magnification of 30 diameters and assigned to major groups such as Testacea, Rotifera, Cladocera etc. The relative abundance of each species of testacean was then estimated by another series of samples in which a variable number of specimens was identified to specific level. It was originally intended to identify a hundred Testacea from each sample, and this was done with most samples, but in some the testaceans were so scarce and other animals so abundant that finding a hundred specimens to identify would have taken an unreasonable length of time. In such samples at least as many testaceans as were present in a sub-sample were identified, usually a multiple of three or four times this number. The minimum number identified was twelve from the May 1956 sample from Fesafari.

The testaceans were identified and counted after a sample of the plankton had been mixed with lactic acid containing Watermans permanent blue ink. This stained the cytoplasm a brilliant blue, and the refractive index of the lactic acid was such that fine details of the structure and sculpturing of the tests were easily visible. Preparations made in this way were examined on a specially ruled glass slide with lines at intervals slightly less than the field diameter of a monocular microscope with a 16 mm objective. The slide was moved with a mechanical stage and each row was systematically searched for testaceans. When a testacean was found it was generally necessary to switch to a parfocal 4 mm objective to make measurements and complete identification. All measurements were made with the same calibrated eyepiece micrometer. The sequence in which Testacea are encountered in this procedure is essentially random, and the counts can be regarded as reasonable estimates of the relative abundance of each species; these estimates will naturally be less reliable when small numbers were identified, but this only applies to a few of the samples. A simple calculation converted the figures for relative abundance into numbers per sub-sample, using the information gathered in the first series of counts.

In presenting the numerical results in histogram form the flood cycle of the river has been imposed on the figures so that it is easy to see when the occurrence of a species in the plankton is entirely due to hydrological conditions, and when the production of light shelled planktonic forms might be suspected.

FLUCTUATIONS IN TOTAL TESTACEA

The abundance of testaceans in relation to the total numbers of zooplankters is shown in Fig. 1. Only when the total zooplankters fall below 1000 per sub-sample do the testaceans begin to form more than 10 per cent of the total. This is an important point to keep in mind; although the relative importance of the Testacea increases greatly during the floods the numbers involved are small. The actual numbers of testaceans per sub-sample are shown in Fig. 2.

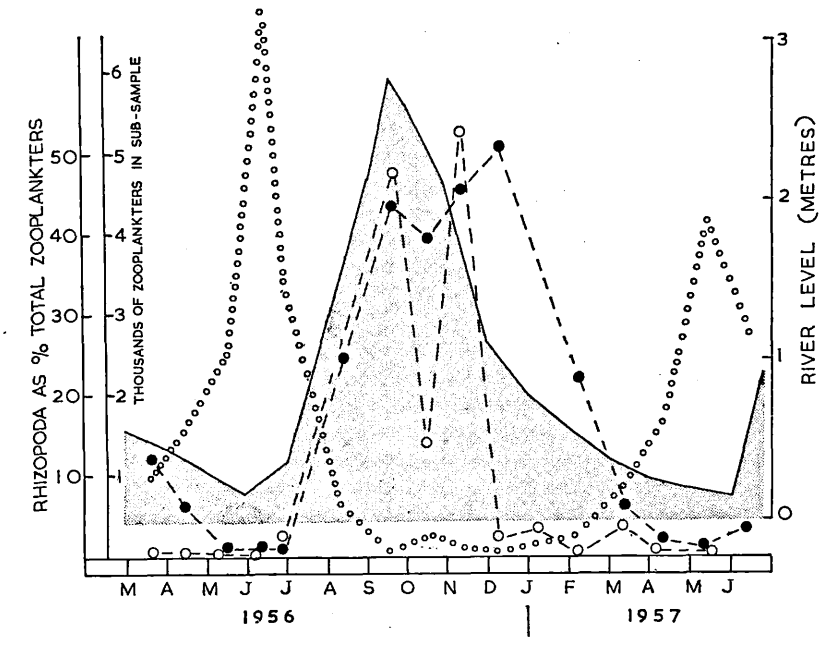


Fig. 1—Seasonal variation in the relative importance of the Testacea as a percentage of the total zooplankton.
oooooooooooo total zooplankton. ●—●—Testacea as percentage total zooplankton in the river. O—O—Testacea as percentage total zooplankton in Fesafari. The continuous line enclosing the stippled area indicates the level of the river.

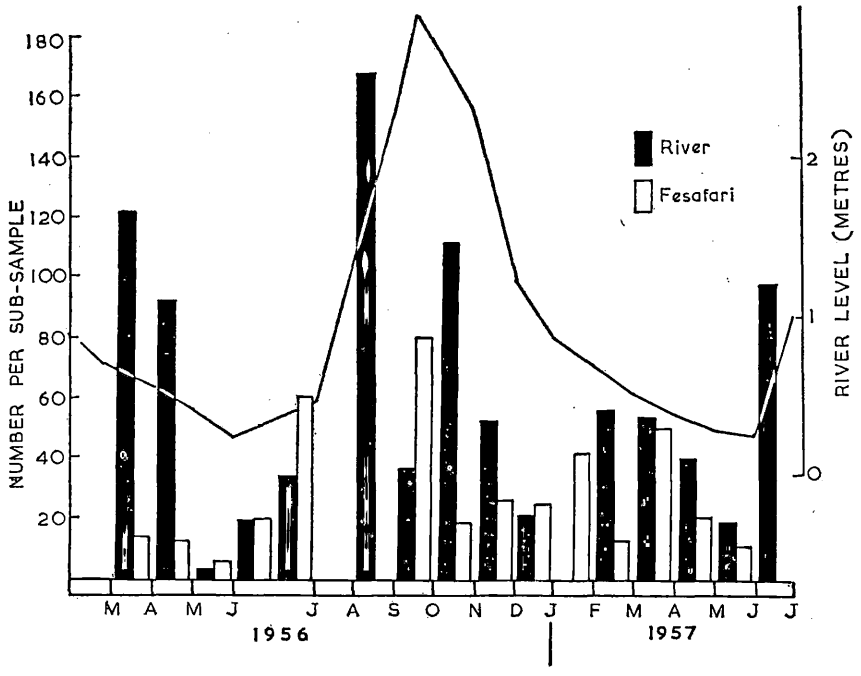


Fig. 2—Seasonal variation in total Testacea in relation to the flood cycle of the river. Notice in this figure that two samples were taken in June 1956, but no sample was taken in July; this also applies to the other diagrams showing seasonal occurrence. Each sub-sample represents the number of individuals in approximately one third of a cubic metre.

They are generally more abundant in the river than in the pool Fesafari, and, as shown in Fig. 1, they maintain their relative importance in terms of percentage of total zooplankton longer in the river than in the pool. This is to be expected from a group which is not primarily planktonic, and whose occurrence in the plankton is governed to a considerable extent by the rate of flow of the river.

SYSTEMATIC SURVEY AND SEASONAL OCCURRENCE

The arrangement of the Testacea into families is a difficult matter, and various authorities have produced different schemes. As the family concept is not relevant to the purpose of the present paper it has been decided that ease of reference should be the overriding consideration. Accordingly, in the following account the genera have been arranged in alphabetical order, and the species within each genus have been treated similarly, as recommended by Moreau (1961).

Arcella dentata Ehrenberg

Most authors (e.g. Penard, 1902; Cash, 1905) regard this as a rare species. Only a few specimens were found in the samples from the river, and a single specimen in the sample from Fesafari in May 1957. The specimens varied both in the number and the length of the spines around the margin of the shell (Fig. 4). The diameter varied from 144 to 180μ, including the spines.

Arcella discoides Ehrenberg

It was not always possible to separate empty shells of this species from *A. polypora*, but most of the specimens still contained cytoplasm, and two nuclei could be distinguished. Most specimens had a diameter between 125 and 140 μ, but one exceptional specimen had a shell diameter of 234 μ and a pseudostome diameter of 69 μ. The diameter of the shell brings this specimen within the size range of *A. megastoma*, but the diameter of the pseudostome, and the presence of only two nuclei clearly indicate that the specimen belongs to *A. discoides*.

Only small numbers of this species were found throughout the year, both in the river and in the pool.

Arcella gibbosa Penard

This species was very rare in Fesafari, and was irregular in occurrence in the river. The occurrences are not particularly related to the flood cycle of the river except that the greatest numbers occurred when the water was rising rapidly in June 1957. The Sokoto specimens lay within the size ranges given by Deflandre (1928) and Decloitre (1953), generally being between 90 and 100 μ in diameter.

Arcella megastoma Penard

This is the largest of the *Arcella* species in the Sokoto, and one of the most frequent. The greatest number recorded, 50 in the sub-sample from the river in August 1956, coincided with rapidly rising water. At other times of year the numbers were much lower.

The diameters of the Sokoto specimens ranged from 133 to 342μ with corresponding pseudostome diameters of 61 and 162 μ.

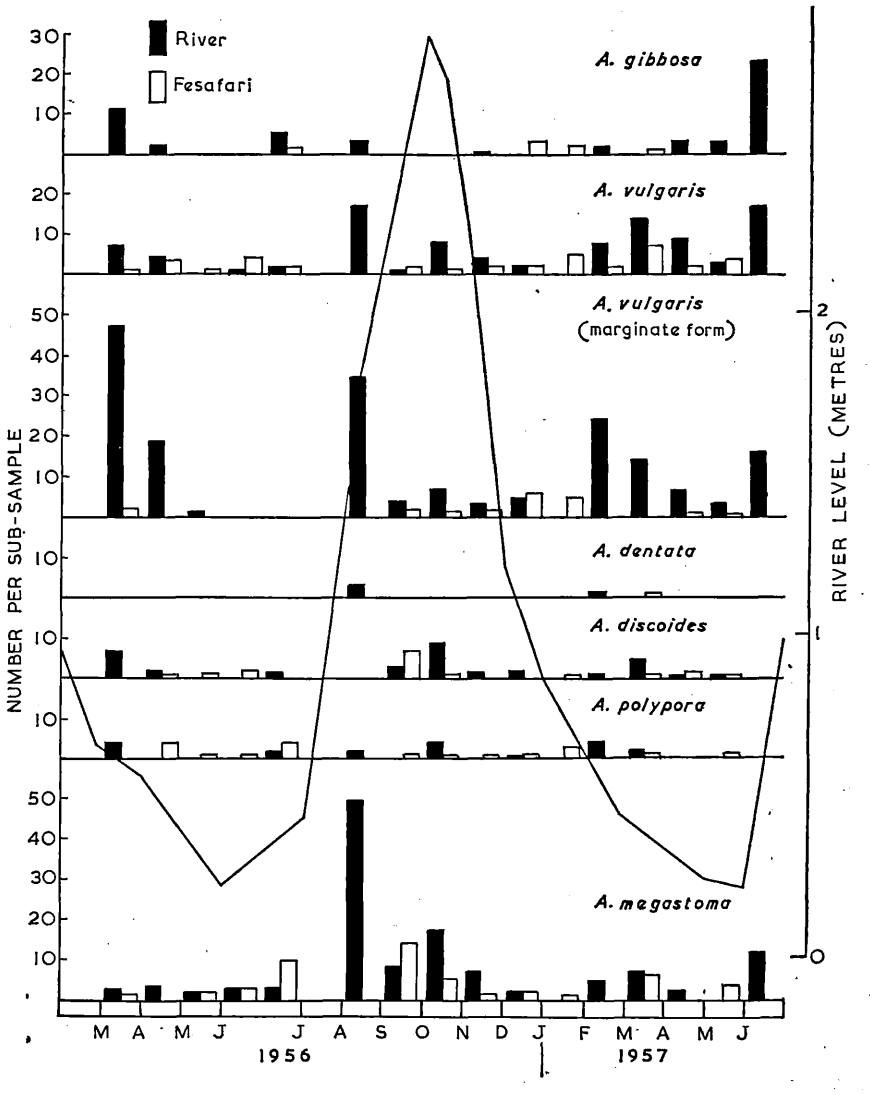


Fig. 3—Seasonal occurrence of *Arcella* spp. in the plankton.

Arcella polypora Penard

In both the pool and the river this species occurred in small numbers throughout the year without showing any special relation to the flood cycle of the river.

The diameter of the pseudostome of this species is usually a little larger than that of a specimen of *A. discoides* of the same size, and there are more than two nuclei; six or more were visible in some of the Sokoto specimens.

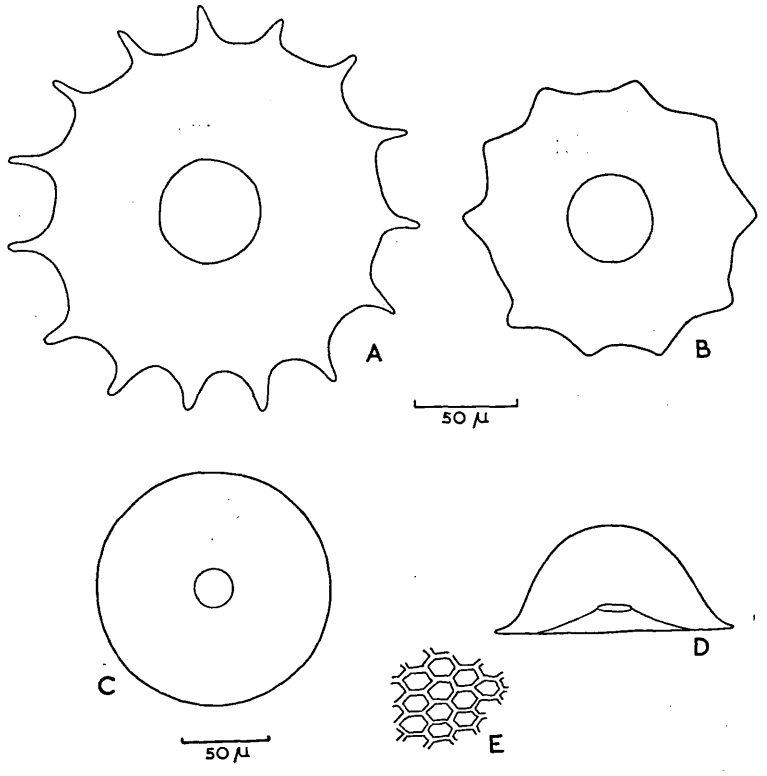


Fig. 4—A & B, extreme forms of *Arcella dentata*; both were taken from the river in August. C. D & E, marginate form of *Arcella vulgaris*. C, from below. D, lateral view. E, detail of reticulation on shell.

Arcella vulgaris Ehrenberg

The typical form of this species (cf. Deflandre, 1928) was most abundant when the river level was rising rapidly (Fig. 3), but in 1957 it was consistently present in the river in March and April, when the river level was very low. Occurrences at other times of year were erratic and involved few individuals.

Kofoid (1908) found this species to be present in the River Illinois throughout the year, but noted that at least half the occurrences were related to rising water levels.

Some of the specimens reached a diameter of 170μ, with a pseudostome diameter of 52 μ, but most were smaller than this.

A form of this species, with a sharp margin to the shell and a small pseudostome (Figs. 4C-E) was even more abundant than the typical form. In general shape this form is similar to *A. brasiliensis* but the whole surface of the test is covered with a slightly irregular hexagonal reticulation. The diameter of the shell was usually about 126 μ, and the diameter of the pseudostome 21-25 μ.

Although it was always scarce in Fesafari this form reached a maximum of 48 in the sub-sample from the river in March 1956. This was at a time when the river was not in flood, and the peak occurrence can be attributed to the production of planktonic forms. The species was evidently reproducing rapidly at this time; many of the shells were very thin and almost colourless. There was also a definite though smaller peak of occurrence in February 1957, again when the river was not in flood.

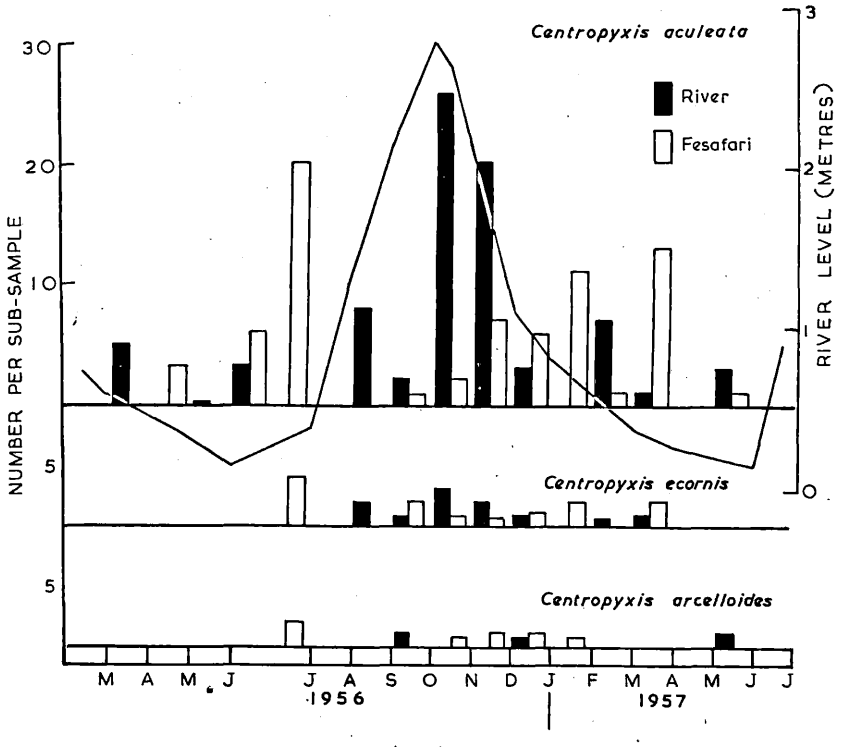


Fig. 5—Seasonal occurrence of *Centropyxis* spp. in the plankton.

Centropyxis aculeata (Ehrenberg) Stein

There is great variation in the size, colour, and number of spines in this species. In counting the sub-samples I have made no attempt to distinguish the various named varieties, though it is certain that some specimens were of the var. *grandis* Deflandre, and others were of the var. *oblonga* Deflandre.

The seasonal occurrence of this form shows some remarkable features (Fig. 5). In the river it is most abundant during the floods, but in Fesafari it is most abundant when the river level is low. This implies a production of

planktonic forms in the pool, though the shells were not noticeably lighter in construction than in other samples.

Centropyxis arcelloides Penard

The occurrence of this species in the samples is very similar to that of the next species; the numbers were always low (Fig. 5).

Centropyxis ecornis (Ehrenberg) Leidy

Small numbers of this large flattened species were found in the river and in the pool, mainly during the period when the river was in flood.

Centropyxis minuta Deflandre

Only a single specimen of this small species was found in the sample from Fesafari in January 1957.

Cucurbitella mespiliformis Penard

A few specimens were found in the samples from Fesafari in October 1956 and from the river in September 1956, December 1956 and April 1957. The maximum length of these specimens, including the collar, was 116 μ. This is at the lower limit of the size range given by Wailes (1919), but it should be noted that Decloitre (1953) has recorded an even smaller specimen (100μ long) from Tanaf, French West Africa.

Difflugia acuminata Ehrenberg

A few specimens were found in the samples taken from Fesafari in June 1956. One of these specimens (Fig. 27) reached a length of 328 μ and belonged to the var. *magna* Deflandre (1926)

Difflugia amphoralis Hopkinson

This species was found only as single specimens without any particular relation to the flood cycle. It occurred in the samples from Fesafari in March 1956 and February 1957, and in the samples from the river in June and August 1956 and April 1957.

The specimens had well developed, reflexed collars (Fig. 11) and ranged in length from 162 to 205 μ.

Difflugia corona Wallich

This species was present in small numbers in the river throughout the year, and occurred at irregular intervals in the samples from the pool. The Sokoto specimens are distinctly smaller than is usual for this species. The diameter of the shell ranged from 90 to 125 μ, not including the spines. Penard (1902) gives the diameter as 200-250 μ, while Leidy (1879) mentions a shell

320 μ in diameter. The diameters of African specimens given by Gauthier-Lièvre and Thomas (1958) are somewhat smaller (120-190 μ), but even so are not so small as some of the Sokoto specimens.

The individuals showed considerable variation in the shape of the shell and the number of spines on the crown (Figs. 6-9). The lobes of the pseudostome also varied slightly in number and depth.

Difflugia difficilis Thomas var. *ecornis* Chardez

This form was only found in the pool Fesafari; two specimens in the sample taken in March 1956, and single specimens in April and June 1956. These specimens (Figs. 15-17) were more variable in size than the dimensions given by Gauthier-Lièvre and Thomas (1958), and ranged in length from 75 to 119 μ, and in diameter from 40 to 90 μ. The diameter of the circular pseudostome ranged from 20 to 36 μ.

The type form of the species was described from France in 1954 by Thomas, and the var. *ecornis* from Belgium by Chardez (1956). Both the type and the variety have since been recorded from West Africa (Gauthier-Lièvre & Thomas 1958).

Difflugia elegans Penard var. *angustata* Deflandre

A single specimen (Fig. 20) of this species was found in the sample taken from the river in April 1958. The total length of the specimen was 100 μ. It has been recorded previously in West Africa by Gauthier-Lièvre and Thomas (1958), and is otherwise known from Algeria, France and Venezuela (van Oye, 1956).

Difflugia globularis (Wallich) Leidy

There appears to be some confusion about the names *Difflugia globularis* and *D. globulosa*. I am here using the specific name in the same way as Gauthier-Lièvre and Thomas (1958).

This is another species showing irregular occurrence in the samples, from both the river and the pool. The length of the specimens ranged from 130 to 165 μ, and the diameter from 108 to 140 μ. The pseudostome was plain and circular (Fig. 21).

Difflugia gramen Penard

This species was found in small numbers at irregular intervals throughout the year, it was slightly more frequent in the pool than in the river, and no clear relation to hydrographic conditions could be traced.

The specimens ranged in length from 60 to 90 μ, and the pseudostome had three rounded lobes. A few specimens of the var. *achlora* Penard were found in the samples taken from the river. This has a small transparent collar around the pseudostome (Figs. 18 &19); the length varied from 63 to 90 μ, thus reaching a much larger size than the specimen originally described by Penard (1902), but agreeing with the dimensions given by Gauthier-Lièvre & Thomas (1958) for African specimens.

Difflugia limnetica Levander

This was one of the commoner species in the samples. It was most abundant during the floods, but it was also fairly abundant in Fesafari in February and March 1957. This may have been due to the production of light shelled planktonic forms. It is worthy of note that this is one of the few species of rhizopods that Rylov (1935) regarded as being planktonic.

This species is closely allied to *D. gramen* and *D. lobostoma*, but it has a collar and the pseudostome has three irregularly formed lobes. Most of the Sokoto specimens were 110 to 120 μ long (Fig. 24).

Difflugia lobostoma Leidy

Typical specimens (Fig. 25) were found in the river at the begining of the floods, but at other times of the year, particularly in March and April 1956, a light shelled form, which sometimes had two to four short horns at the apex of the shell (Fig. 26) was found. The particles adhering to the shell were much smaller and lighter than in the typical form. This bears some resemblance to the var. *cornuta* of Gauthier-Lièvre and Thomas, but the surface of the shell was also slightly tuberculated, recalling their var. *tuberosa*. The pseudostome had three weakly developed lobes, and the length of the shell was usually 120 to 130 μ.

This is the clearest example found in this investigation of the production of a light shelled planktonic form when the river was near its lowest (Fig. 32). It is surprising that the species was so scarce in the samples from Fesafari.

Difflugia oblonga Ehrenberg var. *cylindrus* Thomas

A few specimens of this large variety were found in the samples from Fesafari in May and June 1956. Single specimens were also found in the November 1956 sample from Fesafari and the September 1956 sample from the river.

The shells of these specimens (Figs. 28 & 29) were mostly about 250 μ long, which slightly exceeds the length range, 180-242 μ, given by Gauthier-Lièvre & Thomas (1958).

Difflugia oblonga Ehrenberg var. *parva* Thomas

Single specimens were found in the samples from Fesafari in February and March 1957. These specimens were rather small, one being 140 μ long and the other 162 μ (Fig. 14).

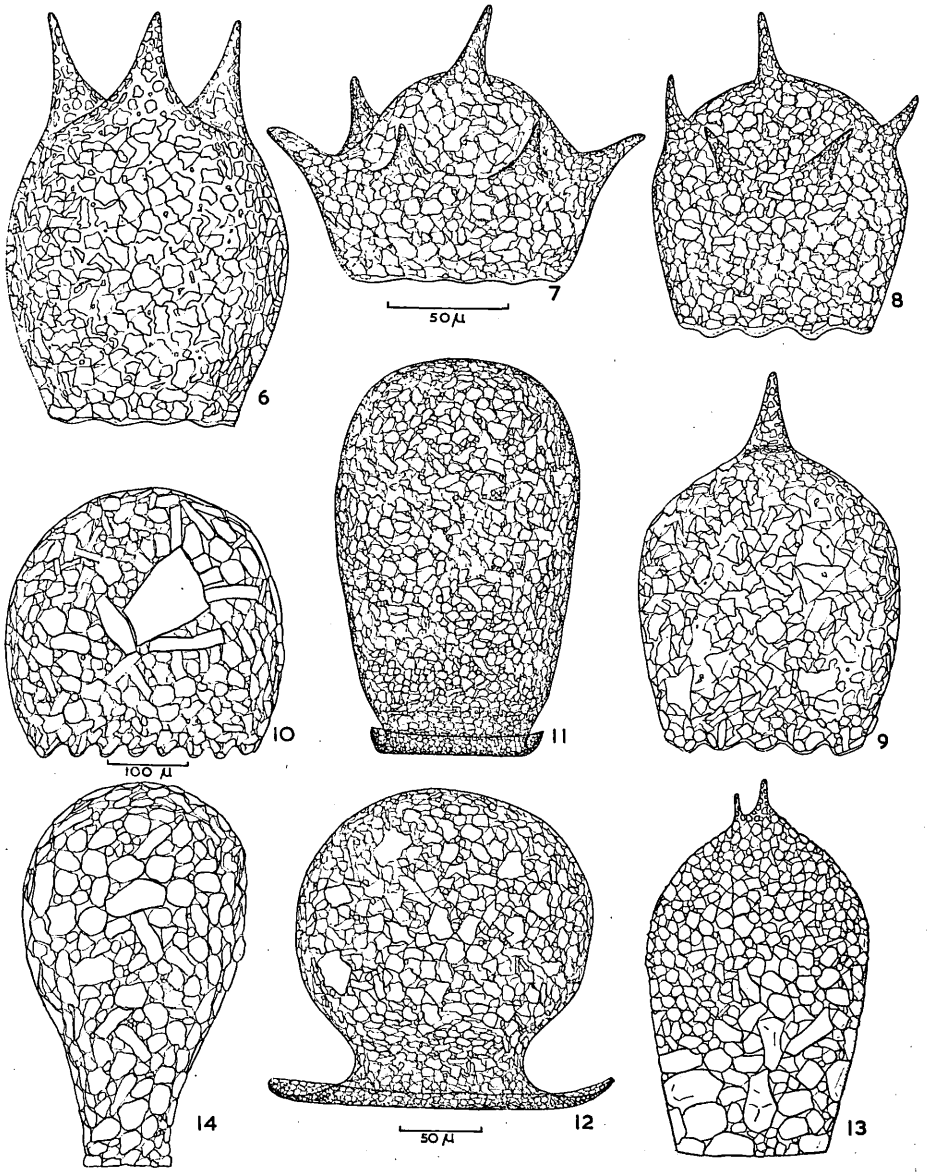
Difflugia penardi Hopkinson

This species was only found in small numbers in the samples taken from the river in April and June 1957. The length of the shell was about 65 μ (Fig. 22).

Difflugia smilion Thomas

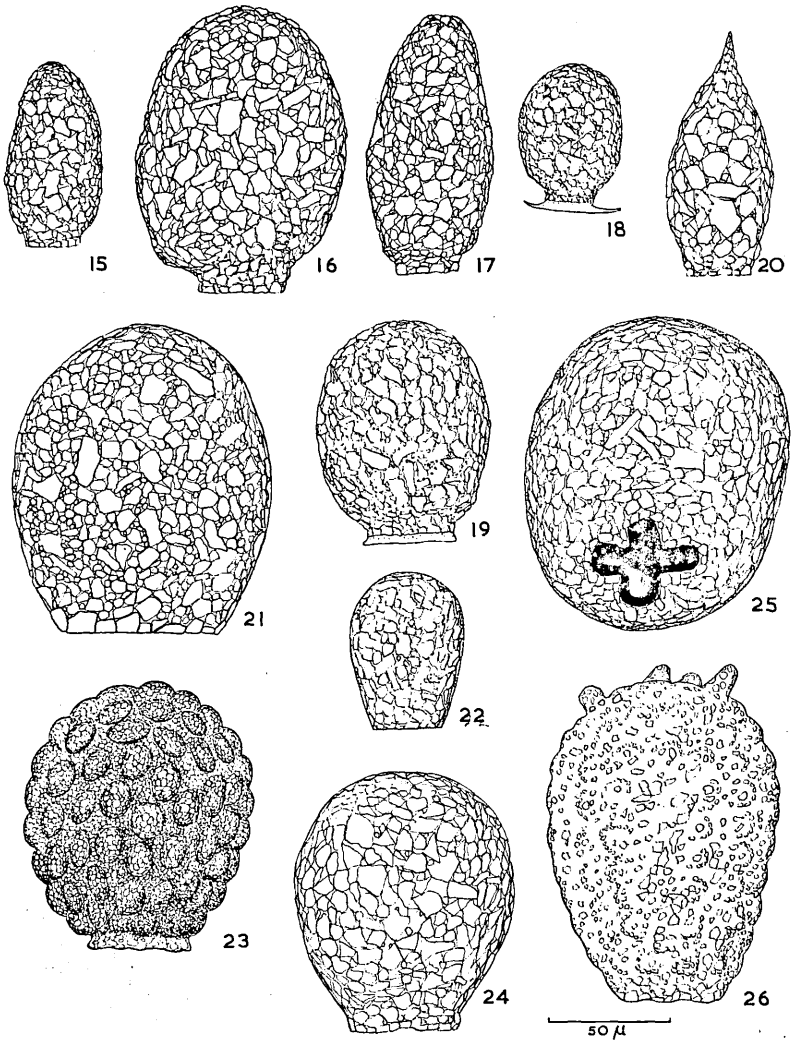
In Holden & Green (1960) this species was listed as *D. scalpellum* Penard, because at that time I had not seen the paper by Thomas (1953) which distinguishes between these closely allied forms. The originally described specimens of *D. scalpellum* came from the profundal region of Swiss lakes, whereas

D. smilion is a shallow water form. It thus seems probable that the record of *D. scalpellum* from the River Illinois (Kofoid, 1908) should be referred to *D. smilion*.



Figs. 6-14—Species of *Difflugia*. 6-9, varieties of *D. corona*, drawn to the same scale. 10, *Difflugia* sp. x, note the scale. 11, *D. amphoralis*.
12, *D. urceolata*, 13, *Difflugia* sp. y. 14, D. *oblonga* var *parva*. Figs. 11, 13 and 14 are drawn to the same scale as Fig. 7.

Single specimens were found in the samples taken from the river in November 1956 and April 1957, and from Fesafari in January 1957. These specimens (Fig. 30) ranged in length from 216 to 260 μ, and were up to 100 μ wide, which is wider than the diameters given by Gauthier-Lièvre & Thomas (1958) for



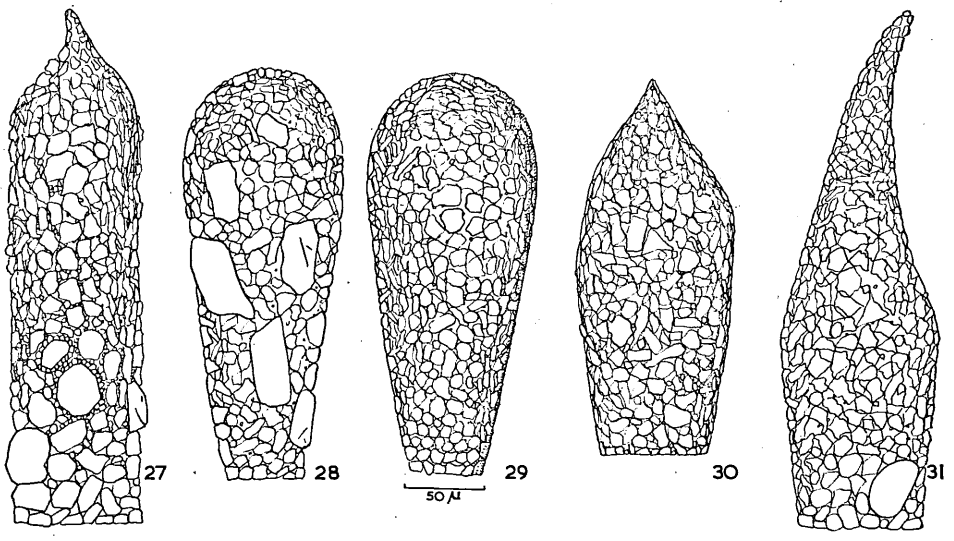
Figs. 15-26—Species of *Difflugia*. 15-17, *D. difficilis* var *ecornis*. 18 & 19, *D. gramen* var *achlora*. 20, *D. elegans* var *angustata*. 21, *D. globularis*. 22, *D. penardi*. 23, *D. tuberculata*. 24, *D. limnetica*. 25, *D. lobostoma*, typical form, oblique view to show the pseudostome. 26, *D. lobostoma*, light shelled planktonic form. All are drawn to the same scale.

specimens from the Ivory Coast. In one specimen the nucleus was visible after crushing the shell; the diameter was 36 μ, and there was a single large spherical nucleolus.

Difflugia tuberculata Wallich

This species had a definite peak of abundance when the floods were at their highest, with only small and erratic numbers at other times.

The distinctive tuberculate shell of this species can be distinguished from the " *tuberosa* " forms of *D. lobostoma* by the presence of a small but distinct collar (Fig. 23). The length of the Sokoto specimens varied between 90 and 108 μ.



Figs. 27-31— Species of *Difflugia*. 27, *D. acuminata* var *magna*. 28 & 29, *D. oblonga* var. *cylindrus*. 30, *D. smilion*. 31, *Difflugia* sp. z. All are drawn to the same scale.

Difflugia urceolata Carter

The occurrence of this large species in the plankton is due entirely to the mechanical effect of the floods. Only single specimens were found in the samples from the river in September to December, and a single specimen from Fesafari in October. The only occurrence outside the flood period was a single specimen from Fesafari in March 1956.

The specimens from the Sokoto (Fig. 12) are similar to the var. *sphaerica* which was originally described by Playfair (1918) from Australia and has since been found in Algeria, various parts of West Africa and the Congo (Gauthier-Lièvre & Thomas, 1958).

Difflugia sp. x (Fig. 10)

This very large form was found in the sample from Fesafari in October 1956 The diameter is about 350 μ. There is a considerable resemblance to *D. corona* var. *ecornis* Gauthier-Lièvre & Thomas, but the diameter is about twice that given by these authors. Possibly it is a giant of the type found in other rhizopods by Fantham & Porter (1945).

Difflugia sp. Y (Fig. 13)

A single specimen was found in the sample from Fesafari in April 1957. The total length was 155 μ. I have not been able to assign this to any of the species of *Difflugia* that I have seen described in the literature, but do not feel justified in creating a new species without knowing any details of the animal which inhabited the shell.

Difflugia sp. z. (Fig. 31)

This single large specimen was taken from Fesafari in June 1956. The total length was 324 μ. It may be an exceptional specimen of *D. smilion*

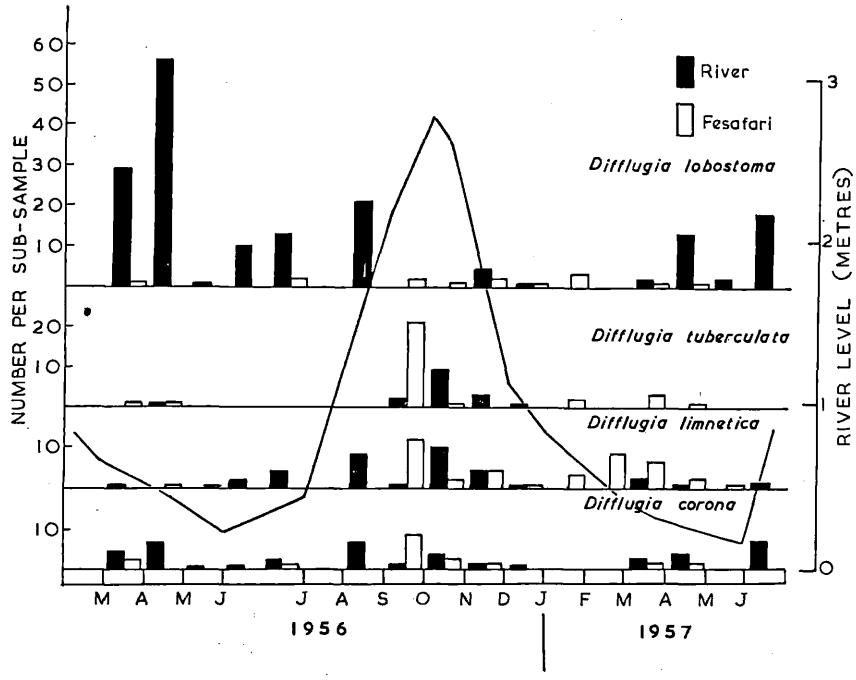


Fig. 32—Seasonal occurrence of the four most abundant species of *Difflugia* in the plankton.

var. *major* Gauthier-Lièvre & Thomas, but this is only put forward as a tentative suggestion.

Diplochlamys vestita Penard

A few specimens were found in the samples taken from Fesafari in September and October 1956.

The identity of these specimens is not absolutely certain; the tests were very large (360 μ diameter) and membranous with a plain circular pseudostome. The covering material of the test contained few very small sand grains, and appeared to consist mainly of dark vegetable detritus. Wailes (1919) states that the usual diameter is about 100 μ, but states that a large form reaching

400 μ diameter is found on submerged mosses. There are several specimens of this large form in the Penard Collection at the British Museum (Natural History), and the structure of the shell of these resembles that of the Sokoto specimens very closely.

Euglypha acanthophora (Ehrenberg)

This form was found only during the flood period of the river. The size is much larger than the normal dimensions given by Wailes (1915). The Sokoto specimens varied between 112 and 126 μ in length, and the spines were up to 72 μ in length. The pseudostome was bordered by a ring of eight scales. The large size of these specimens seems to be another example of the phenomenon noticed by Fantham & Porter (1945) who found inexplicably large forms of certain rhizopods associated with particular situations.

Euglypha tuberculata Dujardin

Single specimens were found in the samples taken from the river in October and November 1956. These specimens were larger than usual, varying between 129 and 147 μ in length.

Lesquereusia modesta Rhumbler

This species was only found in small numbers at irregular intervals both in the river and in the pool; it was most frequently found when the river was in flood.

Lesquereusia spiralis (Ehrenberg)

The seasonal occurrence of this species is shown in Fig. 33. It is quite clear that its occurrence in the plankton is entirely due to the flooding of the river washing the rhizopod from its normal habitat on sub-aquatic vegetation.

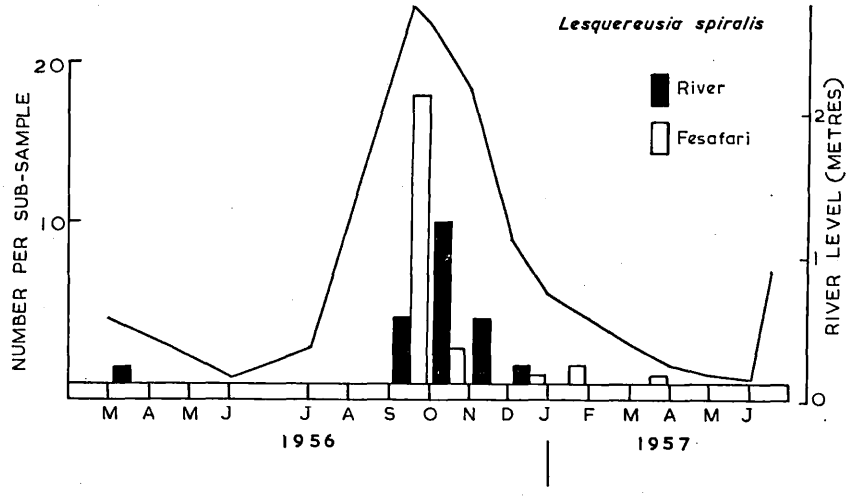


Fig. 33—Seasonal occurrence of *Lesquereusia spiralis* in the plankton.

Nebela collaris (Ehrenberg) Leidy

Only a single specimen was found in the sample taken from the river in August 1956. The length of this specimen was 130 μ.

DISCUSSION

The highest numbers of species, taking the river and the pool together, were found in October and November (Table 1), when the river was in full flood. The lowest numbers of species were found in May and June, when the river was at its lowest. The number of species present in the plankton in March of both years was surprisingly high, and examination of Fig. 2 shows that the total number of individuals was also high at this time. The annual cycle of the Testacea as a whole shows a definite peak of abundance, both in terms of species and individuals, during the floods, with a subsidiary peak centred about March The hydrographic conditions cannot be held responsible for this dry season peak; the production of planktonic forms is the most likely explanation.

Of the species found in this study only four have seasonal occurrences which indicate that they produce planktonic forms; these are : *Arcella vulgaris*, *Difflugia limnetica*, *D. lobostoma* and *Centropyxis aculeata*. The clearest example

Table 1—Seasonal variation in the number of species present in each sub-sample.

Month	Number of species in main channel	Number of species in Fesafari	Total number of species in both samples
March 56	13	11	18
April 56	10	8	15
May 56	5	7	10
June 56 (early)	6	9	11
June 56 (late)	9	14	17
July 56	no sample	no sample	—
August 56	12	no sample	—
September 56	17	15	21
October 56	14	20	22
November 56	17	14	22
December 56	19	13	20
January 57	no sample	17	—
February 57	13	7	17
March 57	13	17	19
April 57	12	10	16
May 57	8	9	12
June 57	9	no sample	—

of a planktonic form is the light shelled form of *Difflugia lobostoma* which was found in its highest numbers in March and April 1956. Another good example is the marginate form of *Arcella vulgaris* which was also abundant in March 1956, as well as showing a peak during the floods.

Although the light shelled form of *D. lobostoma* has smaller particles adhering to its test than the typical form it is not known how this occurs. It may be assumed that during the periods when there is a low rate of flow in the river there is little mineral material in suspension. The structure of the test would then be a reflection of the material available in the immediate environment of the animal once it had assumed a planktonic mode of life. Another

possibility would involve selection of smaller particles by individuals which were about to become planktonic.

One of the factors which may play a part in stimulating the production of planktonic forms is the oxygen content of the water. Bles (1929) has shown that *Arcella discoides* can produce gas vacuoles when subjected to conditions of oxygen shortage, and that these vacuoles make the animal more buoyant. The oxygen content of the river water is at its lowest when the river rises at the begining of the floods and washes stagnant water from the bordering swamps (Holden & Green, 1960). This may well be an additional cause of the peak of abundance of rhizopods in the plankton at this time. The production of gas vacuoles is not restricted to the genus *Arcella*; abundant gas vacuoles were noted in specimens of *Difflugia gramen* which occurred in vast numbers in the plankton of some ponds in Czechoslovakia (Stěpánek & Jiří, 1958).

Some species, such as *Arcella polypora* and *Difflugia corona* were found throughout the year and showed no particular relation in their abundance to the hydrographic conditions. These may well have been producing small numbers of planktonic individuals, but the numbers found were too low to be certain of this.

Most of the other species were adventitious in the plankton. The best example of a species not producing planktonic forms is given by *Lesquereusia spiralis* (Fig. 33), which was absent from the samples from April to August and only appeared when the river was in full flood.

Viewing the list of species as a whole Dr O. W. Heal (personal communication) has remarked that it is typical of neutral waters. It is relevant to note that Holden & Green (1960) found the water of the Sokoto to have a high alkalinity and a pH which rarely fell below 7·0

Some of the Sokoto specimens are larger than is usual for members of their species. This might be due to the relationship between size and water content of the environment which has been noted by Deflandre (1937) and Decloitre (1953), and which has been demonstrated experimentally by Heal (1963) in the genus *Nebela*. Clearly any specimens found in the plankton samples must have originated from habitats in or close to the river; such habitats would obviously tend to have high water contents. A further factor which may have influenced the average size of the specimens in this study is the selective effect of the net. It is possible that other smaller species were actually present, and possibly smaller specimens of the species actually recorded, but these would not have been retained by the net. The Sokoto specimens of *Difflugia corona* are notable in being distinctly smaller than usual; no explanation is available.

ACKNOWLEDGMENTS

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ЗБОРНИК НА РАБОТИТЕ
RECUEIL DES TRAVAUX

J. Green

CRUSTACEA IN LAKE OHRID, WITH SPECIAL REFFERENCE TO THEIR PARASITES AND EPIBIONTS

J. Грeен

КРУСТАЦЕИТЕ НА ОХРИДСКОТО ЕЗЕРО, СО СПЕЦИЈАЛЕН ОСВРТ НА НИВНИТЕ ПАРАЗИТИ И ЕПИБИОНТИ

AUTHOR'S NOTE

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CRUSTACEA IN LAKE OHRID, WITH SPECIAL REFERENCE TO THEIR PARASITES AND EPIBIONTS

by J. Green

Zoology Dept., Bedford College, University of London.

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A detailed review of the fauna of Lake Ohrid has been given by Stanković (1960). The lake is unique in Europe in that it contains a considerable number of endemic species belonging to various animal groups. The Crustacea of the lake include a number of species not found elsewhere, and it was thought worthwhile to make a general survey of the parasites and epibionts of the group to see if they showed any differences from the parasites and epibionts of related Crustacea from other parts of Europe.

The material on which this paper is basad was collected in August 1959. Most of the collections were made from the shore of the lake, but a few dredged samples were examined, and living plankton samples were also taken. The first collections revealed that the Cladocera of the lake were not adequately known, so that this group received particular attention, with the result that a number of additions were made to the known fauna of the lake.

In the following pages the Crustacea which I examined are first noted, and then the parasites and epibionts are described systematically.

CLADOCERA

Sida crystallina (O. F. Müller) — among water lilies, Ohrid Bay, and among vegetation in Ohrid Harbour. No parasites or epibionts found.

Daphnia pulex (De Geer) — planktonic. The form of this species in the lake is slimmer and more transparent than the typical pond form. The fat cells are unusual in that they are coloured with a purple carotenoprotein. This is a feature frequently found in planktonic forms of *D. longispina.*, and if the specimens from Lake Ohrid were only superficially examined they might be misidentified as this species, which might account for earlier records of *D. longispina* in the lake which have not been confirmed. All the specimens which I examined were *D. pulex.*

An interesting feature of the Ohrid population of *D. pulex* is the relatively large size of the eggs in the brood pouches of the females. In a previous study of egg size in the genus *Daphnia* (Green, 1956) it was shown that the eggs

of the planktonic species *D. hyalina* are relatively larger, in proportion to the size of the female, than in pond dwelling species. The eggs of *D. pulex* in Lake Ohrid form an even more striking example. Here we have a planktonic variety producing eggs which are much larger than those produced by the same species when dwelling in ponds. The mean volume of 115 eggs measured from pond dwelling females of *D. pulex* from England was 0.0078 cu. mm. The mean volume of 10 eggs from Lake Ohrid was 0.0169 cu. mm, which is over twice the egg volume from pond dwelling specimens. The females from which the eggs were measured were no larger than pond dwelling forms. It is noteworthy that the large size of the eggs of *D. pulex* in Lake Ohrid is associated wilh a low number of eggs per female. In August 1959, when the measurements were made, each female was carrying only one or two eggs.

The large size and low number of eggs may be a feature which has evolved in association with the low productivity of Lake Ohrid. A young cladoceran may stand a better chance of survival in a poor food situation if it has been well provided with nutrients at the beginning of its life.

About 10 per cent of the mature females were found to be carrying *Colacium vesiculosum* attached to their carapaces. The epibiont was sparse, only three to five individuals per infested host.

A microsporidian, *Plistophora intestinalis*, was unusually abundant in the gut wall of *D. pulex;* about 20 per cent of the adult females were infected.

Ceriodaphnia reticulata (Jurine) — in pools, Ohrid Marsh. This species has not previously been recorded from Ohrid. No parasites or epibionts were found.

Simocephalus vetulus (O. F. Müller) — among vegetation in Ohrid harbour, also in pools, Ohrid Marsh. Not previously recorded from the lake. No parasites or epibionts found.

Scapholebris mucronata (O. F. Müller) — among vegetation, Ohrid Bay, also in pools, Ohrid Marsh. In the latter locality most of the specimens were carrying *Colacium vesiculosum;* some individuals had as many as 50 of the epibionts attached to their carapaces.

Bosmina longirostris (O. F. Müller) — planktonic, and among water lillies, Ohrid Bay. No parasites or epibionts found.

Echinisca tenuicornis (Kurz) — in a pool in Ohrid Marsh. Not previously recorded from Ohrid. No parasites or epibionts found.

Ilyocryptus sordidus (Liéven) — burrowing in mud at depths from 1 to 20 metres. No parasites or epibionts found. Not previously recorded from the lake.

Ilyocryptus acutifrons Sars — burrowing in mud at a depth of 20 metres. No parasites or epibionts found. Not previously recorded from the lake.

Eurycercus lamellatus (O. F. Müller) — in the canal leading from the Hydrobiological Station to the lake. Not previously recorded from the lake. Some specimens carried small numbers of *Cocconeis placentula*, and one was found infected with a microsporidian, *Plistophora obtusa*.

Graptolebris testudinaria (Fischer) — from mud in water lily bed, Ohrid bay, and in canal from Hydrobiological station to lake. Not previously recorded from the lake. No parasites or epibionts found.

Manospilus dispar Sars — in pale brown mud from depth of 20 metres. Not previously recorded from the lake. No parasites or epibionts found.

Camptocercus rectirostris Schödler — in pale brown mud from depth of 20 metres. No parasites or epibionts found.

Acroperus harpae Baird — abundant in the littoral zone, also in pools in Ohrid Marsh. Several specimens from the marsh were found infected with the pink bacterium, *Spirobacillus cienkowskii.*

Alona rectangula Sars — in pools, Ohrid Marsh. Not previously recorded from Ohrid. No parasites or epibionts found.

Alona affinis Leydig — mainly on surface of mud in littoral zone, but also found at depth of 20 metres. Not previously recorded from the lake. No parasites or epibionts found.

Alona guttata Sars — abundant in the littoral zone. No parasites or epibionts found.

Alonella exigua (Lilljeborg) — abundant among vegetation in Ohrid Harbour, males and ephippial females were present. No parasites or epibionts found.

Rhynchotalona rostrata (Koch) — from mud in water lily bed, Ohrid Bay, and in pools in Ohrid Marsh. Not previously recorded from the lake. No parasites or epibionts found.

Pleuroxus trigonellus (O. F. Müller) — from mud in water lily bed, Ohrid Bay, and among vegetation, Ohrid Harbour. Not previously recorded from the lake. No parasites or epibionts found.

Chydorus sphaericus (O. F. Müller) — among vegetation in littoral zone. Large numbers examined but no parasites or epibionts found.

COPEPODA

Macrocyclops albidus (Jurine) — in the canal from the Hydrobiological Station to the lake, and in Ohrid harbour. The following epibionts were found in small numbers: *Epistylis lacustris, Colacium vesiculosum,* and *Tokophyra cyclopum.*

Macrocyclops fuscus (Jurine) — in a small stream flowing into the canal from the Hydrobiological Station to the lake; one specimen from this locality was found infected with the microsporidian *Plistophora cyclopis.*

This copepod was also found in the cool spring water at St. Naum. Not previously recorded from the lake.

Cyclops agilis Koch. Sars (= *C. serrulatus* Fischer) — the commonest of the littoral species. The following epibionts were found: *Epistylis lacustris, E. digitalis, E. zschokkei, Pyxidium henneguyi, Cocconeis placentula* and *Colacium vesiculosum.*

Cyclops viridis (Jurine) — in material from 60 metres depth. Some specimens had a dense even covering of *Cocconeis placentula* and a few specimens of *Colacium vesiculosum.*

Cyclops ochridanus Kiefer — Pranktonic. Specimens from Ohrid Bay had a dense covering of *Colacium vesiculosum,* while the two diaptomids, *Arctodiaptomus steindachneri* and *Eudiaptomus gracilis* which were abundant in the same sample did not carry any specimens of this epibiont.

OSTRACODA

Limnocythere ochridense Klie — common in the littoral zone. *Colacium vesiculosum* was found sparsely on specimens from a small stream flowing into the canal from the Hydrobiological Station to the lake.

Leptocythere prcboscidea Klie — in fine brown mud from a depth of 20 metres. An unidentified peritrich, with a contractile stalk and the zooid enclosed in a gelatinous case covered in detritus was found in small numbers on this ostracod.

Darwinula stevensoni Brady and Roberts — in the littoral zone. No parasites or epibionts found.

Cypridopsis vidua (O. F. Müller) — in the littoral zone. No parasites or epibionts found.

Cyclocypris ovum (Jurine) — in the cool water spring at St Naum. No parasites or epibionts were found.

Cypria ophthalmica (Jurine) — in the cool water spring at St Naum. The following epibionts were found in small numbers: *Pyxidium sp.*, *Lagenophrys sp.* and an unidentified microsporidian was found in one specimen (see p. and 7).

ISOPODA

Asellus aquaticus arthrobranchialis f. *balcanica* Karaman — specimens were examined from the following localities.

1) The canal from the Hydrobiological station to the lake. Most of the specimens had *Cocconeis placentula* on the telson and a *Vorticella sp.* on the legs.

2) A small stream leading into the canal. *Cocconeis* was again found, and *Intrastylum simulans* was abundant on the mouthparts and legs.

3) In the cool springs at St. Naum. Only a few *Cocconeis* were found.

Asellus remyi Monod — a small specimen was found in the littoral zone; it bore a few specimens of *Cocconeis* on its telson.

Asellus gjorgjevići Karaman — specimens from 20 metres depth in Ohrid Bay were found with *Pyxidium aselli* and a *Vorticella sp.* on their legs.

AMPHIPODA

Gammarus ochridensis Schaferna — specimens from two localities were examined.

1) In the littoral zone near St. Stephan the specimens were green in colour. About 20 per cent of the individuals were infected with *Eccrinella gammari*, and all carried *Zoothamnium hyalinum* on their legs.

2) Specimens from 40 metres depth were red in colour; their gills were infected with *Dendrocometes paradoxus*.

Gammarus roeseli Gervais — specimens from two localities were examined.

1) In the canal leading from the Hydrobiological Station to the lake all the animals carried *Zoothamnium hyalinum* on their legs and *Spirochona gem-*

mipara on their gills. One specimen was found with a small colony of *Colacium vesiculosum* near the tip of one leg.

Another was found infected with the larva of an acanthocephalen, *Polymorphus minutus*. Several specimens were infected with *Eccrinella gammari*.

2) Specimens from the cool springs at St. Naum carried *Zoothamnium hyalinum* on their legs, *Spirochona gemmipara* on their gills and *Vorticella incisa* on their telsons.

SYSTEMATIC NOTES ON PARASITES AND EPIBIONTS

BACTERIA

Spirobacillus cienkowskii Metchnikoff 1889 — this bacterium has been found in various species of Cladocera in other parts of Europe. A few specimens of *Acroperus harpae* from pools in Ohrid Marsh were found to be infected. A characteristic feature of the later stages of infection is the bright red colour assumed by the cladoceran. This is due to a carotenoid pigment produced by the bacteria (Green, 1959). The difference between this colour and the redness due to haemoglobin, which is a common pigment in Cladocera (cf. Fox, 1948), is very simple to observe with a microspectroscope. The carotenoid shows a distinct absorption band at 546—550 mμ and a broader, less distinct band at about 500 mμ, while haemoglobin has characteristic absorption bands at 576 and 544 mμ.

ALGAE

Colacium vesiculosum Ehrenberg — this member of the Euglenoidea is a common and widespread epibiont of many different freshwater animals in Europe (cf. Pringsheim, 1953). Although it is recorded from a wide range of hosts it sometimes shows peculiar and at present inexplicable preferences for a certain host when other apparently suitable hosts are available. Thus, for instance, in a plankton sample taken from Lake Ohrid in August 1959 the copepod *Cyclops ochridanus* was found to be densely covered with *Colacium*, while *Diaptomus gracilis* and *Arctodiaptomus steindachneri* completely lacked this epibiont.

Other hosts on which *Colacium* has been found at Ohrid are: *Daphnia pulex*, *Scapholebris mucronata*, *Macrocyclops albidus*, *Cyclops agilis*, *Cyclops viridis*, *Limnocythere ochridense* and *Gammarus roeseli*.

Cocconeis placentula Ehrenberg — this diatom is an abundant epibiont on various aquatic plants, but it occurs on various Crustacea with sufficient regularity to be regarded as more than accidental in occurrence. It may well be that attachment to animals serves as an aid to distribution. At Ohrid *Cocconeis* was found on *Eurycercus lamellatus*, *Cyclops agilis*, *Cyclops viridis*, *Asellus aquaticus* and *Asellus remyi*.

ECCRINALES

(see Manier, 1955, for a classification of this group).

Eccrinelia gammari Leger & Dubosq — this species was originally described from *Gammarus pulex*. At Ohrid it was found in *Gammarus roeseli*

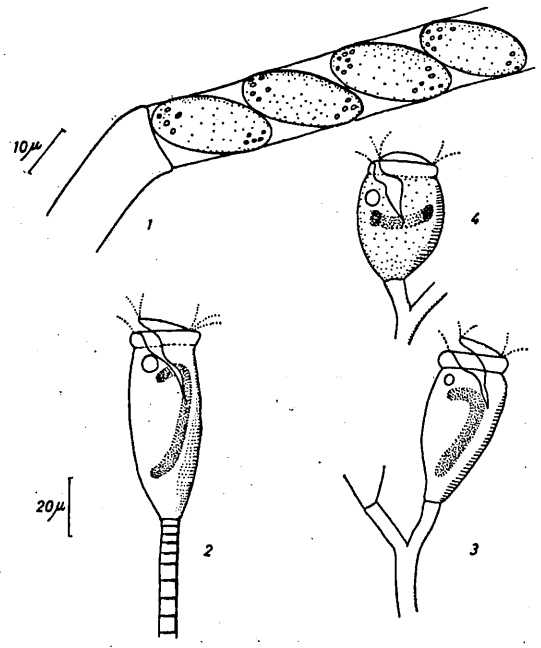
and *Gammarus ocridensis*. A characteristic stage, with the production of microspores is shown in fig. 1.

CILIATA

Epistylis digitalis Ehrenberg (fig. 2) — this peritrich was found only once on *Cyclops agilis*. This is a regular epibiont of *Cyclops spp.* (see for instance: Perty, 1852: Kent, 1880-2; Keiser, 1921, Nenninger, 1948 and Matthes 1950).

Epistylis lacustris Imhoff (fig. 3) — this was the most frequent and abundant peritrich on *Cyclops agilis* and *Macrocyclops albidus*. Although usually present in the form of colonies it was occasionally found in the solitary form. This solitary form might easily be misidentified as a *Rhabdostyla sp.* if not compared closely with the colonial form. This species is usually found on *Cyclops spp.*, but it has been recorded on other arthropods (cf. Matthes, 1950).

Epistylis zschokkei (Keiser) (fig. 4) — was also found on *Cyclops agilis*. It has been recorded previously from both cyclopoid and harpacticoid copepods, and from the cladoceran *Acantholebris curvirostris* (Keiser, 1921, Nenninger 1948).



Figs 1—4. Parasites and epibionts of Crustacea in Lake Ohrid. 1, *Eccrinella gammari*, part of a filament showing microspores, 2, *Epistylis digitalis*. 3, *Epistylis lacustris*. 4, *Epistylis zschokkei*. The three species of *Epistylis* are all drawn to the same scale.

Pyxidium henneguyi Fauré Fremiet (fig. 5) — only a few specimens were found, attached to the abdomen of *Cyclops agilis.*

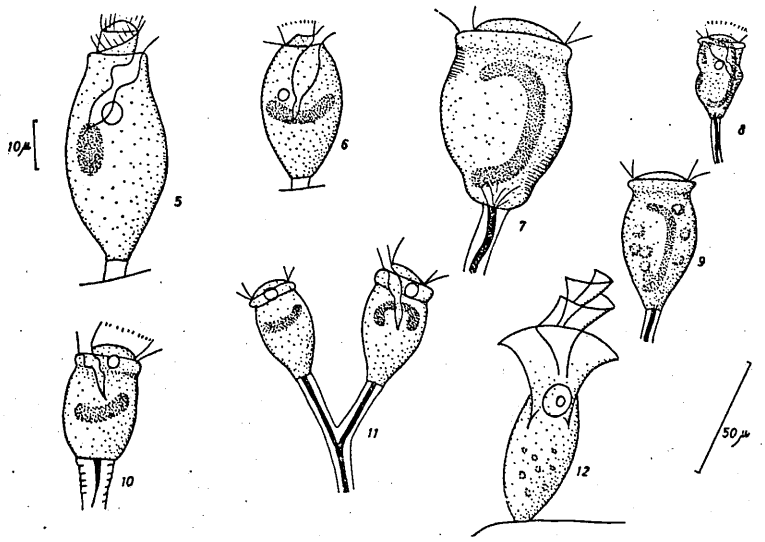
Pyxidium aselli Penard (fig. 6) — was found sparsely on the legs of *Asellus gjorgjevici* from a depth of 20 metres.

Pyxidium sp. — a single specimen of a small *Pyxidium* was found on *Cypria ophthalmica* from the cool springs at St. Naum. The body was 36μ long and 21μ at its greatest width. The stalk was very short. Unfortunately it was nos possible to see any details of the disc, pharynx or nucleus so that it is not possible to asign the specimen to a species.

Vorticella incisa Stiller 1938 (fig. 7) — I have included the specimens found on the telsons of *Gammarus roeseli* in Stillers species on the basis of their body size and shape. Stiller's original figure does not show the form of the nucleus, and I was not able to elucidate the structure of the pharynx in my specimens, so that the identification must be regarded as uncertain. The body length of the specimcns from Ohrid was about 72μ, and the width about 54μ. The stalk reached a lenght of 200μ.

Vorticella spp. — two other *Vorticella spp.* were seen during the survey. These are shown in figs 8 and 9. Only single specimens of each were seen and not enough detail was visible to be sure of identification. The specimen from *Asellus gjorgjevici* kept its constriction in the body for the whole period of observation (about 15 minutes).

Intrastylum simulans Plate (fig. 10) — this species was abundant on the mouthparts and legs of *Asellus aquaticus.*



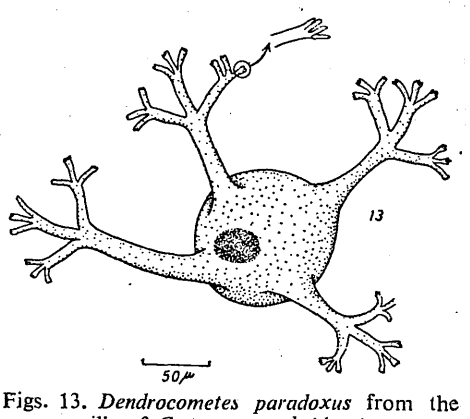
Figs 5—12. Epibionts of Crustacea in Lake Ohrid. 5, *Pyxidium henneguyi,* this specimen is drawn to twice the scale of the other figures, which are all to the same scale. 6, *Pyxidium aselli.* 7, *Vorticella incisa.* 8, *Vorticella* sp. from *Asellus gjorgjevici.* 9, *Vorticella* sp. from. *Asellus aquaticus.* 10, *Intrastylum simulans.* 11, *Zoothamnium hyalinum.* 12, *Spirochona gemmipara.*

Zoothamnium hyalinum Stiller 1953 (fig. 11) — this species was originally described from the legs of *Gammarus roeseli* in Lake Balaton. The specimens from Ohrid agree well with Stiller's description. They were found on the legs of *G. roeseli* and *G. ochridensis.*

Lagenophrys sp. — a single specimen was found on *Cypria ophthalmica* from the cool springs at St. Naum. The shell was ovoid in shape, measuring $72 \times 61\mu$. The case was attached by one flat side to the shell of the ostracod, the other side was rounded and projected about 28μ above the surface of the hosts shell. Because only a single specimen was found, and the details of the case noi adequately observed I have not asigned this specimen to a species.

CHONOTRICHA

Spirochona gemmipara Stein (fig. 12) — typical specimens of this species were found on the gills of *Gammarus roeseli.*



Figs. 13. *Dendrocometes paradoxus* from the gills of *Gammarus ochridensis.*

SUCTORIA

Dendrocometes paradoxus Stein (fig. 13). The number of branches and their subdivisions were slightly variable in the Ohrid material, but the figure is of a typical specimen. The host was *Gammarus ochidensis.*

Tokophyra cyclopum (Claparede & Lachmann) — several young specimens were found on *Macrocyclops albidus.*

MICROSPORIDIA

Plistophora intestinalis Chatton — was found infecting *Daphnia pulex.* The spores were 3μ long and 1.5μ wide. The width of the Ohrid spores is a little less than that given by Jirovec (1936) for Czechoslovakian specimens, but the Ohrid form is identical in other respects, including the site of infection, which is in the anterior part of the hosts midgut.

Plistophora obtusa (Moniez) — was found infecting *Eurycercus lamellatus,* this is the first host recorded outside the *Daphnidae.* The spores were about 3μ long and were somewhat pear shaped, agreeing with the descriptions given by Jirovec (1937) and Weiser (1947).

Plistophora cyclopis Leblanc. Specimens of *Macrocyclops fuscus* were found infected with this species. The spores were mostly $2{,}3\mu$ long and 1.5μ

wide, and were evenly rounded at each end. This species does not appear to have been recorded outside Belgium.

Microsporidian ? indet. — a single specimen of *Cypria ophthalmica* from the cool springs at St. Naum was found with its body cavity full of spores. These were circular with a diameter of 4μ when viewed from one direction and were ovoid with diameters of 4 and 3μ when viewed in a direction at right angles to the first point of view. No pansporoblasts were seen so that it is not possible to asign the spores to a genus.

ACANTHOCEPHALA

Polymorphus minutus (Goeze) — a single pink larva was found in *Gammarus roeseli*. This is a common parasite of *Gammarus spp.;* the adult is a parasite of water birds.

DISCUSSION

The survey described in this paper has added thirteen species to the known fauna of Lake Ohrid and its adjoining marsh. These species are all widespread in Europe, and most of them were collected in the littoral zone. This indicates that the littoral of Lake Ohrid contains a typically European fauna of the smaller Grustacea. It is only among the larger bottom dwelling Crustacea such as amphipods and isopods that endemic littoral Crustacea are found. The ostracods form a partial exception to this, but even in this bottom dwelling group most of the species in the littoral zone are non endemic, whereas those from deep water are almost entirely endemic (cf. Stanković 1960).

The parasites and epibionts found in the presant survey are also typical European species, but it should be emphasised that most of the hosts were collected from the littoral zone.

If the formation of endemic species depends on isolation from surrounding forms then the littoral zone is the least likely place for small endemic Crustacea to be found. The Cladocera and Copepoda inhabiting this region are capable of producing resting stages which are easily transported among bird feathers or on birds feet Their parasites and epibionts might easily be transported in the same way. The chances of endemism occuring in the Crustacea and their parasites in a lake like Ohrid may be summarised as follows.

LITTORAL — small species, easily transported, access to transportation (birds feet etc.), produce resistant stages. Hence low chances of isolation. large species, not so easily transported, do not produce resistant stages. Hence higher chance of isolation.

SUB-LITTORAL (and below) — no access to transportation, isolation from littoral by *Chara* beds. More uniform environment and lack of resistant stages. Very high chance of isolation.

PLANKTON — limited access to transport. Often do not produce resistant stages. Lower chances of isolation than sub-littoral, higher than littoral.

SUMMARY

The following species of Crustacea are recorded from Lake Ohrid for the first time: *Simocephalus vetulus, Ilyocryptus sordidus, I. acutifrons, Eurycercus lamellatus, Graptoleberis testudinaria, Monospilus dispar, Alona affinis, Rhynchotalona rostrata, Pleuroxus trigonellus,* and *Macroyclops fuscus.*

A further three species are recorded from pools in Ohrid Marsh, which lies on the margin of the lake, between the Hydrobiological Station and the town. These species are: *Ceriodaphnia reticulata, Echinisca tenuicornis* and *Alona rectangula.*

A brief survey of the parasites and epibionts of Ohrid Crustacea revealed the presence of at least twenty four species, nineteen of which have been identified as follows.

BACTERIA. *Spirobiacillus cienkowskii.*

ALGAE. *Colacium vesiculosum, Cocconeis placentula.*

ECCRINALES. *Eccrinella gammari.*

CILIATA. *Epistylis digitalis, E. lacustris, E. zschokkei. Pyxidium henneguyi, P. aselli, Vorticella incisa, Intrastylum simulans, Zoothamnium hyalinum.*

CHONOTRICHA. *Spirochona gemmipara.*

SUCTORIA. *Dendrocometes paradoxus, Tokophyra cyclopum.*

MICROSPORIDIA. *Plistophora intestinalis, P. obtusa, P. cyclopis.*

ACANTHOCEPHALA. *Polymorphus minutus.*

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КРУСТАЦЕИТЕ НА ОХРИДСКОТО ЕЗЕРО, СО СПЕЦИЈАЛЕН ОСВРТ НА НИВНИТЕ ПАРАЗИТИ И ЕПИБИОНТИ

(J. Green, Zoology Dept., Bedfort College, University of London)

ЗАКЛУЧОК

(Примено за печатење на 17. II. 1960)

Следните врсти *Crustacea* се забележени прв пат од Охридското езеро: *Simocephalus vetulus*, *Iliocryptus sordidus*, *I. acutifrons*, *Eurycercus lamellatus*, *Graptoleberis testudinaria*, *Monospilus dsipar*, *Alona affinis*, *Rhynchotalona rostrata*, *Pleuroxus trigonellus* и *Macrocyclorps fiscus*.

Други три врсти се забележени од барите во Охридското Блато кое се наоѓа на обалата од Езерото, меѓу Хидробиолошкиот завод и градот. Тие врсти се: *Ceriodaphnia reticulata*, *Echinisca tenuicornis i Alona rectangula*.

Краткиот опис на паразитите и епибионтите на охридските крустацеи открива присуство на најмалу дваесет и четири врсти, деветнаесет од кои се опишани како што следува:

BACTERIA. *Spirobacillus cienkowskii.*

ALGEA. *Colacium vesiculosum*, *Cocconeis placentula.*

ECCRINALES. *Eccrinella gammari.*

CILIATA. *Epitstylis digitalis*, *E. lacustris*, *E. zschokkei*, *Pyxidium henneguyi P. aselli*, *Vorticella incisa*, *Intrastylum simulans*, *Zoothamnium hyalinum.*

CHONOTRICHA. *Spirochona gemmipara.*

SUCTORIA. *Dendrocometes parodoxus*, *Tokophyra cyclopum.*

MICROSPORIDIA. *Plistophora intestinalis*, *P. obtusa. P. cyclopis.*

ACANTHOCEPHALA. *Polymorphus minutus.*

TWO NEW SPECIES OF *PARABATHYNELLA* (CRUSTACEA : SYNCARIDA) FROM LAKE ALBERT, UGANDA

BY

J. GREEN

Bedford College, University of London

[Accepted 12th February 1963]

(With 20 figures in the text)

Parabathynella bakeri sp. n. and *P. ninianae* sp. n. are described and figured. These species were found in the interstitial water of a sand spit on the shore of Lake Albert, and are the second and third species of *Parabathynella* recorded from Africa. A synoptic table of the genus is given, and the genera *Brasilibathynella* and *Thermobathynella* are discussed.

CONTENTS

INTRODUCTION

The interstitial fauna has received a considerable amount of attention in Europe and North America (cf. Delamare Deboutteville (1960) for a recent survey), but in Africa there have been few attempts to study this fauna. During a visit to Lake Albert in October 1962 I had the opportunity to sample the interstices of Kaiso Spit, which was one of the few areas of sandy shore not flooded at that time. The sample was taken by digging a hole about two feet deep in the sand near the water's edge and sweeping a small hand net through the water which collected at the bottom of the hole. This technique yielded thirty specimens of *Parabathynella* belonging to two distinct species, which I name after Sir Samuel Baker and his wife (formerly Florence Ninian von Sass), who first put the Albert Nyanza on the map of Africa.

PARABATHYNELLA BAKERI sp. n.

Adult male. Length up to 1·2 mm. The whole body is elongated and almost uniform in thickness (Fig. 1). The head is nearly twice as long as wide and without any trace of eyes. The body segments are smooth without any tendency to form a dorsal carina. The antennules are slightly longer than the head, each with six podomeres. The endopod of the antennule is reduced to a minute projection bearing three setae. The exopod is formed by three podomeres, the first of which has a distal projection bearing two aesthetascs.

Each antenna is composed of four podomeres, the total length being shorter than the head. The terminal podomere bears two setae and an aesthetasc.

The anterior border of the labrum bears eight teeth which are slightly irregular in size (Fig. 4).

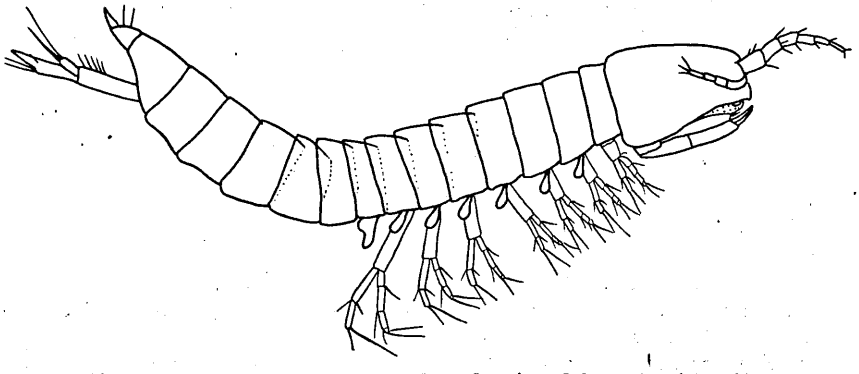


Fig. 1—*Parabathynella bakeri*, adult male, viewed from the right side.

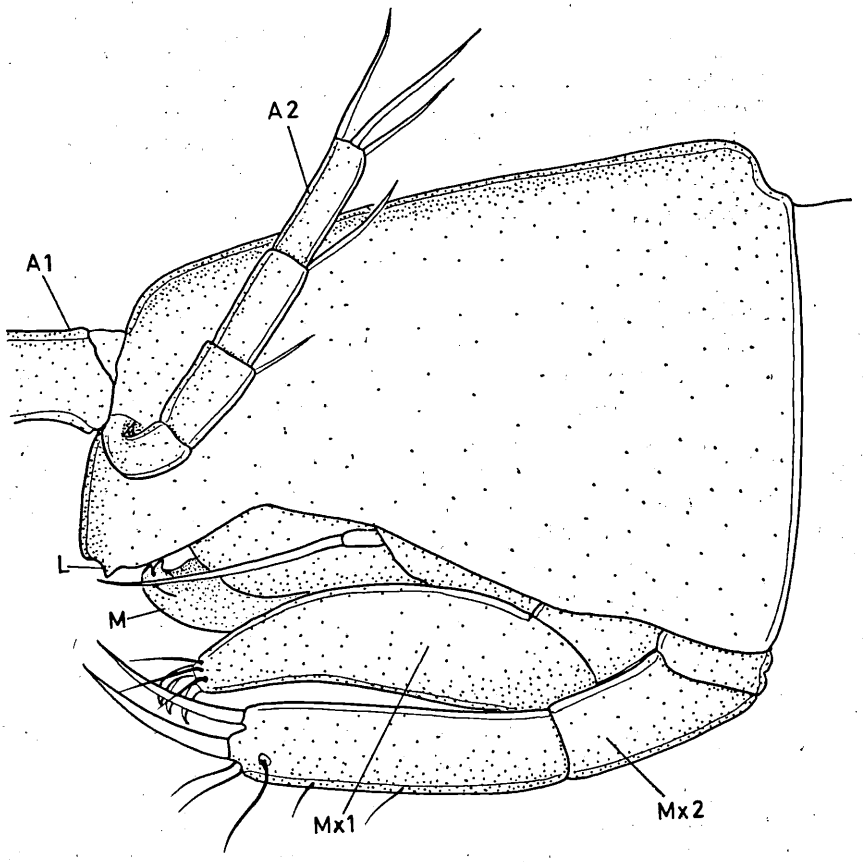
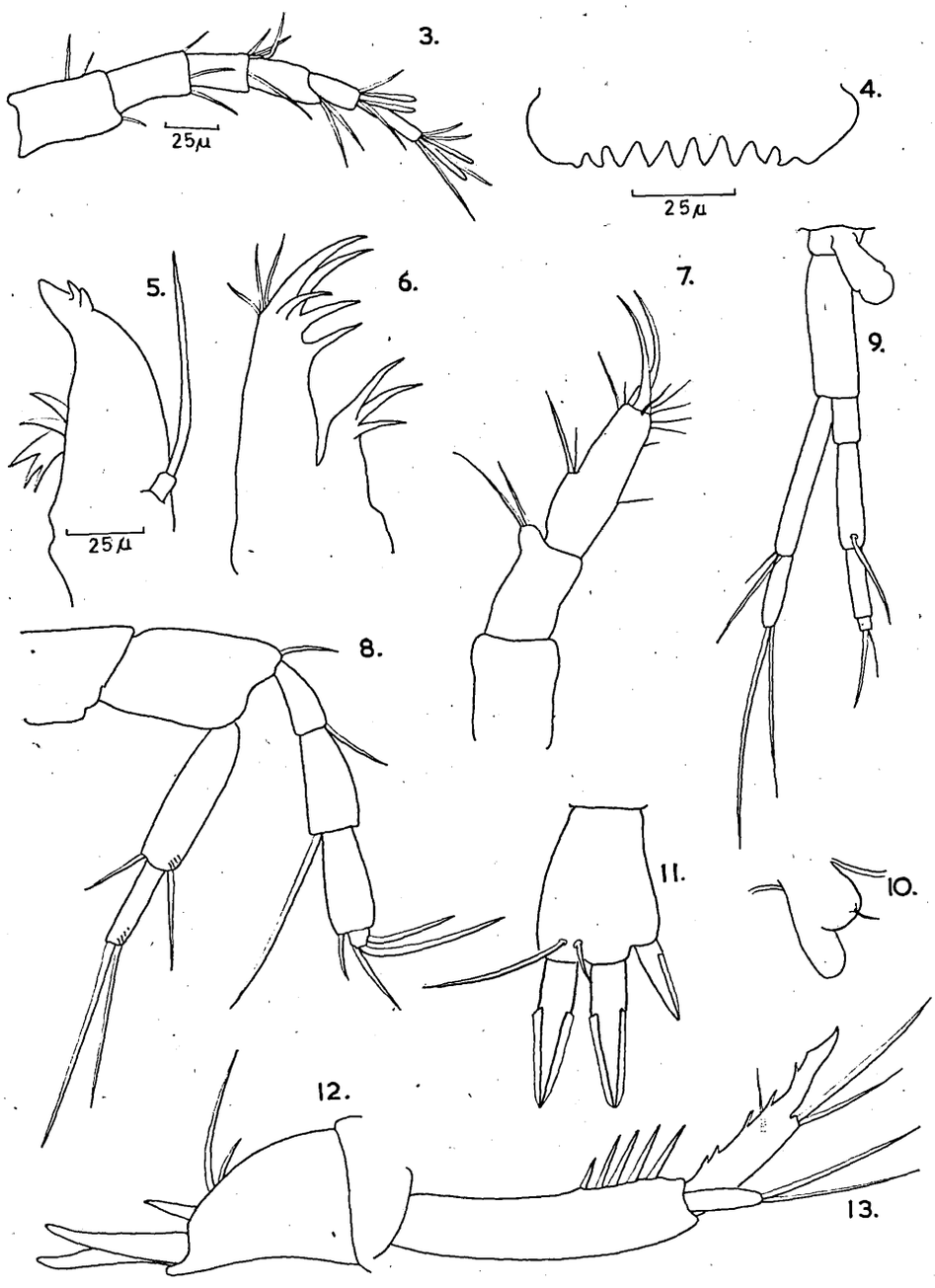


Fig. 2—*Parabathynella bakeri*, head of adult male, viewed from the left side ; only the base of the antennule is shown. A1—antennule ; A2—antenna ; L—labrum ; M—mandible ; Mx1—maxillule ; Mx2—maxilla.



Figs. 3–13—*Parabathynella bakeri*. 3, Antennule. 4, Anterior edge of labrum. 5, Mandible. 6, Maxillule. 7, Maxilla. 8, First leg. 9, Leg 7. 10, Leg 8, adult male. 11, Caudal ramus, dorsal view. 12, Caudal ramus, lateral view. 13, Uropod of female, lateral view. Figs. 3, 7, 9, 10 & 13 are drawn to the same scale. Fig. 5 is drawn to the same scale as Fig. 6. Figs. 8, 11 & 12 are drawn to the same scale as Fig. 4.

The mandible has two accessory teeth near the apex of the incisor process. Proximal to the incisor process is another process bearing five spines ; the homology of this process is uncertain (Fig. 5).

Each mandibular palp is formed by a single podomere with a single long seta which reaches forward to the tip of the labrum (Fig. 2).

The form of the maxillule is shown in Fig. 6. In its natural position the spines at the tip are slightly over-ridden by the spines at the tip of the maxilla (Fig. 2).

The maxilla is formed by three podomeres, the terminal one bearing two long spines and eleven setae (Fig. 7).

The first leg differs from legs 2–7 in lacking an epipodite on the coxopodite. All the legs are similarly constructed, but they increase in length progressively from leg 1 to leg 7, and the proportions of each podomere become progressively more elongated in the same sequence. The endopod is formed by four podomeres, the last of which is small and bears two or three long spine-like setae. The exopod is formed by two podomeres ; the terminal one carrying two long setae.

The eighth leg is a small protuberance with a swelling at its base ; a single small seta is present on the swelling (Fig. 10).

The uropod (Fig. 13) bears five spines near the apex of the protopodite. These spines increase slightly in size from proximal to distal, but the most distal is not markedly larger than the others. The endopod is approximately three times as long as the exopod and bears three setae. The exopod is a simple podomere with two long terminal setae.

Each ramus of the caudal furca bears three stout spines and two setae. The spines have a peculiar thin flange along the margin, except for the basal third (Fig. 11).

Adult female. Similar in external structure to the male, except that there is no trace of the eighth leg. The endopod of the uropod tends to have a more irregular margin on the dorsal side than the male, but this is not a constant feature.

The holotype, a male, and some paratypes have been deposited in the British Museum (Nat. Hist.) collection and registered as 1963.2.6.1–14.

PARABATHYNELLA NINIANAE sp. n.

Adult male. Length about 0·65 mm. The body is relatively more slender than that of *P. bakeri*, but otherwise similar in general form. The body segments are smooth without a dorsal carina.

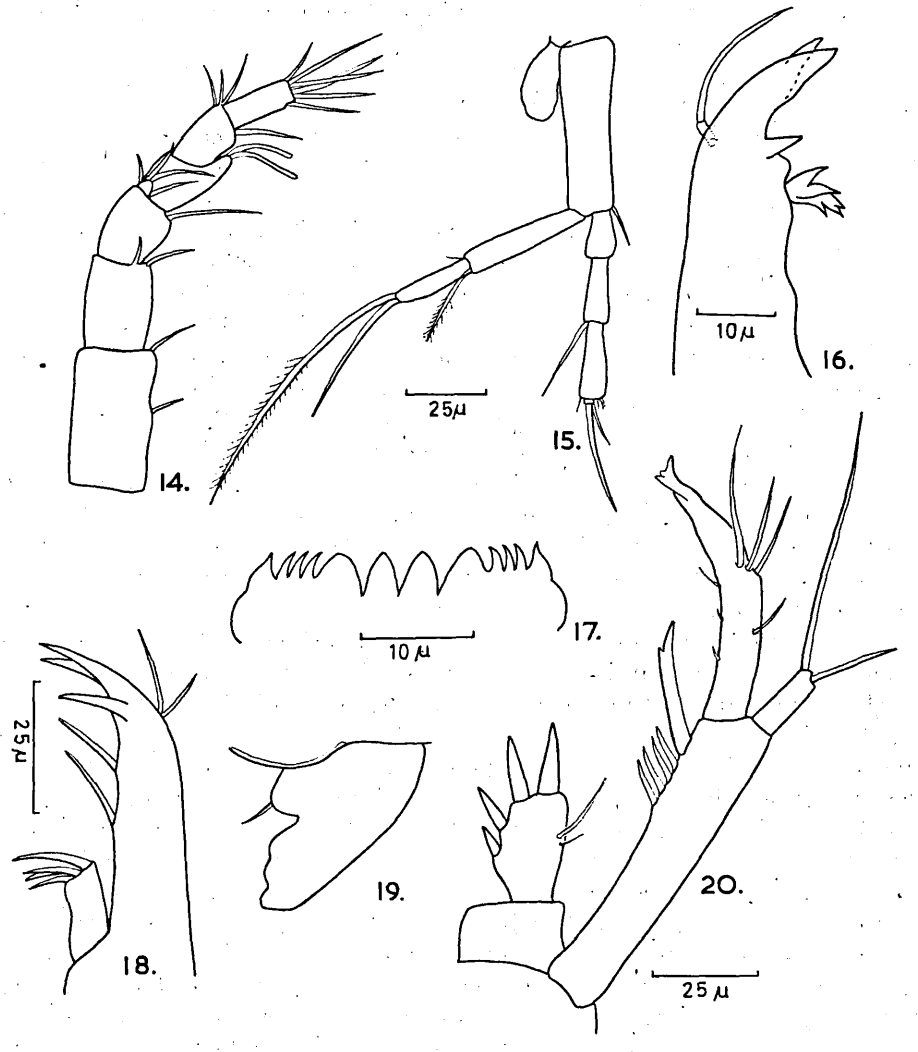
Each antennule is formed by six podomeres (Fig. 14) and each antenna has four podomeres.

The anterior border of the labrum bears twelve teeth (Fig. 17). The four median teeth are much larger than the others.

The incisor process of the mandible has two large terminal teeth. A smaller tooth lies at the same level as the mandibular palp, which is a single podomere with a long seta reaching forwards beyond the tip of the mandible. The mandible also has another process on its median border bearing five small teeth (Fig. 16).

The maxillule is shown in Fig. 18. The general form is similar to that of *P. bakeri*, but the two proximal spines on the median edge are more widely separated from the distal teeth.

The maxilla is formed from three podomeres ; the terminal one bears a single long spine and ten or eleven setae.



Figs. 14–20—*Parabathynella ninianae*. 14, Antennule. 15, Leg 7. 16, Mandible. 17, Anterior edge of labrum. 18, Maxillule. 19, Leg 8. 20, Uropod and caudal ramus. Figs. 14, 19 & 20 are drawn to the same scale.

The legs are similar to those of *P. bakeri*, but the fourth podomere of the endopodite is much smaller, and is very difficult to see on the posterior legs.

The eighth leg is more conical in form than that of *P. bakeri*, but is similar in general structure (Fig. 19).

The uropod (Fig. 20) bears five spines near the apex of the protopodite. The distal spine is much larger than the others. The endopod is about 3·4 times as long as the exopod, and is curved towards the median line. The apex of the endopod bears three small teeth. The exopod is a simple podomere bearing two setae of unequal length.

Each ramus of the caudal furca bears four stout spines which increase in size from proximal to distal. Two setae are also present on the lateral border of each ramus (Fig. 20).

Adult female. Similar in size and external structure to the male, except for the absence of the eighth leg.

The holotype, a male, and a female paratype have been deposited in the British Museum (Nat. Hist.) collection and registered as 1963.2.6.15–16.

AFFINITIES OF THE SPECIES

Parabathynella bakeri is most closely allied to *P. caparti* Fryer (see Table 1), but differs from that species in the following particulars. The body is larger and not so slender. The eighth leg of the male is of a different shape. The endopod of the uropod is longer in relation to the exopod, and the protopodite of the uropod bears five spines instead of four ; the terminal spine of the series is not markedly larger than the others, as it is in *P. caparti*.

Fryer (1957) has pointed out that *P. caparti* has affinities with the species described from Madagascar by Delamare Deboutteville & Paulian (1954). All three of these species have the distal spine of the series on the protopodite of the uropod much larger than the others. The distal spine in *P. caparti* is distinctly larger than the others, but not to the same extent as in the species from Madagascar.

Parabathynella ninianae shows many similarities to the species from Madagascar, particularly in the structure of the uropod, but the ratio of the length of the endopod to the exopod is a clear distinguishing feature.

Table 1 also includes the six species of the genus *Thermobathynella* and the single species of the genus *Brasilibathynella*. This last genus is inadequately described ; and it is very doubtful if it is a valid genus ; the only character which Jakobi (1958) gives as diagnostic is the presence of three instead of four podomeres in the endopods of the legs. This is not a satisfactory character. The numbers of podomeres in various appendages varies considerably within the genus *Parabathynella* (Table 1), so that this character is only of specific value, and Jakobi does not give any other character which would separate *Brasilibathynella* from either *Thermobathynella* or *Parabathynella*. Further, the figures given by Jakobi are on such a small scale that it is quite possible that a small fourth podomere could be overlooked. The fourth podomere on the posterior legs of *Parabathynella ninianae* is so small that it could easily be missed and the endopod be recorded as having only three podomeres.

Uéno (1957) has given a key which separates the genera of the Bathynellidae, but the species of *Thermobathynella* from South America, described by Siewing (1956, 1958) and Noodt (1963), and the species of *Parabathynella* described from Madagascar and Africa (Delamare Deboutteville & Paulian, 1954 ;

Table 1

Species	Number of podomeres			Number of spines		R*	Occurrence
	Antennule	Antenna	Exopod of legs	Ramus	Protopod of uropod		
Parabathynella							
kuma Uéno, 1956	7	5	3	5	14–15	1·3	Japan
carinata Úéno, 1952	6	6	2	6	15	1·3	Japan
lusitanica Braga, 1949	7	?	2	11–13	12	1·0	Europe
miurai Uéno, 1952	6	2	2	6	9	1·4	Japan
gracillimana Uéno, 1956	6	4	2	3	7–8	1·3	Japan
gracillimana ssp. *yaye* Uéno, 1956	7	6	2	3	6–7	1·5	Japan
motasi Dancau & Serban, 1963	7	5	2	5–7	6–8	1·0	Europe
phreatica Chappuis, 1939	6	?	2	4	7	1·0	Europe
fagei Delamare Deboutteville & Angelier, 1950	7	4	3	6–7	5–9	1·0	Europe
malaya Sars, 1929	6	6	3	4	7	1·0	Malaya
bakeri sp. n.	6	4	2	3	5	3·0	Africa
ninianae sp. n.	6	4	2	4	5	3·4	Africa
pauliani Delamare Deboutteville, 1953	6	4	2	4	5	1·8	Madagascar
jeanelli Delamare Deboutteville & Paulian, 1954	6	v. small	2	4	4	1·9	Madagascar
milloti Delamare Deboutteville & Paulian, 1954	6	3	2	4	4	1·7	Madagascar
caparti Fryer, 1957	6	4	2	3	4	2·0	Africa
stygia Chappuis, 1926	6	?	2	4	4	1·0	Europe
Thermobathynella							
adami Capart, 1951	6	5	2	3	1	2·0	Africa
leleupi Delamare Deboutteville & Chappuis, 1955	6	5	2	3	2	2·4	Africa
amyxi Siewing, 1956	5	5	2	3	2	1·0	S. America
jumboli Siewing, 1958	7	3	2	3	6	1·0	S. America
richerti Noodt, 1963	6	5	2	3	2	1·0	S. America
ypacaraiensis Noodt, 1963	6	5	2	3	2	1·0	S. America
Brasilibathynella							
florianopolis Jakobi, 1958	6	5	2	3	2–3	1·0	S. America

* Ratio of the length of the endopod to the exopod of the uropod.

Fryer, 1957) eliminate any possibility of separating these two genera with the characters given by Uéno. The only remaining character is the presence of a distinct tergum and sternum on the abdominal somites of *Thermobathynella*. while in *Parabathynella* these two plates are fused. This alone is scarcely of generic value, and since there appears to be no feature of geographical or ecological distribution associated with this character there seems to be no virtue in maintaining the two genera separately.

ACKNOWLEDGMENTS

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PIGMENTS OF THE HYDRACARINE *EYLAIS EXTENDENS* (ACARI: HYDRACHNELLAE)

J. GREEN

Zoology Department, Bedford College, University of London*

(*Received* 18 *June* 1964)

Abstract—1. Adults of the water mite *Eylais extendens* contain the following carotenoids: β-carotene, a keto-carotenoid, a xanthophyll similar to lutein, and astaxanthin, part of which is esterified.

2. Larvae of the same species contain a keto-carotenoid and astaxanthin, part of which is esterified.

3. Astaxanthin and its esters form over 70 per cent of the total carotenoids in both adult and larva. In the adult, part of the astaxanthin is linked to a protein.

4. A non-carotenoid pink pigment of unknown identity is also found in the adult.

INTRODUCTION

THE water mites are among the most colourful of the Acari, but as yet there has been no attempt to determine the chemical nature of their pigments. The only previous studies on acarine pigments have been concerned with the families Trombidiidae and Tetranychidae, which are terrestrial mites. A summary of this previous work is given by Metcalf & Newell (1962). The present account deals with the pigments present in one species of water mite which is large in size and bright red in colour.

MATERIAL AND METHODS

Adults of *Eylais extendens* (O. F. Müller) were collected from the small overflow pond at the end of the Long Water in Hampton Court Park. A week after the collection of adults there was a remarkable emergence of larvae. The surface of the water around the margin of the pond was coloured scarlet by millions of larvae. The mites and their larvae were carefully examined under a microscope to ensure that no pigmented epibionts were attached to them. Before extracting pigments the adults were kept for several hours in clean water which was changed at intervals. This enabled the mites to finish digesting any food in their intestines.

Extracts of adults and larvae were made by grinding with acetone in a thimble homogenizer. The carotenoids so obtained were transferred to light petroleum (boiling range 40–60°C) after diluting the acetone with water. The crude light petroleum extract was washed with water to remove traces of acetone and was dried over anhydrous sodium sulphate. The pigments were then chromatographed on a

* Present address: Westfield College, Kidderpore Avenue, London, N.W.3.

column of alumina. Separation of the pigments was achieved by washing the column with light petroleum containing gradually increasing percentages of acetone, and the final fraction was eluted with acetone containing a trace of acetic acid.

The fractions eluted from the column were washed with water to remove acetone and then dried over anhydrous sodium sulphate. The final fraction was transferred to light petroleum after dilution of the acetone with water. The absorption spectrum of each fraction was measured in a Unicam S.P. 500 spectrophotometer.

RESULTS

The extract from the adults yielded five fractions:

1. Eluted with 1% acetone in light petroleum; epiphasic in the phase test between light petroleum and 90% methanol. The absorption spectrum in light petroleum showed absorption maxima at 450 and 476 mμ, indicating that the pigment was β-carotene. This fraction formed about 2 per cent of the total carotenoids.

2. Eluted with 5% acetone in light petroleum; epiphasic. A single rounded absorption maximum in light petroleum at 460 mμ. This fraction is similar to the fraction from *Artemia* which was identified as a keto-carotenoid by Gilchrist & Green (1960). About 9 per cent of the total carotenoids were formed by this fraction.

3. Eluted with 10% acetone; epiphasic. Single absorption maximum at 468 mμ in light petroleum. Became hypophasic after saponification, but returned to the epiphase when the hypophase was acidified with acetic acid. This phase behaviour is typical of astacene liberated from an astaxanthin ester by saponification. This fraction formed about 29 per cent of the total carotenoids.

4. Eluted with 60% acetone in light petroleum; hypophasic. Absorption maxima in light petroleum at 448 and 475 mμ. This seems to be a xanthophyll similar to lutein, but the identification cannot be regarded as conclusive without evidence from rechromatography with authentic lutein. This fraction formed about 10 per cent of the total carotenoids.

5. Eluted with acetone and acetic acid; mostly hypophasic. Absorption maximum at 469 mμ in light petroleum. This was the most abundant of the carotenoids, forming about 50 per cent of the total. The chromatographic behaviour and absorption spectrum identify the pigment as astacene derived from astaxanthin.

Another sample of adults was homogenized in distilled water, giving an opalescent orange-pink solution after centrifuging. The orange pigment could be precipitated with 50% ammonium sulphate. The precipitated pigment was treated with acetone, and the resulting solution was diluted with water until the pigment passed into an epiphase of light petroleum. After being dried over anhydrous sodium sulphate, the solution was found to have a simple absorption spectrum with a rounded maximum at 468–469 mμ. This indicates that the pigment obtained in the aqueous extract was astaxanthin linked to a protein.

When larvae were extracted in the same manner as the adults the chromatogram on alumina revealed only three bands:

1. Eluted with 5% acetone in light petroleum; epiphasic, with a single rounded absorption maximum between 456 and 460 mμ in light petroleum. Formed about 25 per cent of the total carotenoids. This seems to be a keto-carotenoid similar to the second fraction obtained from the adults.

2. Eluted with 10% acetone in light petroleum; epiphasic, but became hypophasic after saponification. Single absorption maximum at 468 mμ in light

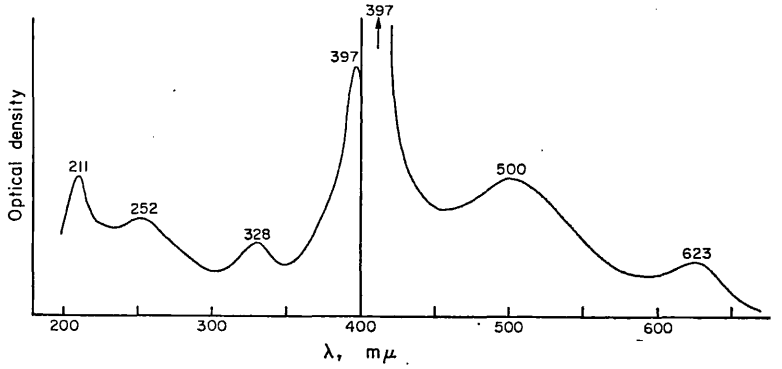

FIG. 1. Absorption spectrum of acid methanol extract of *Eylais extendens* after carotenoids have been removed. The absorption below 400 mμ is drawn to approximately one-tenth of the vertical scale of the spectrum above 400 mμ.

petroleum. This can be identified with the third fraction from the adults. About 35 per cent of the total carotenoids in the larvae belonged to this fraction.

3. Eluted with acetone and acetic acid; mostly hypophasic. Single absorption maximum at 469 mμ in light petroleum. Identical with the fifth fraction from the adults; formed about 40 per cent of the carotenoids in the larvae.

The residue of adults after they had been extracted with acetone still retained a pink colour which would not dissolve on re-extracting with more acetone. The pink colour was also resistant to solution in methanol, but dissolved readily in acid methanol (2% HCl). This gave a pale pink solution with several clearly defined absorption maxima (Fig. 1). There are probably several different substances in this solution, and it has not yet been possible to purify the pink pigment. The colour is destroyed if a small amount of potassium borohydride is added to the solution. It is hoped that when further material becomes available this pink pigment will be subjected to further analysis.

DISCUSSION

There are two points of interest in the results. The first concerns the occurrence of a range of carotenoids in the adults. *Eylais* feeds to a considerable extent on Cladocera, such as *Daphnia*, which often contains large amounts of carotenoids derived and modified from those in the minute algae which form its food (Green,

1957). It is remarkable that the xanthophyll resembling lutein should still persist as far as the primary predator stage of the food chain, when there is no trace of any of the other plant pigments such as the chlorophylls.

The second point of interest is the presence of a keto-carotenoid in both adults and larvae. This indicates that the metabolic route used in the formation of astaxanthin is similar to that in other animals. Keto-carotenoids have been found as possible intermediates in the formation of astaxanthin in echinoderms (de Nicola, 1954), Crustacea (Gilchrist & Green, 1960; Lenel, 1961), polychaetes (Dales, 1962) and among the Eugleninae in the Protozoa (Krinsky & Goldsmith, 1960; Green, 1963). Some similar fractions have been found in flamingoes (Fox, 1962) which also make astaxanthin.

The results given above are similar in some respects to those obtained by paper chromatography of the pigments of spider mites (Metcalf & Newell, 1962). Astaxanthin and β-carotene were found in *Panonychus citri* (McGregor), but the small amount of material did not permit the detection of any keto-carotenoid which may have been present. The spider mites are herbivorous, and the extracts from them yielded easily detectable amounts of chlorophylls and their derivatives. The extracts from *Eylais* did not show any trace of chlorophyll, which is what one would expect from a predator which has been allowed time to digest its food before pigments were extracted.

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Two species of *Peritrichous Ciliata* epibiotic on parasitic Crustacea from Lake Albert, Uganda

By J. GREEN
*Bedford College, University of London**

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The two peritrichs described below were found in September and October 1962 while collecting parasitic Crustacea in Lake Albert. They were studied alive within a short time of the removal of their crustacean hosts from the fish which they parasitized. Both species belong to the family Epistylidae, as defined by Kahl (1935).

Rhabdostyla elongata sp.nov. (Fig. 1A–C)

The body is cylindrical in shape, reaching a maximum length of 520μ, and a maximum width of 80μ. The pellicle is smooth, without striations. The disk is flat and projects very slightly beyond the peristome, even when the cilia are functioning and the animal is apparently in a fully open feeding condition. The peristomal bulge is slight. The contractile vacuole lies at the inner end of the cytostome, and when fully expanded reaches a diameter of 20μ. The cytoplasm is finely granular and colourless. The macronucleus has the shape of a curved sausage and lies approximately in the middle of the body. Under low magnification of living specimens the stalk appears to be homogeneous. When formalin-preserved specimens are cleared in lactic acid and examined under high magnification, the stalk is seen to have fine longitudinal striations which become suddenly more marked about 3μ from the cell body (Fig. 1C).

A remarkable feature of this species is its ability to bend and twist in a worm-like manner. It is readily distinguished from other members of its genus by the combination of large size, cylindrical shape, slight peristomal bulge and slight protrusion of the disc.

Host. *Lernaea barnimiana* (Hartmann), parasitic on the Nile Perch, *Lates niloticus* (L.) from Lake Albert. The peritrich was found in large numbers forming a girdle around the body of the host at the point where the copepod emerged through the skin of the fish. A girdle of peritrichs in this position has been noted by several workers (Cunnington, 1914; Capart, 1944; Fryer, 1956).

Designation of type specimen. It is not possible at present to preserve peritrichs well enough to retain their specific characters, so that the designation of type specimens is not practicable. Vavra (1962) has suggested that photographic documentation should be obligatory in describing new species. This suggestion is

* Present address: Department of Zoology, Westfield College, Kidderpore Avenue, Hampstead, London, N.W. 3.

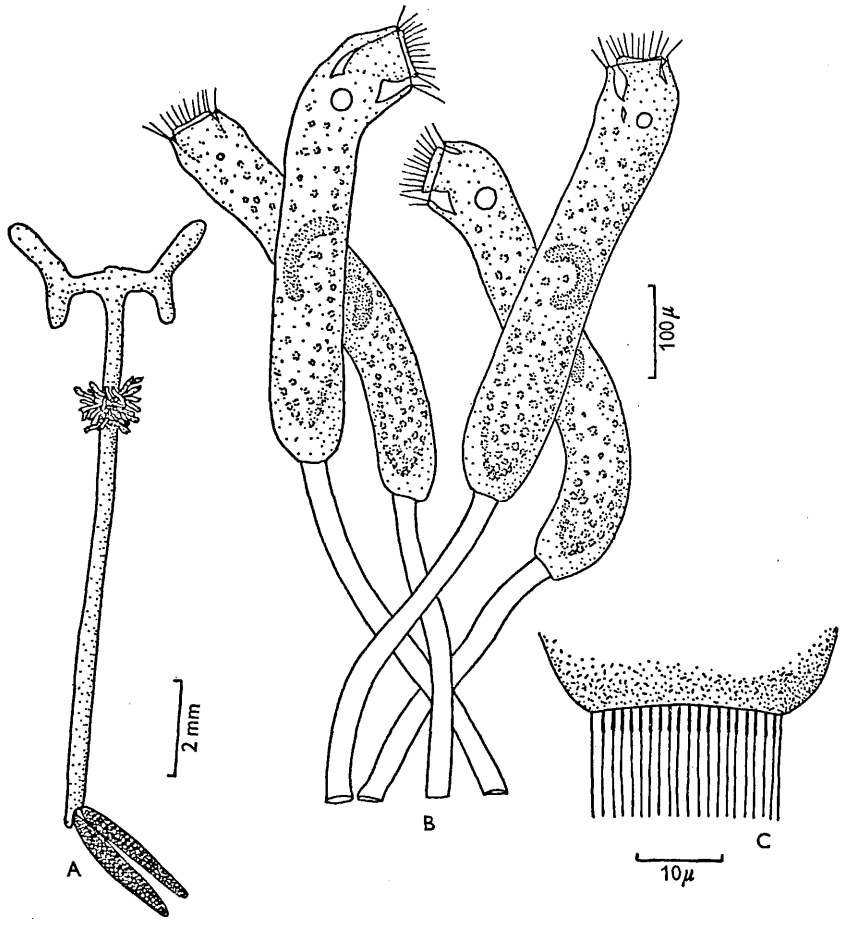

Fig. 1. A, Adult female of *Lernaea barnimiana* with a girdle of *Rhabdostyla elongata*; B *Rhabdostyla elongata*, zooids in open feeding condition; C, junction of stalk and cell body.

worthy of serious consideration, but it does present difficulties when working in the field in under-developed countries.

Epistylis articulata Fromentel (Fig. 2A–C)

The general shape of the body is shown in Fig. 2B. The length varies from 100 to 200μ and the width from 43 to 81μ. The peristomal bulge is poorly developed. The disk is almost flat and projects only slightly through the peristome. The contractile vacuole lies within the peristome and reaches a maximum diameter of 20μ. The cytoplasm contains many globules and is colourless. The macronucleus is broad and band shaped, lying somewhat nearer to the disk than to the stalk. The pellicle is very finely cross striated. The stalk at the base of each individual has a diameter of 18μ, while at the base of the colony the diameter reaches 25μ. Distinct longitudinal striations are visible on the basal part of the stalk and there are fine cross bars at intervals (Fig. 2A). The colony stalk branches in equal dichotomy. Most of the colonies have six or eight individuals.

There are some differences between the description given above and the diagnosis of this species given by Kahl (1935). The maximum length given by Kahl is only 75μ, and the body is described as being more conical, with the disk protruding more than in the present specimens. These differences do not seem to be large

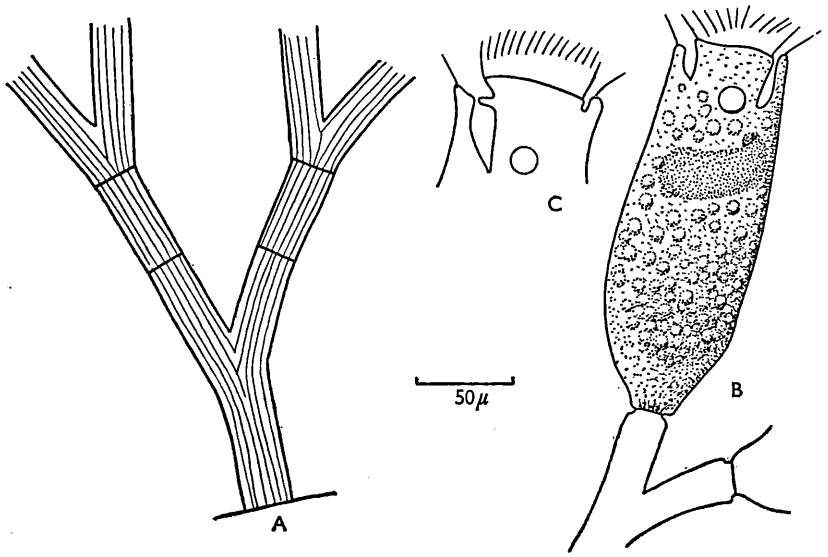

Fig. 2. *Epistylis articulata.* A, Stalk at base of colony; B, zooid in open feeding condition; C, apex of zooid showing the cytostome.

enough to warrant the creation of a new species, particularly in view of the recent work of Vavra (1963) which shows that the shape and size of a peritrich can vary greatly under the influence of environmental factors.

Table 1. *Occurrence of* Epistylis articulata *in Lake Albert*

Fish	Branchiuran parasite	Presence of *Epistylis*
Lates niloticus (L.)	*Dolops*	+
	Argulus	+
Clarias lazera Cuv. & Val.	*Dolops*	+
Auchenoglanis occidentalis (Cuv. & Val.)	*Dolops*	+
	Argulus	−
Bagrus docmac (Forsk.)	*Dolops*	+
	Argulus	−
Labeo horie Heckel	*Argulus*	+
Distichodus niloticus (L.)	*Argulus*	−

HOSTS. Branchiurans of the genera *Argulus* and *Dolops*, which parasitize a wide range of fish in Lake Albert. The species of *Argulus* in Lake Albert are at present being revised by Dr G. Fryer.

There is some correlation between the length of the stalk of the peritrich and the fish host of the branchiuran. The colonies found on *Dolops ranarum* Stuhlmann

taken from the Nile Perch, *Lates niloticus*, which is a well-scaled fish, had much shorter stalks than colonies found on *Dolops* taken from the catfish *Clarias lazera*, which is a very slimy fish. The longer stalks would carry the peristome and disk clear of the thick layer of mucus which covers *Dolops* when it occurs on *Clarias*.

Dolops was generally more heavily infested than *Argulus*, and on several occasions it was found that *Dolops* was infested while *Argulus* from the same fish was not (Table 1). This was so on specimens of *Auchenoglanis occidentalis* and *Bagrus docmac* which were parasitized by both *Argulus* and *Dolops*. The difference in degree of infestation of these two genera might be related to their different habitat preferences. *Dolops* is often found under the operculum of a fish, while *Argulus* is more frequently found on the external surface of a fish.

DISCUSSION

The occurrence of peritrichs on freshwater parasitic copepods has been noted by several authors, notably Cunnington (1914), Monod (1932), Capart (1944), Fryer (1956) and Dollfus (1960), but there have been few attempts to identify the species of peritrich concerned. Recently, Yin, Ling, Hsü, Chen, Kuang & Chu (1963) have identified some of the species found on parasitic copepods in China. They found species of *Epistylis*, *Carchesium* and *Glossatella*. The present study adds another genus, *Rhabdostyla*, to those now known to occur on parasitic crustaceans.

The location of the peritrichs on a parasitic copepod is not always as restricted as shown in Fig. 1A. The more posterior regions of the body are sometimes heavily infested. Through the kindness of Dr G. Fryer, I have been able to examine two species of *Opistholernaea* from the River Niger. These copepods were infested with at least two species of *Epistylis*, and on *Opistholernaea longa*, a parasite of the Nile Perch, *Lates niloticus*, a large species of *Epistylis* infested the anterior part of the copepod, while a smaller species with a different colony form was found more posteriorly.

It is not possible at present to fit these peritrichs into any scheme of specificity, such as that put forward by Nenninger (1948). Even with the comparatively well-worked European peritrichs several of the species in Nenninger's scheme have been moved from one group to another (Matthes, 1950). *Epistylis articulata* is probably not very specific in its choice of host; it has been recorded by Stiller (1941) on a species of *Cyclops* from Lake Balaton in Hungary.

SUMMARY

Rhabdostyla elongata sp.nov. is described and figured. This peritrich was found as an epibiont on *Lernaea barnimiana*, a copepod parasitic on the Nile Perch, *Lates niloticus* in Lake Albert.

Epistylis articulata Fromentel is recorded as an epibiont of *Dolops* and *Argulus*, which parasitize a wide range of fish in Lake Albert.

My thanks are due to Mr M. J. Holden who identified the fish as they were removed from the nets, and to Dr G. Fryer, who identified *Lernaea barnimiana*

and has helped with the literature. I am also grateful for a grant from the Leverhulme Trust which made possible the collection of this material.

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ZOOPLANKTON OF LAKES MUTANDA, BUNYONYI AND MULEHE

BY

J. GREEN

*Bedford College, University of London**

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(With 3 figures in the text)

Quantitative samples of zooplankton from three lakes in the Kigezi District of Uganda have been studied. The systematics of the zooplankton are considered, and some of the identifications given by Worthington & Ricardo (1936) in a previous study of one of these lakes are revised.

Lake Mulehe is the shallowest of the three lakes and contains the largest standing crop of zooplankton. This is in agreement with chemical data which indicate that the supply of nutrient salts in Lake Mulehe is higher than in the other two lakes.

In October 1962 the zooplankton of Lake Mutanda was characterized by the relative abundance of three species of *Daphnia* which were not found in the samples from the other lakes, although two of these species were present in Lake Bunyonyi in 1931. Rotifers were sparse in Lake Mutanda, but were dominated by *Tetramastix opoliensis*. Lake Bunyonyi was richer in rotifers, but here the dominant species was *Keratella tropica*, while in Lake Mulehe the dominant rotifer was *Synchaeta pectinata*.

The zooplankton of Lake Mutanda in October 1962 was similar in composition to that of Lake Bunyonyi in 1931, but in 1962 the zooplankton of Lake Bunyonyi was more like that of Lake Mulehe. The possible causes of this change are discussed.

CONTENTS

INTRODUCTION

This study was undertaken with the cooperation of the Uganda Fisheries Department, Entebbe, and was intended to provide a quantitative comparison of the standing crops of zooplankton in three lakes in the mountainous district of Kigezi which occupies the southwestern corner of Uganda. The aim of this comparison was to gain some insight into the availability of zooplankton as food for fish in these lakes, but the samples have also yielded results of systematic and zoogeographic interest. The only previous quantitative study of the zooplankton of any of these lakes was that of Worthington & Ricardo (1936) on Lake Bunyonyi.

*Present address : Westfield College, Hampstead, London, N.W.3.

Lakes Mutanda, Bunyonyi and Mulehe lie a little over 1° S of the Equator. The three lakes are connected by swamps and rivers (Fig. 1). The Ruhuma

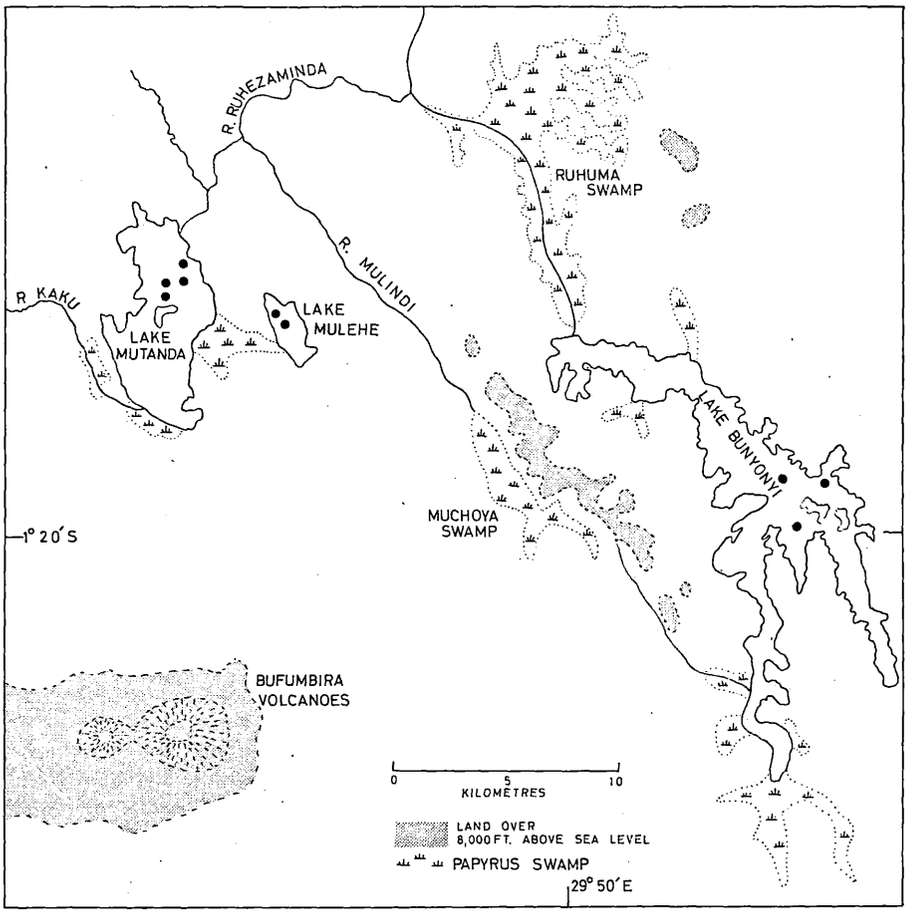

Fig. 1—Map of Lakes Mutanda, Bunyonyi and Mulehe. Many small areas of swamp have been omitted. Dots indicate the positions of sampling stations.

River runs from Lake Bunyonyi into the Ruhuma Swamp. This swamp is connected to Lake Mutanda by the Ruhezaminda River. A few miles before it enters Lake Mutanda the Ruhezaminda is joined by the Mulindi which drains the Muchoya Swamp. Lake Mulehe lies to the East of Lake Mutanda, and its surface is about 47 feet higher ; water drains from Lake Mulehe through a swamp into Lake Mutanda.

The altitudes of these lakes vary between 5,877 feet (Lake Mutanda) and 6,474 feet (Lake Bunyonyi) above sea level. Lake Bunyonyi is the largest in area (56 sq km) and Lake Mulehe is the smallest (5 sq km) ; the greatest depths of the lakes are 56, 40 and 6 metres, respectively.

These lakes were formed when volcanic larva flows blocked the valleys in which they lie. This seems to have happened fairly recently in a geological sense, because there are no signs that the water levels were ever much higher

than they are at present. This means that the outlets have not been in existence long enough to be cut down to any significant extent. This relatively recent origin is probably the reason for the lack of indigenous fish. Several fish, notably species of *Tilapia* and *Clarias* have been introduced by man, both by design and accident. Lake Mulehe is generally regarded by the fisheries authorities as the most productive of the three.

The chemistry of these lakes has been studied by Visser (1962), and a summary of this and other work on this aspect is given by Talling & Talling (1965).

MATERIAL AND METHODS

The samples were taken with standard zooplankton and phytoplankton nets as supplied by the Windermere Laboratory of the Freshwater Biological Association. Vertical hauls from the bottom to the surface were made after the depth had been measured with a weight on a cable running over a Bergen Nautic metre wheel. The winch was operated from a small fisheries launch on Lake Bunyonyi, and from dugout canoes on the other lakes. A violent storm caused the sampling of Lake Mulehe to be abandoned earlier than was intended.

Qualitative samples were also taken with a small hand net from the shores of Lakes Mutanda and Bunyonyi. The non-planktonic Crustacea found in these samples are recorded in the Appendix (pp. 400–402).

In addition to this material I have examined similar quantitative samples from Lake Bunyonyi, collected in July 1962 by Mr M. J. Holden, and some samples originally collected by E. B. Worthington from Lake Bunyonyi in August 1931, and now housed at the British Museum (Natural History).

All the *Chaoborus* larvae in each sample were counted. If the remaining zooplankters were sparse then all were counted, but if the zooplankton was abundant then subsamples were taken with a broad mouthed graduated pipette. Counting was done in two stages. In the first stage the zooplankters were assigned to major groups such as Cladocera, Cyclopoida, Calanoida and Rotifera. In the second stage the relative abundance of each species within the major groups was estimated by identifying a hundred specimens to specific level. If less than a hundred specimens of a major group were present in the whole sample then all were identified.

In the tables the term Cyclopoida is used instead of giving a precise specific name, even though all the adults were identified as *Thermocyclops schuurmanae* and, in Lake Bunyonyi, *Mesocyclops leuckarti*. This is done because it was not possible to identify all the smaller copepodid stages, and these may have included immature forms of species found as adults at the margins of the lakes (see Appendix).

In compiling the tables the data from the zooplankton nets have been used as estimates of the abundance of Cladocera, Calanoida, *Caridina* and *Chaoborus* larvae, while the counts from the phytoplankton nets have been used for the Cyclopoida, nauplii and Rotifera. In order to compare the standing crops of the three lakes the numbers under one square metre of lake surface have been calculated. The percentage composition of the zooplankton has been calculated from the tables of numbers under one square metre. An exception to this is

Table 6 where the percentages are calculated from the zooplankton nets only, to afford direct comparison with the data of Worthington & Ricardo (1936).

SYSTEMATIC ACCOUNT OF THE ZOOPLANKTON

Class Insecta

Order Diptera

Family Chaoboridae

Chaoborus anomalus Edwards

Larvae of this dipteran were found in all three lakes, but most abundantly in Lake Mutanda.

The larvae of *C. anomalus* and *C. ceratopogones* have been described in detail by Verbecke (1957), and using the characters given by him all the larvae could be assigned without doubt to *C. anomalus*. These records of this species from the Kigezi lakes are of particular interest. Verbecke (1958) has summarized the known distribution of the two species named above, and from his geographical data one might reasonably expect to find either of the two species in Kigezi. Now Verbecke regards *C. anomalus* as characteristic of the great lakes and gives *C. ceratopogones* as the species found in the smaller lakes in craters of the Virunga volcanoes and in the lakes of Ruanda. Thus one might expect to find *C. ceratopogones* in the Kigezi lakes. Instead we find *C. anomalus*. Although Lakes Mutanda and Bunyonyi are relatively small lakes their depths are similar to that of Lake Albert, where *C. anomalus* is also found. Lake Mulehe is much shallower than the other two lakes, but since if forms part of the same system it is not surprising that it contains the same chaoborid larva, even though it forms an exception to the general rule formulated by Verbecke (1958).

Class Crustacea

Subclass Branchiopoda

Order Cladocera

Family Daphniidae

Daphnia curvirostris Eylmann

Although described in 1887 by Eylmann this species was not clearly recognized in Europe until it was redescribed by Johnson (1952) in Britain, and later by Hrbáček (1959) in Czechoslovakia. The specimens from Lake Mutanda are quite typical with reduced antennules and few spinules on the dorsal margin of the carapace.

This species was not present in the samples which I collected from Lake Bunyonyi, or in the samples collected by Mr Holden, but it was present in the samples collected by Worthington in 1931. These specimens were identified by Lowndes (1936) as *Daphnia pulex*, and were recorded as such by Worthington & Ricardo (1936). This is an understandable error in view of the close similarity of the two species and the fact that they were not at that time clearly separated by taxonomists in Europe.

This species has been recorded in Africa previously from Lake Kivu (Harding, 1957 a) ; the statement by Harding (1957 b) that this species has been recorded from Lake Tanganyika is an error (Harding, personal communication).

Several specimens of this and the other two species of *Daphnia* in Lake Mutanda were found to be infected with *Spirobacillus cienkowskii* Metchnikoff, which is characterized by its brilliant red colour due to carotenoid pigments (Green, 1959).

Daphnia longispina O. F. Müller

This species was more abundant in the shallower regions of Lake Mutanda than in the deeper areas (Table 1). It was not found in the other two lakes.

The general form of the head and carapace resembled *D. curvirostris*, but the claw of the post abdomen lacked a coarse comb and was dark in colour, which is a feature typical of European specimens.

Daphnia laevis Birge

The principal diagnostic features of this species are the small size of the second abdominal process in the mature female, and the elongated head, which usually has a convex ventral margin. A detailed description is given by Brooks (1957).

This species was abundant in the samples from Lake Mutanda, but was not found in the samples from the other two lakes. Worthington & Ricardo (1936), in their study of the vertical distribution of the zooplankton in Lake Bunyonyi record '*Hyalodaphnia barbata*'. I have examined some of their samples and cannot find any specimens of *Daphnia barbata*, although specimens of *D. laevis* are present. The slight resemblance between the two species seems to have mislead Lowndes (1936) when he identified the specimens collected by Worthington.

Daphnia laevis is probably widespread in East and South Africa. Wagler (1936) illustrates forms from South Africa which can be referred to this species, and Harding (1961) figures some specimens which may be *D. laevis*. I have seen specimens of this species from Lake MacIlwaine in Southern Rhodesia, collected by Mr J. L. Munro.

This species was originally described from North America by Birge (1878), and the recent study by Brooks (1957) has shown that it is widely distributed in the Southern States.

Ceriodaphnia reticulata (Jurine)

The specimens found in Lake Mutanda and Lake Mulehe had prominent serrated processes on each fornix and could all be assigned to the var. *serrata* (Sars). This species was not found in the samples from Lake Bunyonyi, but it was present in the samples collected by Worthington in 1931.

Ceriodaphnia cornuta Sars

This species was not found in the samples from Lake Mutanda, but it was present in the other two lakes. In Lake Bunyonyi it formed about one per cent

of the total zooplankton. In Lake Mulehe it formed only 0·02 per cent of the zooplankton, while *C. reticulata* was eighty times as abundant, forming 1·6 per cent of the zooplankton.

Moina dubia De Guerne & Richard

This species was not found in the samples from Lake Mutanda; in Lake Bunyonyi it formed a little over one per cent of the total zooplankton, and in Lake Mulehe it was much more abundant forming about 10 per cent of the total zooplankton.

Worthington & Ricardo (1936) record *Moina brachiata* (Jurine) as forming 0·5 per cent of the coarse net zooplankton in Lake Bunyonyi. They give no details of the criteria used in their identification, and it seems as if it rests on the table of occurrence given by Lowndes (1936), where a cross against the name *M. brachiata* is given under Lake Bunyonyi. Lowndes does not give any further details of the specimens. All the specimens from Lake Bunyonyi and Lake Mulehe that I have examined have agreed with Gauthier's (1954) detailed description of *M. dubia*.

Subclass Copepoda

Order Calanoida

Family Diaptomidae

Metadiaptomus aethiopicus Daday

This species was redescribed in detail by Lowndes (1936) using specimens from Lake Bunyonyi.

In Lake Mutanda this species was most abundant, forming between 25 and 44 per cent of the total zooplankton. A few specimens were found in the samples from Lake Mulehe, but none were found in the Bunyonyi samples. When Worthington sampled Lake Bunyonyi in 1931 this species was abundant.

Order Cyclopoida

Family Cyclopidae

Mesocyclops (*Thermocyclops*) *schuurmanae* Kiefer

This species is very closely allied to *T. hyalinus* (Rehburg), but may be distinguished by the form of the receptaculum. Specimens from all three lakes were remarkably constant in the form of their receptacula, which agreed with the figures given by Kiefer (1929). Worthington & Ricardo (1936) record this species in Lake Bunyonyi as *T. hyalinus*. Some authors might not regard *T. schuurmanae* as a distinct species, but I have used the name because of the constancy of the form of the receptaculum in the specimens from these lakes. The lateral arms of the receptaculum are longer and more curved than those of *T. hyalinus*.

This was the dominant cyclopoid in all three lakes, but in Lake Bunyonyi it was accompanied by the following species.

Mesocyclops leuckarti (Claus)

This cosmopolitan species was found in the samples from Lake Bunyonyi, but not from the other two lakes.

Subclass Malacostraca
Order Decapoda
Family Atyidae
Caridina nilotica (Roux)

This small shrimp was found in all three lakes. It was most abundant in Lake Mutanda, less so in Lake Mulehe, and in Lake Bunyonyi it was not found in the plankton samples, but a single specimen was taken from the shore with a hand net. Worthington & Ricardo (1936) found that this species formed about 0·4 per cent of the zooplankton caught in Lake Bunyonyi with the coarse net in 1931. This is very close to the figure for Lake Mutanda in 1962 (Table 6).

The specimens caught in the deep water of Lake Mutanda were all immature. Those taken from the shore with a hand net included many mature females with eggs. The form of the rostrum in these mature specimens was

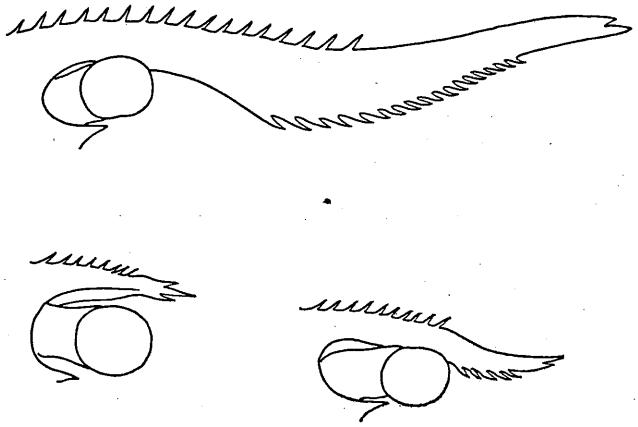

Fig. 2—Variation in the rostrum of *Caridina nilotica* from Lake Mutanda. All the specimens illustrated were mature females carrying eggs.

very variable. The range is illustrated in Fig. 2. In some the tip of the rostrum reached only a short distance beyond the eye ; in others it was very much longer. This range of variation is similar to that described by Gordon (1933) in specimens collected from Lake Bunyonyi by Worthington in 1931. This degree of variation in the development of the rostrum is much greater than has been recorded elsewhere in Africa.

Class Rotifera
Order Monogononta
Family Brachionidae
Anuraeopsis fissa (Gosse)

A few specimens were found in the samples from Lake Bunyonyi. They agreed with Evens's (1947) description of *A. congolensis*, but Gillard (1948) does not regard this as sufficiently distinct to be regarded as a separate species. The length of the lorica of two measured specimens was 83 μ.

Brachionus calyciflorus Pallas

This species formed about 8 per cent of the total rotifers in Lake Mulehe, but was not found in the samples from the other two lakes.

Brachionus caudatus Barrois & Daday

This species was abundant in Lake Bunyonyi, where it formed about 30 per cent of the total rotifers. Most of the specimens were very small, with a lorica 100–110 μ long. These specimens lacked posterior spines on the lorica, but a small percentage of larger forms were found bearing well developed posterior spines. The relationship between the length of the lorica and the length of the posterior spines in this species has been studied in detail in a population from West Africa (Green, 1960).

In Lake Mulehe this species was also abundant, but only formed 5 or 6 per cent of the total rotifers in the samples. No specimens were found in the samples from Lake Mutanda.

Brachionus falcatus Zacharias

In Lake Bunyonyi this species was sparse, forming up to 2 per cent of the total rotifers. It was somewhat more abundant in Lake Mulehe, where it formed 3 per cent of the rotifers in one of the samples and 18 per cent in the other. It was not found in the samples from Lake Mutanda.

Brachionus quadridentatus Hermann

A few specimens with poorly developed anterior spines and no posterior spines were found in the samples from Lake Bunyonyi.

Keratella tropica (Apstein)

This was the dominant rotifer in Lake Bunyonyi, forming up to 68 per cent of all the rotifers. About 9 per cent of the rotifers in the samples from Lake Mutanda were of this species, but as the total numbers of rotifers were low it was much less numerous than in the samples from Lake Mulehe where it formed under one per cent of the total rotifers.

Family Lecanidae

Lecane bulla (Gosse)

A few specimens were found in the samples from Lake Bunyonyi, but the species was not sufficiently numerous to enter in Table 7.

Family Trichocercidae

Trichocerca chattoni (de Beauchamp)

Specimens of this rarely recorded species were found in the samples from Lake Mutanda. It has previously been recorded from France, Bali, South Africa and the Congo (Evens, 1949). This was originally described as a variety of *T. cylindrica*, but more recent workers regard it as a distinct species (Voigt,

1957). The length of the body in the specimens from Lake Mutanda was 300 μ ; the anterior spine reached a length of 54 μ, and the longest of the toes reached a length of 116 μ.

Trichocerca pusilla (Jennings)

This species formed a little over one per cent of the rotifers in the samples from Lake Bunyonyi. It was not found in the other two lakes. The specimens from Lake Bunyonyi are slightly larger than usual, and range up to 170 μ in length. The form of the body is however quite typical, with no teeth or spines on the anterior border of the lorica, and no crests on the dorsal surface. The longer (left) toe has a characteristic gentle curvature and reaches a length of 72 μ, while the shorter toe is very small and difficult to see.

Family Synchaetidae

Synchaeta pectinata Ehrenberg

This was the dominant rotifer in Lake Mulehe, but it was not found in the other lakes. This species is often abundant in the early spring in Europe, but Ruttner (1929/30) found a form of the species in the Lunzer Untersee which was abundant in the summer. The foot was smaller than usual in the specimens from Lake Mulehe, but a form with a reduced foot has also been recorded from Lake Lukula in the Congo (Beauchamp, 1939).

Polyarthra dolichoptera (Idelson)

This species formed about 4 per cent of the total rotifers in the samples from Lake Mulehe, but was not found in the other two lakes. The length of the body was about 104 μ and the lateral spines (or fins) were 108 μ long and 12–14 μ wide. The lateral spines are somewhat shorter than the dimensions usually given for European specimens, though it should be noted that Bartos (1950) has described a form of *P. dolichoptera* with spines that are much shorter than usual, and Pejler (1957) has given details of several populations which show characters intermediate between *P. dolichoptera* and *P. vulgaris* Carlin. This species has previously been recorded in Africa from the Congo (Gillard, 1957).

Family Testudinellidae

Pedalia mira (Hudson)

About 27 per cent of the rotifers in the samples from Lake Mutanda belonged to this species. In Lake Bunyonyi the highest percentage in any sample was only 2. No specimens were seen in the samples from Lake Mulehe. The specimens from Lake Mutanda and Lake Bunyonyi all carried the characteristic caudal appendages and had six teeth on the uncus of the mastax.

Filinia longiseta (Ehrenberg)

This species was found in small numbers in the samples from Lake Mutanda. I am following Pejler (1957) in regarding *F. limnetica* (Zacharias) as a synonym of *F. longiseta*.

Tetramastix opoliensis Zacharias

This was the dominant rotifer in Lake Mutanda, forming about 41 per cent of all the rotifers in the samples, but the total number of rotifers in this lake was so low that the numbers under one square metre were much lower than in Lake Bunyonyi where it formed only 6 per cent of the rotifers. No specimens were found in the samples from Lake Mulehe.

QUANTITATIVE DATA

There are two different methods of expressing the quantitative data derived from vertical hauls. The first is in terms of numbers per unit surface area of the lake. This is a measure relevant to production ecology because solar radiation, which is the energy source of biological production, is measured in terms of energy per unit surface area. The numbers of zooplankters per unit surface area enable one to compare the standing crops of different lakes irrespective of their depths. The disadvantage of this method is that it gives no idea of the volume of water occupied by each zooplankter, and so does not indicate how much searching a fish may have to do before it encounters food. Feeding would be easier for a fish in a shallow lake compared with a deep lake if both had the same standing crop zooplankton per unit surface area. Complications and even partial exceptions to the last general statement might be brought about by the diurnal vertical migrations of zooplankters, but in general terms the rule will be valid.

The second method is to record the numbers per unit volume. Data expressed in this way are then in a form which can be compared with chemical data (usually expressed as milligrams per litre) and with other types of plankton samples which take known volumes from particular depths without necessarily sampling the whole depth of the lake. Many zooplankton studies express their results in terms of numbers per cubic metre.

Care must be taken when comparing different lakes in terms of numbers of zooplankters per unit volume. If two lakes receive a similar amount of solar radiation and have similar supplies of nutrient salts then the shallower lake must produce higher concentrations of zooplankton per unit volume than the deeper lake. This may be in spite of the fact that the numbers of zooplankters per unit surface area may be larger in the deeper lake.

A similar effect is seen within one lake, such as Lake Mutanda (Tables 1 and 4). Here the numbers per unit surface area are greater in the deeper water, but the numbers per unit volume are greater in the shallower water.

In this study the numbers per unit surface area are regarded as the basic form of information. Data from all the samples are given in Tables 1–3. The numbers per cubic metre have also been calculated. They are given in full for Lake Mutanda, because the stations on this lake differed so much in depth. For the other two lakes, where the stations within one lake did not differ so much, a mean figure has been taken. These data are displayed in Table 4.

Lake Mutanda

This is the deepest of the three lakes. Samples were taken from four stations where the depths were 55·5, 22·5, 12, and 11 metres; vertical hauls

of 55, 22, 12 and 10·5 metres were taken. It will be noted that the depths of the hauls were half a metre less than the actual depth, except in the case of the third station, where the mouths of the nets touched the bottom of the lake and there was some contamination with bottom dwelling animals. The nets did not seem to be clogged when they reached the surface and the counts of zooplankters are similar to those from the fourth station. The results are given in Table 1 and Table 4. These tables show that the density of all zooplankters per unit volume increases towards the shallow water, but if the numbers of zooplankters under one square metre are considered then there is variation between different groups and species. *Metadiaptomus aethiopicus* has a fairly constant number of individuals under a unit surface area, while the cyclopoids have fewer individuals under one square metre in shallow water than in deep water. In contrast to this *Daphnia longispina* shows an increase in number per unit surface area in shallow water compared with the deeper water ; the other two species of *Daphnia* show a distribution more like that of the cyclopoids.

Lake Bunyonyi

It was very difficult to study the distribution of the zooplankton in this lake by means of vertical hauls from close to the bottom because the bottom sloped so steeply that any slight movement of the launch between the measurement of depth using the weight and the lowering of the nets to take the sample meant that one was sampling a different depth of water from that originally measured. The three sets of clean samples taken in October 1962 were the result of many more attempts which were ruined by the mouths of the nets coming into contact with the bottom. The results are shown in Table 2. The plankton is dominated by cyclopoids and their nauplii, and the Rotifera are much more abundant than in Lake Mutanda, but not so abundant as in Lake Mulehe. There has been a considerable change in the composition of the zooplankton since it was examined by Worthington & Ricardo (1936), this is discussed on p. 397.

Lake Mulehe

Only two sets of samples were taken from this lake, both from a depth of six metres. The results are shown in Table 3. Striking features are : the great abundance of rotifers ; the absence of the genus *Daphnia* ; abundance of *Moina dubia* ; low numbers of *Chaoborus anomalus* larvae ; and high numbers of cyclopoids. In terms of total numbers per unit surface area and in numbers per unit volume this is by far the richest of the three lakes (Fig. 3).

DISCUSSION

Some chemical data concerning these lakes are summarized in Table 5. These data indicate that Lake Mulehe is richer in nutrient salts than the other two lakes. Further, because of the shallowness and lack of any lasting stratification these nutrients are available throughout the whole depth. The large standing crop of zooplankton is a reflection of the mineral nutrient supply.

The production of fish, as estimated by catch per unit effort is higher in Lake Mulehe than in Lake Mutanda. There are few precise data on this point,

Table 1—Numbers of zooplankters in Lake Mutanda, 29th October 1962

Station	1		2		3		4	
Depth of haul (m)	55		22		12		10·5	
	No. under 1 sq m	%	No. under 1 sq m	%	No. under 1 sq m	%	No. under 1 sq m	%
Daphnia curvirostris	22,000	10·2	11,425	7·5	6,335	5·1	13,235	11·1
Daphnia longispina	1,048	0·5	2,856	1·9	2,715	2·2	3,309	2·8
Daphnia laevis	10,606	4·9	6,292	4·1	3,111	2·5	5,939	5·0
Ceriodaphnia reticulata	5,374	2·5	2,828	1·9	2,121	1·7	3,676	3·1
Metadiaptomus aethiopicus	54,439	25·2	41,289	27·1	40,299	32·2	52,601	44·0
Cyclopoida	52,177	24·1	31,037	20·4	28,280	22·6	13,744	11·5
Nauplii	60,802	28·1	47,651	31·3	35,632	28·4	21,606	18·1
Caridina nilotica	594	0·3	85	0·06	283	0·2	57	0·05
Chaoborus anomalus lv.	7,686	3·6	7,636	5·0	5,373	4·3	4,383	3·7
Hydracarina	141	0·07	0	—	0	—	0	—
Rotifera	1,414	0·7	990	0·7	1,131	0·9	1,018	0·9
Total	216,281		152,089		125,280		119,568	

Table 2—Numbers of zooplankters in Lake Bunyonyi

Station	1		2		3		4	
Date	9 July 62		30 Oct. 62		30 Oct. 62		30 Oct. 62	
Depth of haul (m)	40		38		38		24	
	No. under 1 sq m	%	No. under 1 sq m	%	No. under 1 sq m	%	No. under 1 sq m	%
Moina dubia	127	0·04	2,969	1·9	2,757	1·2	5,373	1·7
Ceriodaphnia cornuta	990	0·3	1,852	1·2	1,654	0·7	3,846	1·2
Cyclopoida	110,151	31·4	61,650	39·6	104,353	45·4	128,108	41·5
Nauplii	187,638	53·5	67,448	43·3	93,607	40·7	138,289	44·6
Chaoborus anomalus lv.	1,315	0·38	2,376	1·5	2,291	1·0	113	0·04
Rotifera	50,056	14·3	19,372	12·4	25,028	10·9	33,795	10·9
Rhabdocoela	141	0·04	141	0·09	283	0·12	283	0·09
Arcella	0	—	0	—	0	—	141	0·05
Total	350,418		155,808		229,973		309,948	

Table 3—Numbers of zooplankters in Lake Mulehe, 29th October 1962

Station	1		2	
Depth of haul (m)	6		6	
	No. under 1 sq m	%	No. under 1 sq m	%
Moina dubia	84,133	11·1	51,611	10·1
Ceriodaphnia reticulata	12,302	1·6	8,233	1·6
Ceriodaphnia cornuta	141	0·02	110	0·02
Cyclopoida	174,205	23·1	116,938	22·9
Nauplii	195,980	25·9	176,043	34·5
Caridina nilotica	283	0·04	424	0·08
Ostracoda	0	—	424	0·08
Chaoborus anomalus lv.	85	0·01	156	0·03
Rotifera	288,456	38·2	155,823	30·6
Total	755,585		509,662	

Table 4—Numbers of zooplankters per cubic metre in October 1962

	Lake Mutanda				Lake Bunyonyi	Lake Mulehe
Depth of haul (m)	55	22	12	10·5	(24 & 38)	6
Daphnia curvirostris	400	519	528	1,260	0	0
Daphnia longispina	19	130	226	315	0	0
Daphnia laevis	193	286	259	566	0	0
Moina dubia	0	0	0	0	125	11,312
Ceriodaphnia reticulata	98	129	177	350	0	1,711
Ceriodaphnia cornuta	0	0	0	0	84	21
Metadiaptomus aethiopicus	990	1,877	3,358	5,010	0	0
Cyclopoida	949	1,411	2,357	1,309	3,235	24,262
Nauplii	1,105	2,166	2,969	2,058	3,333	31,002
Caridina nilotica	11	4	24	5	0	59
Ostracoda	0	0	0	0	0	36
Chaoborus anomalus lv.	140	347	448	417	43	20
Hydracarina	3	0	0	0	0	0
Rotifera	26	45	94	97	859	37,023
Rhabdocoela	0	0	0	0	7	0
Arcella	0	0	0	0	2	0
Total	3,934	6,914	10,440	11,387	7,688	105,446

Table 5—Some chemical characteristics of the Kigezi Lakes (from Talling & Talling 1965)

	Mutanda	Bunyonyi	Mulehe
Conductivity	200–235	233–275	252–275
Phosphate (μg/l)	25–93	10–93	220–228
Nitrate (μg/l)	16–27	15–17	22
Sulphate (mg/l)	6–12	2–6	21–31
Stratification with deoxygenation of hypolimnion	yes	yes	no

but in August 1962 the Fisheries Department of Uganda recorded nightly landings of *Tilapia* from the two lakes. Lake Mutanda was worked by an average of 29 canoes each carrying three nets, and Lake Mulehe was worked by 30 canoes, again each was carrying three nets. The average weight of fish landed per night from Lake Mulehe was 613 pounds, while from Lake Mutanda 336 pounds were landed per night. This represents a substantially larger yield from the smaller lake for a similar expenditure of effort.

The zooplankton of Lake Bunyonyi in 1962 was very different from that of Lake Mutanda, but the samples taken from Lake Bunyonyi in 1931 by E. B. Worthington were remarkably similar to those taken from Lake Mutanda in 1962. Table 6 gives a detailed comparison. The composition of the zooplankton in Lake Bunyonyi in 1962 was in some respects intermediate between that of Lake Mutanda and Lake Mulehe. The numbers of *Chaoborus* larvae, cyclopoids and rotifers in Lake Bunyonyi lay between the numbers found in the other lakes (Table 4). But Lake Bunyonyi also had its peculiarities : *Ceriodaphnia cornuta* was more abundant than in the other two lakes ; *C. reticulata* was absent ; and *Mesocyclops leuckarti* accompanied *Thermocyclops schuurmanae* in the samples.

Each of the three lakes had its characteristic rotifer plankton (Table 7). In Lake Mutanda rotifers were very sparse, but of those present the majority belonged to two species : *Tetramastix opoliensis* and *Pedalia mira*. Lake Mutanda was further characterized by the presence of *Filinia longiseta* and *Trichocerca chattoni*, and negatively by the absence of the genus *Brachionus*. Lake Mulehe was characterized by an abundance of rotifers, the majority of which were *Synchaeta pectinata*, but *Brachionus* species were also abundant and *Polyarthra dolichoptera* was present. In Lake Bunyonyi the dominant rotifer was *Keratella tropica*. *Brachionus* species were also present, but not so abundantly as in Lake Mulehe. Viewing Table 7 as a whole there is a strong impression that at the time of sampling the rotifer plankton of Lake Bunyonyi was intermediate between that of Lake Mutanda and Lake Mulehe.

It is possible to regard the three lakes as forming a series which may indicate stages in lake evolution, with Lake Mutanda as an early stage, and Lake Mulehe as a later stage. The change in the zooplankton of Lake Bunyonyi during the last thirty years from a composition like that of Lake Mutanda to a composition more like that of Lake Mulehe lends some support to this idea.

The cause of the change in the zooplankton of Lake Bunyonyi between 1931 and 1962 is not known. It is not a simple seasonal difference. The water temperature of Lake Bunyonyi is probably near 20° C throughout the year. Rainfall is seasonal and may cause some change in the plankton, but it should be noted that the samples taken in July 1962 yielded results similar to those taken at the end of October 1962. Worthington's samples were taken in August 1931. The change may have been gradual and progressive. A long

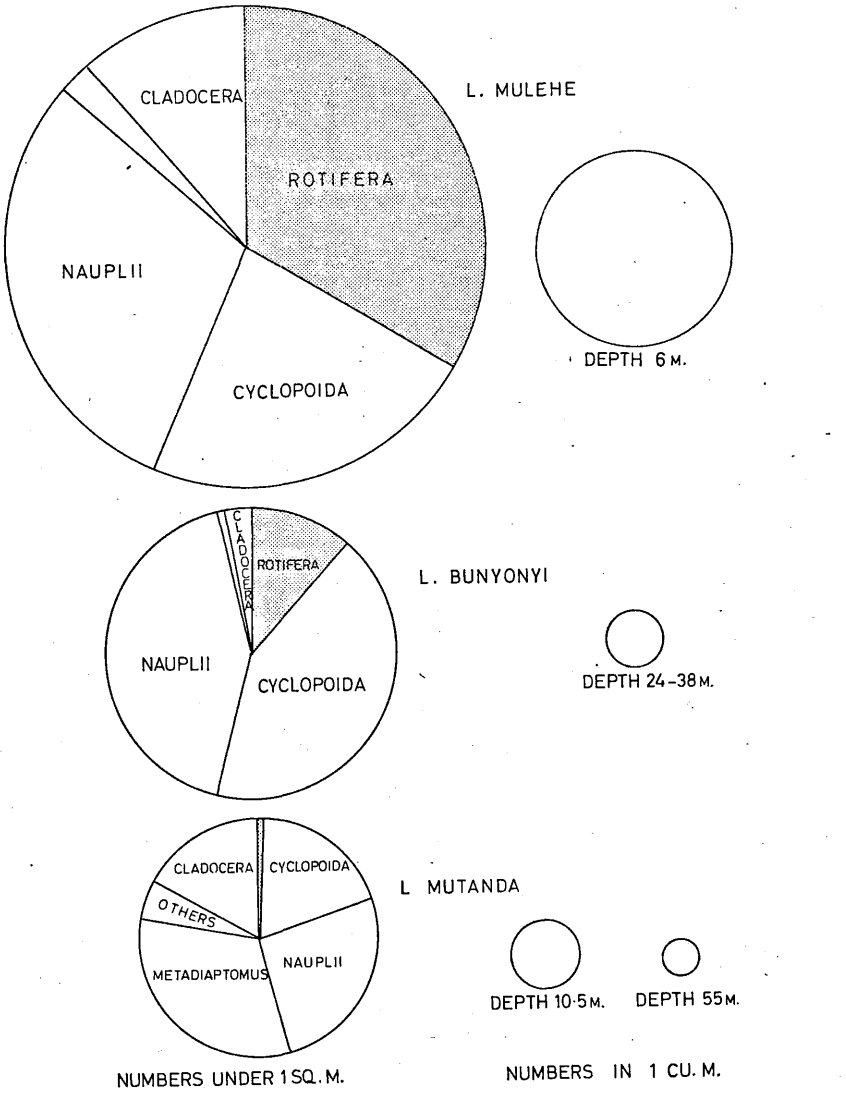

Fig. 3—Comparison of the standing crops of zooplankton in Lakes Mutanda, Bunyonyi and Mulehe. The area of each circle is proportional to the total number of zooplankters. The circles representing the numbers under one sq m of surface have been divided according to the percentage of the total formed by each group.

Table 6—Percentage composition of the zooplankton of Lake Mutanda in October 1962 compared with that of Lake Bunyonyi in August 1931, July 1962 and October 1962. (Percentages calculated from zooplankton net samples only.)

	Mutanda	Bunyonyi		
	Oct. 1962	Aug. 1931*	July 1962	Oct. 1962
Daphnia curvirostris	20·8	23·0	—	—
Daphnia longispina	1·0	—	—	—
Daphnia laevis	10·0	0·8	—	—
Ceriodaphnia reticulata	5·1	4·3	—	—
Ceriodaphnia cornuta	—	—	37·8	24·7
Moina dubia	—	0·5	4·9	39·6
Cyclopoida	3·5	8·6	7·0	4·0
Chaoborus larvae	7·0	2·4	50·3	31·7
Caridina nilotica	0·6	0·4	—	—
Metadiaptomus aethiopicus	51·5	59·6	—	—

* Data from Worthington & Ricardo (1936) with identifications corrected according to the systematic section of this paper (pp. 386–392).

Table 7—Numbers and percentage occurrence of Rotifera in Lakes Mutanda, Bunyonyi and Mulehe

	Mutanda		Bunyonyi		Mulehe	
	No. under 1 sq m	% total Rotifera	No. under 1 sq m	% total Rotifera	No. under 1 sq m	% total Rotifera
Anuraeopsis fissa	0	—	129	0·6	0	—
Brachionus calyciflorus	0	—	0	—	16,329	7·5
Brachionus caudatus	0	—	7,873	29·0	11,886	5·5
Brachionus falcatus	0	—	325	1·3	18,351	10·5
Brachionus quadridentatus	0	—	83	0·3	0	—
Keratella tropica	102	9·0	15,407	61·0	779	0·5
Trichocerca chattoni	159	14·0	0	—	0	—
Trichocerca pusilla	0	—	451	1·3	0	—
Synchaeta pectinata	0	—	0	—	165,245	72·0
Polyarthra dolichoptera	0	—	0	—	9,549	4·0
Pedalia mira	308	27·0	166	0·6	0	—
Filinia longiseta	68	6·0	0	—	0	—
Tetramastix opoliensis	467	41·0	1,631	6·0	0	—
Total	1,138		26,065		222,139	

*Mean numbers for each lake based on samples taken in October 1962.

term change of this type has recently been recorded in the Lago Maggiore (Tonolli, 1962). A change in the condition of Lake Bunyonyi may well have been brought about by an increase in the native population around its shores. The Kiga are a vigorous people, with a highly developed system of terrace agriculture, and the population has been increasing steadily over the last fifty years. This may have lead to increased polution and eutrophication of the lake.

Another possibility is that the change is part of a shorter term cycle, which may take several years and is not necessarily related to any annual

climatic cycle. There is no information available concerning such cycles in tropical lakes, but they should be borne in mind as a possibility. The detection of such cycles would require continuous sampling of a lake at regular intervals for several years.

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APPENDIX

Non-planktonic Crustacea collected from the shores of Lakes Mutanda and Bunyonyi

Subclass Branchiopoda

Order Cladocera

Family Macrothricidae

Ilyocryptus agilis Kurz

A single specimen was taken from Lake Mutanda. The specimen was a mature female, and the proximal curve of the dorsal edge of the post abdomen was short and had eight spines. This seems to be the first record of this species in Africa.

Family Chydoridae

Chydorus globosus Baird

A single specimen was taken from Lake Mutanda.

Chydorus eurynotus Sars

Several specimens were taken from Lake Bunyonyi. The carapaces of these specimens lacked any sculpturing of the type depicted by Gauthier (1939), but it has already been noted that the sculpturing is not a constant feature of this species (Harding, 1957 a ; Green, 1962).

Alona pulchella King

Typical specimens of this cosmotropical species were taken from Lake Mutanda.

Alona diaphana King

A few specimens were taken from Lake Bunyonyi.

Subclass Ostracoda

Family Cypridae

Cypridopsis cf. *trigonellus* Sars

Several specimens which may be provisionally assigned to this species were taken from Lake Mutanda. The shape of the shell agrees with Sars (1924) figure, but since he does not describe the appendages the identification cannot be regarded as certain.

Subclass Copepoda

Order Cyclopoida

Family Cyclopidae

Macrocyclops albidus (Jurine) *oligolasius* Kiefer

Typical specimens with relatively short caudal rami were taken from Lake Mutanda.

Eucyclops serrulatus (Fischer)

This cosmopolitan species was found in Lake Mutanda.

Eucyclops euacanthus (Sars)

Several specimens were taken from Lake Bunyonyi. Kiefer (1929) regarded *E. cognatus* Kiefer as a separate species, but Rylov (1948) indicates that the two are synonyms.

Ectocyclops phaleratus rubescens Brady

I am following the nomenclature of Fryer (1955) in giving this name to the specimens taken from Lake Mutanda.

Tropocyclops prasinus (Fischer) sens. lat.

Specimens similar to *T. onabamiroi* Lindberg (1950) were taken from Lake Mutanda. The status of the various forms of *Tropocyclops* recorded from

Africa is still uncertain, so that I prefer to leave the identification in the wider sense until a critical revision of the genus has been made. Ideally such a revision should include breeding experiments.

Subclass Malacostraca

Order Decapoda

Family Potamonidae

Potamonautes emini (Hilgendorf)

This small crab was common around the shores of Lake Mutanda. Two specimens were collected from the canoe used to sample the plankton. Chase (1942) described specimens from Lake Mutanda as a separate species *P. mutandensis*, but I am following Bott (1955) in regarding this as a synonym of *P. emini*.

CHEMICAL EMBRYOLOGY OF THE CRUSTACEA

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By J. GREEN

Westfield College, University of London

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CONTENTS

I. INTRODUCTION

In 1931 Needham surveyed the chemical embryology of the whole animal kingdom and provided a detailed account which has served as a foundation for more recent work. The purpose of the present article is to examine advances made since 1931 in the study of chemical changes which accompany embryonic development in crustaceans. The scope has been limited to changes occurring between laying of eggs and emergence of free-living young. Oogenesis has not been included; a recent review of this subject has been given by Raven (1961).

The Crustacea offer certain advantages which facilitate the study of their embryos. The eggs of many Crustacea are carried by the females for the whole of the incubation period, and it is only necessary to collect a large number of adults to obtain numerous eggs in all stages of development. All the eggs carried by a given female are normally uniform embryologically, being laid in a short period of time. Some exceptions have been found, for instance in three species of the isopod genus *Limnoria* where 24% of the females were found to be carrying more than one developmental stage (Eltringham & Hockley, 1961). These exceptions are not sufficiently numerous to invalidate the general rule that the majority of the Crustacea produce separate broods in which all the embryos are at a similar stage of development.

II. STAGES OF EMBRYONIC DEVELOPMENT

The study of chemical change during development is facilitated if the duration of development is known and if a number of easily recognizable stages have been defined and timed. The embryology of Crustacea has received a considerable amount of

attention, but the embryologist correctly regards embryogenesis as a continuous process and is not necessarily concerned with the definition of developmental stages.

The following account gives selected references to descriptions of embryonic development, and for some groups the practical subdivisions used by physiologists and ecologists. This is not intended to be a complete bibliography of the embryology of the Crustacea, but references have been selected which are either recent or which give figures that may be used to determine embryonic stages.

(1) *Subclass Branchiopoda*

Most Notostraca, Conchostraca and Anostraca produce eggs with opaque shells, so that it is not possible to observe stages of development in the intact egg. Eggs produced by these groups are often resistant to desiccation, and a convenient method of subdividing the developmental period is to use time after the dried eggs have been immersed in water. This is the usual procedure with the eggs of *Artemia salina* (L.). This anostracan can produce two types of egg. One is retained in the brood pouch of the female until the nauplius larva emerges after a period of several days. The other type becomes enclosed in a tough shell and is released from the brood pouch. When the latter type of egg is dried it can be kept for months or even years and still give rise to a viable nauplius when immersed in water. The embryo within the dried shell contains about 4000 nuclei, and this number does not change significantly until the nauplius hatches (Nakanishi, Iwasaki, Okigaki & Kata, 1962). The term cyst is frequently applied to this commercially available stage in the development of *Artemia*. In sea water at 20° C. the eggs swell and after 50 hr. the shell splits and the embryo emerges enclosed in a thin membrane. The nauplius swims out of this membrane about 90 hr. after the egg has been put into sea water. At this stage the nauplius still has some yolk and does not feed until it has been free-swimming for about 30 hr. (Urbani, 1962).

A considerable amount of histochemical and ultrastructural information is available concerning oogenesis and the early cleavage stages of *Artemia* (Fautrez, 1951; Fautrez & Fautrez-Firlefyn, 1951, 1961, 1963, 1964; Fautrez-Firlefyn, 1957; Fautrez-Firlefyn & Fautrez, 1953, 1962, 1963; Fautrez-Firlefyn, Fautrez & Labis, 1963; Anteunis & Fautrez-Firlefyn, 1961; Anteunis, Fautrez-Firlefyn & Fautrez, 1961; Anteunis, Fautrez-Firlefyn, Fautrez & Lagasse, 1964; Roels, Fautrez-Firlefyn & Fautrez, 1964), but these studies do not extend to a quantitative comparison with the later stages.

Cyclestheria hislopi (Baird) is remarkable among the Conchostraca for producing eggs which develop into miniatures of the adult instead of hatching as nauplii. These eggs have transparent membranes and stages of development can be seen in the intact egg. A series of coloured figures showing stages of embryonic development has been given by Sars (1887).

The Cladocera produce two types of egg. One develops in the brood pouch of the female and becomes free-swimming after a few days. The development of this type of egg has been subdivided by Fox (1948) into eight stages. I have adapted and timed these stages using *Daphnia magna*, as shown in Table 1 (Green, 1956*a*).

Subdivisions of embryonic development in the Cladocera using fewer stages have

been given by Jančařík (1947) and Hoshi (1950*a*). A detailed account of the embryology of *Daphnia* is given by Baldass (1942).

The eight stages given in Table 1 have been used successfully with a wide range of Cladocera, such as *Sida*, *Simocephalus*, *Eurycercus* and *Chydorus*. These stages probably need some modification if an attempt is made to apply them to those Cladocera which secrete a nutritive fluid into the brood pouch (*Moina* among the Daphniidae, and all members of the Polyphemidae).

Table 1. *Stages in embryonic development of* Daphnia magna

stage	description	duration in hr. at 22° C.
1	eggs opaque, or translucent with transparent edges	3
2	with markedly granular transparent edges; fat cells forming	17
3	headless embryos; egg membrane cast off	10
4	embryos with heads but no eyes; antennae short	10
5	embryos with very small pink eyes; antennae longer	4
6	embryos with two distinct red eyes	2–3
7	embryos with two large black eyes	8
8	embryos with one large black eye	15

(2) *Subclass Cephalocarida*

Only the post-embryonic development of this group has so far been described (Sanders, 1963).

(3) *Subclass Copepoda*

Studies on the embryonic development of cyclopoid copepods have been made by Fuchs (1914) and Jacobs (1925). Descriptions of development of embryos of parasitic copopods are given by Wilson (1911) and Heegaard (1947). The development of *Calanus* has been summarized by Marshall & Orr (1955). Lang (1948) has summarized such knowledge as is available concerning the Harpacticoida.

(4) *Subclass Ostracoda*

A detailed account of the embryology of *Cyprideis* has been given by Weygoldt (1960). The total duration of development is about 2 weeks.

(5) *Subclass Mystacocarida*

As yet only the post-embryonic development of this group has been described (Delamare-Deboutteville, 1954).

(6) *Subclass Cirripedia*

Groom (1894) described a series of embryonic stages of barnacles, and these were used by Barnes & Barnes (1959) in their study of oxygen consumption by embryos of *Balanus balanoides* (L.) and *Pollicipes polymerus* Sowerby.

Crisp (1954) studied *Balanus balanus* (L.) (= *B. porcatus* Da Costa) and described a series of stages which have certain advantages over those described by Groom. The main advantage is that Crisp subdivides Groom's last stage which occupied half the total duration of development. Crisp's stages are given in Table 2.

Detailed data concerning the rates of development of seven species of barnacles over a range of temperatures are given by Patel & Crisp (1960).

Table 2. *Stages in embryonic development of* Balanus balanus (*after Crisp*, 1954)

stage	description	duration in days at 6–7·5° C.
1	unsegmented egg in oval or pyriform case	1·1
2	two simple blastomeres	0·8
3	upper blastomere divided, yolk not completely covered, 3 to 32 blastomeres	1·3
4	yolk all undivided, completely or nearly completely covered by blastoderm cells	1·0
5	yolk cell divided in two by an oblique furrow and completely covered by blastoderm cells	2·0
6	yolk cell divided into 3 to 5 cells enclosed in blastoderm	1·9
7	six or more yolk cells, posterior thickening of mesoblast present	3·0
8	embryo divided by two or more constrictions between rudimentary swellings giving rise to the appendages	3·2
9	appendages clearly visible as short swellings, setae absent or not evident	5·2
10	appendages with distinct setae, no eye visible	2·3
11	median eye red or poorly pigmented, mass of yolk cells present	2·2
12	eye darkly pigmented, black or reddish brown; endoderm forms a clearly defined gut. Not hatching within a few minutes of placing in sea water	7·3
13	as 12 but more strongly pigmented. Hatching within a few minutes of placing in sea water	12·8
	total time to hatching	44·0

(7) *Subclass Malacostraca*

This group is so large and diverse that it is necessary to give references to each of the divisions.

Division Phyllocarida. The embryology of *Nebalia bipes* (Fabr.) has been described in detail by Manton (1934), who gives a series of drawings of living embryos which could serve as useful reference points in chemical studies.

Division Hoplocarida. Accounts of the embryology of *Squilla* have been given by Nair (1941) and Shiino (1942).

Division Syncarida. Hickman (1937) describes the embryology of *Anaspides*.

Division Peracarida. Manton (1928) gives a thorough account of the embryology of the mysidacean *Hemimysis lamornae* (Couch) and gives references to important earlier papers on malacostracan embryology. Some more recent accounts include those of Nair (1956) on *Irona* (Isopoda), Weygoldt (1958) on *Gammarus* (Amphipoda), Barker (1962) on *Thermosbaena* (Thermosbaenacea), and Scholl (1963) on *Heterotanais* (Tanaidacea). Kinne (1954) figures five stages in the development of the isopod *Sphaeroma hookeri* Leach, and Naylor (1955) describes four easily recognizable stages of another isopod, *Idotea*.

Division Eucarida. A detailed account of the embryology of a decapod (*Palaemonetes varians* Leach) is given by Weygoldt (1961). A simple division of the embryonic

stages of *Carcinus maenas* (L.) was given by Needham (1933) as follows: stage 1, cleavage; stage 1*a*, a transparent crescent just visible; stage 2, the yolk occupying four-fifths of the diameter of the egg; stage 3, intermediate between stages 2 and 4; stage 4, yolk occupying one-third of the diameter of the egg; stages 5 and 6, intermediate between stages 4 and 7; stage 7, yolk nearly all gone; stage 8, yolk completely gone, zoea ready to hatch.

Boolootian, Giese, Farmanfarmaian & Tucker (1959) give ten stages which they applied to both brachyuran and anomuran crabs (Table 3).

Table 3. *Stages in development of decapod crustaceans (after Boolootian* et al. 1959)

stage	description
1	no cleavage visible
2	cleavage has taken place
3	a yolk-free (transparent) part becomes visible. This stage coincides with the appearance of endoderm cells and the beginning of invagination
4	a more distinct division into a yolk-free and a yolk-containing part becomes clearly visible
5	eye pigment of embryo becomes visible
6	pigment bands of embryo become visible
7	embryo becomes strongly pigmented but still contains much yolk
8	the yolk is reduced to two small separate patches
9	zoea larva becomes recognizable
10	swimming larvae appear

III. CHANGES IN DRY WEIGHT DURING EMBRYONIC DEVELOPMENT

Measurements of dry weight provide a simple means of studying changes in constitution during development. This can give, under certain conditions, an indication of the amount of material used by the developing embryo. The method is most likely to be successful with freshwater and terrestrial Crustacea, which have limited possibilities of salt intake. The dry weight of the embryos of such Crustacea should diminish as respiratory substrates are oxidized. The embryos of Crustacea in saline habitats may be able to maintain their dry weight by taking up salts. Dutrieu (1960) found that the dry weight of *Artemia* embryos did not change during the course of development and attributed this to uptake of salts. In contrast the dry weight of the parthenogenetic egg of the freshwater cladoceran *Daphnia magna* decreases by 16–25 % during embryonic development, and a similar decrease is found in *D. curvirostris* Eylmann (Green, 1956*a*).

Saudray (1954) found that the eggs of the lobster *Homarus vulgaris* Milne-Edwards lost 7 % of their dry weight by time the larvae hatched. This lower percentage loss compared to freshwater Cladocera may be due to salt uptake.

A special feature is encountered when eggs are enclosed in a brood pouch and receive nutrients secreted by the mother. The dry weight can then give an estimate of the material secreted by the mother, although in marine Crustacea there is always the complicating factor of salt uptake. Newly hatched young of *Ligia oceanica* (L.) have

a dry weight 36% in excess of the initial dry weight of the egg (Saudray, 1954). If one accepts that salt uptake may compensate for the loss of dry weight caused by utilization of respiratory substrates, as in *Artemia*, then the extra 36% dry weight of the neonate *Ligia* represents material supplied by maternal secretions.

IV. OXYGEN CONSUMPTION

The amount of oxygen consumed by an embryo provides a measure of metabolic rate, and if combined with an estimate of carbon dioxide output will provide a respiratory quotient (R.Q.) which may indicate the substrate used to supply energy to the developing embryo. The respiratory quotient can be modified by factors which influence the release of carbon dioxide, so that caution is always necessary in drawing conclusions from respiratory studies. Nevertheless, the measurement of respiratory quotients can provide useful information which may help to confirm conclusions drawn from other types of data.

Hoshi (1950*a*) measured the oxygen consumed by embryos of *Simocephalus vetulus* (O. F. Müller). In the early stages, when the embryos were 10 or 12 hr. old, the amount of oxygen consumed at 27° C. was 0·019 μl. per embryo per hour, while in the later stages the figure increased to 0·056 μl. per embryo per hour. The latter figure was doubled when the young were released from the brood pouch and became free-swimming, so that the overall increase in oxygen consumption was approximately sixfold. Hoshi also measured carbon dioxide production and calculated the respiratory quotient. This rose from 0·74 when the embryos were 10 hr old to 0·99 when the young were released from the parental brood pouch. These results suggest that fat is utilized to supply energy in the early stages of development and carbohydrate in the later stages.

An important point to note when comparing Hoshi's results with those of other workers is that the earliest stages were 10 hr. old. This is about 20% of the duration of development of *Simocephalus* at the temperature used by Hoshi, so that the earliest stages were not studied. Needham (1933) studied embryos of the shore crab, *Carcinus maenas*. His earliest stages were in the process of cleaving and had an R.Q. close to unity. By the time stage 2 (see p. 584) was reached the R.Q. had fallen to 0·72. The later stages showed a gradual increase in R.Q. to 0·83 at the time of hatching. The durations of the stages used by Needham are not known, but it seems likely that the low R.Q. in stage 2 corresponds to the similar figure found by Hoshi in *Simocephalus* when one-fifth of the duration of development had passed.

The early embryos of *Balanus balanoides* consume less oxygen per unit weight than the later stages, and do not increase this consumption with increasing temperature to the same extent as the later stages (Barnes & Barnes, 1959). The lower respiratory rate of the early stages is attributed to the smaller amount of active protoplasm compared to yolk. The effect of size on oxygen consumption is shown well by a comparison of the eggs of *Balanus balanoides* and *Pollicipes polymerus*. The volume of one egg of *P. polymerus* is about one-twelfth that of an egg of *B. balanoides*, but the rate of oxygen consumption lies between one-third and one-sixth of the oxygen consumed by

an egg of *B. balanoides*. In terms of oxygen consumed per unit wet weight, the amount used by eggs of *P. polymerus* is between two and four times that consumed by eggs of *B. balanoides*.

When the encysted embryos of *Artemia salina* have been hydrated for 4 hr., they consume oxygen at a rate of 200 μl./hr./100 mg. dry weight at 30° C. The free-swimming nauplii consume oxygen at a rate of 1740 μl./hr./100 mg. dry weight (Dutrieu, 1960). This increase of over eightfold is somewhat greater than that found by Hoshi (1950*a*) when comparing newly liberated young with the early embryonic stages of *Simocephalus vetulus*.

The consumption of oxygen by developing embryos of *Artemia* has been studied in more detail by Clegg (1964). When incubated in 0·25 M sodium chloride solution at 30° C., the embryos consume oxygen at a rate of 60 μl./30 mg. of cysts between the third and fourth hours after being wetted. If the concentration of sodium chloride is increased, the consumption of oxygen falls. A similar decrease in oxygen consumption is found when other substances, such as glucose, sucrose and mannitol, are used to increase the external osmotic presure. With external osmotic pressures up to 30 atmospheres, the decrease in oxygen consumption is not caused by any reduction in the number of cysts respiring or by any decrease in the final percentage of emergence, but by a reduction in the rate of embryonic development. At external osmotic pressures above 30 atmospheres the percentage emergence begins to fall, and at 65 atmospheres no emergence take place. This relationship between external osmotic pressure and metabolism is discussed further in the next Section.

V. CARBOHYDRATES

Variation in glycogen content during development of the parthenogenetic egg of *Simocephalus vetulus* has been studied by Hoshi (1951, 1953, 1954). Glycogen is confined to the cytoplasmic parts of the developing embryo and is not found in the yolk. As development proceeds glycogen appears in the muscles, the wall of the alimentary canal and in the wall of the maxillary gland. It is formed rather than utilized in the early stages of development. The normal glycogen content of the gastrula is 0·7 % of the wet weight. Later the content rises to 0·91 % and when the young are released from the maternal brood pouch the glycogen falls to 0·71 % of the wet weight. If the embryos are kept in anaerobic conditions there is no change in the glycogen content, which remains at 0·5–0·6 % of the wet weight.

In anaerobic conditions at 20° C. the embryos of *Simocephalus vetulus* die after 11 hr., when about 28 % of the total glycogen has been utilized. When a small amount of oxygen (0·02 ml./l.) is present in the medium the amount of glycogen used in 11 hr. is increased to 47 % of the total available. At higher oxygen concentrations less glycogen is used and the metabolism of the embryo is more like that of embryos in well aerated water. Hoshi (1951, 1953, 1954) makes it clear that in well-aerated conditions the embryos of *Simocephalus* make glycogen in the early stages and utilize a small amount during the later stages.

Carbohydrate metabolism may be different in other Cladocera, particularly those

with closed brood pouches. Brammertz (1913) found no glycogen in the eggs of *Moina*.

The formation of glycogen in *Simocephalus* has a parallel in the encysted embryos of *Artemia*, where the main carbohydrate reserve is trehalose (Dutrieu, 1960; Clegg, 1964). During the course of development, the trehalose content falls from 15·3% of the dry weight of the egg to 0·6% of the dry weight of the newly hatched nauplius. At the same time the glycogen content shows an increase from 1·5 to 5·0% of the dry weight.

Metabolism of carbohydrates during the development of *Artemia* is influenced strongly by the osmotic pressure of the external medium. The normal desiccated cysts contain 34 μg. glycerol per mg. dry weight. When incubated in 0·25 M sodium chloride the glycerol content increases to 56 μg./mg. dry weight just before emergence. If the external medium is 0·75 M sodium chloride the glycerol content increases to approximately 80 μg./mg. dry weight (Clegg, 1964). In contrast, less glycogen is formed at high than at low external osmotic pressures.

High external osmotic pressure thus appears to stimulate the formation of glycerol at the expense of glycogen. The source of both these substances is trehalose which is the main carbohydrate reserve of the embryo of *Artemia*. The formation of glycerol by *Artemia* is an adaptation to increase the internal osmotic pressure above the external osmotic pressure, so that osmotic rupture of the hard outer shell may be facilitated. The presence of a relatively high concentration of glycerol in the desiccated cysts may also be an adaptation enabling the embryos to resist drying and freezing. The hygroscopic properties of glycerol may aid in the retention of water when the cysts are subjected to desiccation. The role of glycerol in the cold hardiness of insects is now well established (Salt, 1961), but *Artemia* may also gain advantage by utilizing trehalose as the main carbohydrate reserve. Asahina & Tanno (1964) have shown that the frost-resistant prepupal larva of the sawfly *Trichiocampus populi* Okamoto contains a high concentration of trehalose, but no glycerol. The cysts of *Artemia* are exceptionally resistant to cold and heat (Mathias, 1934; Whitaker, 1940; Hinton, 1954), and it is probable that the combined high concentrations of trehalose and glycerol are involved in maintaining this resistance.

Trehalose in the encysted egg of *Artemia* is formed within the egg and not supplied as trehalose by the parent. Glucose is the only sugar in the blood of female *Artemia* (Clegg, 1965). The type of egg which is retained in the brood pouch of the female until the nauplius larva emerges shows a difference in carbohydrate metabolism from the egg which becomes encysted and dormant. The glycogen contents of both types are similar after 10 hr. of development, but in the egg destined to become dormant the glycogen content decreases to the low level found in the encysted embryo. Non-dormant embryos of the same age have a much higher glycogen content, but a hardly detectable trehalose content. This difference between the dormant and non-dormant eggs of *Artemia* is another indication that trehalose plays a significant role in the dormant state (Clegg, 1965).

VI. LIPIDS

Direct observation of the centrifuged egg of *Simocephalus vetulus* reveals that the oil droplet which is so conspicuous near the beginning of development is broken up and decreases in volume as development proceeds. Utilization of lipids seems to be greatest early in development, and this is supported by the respiratory quotient (Hoshi, 1950*a*, *b*).

There is some disagreement concerning changes in the lipid content of developing *Artemia* embryos. Dutrieu (1960) states that the total lipids increase slightly during the transition from egg to nauplius, from 20 to 24% of the dry weight. Urbani (1962) shows the total lipid content decreasing during the course of embryonic development, and from the slope of the line on the graph it would appear that the rate of decrease is more rapid towards the end of development. Bellini & Lavizzari (1958) measured lipase activity in *Artemia* embryos, and found it to be greatest just before the nauplius became free-swimming; the increase was threefold or fourfold between immersion and emergence. This may not necessarily be connected with utilization of lipids, but could indicate a reorganization of lipids. Dutrieu (1960) records a change in the composition of lipids during the development of *Artemia*: the unsaponifiable fraction increases from 4·8 to 12% of the total lipids.

Developing embryos of *Ligia oceanica* and *Homarus vulgaris* utilize considerable amounts of lipid. In *Ligia* 32% of the initial content of the egg disappears by the time the neonate emerges, and in *Homarus* the corresponding figure is 60% (Saudray, 1954). The lower figure for *Ligia* may be caused by the embryos receiving nutrients while in the maternal brood pouch.

VII. AMINO ACIDS AND PROTEINS

The amino acid composition of the proteins in ovarian eggs of the Japanese Spiny Lobster, *Panulirus japonicus* (von Siebold), has been analysed by Suyama (1959). Glutamic acid and aspartic acid each form about 12% of the total. Alanine, arginine, glycine, histidine, isoleucine, leucine, lysine, phenylalanine, proline, serine, threonine, tryosine and valine are all present in percentages ranging from approximately 4 to 7. Much smaller amounts of cystine, hydroxylysine, hydroxyproline, methionine and tryptophan are also present. The possibility of changes in these percentages during development has not yet been studied in *Panulirus*, but some changes in relative proportions of amino acids during development have been found in *Artemia* (Dutrieu, 1960).

The six most abundant free amino acids in *Artemia* eggs are, in order of abundance, aspartic acid, cystein, serine, glycine, alanine and glutamic acid. In the hatched nauplius the order becomes alanine, serine, glycine, proline, tyrosine and glutamic acid. There is an increase in the proportion of proline and a marked decrease in aspartic acid. Of the less abundant amino acids, arginine and threonine were scarcely detectable in eggs but were much more in evidence in nauplii.

There are very small changes in the total nitrogen and protein nitrogen during the course of development of *Artemia*, so that protein is not apparently used to any

significant extent as a respiratory substrate. The changes in relative abundance of amino acids noted above indicate that there must be some reorganization of the proteins during the course of development. Studies on enzymes concerned with amino acids and proteins indicate that there is a peak of activity at the time when the embryo is emerging from the outer shell. Dipeptidase activity increases fourfold in the first hour after the egg has been immersed in water, then rises by half as much again at the time of emergence. Proteinase activity does not increase in the first hour after immersion, but later rises to a peak at the same time as dipeptidase activity (Bellini, 1957). The latter is greater in tetraploid than in diploid nauplii (Coromaldi & Urbani, 1959).

Ammonia is the end product of nitrogen metabolism in both diploid and tetraploid embryos of *Artemia* (Bellini & De Vincentiis, 1960). The tetraploid embryos have a higher rate of ammonia elimination than the diploids, and this may be correlated with the greater dipeptidase activity of the tetraploids.

VIII. ENZYME ACTIVITY DURING EMBRYONIC DEVELOPMENT

Most of the work on enzymes in crustacean embryos has been concerned with the encysted embryos of *Artemia*. The properties of some of the enzymes found in these eggs are summarized in Table 4 and variation in activity during development are shown in Fig. 1.

Table 4. *Enzymes in* Artemia salina *embryos*

enzyme	optimum temperature (°C.)	optimum pH	authority
proteinase	38	4·8	Urbani & de Cesaris-Coromaldi (1953)
dipeptidase (alanine-glycine)	40	7·4	Urbani, Rognone & Russo (1952)
amylase	49–56	6·6	Urbani, Russo & Rognone (1953)
alkaline phosphatase	38	9·8	Urbani & Urbani-Mistruzzi (1953)

As noted in Section VII, there is a peak of proteinase activity as the embryo emerges from the shell. If this feature is found in other Crustacea it may support the idea that carotenoproteins are held in reserve until a specific stage of development has been reached. The increased proteinase activity may coincide with the period in which the link between carotenoid and protein is broken.

There are no comparable data on enzymes in other Crustacea. One might expect considerable variation between embryos of different Crustacea. Within the genus *Marinogammarus* it has been shown that *M. obtusatus* (Dahl) maintains a constant high level of esterase activity throughout embryonic development, while *M. finmarchicus* (Dahl) starts with a low esterase activity which suddenly increases tenfold when the embryo is 8 days old (at 15 °C., the total duration of development being 19 days). This sudden increase is coincident with an acceleration of organogenesis (Doyle, Rappaport & Doyle, 1959).

IX. PIGMENTS

(1) *Haem pigments*

The presence of haemoglobin in the parthenogenetic eggs of *Daphnia* was recorded by Teissier (1932) and confirmed by Fox (1948) who found that the amount diminishes as development proceeds. Haemoglobin passes from the maternal blood into the eggs while still in the ovary during a few hours before the eggs are laid (Dresel, 1948). The amount of haemoglobin passed into each egg depends on the state of nutrition of the

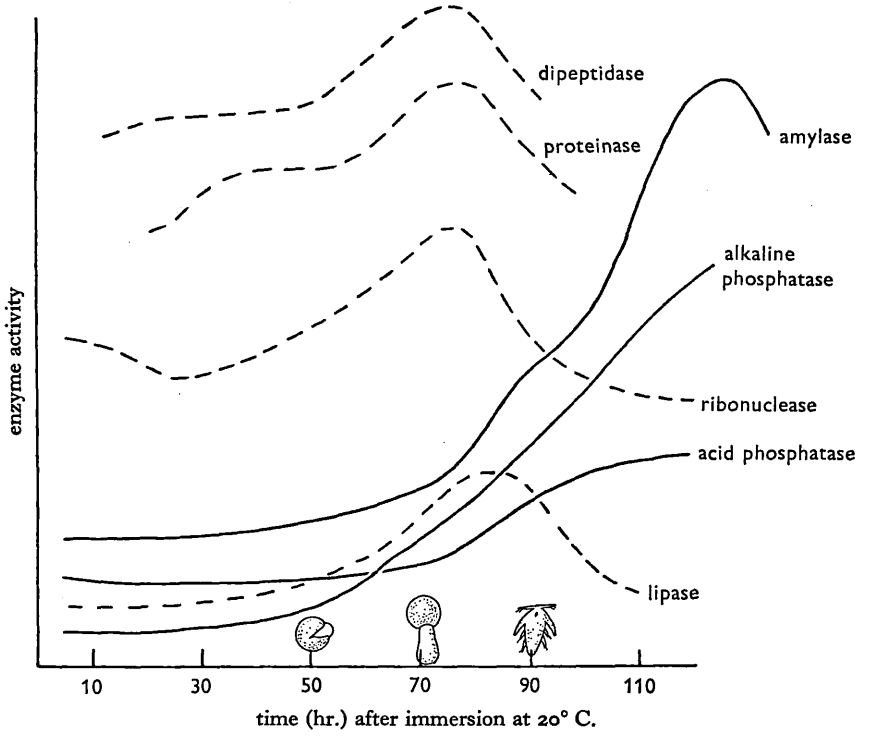

Fig. 1. Enzyme activity in embryos of *Artemia salina*. The small drawings give an indication of the progress of emergence. Each curve gives the proportional increase in activity of a particular enzyme, but all the curves are not drawn to the same vertical scale. (Based on data from Bellini, 1957, 1958; Bellini & Lavizzari, 1958; Urbani & Bellini, 1958; Urbani, 1962.)

mother and the oxygen content of the water in which she lives (Green, 1956*b*). High concentrations of haemoglobin are found in eggs laid by females of *D. magna* producing few eggs in poorly aerated water when food is abundant. Low concentrations are found in eggs laid by a female in well aerated water. When the concentration of haemoglobin in the egg is high, most of the pigment is taken into the fat cells of the embryo as soon as these are formed. The haemoglobin in these cells is then broken down during the course of embryonic development (Green, 1955). When the concentration of haemoglobin in the egg is not so high, the fat cells are not coloured by

haemoglobin, but the concentration still diminishes during the course of embryonic development (Fox, 1948; Phear, 1955), so that the neonatae are not visibly coloured by the pigment.

When embryos of *Simocephalus vetulus* are treated with carbon monoxide to inactivate haemoglobin their rate of oxygen consumption does not differ significantly from that of normal embryos (Hoshi, 1957). Embryos treated with carbon monoxide take a longer time to complete their development, so that haemoglobin serves a function in accelerating development, particularly in poorly aerated water (Table 5). A similar effect had been found earlier in *Daphnia* (Fox, 1948).

Table 5. *Effect of carbon monoxide on development of embryos of* Simocephalus vetulus (*after Hoshi*, 1957)

temperature °C.	oxygen concentration c.c./l.	medium	time in hr. for 50% hatching
25	5·4	without CO	18·5
25	5·4	with CO	22·1
25	1·5	without CO	20·6
25	1·5	with CO	25·0
28	1·9	without CO	19·2
28	2·0	with CO	23·5

It may be that haemoglobin is passed into the eggs of Cladocera merely as a supply of protein, and carbon monoxide may render it unsuitable for use by the embryos. The greater difference in rate of development between treated and untreated embryos in poorly aerated water may be due to the slower rate of decomposition of carboxy-haemoglobin in these conditions. The decrease in haemoglobin during the course of development supports the view that its function is to supply protein for embryogenesis. Haemoglobin may have an advantage in this respect over proteins not attached to a prosthetic group in that it may be stabilized against the action of the less specific proteases and so have the possibility of being retained until a specific stage of development is reached. This suggestion is the same as that made in relation to carotenoproteins in crustacean embryos (p. 593).

The gut of late-stage embryos of *Daphnia* contains a haemochromogen named daphniarubin by Fox (1948). In eggs and early embryos no haemochromogen can be detected, so that the pigment found in the later stages must be formed during the course of embryonic development. The total haem contents of early and late stages are similar, suggesting that the haemochromogen is formed from some other haem compound (Phear, 1955). There is a decrease in the haemoglobin content of the embryo during development (Fox, 1948; Phear, 1955) and this suggests that part at least of the haemochromogen formed in late embryos is derived from haemoglobin present in the egg.

(2) *Bile pigments*

Although the developing embryos of *Daphnia* can destroy haemoglobin during the course of development, no bile pigments have been detected in embryos or adults (Fox, 1955; Smaridge, 1956). This lack of bile pigments indicates that the metabolic pathway of haemoglobin breakdown in *Daphnia* is different from that found in the vertebrates

where bile pigments are a normal end product. It has been suggested that the breakdown of haemoglobin in *Daphnia* involves a coupled oxidation with unsaturated fatty acids such as linoleic acid (Green, 1957*a*). The fat cells, which are the main site of haemoglobin breakdown, usually contain droplets with a complex mixture of lipids (Jäger, 1935), so that the materials for such an oxidation are available in both embryos and adults.

The normal parthenogenetic eggs of *Polyphemus pediculus* (L.) are colourless, but as the embryo develops the eye rudiments are seen to be green in colour. This colour is due to biliverdin (Green, 1961). The pigment seems to be synthesized in the developing eyes and is not found elsewhere in the body. Later in embryonic development the green pigment is obscured by a high concentration of an ommochrome which progressively darkens the compound eye until it appears black.

Bloch-Raphael (1948) indicated that bile pigments were present in eggs and embryos of the parasitic cirripede *Septosaccus cuenoti* (Duboscq), but did not give any precise chemical data.

(3) *Carotenoids*

The eggs of *Artemia* contain a carotenoid which has been identified as an ester of astaxanthin (Gilchrist & Green, 1960). A change in carotenoid was found during the course of development. More pigment was extractable from the nauplii, and the extract from nauplii gave two bands when chromatographed on alumina, in contrast to the single band given by extracts from eggs. Recent work by Krinsky (1964) indicates that the pigment in the nauplii is canthaxanthin, which is closely allied to astaxanthin (see p. 593). We found that all the extracts from both eggs and nauplii gave similar absorption spectra, and both of the bands which separated when the extract from nauplii was chromatographed had the same spectrum (Gilchrist & Green, 1960). If, as seems probable, the pigment in the nauplii is the same as that in the eggs, the original identification as astaxanthin must fall.

Earlier work had indicated that there may be a net synthesis of carotenoid during the development of *Artemia* (Needham & Needham, 1930). Reinvestigation revealed that the difference between quantitative extracts from eggs and nauplii was approximately 8%, and it was considered that this could be caused by differing degrees of adsorption on to embryonic tissues and by slightly different extinction coefficients of the pigments from eggs and nauplii (Gilchrist & Green, 1960), so that the synthesis of carotenoid during embryonic development of *Artmeia* must be considered as unproven.

Eggs of the crab *Carcinus maenas* contain a much wider range of carotenoids than the eggs of *Artemia*. Lenel (1961) found β-carotene, cryptoxanthin, a free xanthophyll, hypophasic astaxanthin and traces of a monohydroxy ketocarotenoid. The rhizocephalan parasite *Sacculina carcini*. Thompson, which commonly occurs on *Carcinus*, accumulates only β-carotene and this is the only carotenoid in its embryos.

The eggs of lobsters (*Homarus* spp.) contain a green carotenoprotein named ovoverdin by Stern & Solomon (1937). When this pigment is treated with agents to denature protein the colour changes to red, owing to liberation of astaxanthin (Kuhn &

Sorensen, 1938*a*, *b*). This change in colour also occurs naturally towards the end of embryonic development. The total carotenoid content of the embryo does not change, but the link between carotenoid and protein is broken (Goodwin, 1951). Similar carotenoproteins, differing in colour, are found in the eggs of the cirripede *Lepas* (Ball, 1944), the copepods *Idya furcata* (Baird) (Lwoff, 1925, 1927) and *Cyclops* (Dupraw, 1958), and in a wide range of Cladocera (Green, 1957*b*). In the Cladocera the carotenoprotein becomes restricted to the embryonic fat cells as soon as these are formed. When the link with the protein is broken, the freed carotenoid passes into fat droplets and intensifies their colour. The function of carotenoproteins in crustacean eggs is not known, but breakdown of the pigment towards the end of embryonic development suggests that if there is a function it is more important in the early stages than later. There is the possibility that linking a protein with a carotenoid may remove the protein from the possibility of attack by certain enzymes. Stabilization of a protein by linkage to a carotenoid is found in an extreme form in the eggs of the tropical snail *Pomacea canaliculata* (d'Orbigny). The carotenoprotein, ovorubin, is not readily coagulated by heat and resists attack by trypsin, but the apo-protein is readily coagulated by temperatures above 70° C. and can be digested by trypsin (Cheesman, 1958). In a less extreme form such a linkage with a carotenoid might enable a protein to be held in reserve until a specific stage of development.

Carotenoproteins may also function by absorbing light and protecting embryos from injury by solar radiation. This function is suggested by the increased deposition of carotenoproteins in the eggs of *Daphnia magna* when exposed to light (Green, 1957*b*),

The colour of the carotenoprotein in the eggs of *Cyclops vernalis* Fischer varies according to the food eaten by the copepod. This variation appears to be due to differences in proportions of carotenoprotein linkages giving purple and red colours. The carotenoid does not vary, but is linked in at least two ways so that a range of colours can be produced by varying the relative proportions of each linkage (Dupraw, 1958).

There is some evidence that β-carotene is the main precursor of the other carotenoids in the Crustacea. For instance, Teissier (1932) fed *Daphnia* with a variety of carotenoids, but only 'carotene'—presumably mostly β-carotene—was effective in producing green carotenoprotein pigmentation of the eggs. If the full transition from β-carotene to astaxanthin is made, then the following carotenoids are possible intermediates:

** β-carotene
** cryptoxanthin—3-hydroxy β-carotene
* echinenone—4-keto β-carotene
* hydroxyechinenone—3-hydroxy-4-keto β-carotene
zeaxanthin—3,3′-dihydroxy β-carotene
euglenanone—3,4-diketo β-carotene
** canthaxanthin—4,4′-diketo β-carotene
* crustaxanthin—4,4′-3,3′-tetrahydroxy β-carotene
(**) astacene—4,4′-3,3′-tetra keto β-carotene
** astaxanthin—3,3′dihydroxy-4,4′-diketo β-carotene

This does not exhaust the list of possible intermediates, but indicates those which are most likely to be involved from the point of view of their structure, or are of known occurrence in Crustacea * and their eggs **.

Some possible steps in the formation of astaxanthin from β-carotene are shown in Fig. 2. These steps are hypothetical as yet, but several organisms are known which contain three or more of the carotenoids shown in this scheme. For instance, *Euglena gracilis* Klebs contains β-carotene, echinenone, euglenanone, cryptoxanthin, hydroxyechinenone, and zeaxanthin in addition to other carotenoids (Krinsky, Gordon & Stern, 1964). A recent study by Bodea, Nicoara, Illyes & Serban (1964) has shown that the calanoid copepod *Arctodiaptomus salinus* Daday contains β-carotene, hydroxyechinenone, crustaxanthin and astaxanthin.

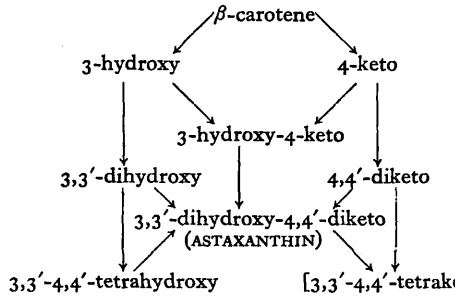

Fig. 2. Possible steps in the formation of astaxanthin. Note that 3,3′-4,4′- tetraketo β-carotene (astacene) is generally regarded as not being of natural occurrence, but as an oxidation product of astaxanthin, for this reason it is placed in square brackets, and the occurrence in eggs is bracketed in the list on p. 593.

X. PHOSPHORUS METABOLISM

Studies on various Crustacea have shown that the total phosphorus content of embryos does not change during the course of development. This is so in *Artemia* and *Emerita* (Needham & Needham, 1930) and in *Calanus* (Marshall & Orr, 1961). In the latter study radioactive phosphorus was fed to females and the radioactivity in eggs laid by these females was compared with that of nauplii hatched from such eggs. No difference was found.

Although the total phosphorus content does not change, there may be some changes in distribution of phosphorus. In *Emerita* the ether-soluble phosphorus declines as development proceeds, but in *Artemia* this fraction rises slightly during development (Needham & Needham, 1930). The energy-rich phosphate in *Artemia* eggs shows negligible changes during the first 12 hr. of incubation (Muramatsu, 1962).

XI. DISCUSSION AND CONCLUSIONS

Recent studies on the resting eggs of *Artemia* have given some insight into the maintenance of viability during dormancy and the chemistry of osmotic hatching. There is no detectable uptake of oxygen by the dried cyst (Muramatsu, 1960), but 2 or 3 hr. after immersion in water uptake of oxygen becomes measurable. Many enzymes are preserved within the cyst and their activities are evident immediately the

cyst is rehydrated (Urbani & De Cesaris-Coromaldi, 1953; Urbani & Urbani-Mistruzzi, 1953). The outer shells of the resting eggs of different branchiopods are very variable in composition, so that it is evident that the resistance to freezing and drying which they exhibit is not dependent on a particular type of covering, but on the organization of the cytoplasm of the egg. The work of Clegg (1964, 1965) indicates that the use of trehalose as the main carbohydrate reserve is related to the dormant state. The presence of glycerol in the dry cysts of *Artemia* may also be related to resistance against freezing and drying, but a further role of glycerol in raising the internal osmotic pressure to facilitate hatching has also been demonstrated (Clegg, 1964).

In Section IX the view was put that haemoglobin in the eggs of Cladocera and the carotenoproteins found in eggs of many other Crustacea are present in the role of proteins rather than pigments. The linkage with an iron porphyrin or with a carotenoid is thought to act as a stabilizing factor which removes the particular protein from the sphere of action of certain proteinases. The protein may then be retained until a specific stage in development is reached. In *Daphnia* the amount of haemoglobin diminishes as embryonic development proceeds, and the link between carotenoid and protein is broken towards the end of embryonic development. These facts are in agreement with the idea that the proteins are retained until a certain stage of development. The finding by Hoshi (1957) that there is no change in oxygen uptake when embryos of *Simocephalus* are treated with carbon monoxide is an indication that haemoglobin is not passed into the eggs in the role of a respiratory pigment. The fact that development is delayed in poorly aerated water when haemoglobin is converted to carboxyhaemoglobin can be explained by the slower rate of decomposition of the latter compound in poorly aerated conditions.

XII. SUMMARY

1. The dry weight of a neonate crustacean is sometimes lower than the initial dry weight of the egg, owing to utilization of respiratory substrates, as in *Daphnia*, where the dry weight decreases by 16–25 %. In *Artemia* any loss is compensated by an uptake of salts so that there is no change in dry weight during the course of embryonic development. The embryos of *Ligia* receive nutrients secreted into the maternal brood pouch, and the dry weight of the neonate is greater than the initial dry weight of the egg.

2. Early embryos of *Simocephalus*, *Artemia* and *Balanus* consume less oxygen per unit dry weight than older embryos. The increase in oxygen consumption from early to late embryos is approximately six to eightfold.

3. Oxygen consumption decreases when the rate of embryonic development of *Artemia* is reduced by increasing osmotic pressures in the external medium.

4. Glycogen is formed rather than utilized in the embryos of *Simocephalus* and *Artemia*. In the latter the main carbohydrate reserve is trehalose, which is transformed to glycogen and glycerol. The relative formation of these two substances is influenced by external osmotic pressures. At high external osmotic pressures more glycerol is formed, and this aids osmotic rupture of the tough outer shell of the egg.

5. Trehalose seems to be important in relation to the dormant state of *Artemia* embryos. The embryos which develop rapidly in the maternal brood pouch contain much less trehalose than those which become encysted and dormant.

6. The proportion of the initial lipid content of a crustacean egg that is utilized during development is very variable. In *Homarus* 60 % disappears by time the neonate emerges, in *Ligia* 32 %, and in *Artemia* the decrease is very slight, or there may even be a slight increase.

7. Haemoglobin in the eggs of Cladocera does not appear to act as a respiratory pigment, but does serve to accelerate development in poorly aerated water. It is suggested that the main function is to act as a supply of stable protein.

8. The haemochromogen found in the gut of late embryos of *Daphnia* appears to be formed in part at least from haemoglobin, which diminishes in concentration as development proceeds.

9. Breakdown of haemoglobin during embryonic development of *Daphnia* is not accompanied by formation of bile pigments.

10. Biliverdin is synthesized in the embryonic eyes of *Polyphemus* but not elsewhere in the embryo.

11. The carotenoproteins in the eggs of many different crustaceans break down towards the end of embryonic development, liberating free carotenoid. It is suggested that these proteins are stabilized and held in reserve by linkage to a carotenoid until a particular stage of embryonic development has been reached.

12. Metabolic pathways in the formation of astaxanthin from β-carotene may involve monoketo and diketo carotenes, or by an alternative route monohydroxy and dihydroxy carotenes.

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XIV. ADDENDUM

Barnes (1965) has studied chemical changes during the development of *Balanus balanoides* (L.) and *B. balanus* da Costa from a locality in Scotland, where the former takes 105 days to reach the stage of hatching and the latter 32 days. The respiratory rate of the faster developing embryos is approximately twice as high as that of the embryos of the slower developing species. Both species utilize over 74% of their initial carbohydrates, over 25% of their initial nitrogen and between 11 and 17% of their initial lipids. Glycogen decreases steadily during the course of development, but glucose shows an increase during the first half of development, followed by a decrease in the second half. The main pigment in the eggs appears to be a chromolipid; there is also evidence of one or more carotenoids.

Dr B. M. Gilchrist (personal comm.) has confirmed that the pigment in the eggs of *Artemia* is canthaxanthin. The same pigment, together with echinenone, has been found in *Daphnia magna* Straus and *D. longispina* O. F. Müller (Thommen & Wackernagel, 1964). My own unpublished work confirms this for *Daphnia magna* and shows that the same pigments are present in *Scapholeberis mucronata* (O. F. Müller) together with astaxanthin and other carotenoids, but it is not yet known how many of these pigments pass into the eggs.

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SEASONAL VARIATION IN EGG PRODUCTION BY CLADOCERA

BY J. GREEN

*Westfield College, Hampstead, London, N.W.*3

The number of eggs produced by a female cladoceran is influenced by a variety of environmental factors. A review of these factors, based largely on laboratory experiments, has been given by Green (1956). The factors which are most likely to be of importance in natural populations are food, temperature, oxygen content of the water, and population density. The relative importance of each factor under natural conditions is unknown. The aim of the present work is to study seasonal variation in egg production in natural populations of Cladocera and to relate this variation to the known effects of environmental factors which have been studied in the laboratory.

The Long Water in Hampton Court Park was chosen as the habitat for the study of seasonal cycles. Observations on latitudinal variation were made in Greenland (Disko Island) and Northern Italy (Pallanza).

METHODS

Samples of Cladocera were collected with a fine-meshed bolting silk hand net. The mesh of this net (180/in., 7·2/mm) was fine enough to retain all stages of the smallest cladoceran. All observations were made on living animals so that artifacts due to fixation and preservation were avoided. Each sample was examined on the day of collection.

The frequency of sampling was somewhat variable, but during the spring and autumn, when conditions were likely to be changing most rapidly, samples were taken at weekly intervals, sometimes more frequently. The number of visits to the Long Water in each year were as follows: 1959, 34; 1960, 44; 1961, 41; 1963, 42.

The lengths of females with eggs were measured with a calibrated eyepiece micrometer from the crown of the head to the posterior border of the carapace. If a spine was present on the carapace, as in *Scapholeberis*, the length of the spine was not included.

Egg numbers were counted after dissection of the eggs from the brood pouch under a low-powered dissecting microscope. The term 'egg number' will be used irrespective of whether eggs or embryos were counted. This usage is possible because the eggs are retained in the brood pouch until the young are liberated. The duration of brooding varies inversely with increasing temperatures. The egg number will thus reflect the environmental conditions a few days before the time at which each sample was taken. The mean egg number was calculated from a sample of ten or twenty females with eggs in their brood pouches, so that it represents the mean number of eggs per mature and fertile female, not the mean egg number carried by the total female population.

Eggs were measured in the early second stage of development (cf. Green 1956, p. 179); they had swollen to the limit within the egg membrane, but had not yet cast this membrane. Freshly laid eggs are not suitable for measuring because they swell considerably during the first few hours in the brood pouch, and the membrane is delicate and liable to burst when the eggs are dissected from the brood pouch. Measurements were made with the eggs covered in water. This supported the eggs and reduced the danger of

compression due to their own weight. Two diameters were measured: the greatest and the least.

When some of the females were producing resting eggs the percentage was estimated by taking a large random sample and counting them on a glass plate under a dissecting microscope.

No attempt was made to obtain numerical estimates of population density. It was clear at the beginning of this study that the Cladocera in the Long Water were distributed patchily, and often formed swarms varying in diameter from a few inches to several feet. The density of population could vary a hundred-fold over a distance of 1 m.

Water temperature was measured at 09.00 hours (GMT or BST according to season) on days when collections were made. A total immersion thermometer was held about 10 cm below the surface of the water and the temperature read after 2 minutes with the thermometer completely immersed.

The chlorophyll content of the water was estimated from 100 ml samples, using the technique of Green (1963). A slight modification was made during 1963 in that the acetone solution was not filtered but was centrifuged to clear before examination in the spectrophotometer. On a few occasions there was incomplete extraction of the pigments. When this occurred the residue was soaked for 2 days in fresh acetone and the absorption of the second extract was added to that of the first. Absorption of light by the acetone solution was measured at 665 and 750 mμ. The latter measurement was used as an estimate of scattering and residual absorption; the estimate of chlorophyll was obtained by deducting this reading from that at 665 mμ.

The oxygen content of the water was not measured, but a close watch was kept on the colour of the blood of the Cladocera. The amount of haemoglobin in the blood of many Cladocera varies inversely with the oxygen content of the water in which they live (Fox 1955). The colour of the blood can be used as a rough guide to the oxygen content of the water. There were some slight variations in the haemoglobin content of the blood of *Simocephalus*, but it never became so red that it indicated a severe reduction in the oxygen content of the water. For this reason it is thought that the seasonal variation in egg production by Cladocera in the Long Water was not influenced to any significant extent by variation in the oxygen content of the water.

HAMPTON COURT LONG WATER: SEASONAL VARIATION IN ENVIRONMENTAL FACTORS

The Long Water is rectangular in shape, about 1000 m long and 30 m wide. The greatest depth is about 2 m. A controllable overflow enables the water level to be maintained fairly constant. This gives great stability to the margins and allows the growth of a narrow but rich band of vegetation along the banks. Patches of water-lilies (*Nymphaea*) in the deeper water provide an ideal habitat for *Sida crystallina*, which attaches itself to the underside of the leaves. At intervals of 2 or 3 years the marginal vegetation is cut down and partly dug out, and this was done in August 1963. This is an important factor in maintaining the character of this body of water and keeping it in a condition favourable to the development of large populations of Cladocera.

The chlorophyll content of the water was used as a quantitative estimate of the phytoplankton available as food for Cladocera. This is not an ideal measure because some species such as *Eurycercus lamellatus* are deposit feeders and periphyton feeders (Fryer 1963), and different species of planktonic algae vary in value as food for Cladocera

(Lefèvre 1942). A quantitative survey of the seasonal variation in phytoplankton and periphyton, coupled with an examination of the gut contents of Cladocera, would be the ideal method, but failing this the measurement of chlorophyll provides at least a rough measure of the available food.

The Long Water is very productive; the highest values for chlorophyll content shown in Fig. 1 are equivalent to 400 mg/m^3. Fig. 1 shows great differences between the two years in which estimates were made. The chlorophyll content was high in the early part of 1961, but fell to a low level in May and remained low throughout the summer, finally rising to very high levels during the autumn. In 1963 there were large fluctuations in the chlorophyll content of the water through the summer, reaching separate high peaks in

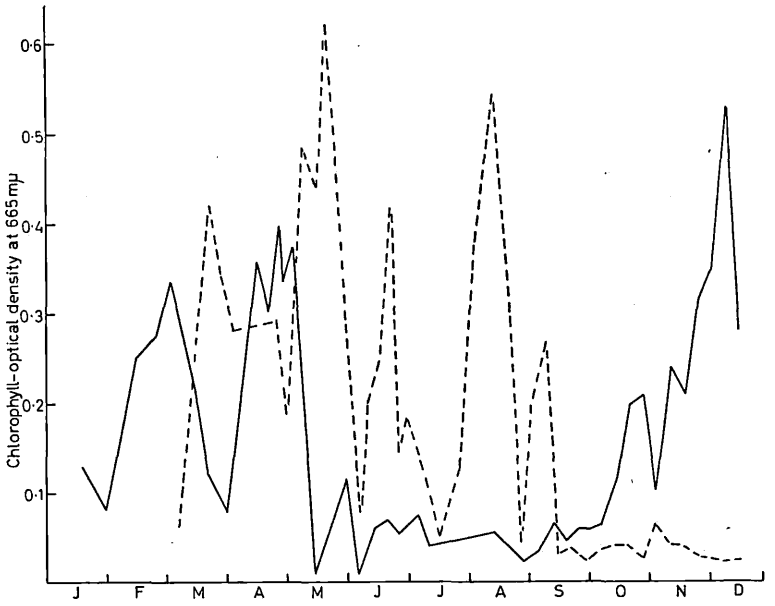

FIG. 1. Variation in the chlorophyll content of the water in Hampton Court Long Water during 1961 (———) and 1963 (– – –).

May, June and August, with a smaller peak in the first week of September. In the middle of September the chlorophyll fell to a low level and remained low for the rest of the year. These two years thus provide an opportunity to analyse egg production by Cladocera under different conditions of available food.

The phytoplankton responsible for the production of chlorophyll was dominated during the years 1959–61 by the genus *Stephanodiscus*, particularly by *S. hantzschii* Grunow. A number of other genera played subsidiary roles. The following were noted as being among the most abundant: *Actinastrum*, *Ankistrodesmus*, *Asterionella* (particularly abundant in February 1959), *Chlamydomonas*, *Gonium*, *Phacus*, *Scenedesmus* and *Synura*. During 1963 *Stephanodiscus* was not so conspicuous in the plankton. After the ice had melted in March the phytoplankton was dominated by *Chlamydomonas*, which was accompanied by *Cryptomonas*, *Euglena*, *Phacus* and *Scenedesmus*. Later in the year the peaks of phytoplankton were dominated by *Dictyosphaerium*, which was accompanied by *Ankistrodesmus*, *Phacotus*, *Oocystis*, *Pediastrum*, *Stephanodiscus* and *Stichococcus*. When the phytoplankton fell to a low level during the autumn there was a

considerable growth of *Coccochloris microscopica* (Naeg.), large colonies of which were found lying on the bottom of the Long Water. These might in part have been responsible for the failure of the autumn crop of phytoplankton, by competing for nutrient salts. The year 1963 thus differed from previous years in that diatoms were much less important in the plankton, and that the autumn crop of phytoplankton failed at the same time as *Coccochloris* developed extensively on the bottom of the Long Water.

The temperatures measured during the four years are recorded in Fig. 2. Each year is compared with the smoothed 3 years' mean for 1959–61. The years 1961 and 1963 show contrasting departures from the mean in the early part of the year. In 1961 the water temperature remained above average throughout February and March and reached

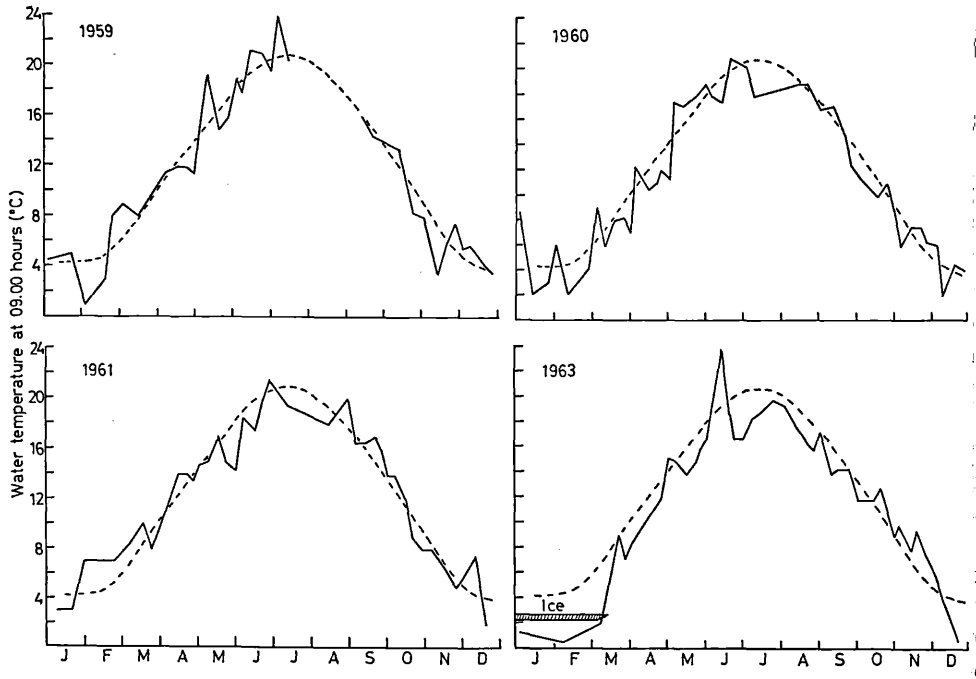

FIG. 2. Variation in the temperature of the water at 09.00 hours in Hampton Court Long Water during 1959, 1960, 1961 and 1963. The broken line in each graph gives the smoothed mean for the years 1959–61.

10° C in March, which was not so in any of the other years. This early warmth was associated with an early appearance of summer forms such as *Polyphemus pediculus* and *Scapholeberis mucronata*. In 1963 the temperatures in the early part of the year were well below normal, and there was a thick cover of ice from the last week of December 1962 to the end of the first week in March 1963. In previous years the longest period of ice cover was found at the end of 1961, when the surface of the Long Water was frozen from 18 December until 7 January 1962. In March 1963, after the ice had melted, the temperature rose rapidly to 9° C, but thereafter the temperatures were below normal, apart from two short periods at the beginning of May and in June when the temperatures rose above the average. For the rest of the summer the water temperature remained below the 3-year mean, but became normal, or slightly above, during the autumn.

A biotic factor which may influence the populations of some Cladocera is the spawning

of fish and the occurrence of shoals of small fish in the marginal region. The fish which have been found spawning in the shallow water are perch (*Perca fluviatilis* L.), tench (*Tinca tinca* L.) and roach (*Rutilus rutilus* (L.)). Shoals of young fish have been seen most frequently in shallow water in the first 2 weeks of June.

Many of the invertebrates in the Long Water take Cladocera as part of their food. The seasonal influence of these predators has not been studied, but one clear relationship has been found. The small oligochaete *Chaetogaster diaphanus* Gruithuisen becomes particularly abundant in the Long Water in April when the population of *Chydorus sphaericus* is at its maximum. This oligochaete is known to feed on chydorids to a large extent (Green 1954b). In 1961 *Chaetogaster diaphanus* became abundant in March. The population of *Chydorus sphaericus* also reached its peak earlier than usual, probably because there were no prolonged cold spells in February and March.

SEASONAL OCCURRENCE OF CLADOCERA IN HAMPTON COURT LONG WATER

Each collection of Cladocera was examined to determine which species were present and which were producing eggs. During the course of the 4 years thirty-six species were found. Some of these were found only once or twice, and others were not regularly found because their collection involved special techniques. The main collecting effort was directed towards those species which occur among vegetation near the margins of the Long Water.

The seasonal occurrences of the nineteen species most frequent in the Long Water are shown in Fig. 3, which shows the months in which females were found producing either parthenogenetic eggs or resting eggs.

As shown in the diagram, some species can continue reproducing throughout the year while others do not reproduce during the winter. The overwintering species were severely reduced during the early part of 1963. Collecting was made difficult by a layer of ice about a foot (30 cm) thick, but on 13 February 1963 a collection was made from the overflow pond at the end of the Long Water. The temperature of the water under the ice was 0·5° C. A few specimens of the following Cladocera were found: *Simocephalus vetulus* (without eggs), *Chydorus sphaericus* (with parthenogenetic eggs), *Alona affinis* (with parthenogenetic eggs), *Leydigia leydigi* (with parthenogenetic eggs) and *Pleuroxus aduncus* (without eggs). All these species are among those found regularly throughout the winter in other years.

A complete list of the Cladocera found in the Long Water is given below, together with notes on their seasonal occurrence and production of eggs.

Family Sididae

Sida crystallina (O. F. Müller). Details of the seasonal cycle of this species are given in the following section. It usually persists throughout December, and a few females with resting eggs may be found in January.

Diaphanosoma brachyurum Liéven has been found on relatively few occasions in the Long Water. Specimens with eggs were found in September 1961 and August 1963.

Family Daphniidae

Daphnia longispina O. F. Müller occurs throughout the year and has been found with parthenogenetic eggs under ice.

D. ambigua Scourfield. This species has probably been introduced with aquatic plants

from America, where it is widespread (Brooks 1957). In Britain it is known only from the London Area, with records from Kew Gardens (Scourfield 1946), Regents Park (Fox 1948), the Serpentine (Fox 1960, personal communication) and the small ornamental lake in the gardens of Buckingham Palace (Evans, Gilchrist & Green 1964). It was found in the Long Water in May 1962.

Scapholeberis mucronata (O. F. Müller). The seasonal cycle of this species in the Long Water has been described in detail (Green 1963). Some additional data are given in the following section.

S. aurita (Fischer). The only previous record of this species in Britain appears to be that of Gurney (1903) from Norfolk. Single specimens were taken from the Long Water on 13 and 28 August 1963, both specimens were females and each carried ten eggs.

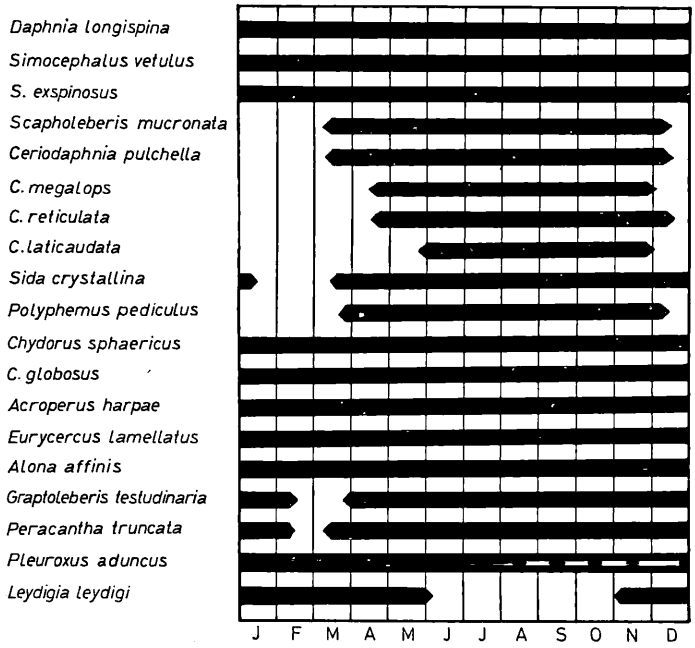

FIG. 3. Seasonal occurrence of reproducing females of the nineteen most frequent species of Cladocera in Hampton Court Long Water.

Simocephalus vetulus (O. F. Müller). This is one of the most abundant Cladocera in the Long Water. In mild winters the population persists and reproduces by parthenogenesis, but when conditions are more severe some of the females produce resting eggs. The percentage of females which lay resting eggs is always very low. This species was chosen for special study because of its abundance and persistent parthenogenesis through most winters.

S. exspinosus (Koch) also reproduces parthenogenetically throughout the year, but is usually much less abundant than *S. vetulus*.

Ceriodaphnia reticulata (Jurine). Members of the genus *Ceriodaphnia* are all summer forms, which overwinter as resting eggs and become active in March or April. A detailed account of the reproductive cycle of the species in the Long Water during 1963 is being prepared by Miss M. J. Burgis. The notes which follow are based on collections made

between January 1959 and December 1961. *C. reticulata* becomes active towards the end of April, and females with parthenogenetic eggs can be found from the last week in April until the middle of December. A few resting eggs are produced in June, but the main production of resting eggs occurs from October to December.

C. megalops Sars is much more erratic in occurrence than *C. reticulata* in the Long Water. It was not found in the Long Water in 1959, but became abundant during the summer of 1960. Males and resting eggs were found in October and November.

C. pulchella Sars is sometimes found reproducing by parthenogenesis in March and continues breeding until the middle of December. Resting eggs have been found in October, November and December.

C. laticaudata P. E. Müller has not been collected with the same regularity as the three preceding species. In the Long Water it has been found reproducing by parthenogenesis from June to November.

C. quadrangula (O. F. Müller) is rare in the Long Water, only a few isolated specimens have been taken during the summer months.

Family Bosminidae

Bosmina longirostris (O. F. Müller) lives in the open water away from the margins of the Long Water, and was not sampled regularly, but it was found to be capable of reproducing by parthenogenesis through mild winters, but not through the severe winter of 1963.

Family Macrothricidae

Ilyocryptus sordidus (Liéven). The annual cycle of this species has not been studied in sufficient detail to be certain about it overwintering in an active state, although specimens have been collected in January.

I. agilis Kurz is somewhat rarer than *I. sordidus*, but I have records of it breeding by parthenogenesis in the Long Water from March until November.

Macrothrix laticornis (Jurine)—only a few isolated specimens have been found.

Family Chydoridae

Eurycercus lamellatus (O. F. Müller) fluctuates greatly in abundance without any obvious relation to the seasons. It has been found reproducing by parthenogenesis through some winters, but then became very sparse in the late spring. Further details concerning this species are given in the following section.

Graptoleberis testudinaria (Fischer). This small transparent species is not easy to find regularly, but I have been able to establish that it breeds by parthenogenesis in the Long Water from the end of March to the beginning of February. The few specimens found after the middle of February were without eggs, so I believe that this species usually stops reproducing for a very short period from about the first week of February to the end of March.

Peracantha truncata (O. F. Müller) reproduces by parthenogenesis for the greater part of the year, but I have not been able to find reproducing females for a short period in late February and early March. The females taken in January are usually without eggs. This species becomes very abundant in the Long Water in autumn, and resting eggs have been seen in October and November.

Camptocercus rectirostris Schödler has been recorded only twice in the Long Water, in November 1961 and January 1962.

Acroperus harpae Baird reproduces by parthenogenesis throughout the year, and bisexual reproduction has been observed in January, February, June and December.

Leydigia leydigi Schödler reproduces by parthenogenesis throughout the winter, but becomes scarce in the summer, and I have not found any active specimens during the late summer in the Long Water.

Alona affinis Leydig reproduces by parthenogenesis throughout the year, and often becomes abundant during the winter.

A. rectangula Sars has also been found reproducing by parthenogenesis through most winters, but it was not found in the severe winter of 1963.

A. quadrangularis (O. F. Müller) has not been collected with sufficient regularity to establish its breeding period, but females with parthenogenetic eggs have been found in November, January, February, March and April.

A. guttata Sars seems to breed by parthenogenesis through mild winters. This species has not been collected in every month from the Long Water, but females with parthenogenetic eggs have been taken in January, February, March, June, November and December.

A. costata Sars has been recorded only twice in the Long Water, once in March and once in September; it may well have been overlooked in other months.

Alonella rostrata (Koch) (syn., *Rhynchotalona rostrata*) has been collected on a few occasions during the summer months.

Pleuroxus aduncus (Jurine) breeds parthenogenetically throughout the year, but specimens collected in midwinter 1963 were without eggs. Bisexual reproduction has been observed in February.

P. trigonellus (O. F. Müller) has not been studied in all months, but has been found reproducing parthenogenetically in January, February, November and December.

P. uncinatus Baird has been collected on relatively few occasions. Parthenogenetic reproduction has been seen in May, June, October and November.

P. denticulatus Birge. A single specimen was collected in June 1961. This appears to be the second record of this species in Britain, the first being from Exminster, Devon (Scourfield 1907).

Chydorus sphaericus (O. F. Müller) breeds parthenogenetically throughout the year, and becomes very abundant in March and April. Bisexual reproduction has been observed in the Long Water in December.

C. globosus Baird is always much scarcer than *C. sphaericus*, but it has been found breeding parthenogenetically throughout the year.

Family Polyphemidae

Polyphemus pediculus (L.) emerges from resting eggs towards the end of March and reproduces parthenogenetically until the beginning of December. Bisexual reproduction begins in June and reaches a peak in October (details in the following section).

SEASONAL VARIATION IN EGG PRODUCTION

Simocephalus vetulus

Seasonal variations in the length of mature females and the number of eggs carried by them are shown in Fig. 4. The years 1959–61 were very similar, with distinct peaks of egg production in spring and autumn. In 1961 the spring peak of egg production was higher than in previous years, even though the mean length of the females carrying the

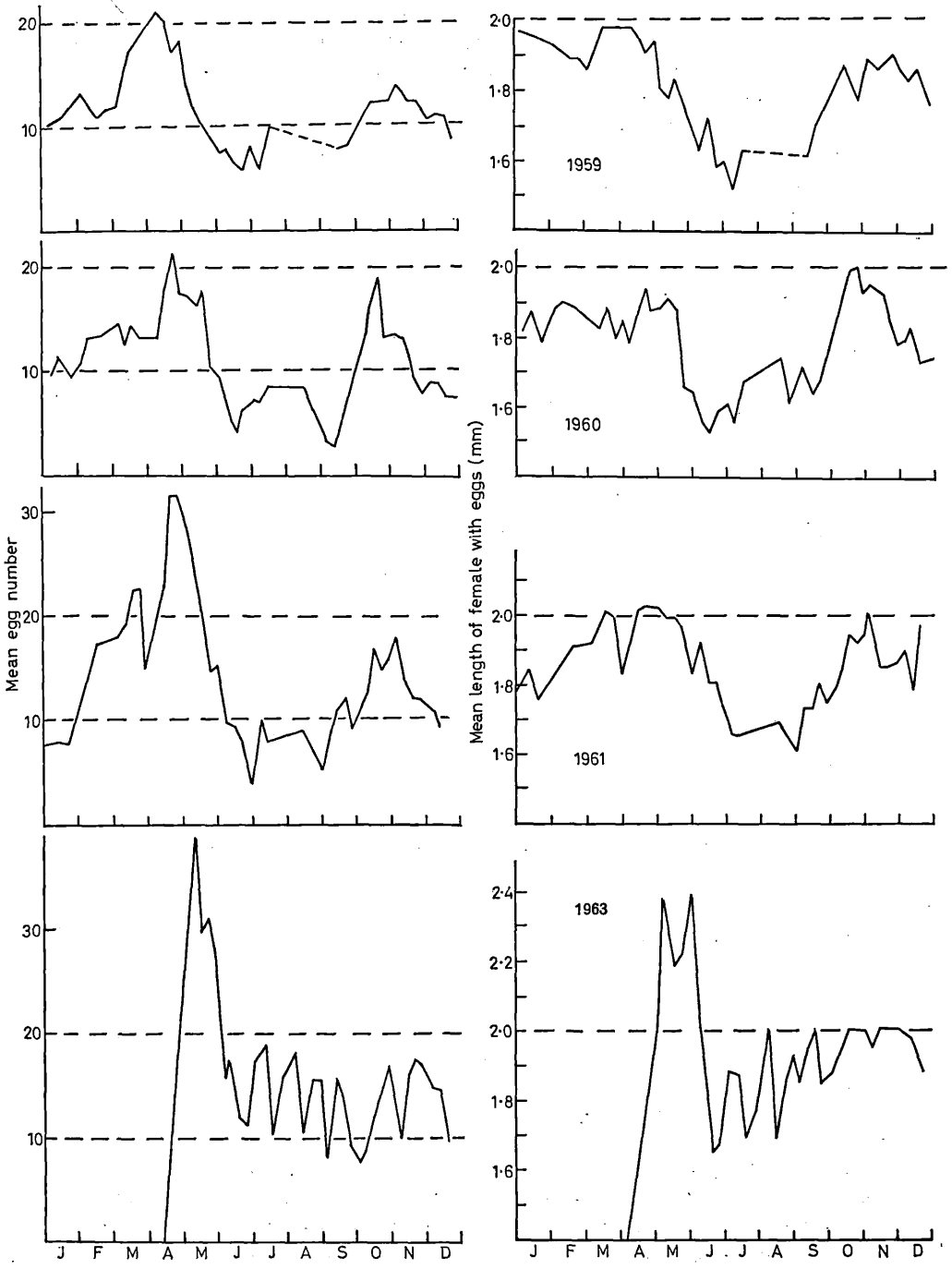

FIG. 4. *Simocephalus vetulus*: seasonal variation in mean egg number and in mean length of mature females in Hampton Court Long Water. The broken lines at a mean length of 2·0 mm and mean egg numbers of ten and twenty have been added to facilitate comparison of the four years.

eggs was only slightly greater. Seasonal variation in 1963 was quite different from that in the other three years. The species did not begin reproducing until April, and then the mean egg number rose to a very high level. The females were also much larger than usual. During the summer there were fluctuations in the mean egg number, but the number never fell to the low values recorded in the summers of other years, nor was there a clearly defined autumn peak. This can be related to the fluctuating but relatively high concentration of chlorophyll in the Long Water throughout the summer. The egg number was low in the first week of November when the mean fell to 10·1, otherwise the mean egg numbers were similar to those of November 1961. This is remarkable in view of the

Table 1. Simocephalus vetulus: *variation in the percentage composition of the population in Hampton Court Long Water*, 1959

Date	Females with parthenogenetic eggs	Females of mature size without eggs	Immature females	Ephippial females	Males
30 April	43	1	56	0	0
5 May	34	1	65	0	0
12 May	20	0·5	79·5	0	0
20 May	29	1	69	0·5	0·5
27 May	36	1	62	1	1
4 June	42	2	56	0	0
9 June	49	3	48	0	0
17 June	48	2	50	0	0
26 June	46	1	53	0	0
1 July	48	1	51	0	0
7 July	37	2	61	0	0
15 July	46	1	53	0	0
16 Sept.	34	1	65	0	0
24 Sept.	10	2	88	0	0
15 Oct.	30	1	69	0	0
29 Oct.	21	1	76	1	1
5 Nov.	9	1	89	0·5	0·5
12 Nov.	28	1	67	1	3
19 Nov.	30	1	69	0·5	0·5
27 Nov.	32	1	65	1	1
3 Dec.	35	1	64	0	0
10 Dec.	37	1	62	0	0

great differences in chlorophyll content of the water towards the ends of the two years. Decreasing temperatures are probably responsible for restricting egg production in the presence of abundant food (see following section, and Fig. 10).

The percentage composition of the population was studied from 30 April 1959 to 10 December 1959 to determine when bisexual reproduction occurred and the extent to which females changed from parthenogenesis to producing resting eggs. Table 1 shows two short periods of bisexual reproduction. One, at the end of May, lasted 2 weeks. The second occurred during November and lasted about 4 weeks. The percentage of females with resting eggs did not rise above 1% of the total population during either period, but the percentage of males rose to 3 on 12 November. An interesting feature of Table 1 is the stability of the population structure during June and July, and the low percentage of females of mature size which were not producing eggs. No females with resting eggs were found in 1960, although each sample was searched carefully.

Scapholeberis mucronata

A detailed account of the reproductive cycle of this species in the Long Water during 1960 and 1961 has already been given (Green 1963). The cycle in 1963 was studied to see whether the events of the previous years were repeated and whether the severe winter caused any delay in the first appearance of females carrying eggs. Some samples were also taken in 1964 to determine the first appearance of females with eggs. The dates when the first females with eggs were found were as follows: 1960, 14 April; 1961, 15 March; 1963, 25 April; 1964, 15 April. Thus the first appearance of reproducing females in 1963 was about 10 days later than in a normal year, and over a month later than in 1961 which had above normal temperatures during February and March (see Fig. 2). In 1963 several immature females were found as early as 27 March. They had probably emerged from resting eggs in response to the stimulus provided by the sudden rise in temperature after the ice had melted, but the low temperatures in the early part of April seemed to prevent

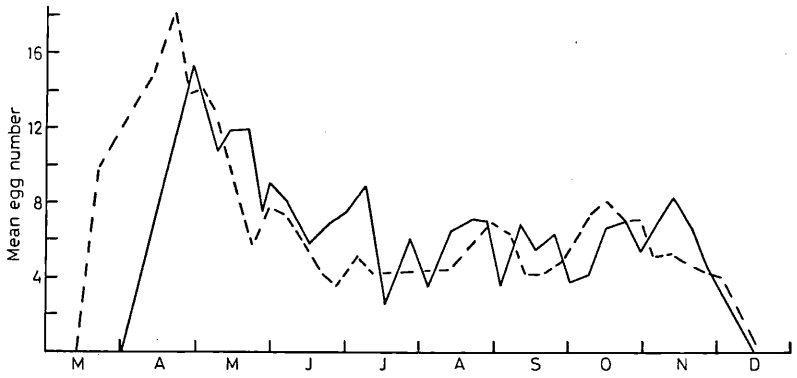

FIG. 5. *Scapholeberis mucronata*: seasonal variation in egg production, 1961 (– – –) and 1963 (———).

these early emergers from becoming mature until near the end of the month. The seasonal variation in egg production during 1963 is compared to that of 1961 in Fig. 5. The two sets of data are basically similar except that the spring peak is later in 1963 and there are greater fluctuations during the summer compared to 1961. This species did not respond so markedly as *Simocephalus vetulus* and *Sida crystallina* to the differences in chlorophyll content of the water in the two years.

Polyphemus pediculus

The reproductive cycle of *Polyphemus* in the Long Water during 1963 is shown in Fig. 6. Reproductive activity began at the end of April. There was a marked spring peak in egg production which then declined rapidly during May. The production of resting eggs began in June, but only a small percentage of females was involved. A small peak in this percentage was found at the end of June, but it then declined to only 1 or 2% during the rest of the summer until the end of September. During October there was a very high peak in the percentage of females carrying resting eggs, but this declined rapidly at the beginning of November. The mean number of resting eggs carried by each female varied between two and four during the period from June to December (Fig. 6).

Sida crystallina

Fluctuations in egg production by *Sida* during 1961 and 1963 are shown in Fig. 7. In 1961 the reproductive season began in March, but in 1963 the onset of reproduction was delayed until the middle of April. In 1961 there was a very sharp peak of egg production in spring, falling to a low level through the summer, then rising to a high level

Table 2. *Highest and lowest mean egg numbers and lengths of adult female Cladocera in Hampton Court Long Water*

Species	Date	Mean egg number	Mean length (mm) of females with eggs
Simocephalus vetulus	8 May 1963	39·2±3·38	2·38±0·07
	13 Sept. 1960	2·9±0·32	1·64±0·03
Scapholeberis mucronata	10 May 1962	21·0±1·30	1·12±0·03
	17 July 1963	2·6±0·56	0·79±0·02
Sida crystallina	20 April 1961	38·2±1·63	2·97±0·04
	12 July 1961	4·0±0·72	1·88±0·04
Eurycercus lamellatus	20 April 1961	20·2±2·41	2·51±0·06
	12 July 1961	3*	1·81*
Polyphemus pediculus	30 April 1963	28·3±2·31	1·44±0·03
	3 Sept. 1963	3·9±0·23	1·03±0·03

* Only a single specimen found.

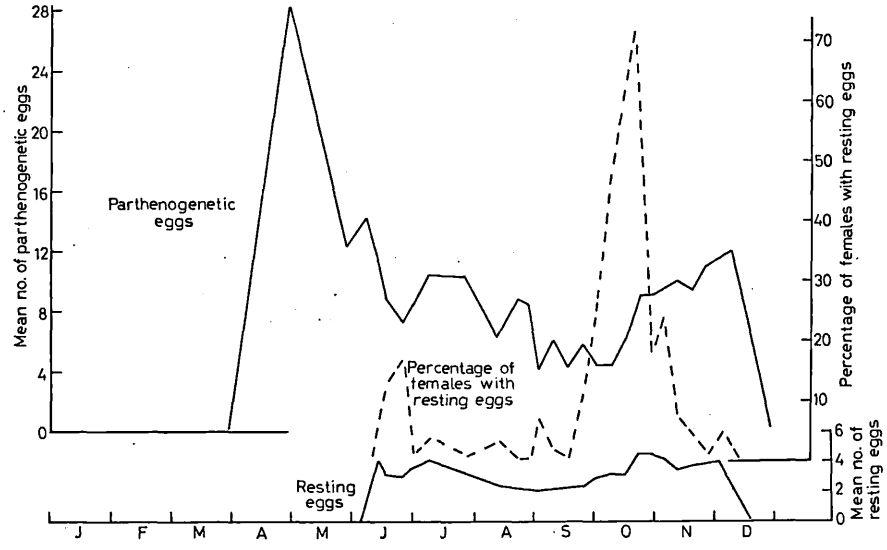

FIG. 6. *Polyphemus pediculus:* seasonal variation in egg production during 1963. The positions of the axes have been adjusted to prevent excessive overlapping of the sets of data.

again in the autumn. Most of the eggs produced in autumn were resting eggs, the change from parthenogenesis taking place in October. In 1963 there was not such a sharply defined spring peak and the number of eggs per female did not fall very low during the summer, but fluctuated in a manner similar to that noted in *Simocephalus vetulus* in the same year. There was a well-marked rise in egg number towards the end of 1963, with

the change to resting egg production occurring again in October. This change was completed somewhat earlier in 1963 than in 1961.

The almost complete change to production of resting eggs during October influences the structure of the population during November and December. The resting eggs do not hatch until the following spring, so that there are very few new recruits to the autumn population. The females which remain increase gradually in size (Fig. 8). This increase took place in 1963 when food was sparse, and in 1961 when food was abundant (cf. Fig. 1). Very large females were found in November 1963. This can be related to the earlier change to production of resting eggs in 1963 (Fig. 7) which would lead to an earlier reduction in the number of recruits to the population. The remaining females then grew to a large size.

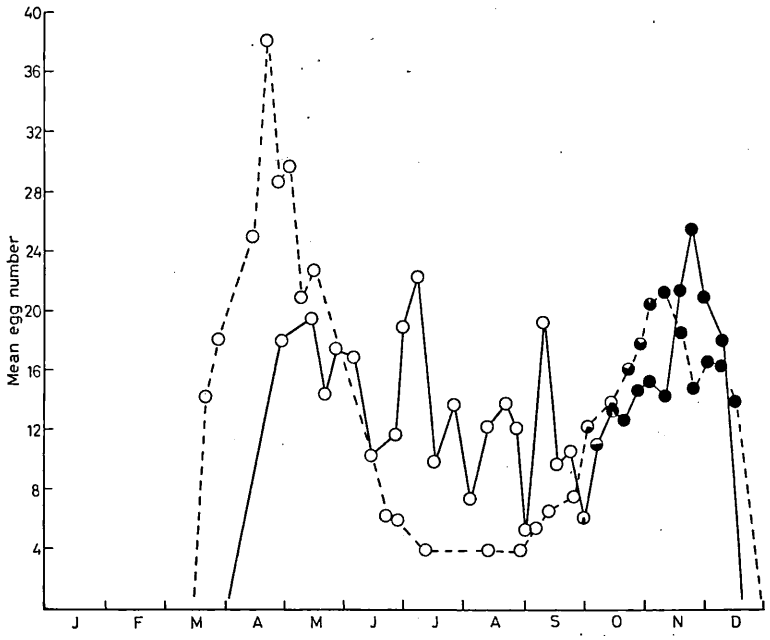

FIG. 7. *Sida crystallina*: seasonal variation in egg production, 1961 (– – –) and 1963 (———). Some circles are partially filled in to show the approximate percentage of mature females with resting eggs.

Eurycercus lamellatus

This species was not studied for a complete year because it becomes scarce in the shallow water during the summer. This may be due to movement to deeper water. Berg (1938) found that *Eurycercus* was not common in very shallow water at the margin of Lake Esrom, but found that it reached a density of 300/m^2 at a depth of 1 m. Samples of this species were taken from the Long Water from 9 November 1960 to 9 May 1961. At the end of November and the beginning of December the population was dense and some of the females were producing resting eggs. At the end of December the population declined and remained small throughout the rest of the sampling period, apart from a slight increase at the beginning of May. This description applies only to the population in shallow water. It is probable that the population in deeper water showed a considerable increase during March, April and May, because the females found in shallow water were

producing large numbers of eggs during this period (Fig. 9). A few specimens were found in shallow water in June 1961. They were small and each female was carrying only three or four eggs.

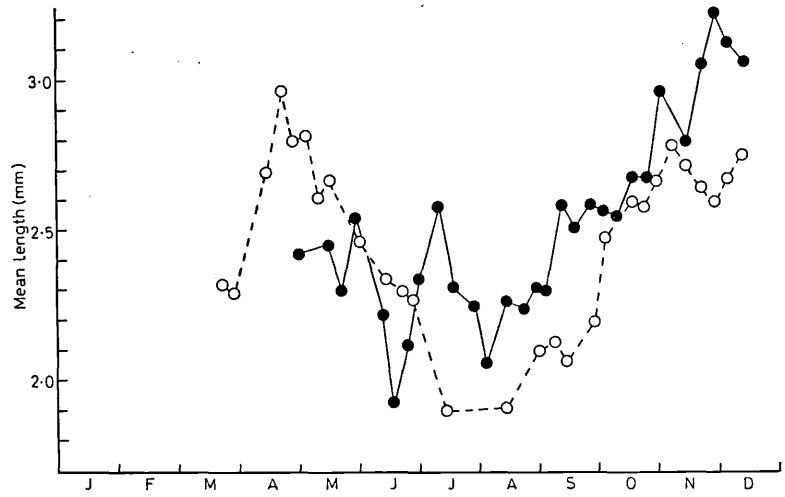

FIG. 8. *Sida crystallina*: seasonal variation in the mean length of females with eggs, 1961 (– – –) and 1963 (———).

EGG PRODUCTION IN RELATION TO TEMPERATURE AND FOOD

Comparison of the data concerning egg production by *Simocephalus vetulus* during 1961 and 1963 (Fig. 4) with the chlorophyll content of the water (Fig. 1) gives a clear indication of the importance of food, particularly during the spring peak of egg production. There

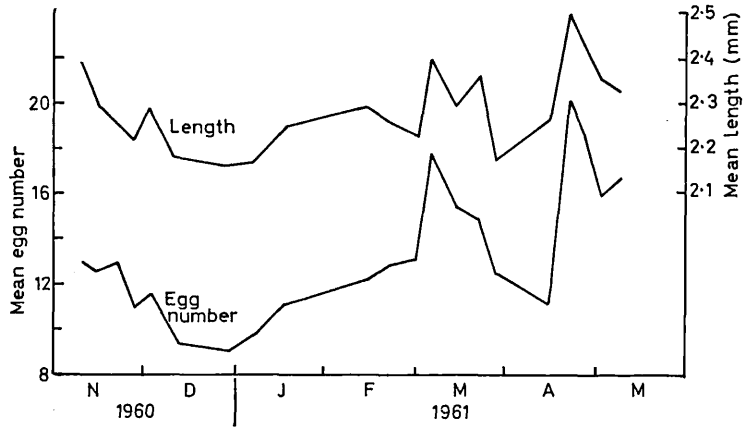

FIG. 9. *Eurycercus lamellatus*: seasonal variation in egg production and mean length of females with eggs.

is also some interaction with temperature because during the summer the egg numbers remain lower than the spring peak even when food is abundant. In 1963 the succession of chlorophyll peaks throughout the summer prevented the mean egg number from falling to the low levels recorded in previous years.

The low mean egg number found in most summers is due to a combination of factors. At high temperatures a female cladoceran becomes mature at a smaller size than at lower temperatures (MacArthur & Baillie 1929). This smaller size will naturally restrict the egg laying capacity of the female (Green 1954a, 1956). The density of the population and of populations of allied species of Cladocera are usually high during the summer, so that for a given amount of food there will be less for each individual; this will also restrict egg production.

Fig. 10 shows that low temperatures also restrict egg production. In this figure all the data for egg production in three ranges of temperature and at known chlorophyll content of the water have been grouped together. Means have been taken for each 100 units of chlorophyll. Each point is based on at least twenty females with eggs, and the points for the lower concentrations of chlorophyll are based on several hundred. It is

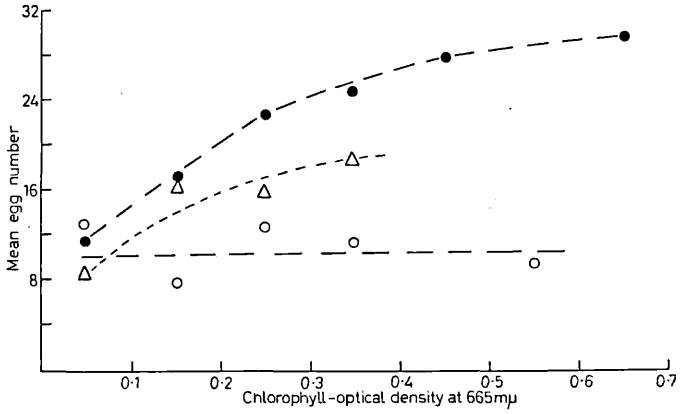

FIG. 10. *Simocephalus vetulus*: relationship between egg production, temperature and food. Each point is based on the mean value of several samples of twenty females grouped together at intervals of 100 spectrophotometer units of chlorophyll (an optical density of 0·1 is equal to 100 units). ○, 2–7° C; ●, 12–17° C; △, 18–22° C.

clear that in the lowest temperature range there is no increase in egg production with increasing food. In the middle temperature range the number of eggs per female increases with increasing food. At temperatures above 18° C the mean egg number also increases with increasing food, but not to the same extent as it does when the temperature lies between 12 and 17° C. From these data it would seem that the optimum temperature for egg production per female in the population of *S. vetulus* in the Long Water lies somewhere between 12 and 17° C. If the data for the four years are grouped together and the mean egg number for each interval of 3° C is calculated a smooth curve results (Fig. 11) and supports the conclusion reached from Fig. 10.

When the data for *Scapholeberis mucronata* and *Sida crystallina* are treated in the same way it is found that *Scapholeberis mucronata* has an optimum temperature for egg production per female similar to that of *Simocephalus vetulus*. *Sida crystallina* shows a humped curve with the largest egg number produced at a surprisingly low temperature. The explanation of this peculiar curve can be derived from the nature of the annual cycle of *Sida* (see previous section). The change-over to production of resting eggs in October results in the growth of very large individuals capable of producing large numbers of resting eggs when the temperature of the water is falling in the autumn. The first three

points from the left in the curve for *Sida* all apply to resting eggs, while the others apply to parthenogenetic eggs. The curves for *Simocephalus* and *Scapholeberis* are drawn entirely from parthenogenetic eggs.

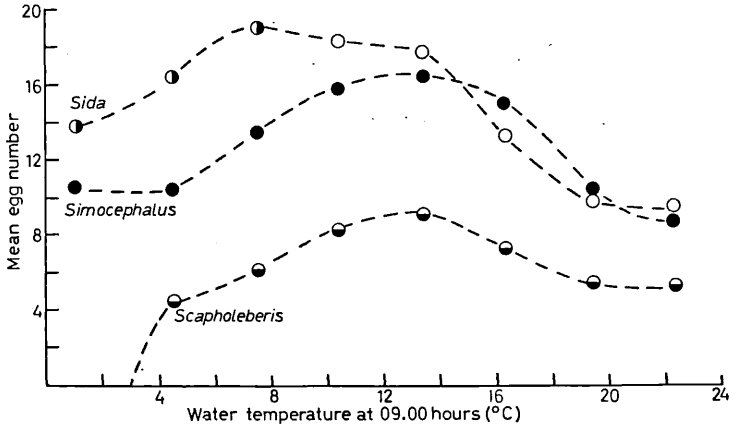

FIG. 11. The relationship between temperature and egg production in *Simocephalus vetulus* (4 years' data), *Scapholeberis mucronata* (3 years' data) and *Sida crystallina* (2 years' data). The data have been grouped at intervals of 3° C to provide a large number of counts for each point. The first three points from the left on the curve for *Sida* refer to resting eggs; the others refer to parthenogenetic eggs.

The analysis given above is based on the mean number of eggs produced by each female. Now Figs. 4 and 18 show that size and egg number are closely related and tend to fluctuate together, so that variation in egg production might be caused simply by variation in size. This would be so only if the regression of egg number on size were

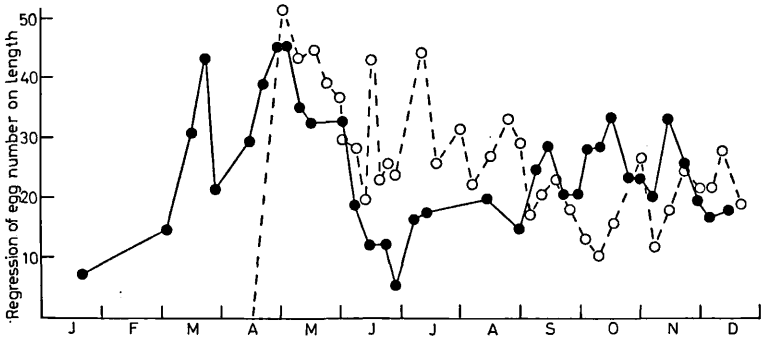

FIG. 12. *Simocephalus vetulus:* seasonal variation in the regression of egg number on length. Each point gives the value of b which indicates the slope of the relationship between egg number (Y) and length (X). The value of Y can be estimated from the equation $Y-\bar{y} = b(X-\bar{x})$, where $\bar{y}$ is the mean egg number and $\bar{x}$ is the mean length. ●, 1961; ○, 1963.

similar in all samples. But it is not. If regression coefficients are calculated for each sample a wide range is found. When these coefficients are plotted on a seasonal basis (Fig. 12) the fluctuations are similar to and follow those of the chlorophyll content of the water. This is well shown by a comparison of *Simocephalus vetulus* in the summers of 1961 and 1963. The chlorophyll content of the water was much higher in 1963 and

showed more violent fluctuations than in 1961. The regression of egg number on length shows similar fluctuations.

The regression coefficients may also be used to calculate egg production corrected to a grand mean size. These corrected egg numbers can then be used to study the effects of environmental conditions on egg production at a given size. The results of correcting the

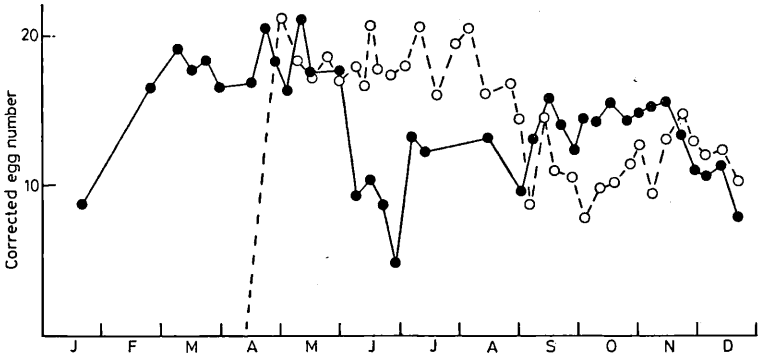

FIG. 13. *Simocephalus vetulus*: egg production per female corrected for the size of the females, 1961 (●) and 1963 (○).

data for *S. vetulus* are shown in Fig. 13. When these results are compared with the crude egg numbers (Fig. 4) it is seen that the spring peak is levelled down by the correction procedure. This indicates that the large number of eggs per female in the spring is due to the large size of the females. Further, the smaller number of eggs per female in the summer is mainly due to the small size of the females. The relation between temperature

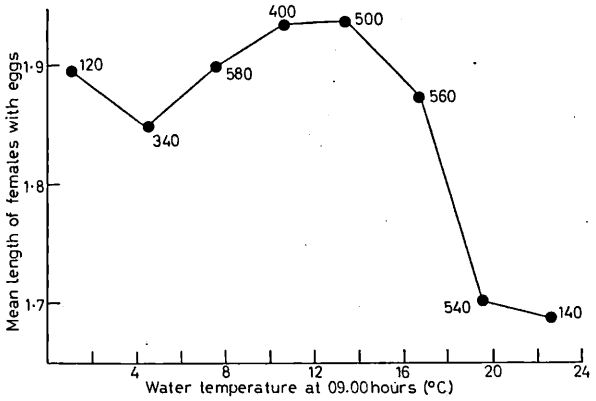

FIG. 14. *Simocephalus vetulus*: mean length of adult females in relation to temperature. The figures at the points give the number of females on which each point is based.

and mean length of adult females is shown in Fig. 14. It is clear that when the temperature rises above 18° C the mean length of the adult females falls steeply. There is also a clear effect of food, because the corrected egg numbers for 1963 are higher than in 1961 during June, July and August when the chlorophyll content of the water was also higher than in 1961. In September, October and November the situation is reversed; the chlorophyll content was higher in 1961 than in 1963 and the corrected egg numbers show corresponding changes. In December both years show a decrease in the corrected egg number which may be attributed to low temperatures. If the procedure used to construct Fig. 10

is repeated using the corrected egg numbers for each sample an interesting result is obtained (Fig. 15). The difference between the middle group (12–17° C) and the high temperature group (18–22° C) disappears and above a certain level of chlorophyll there is no further increase of egg production. This shows that the increase in egg production

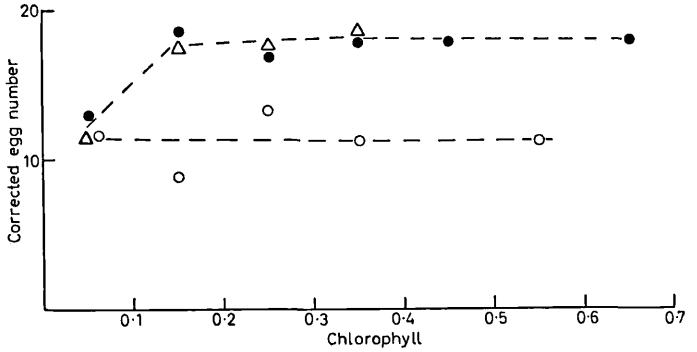

FIG. 15. *Simocephalus vetulus*: relationship between corrected egg number, temperature and food (compare with Fig. 10). ○, 2–7° C; ●, 12–17° C; △, 18–22° C.

with increasing chlorophyll shown by the 12–17° C group in Fig. 10 is due to the extra food causing an increase in size. At low temperatures (2–7° C) the corrected egg numbers remain below those of the two other groups, so that even in the presence of abundant food the production of eggs is restricted.

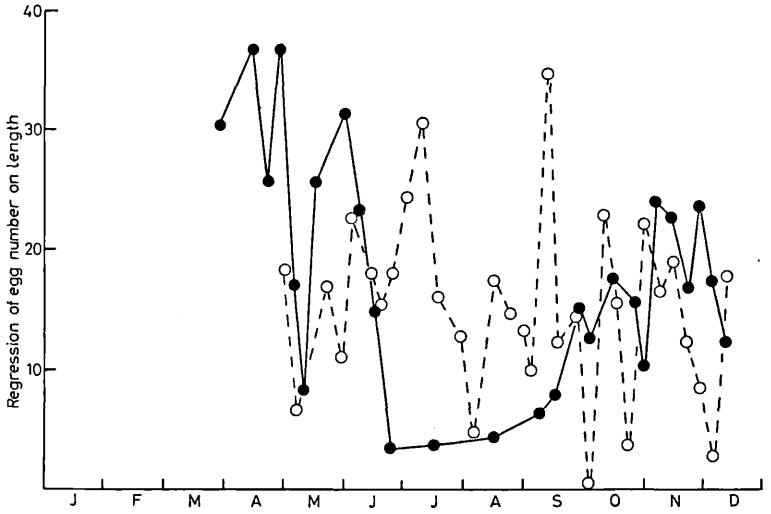

FIG. 16. *Sida crystallina*: seasonal variation in the regression of egg number on length. Each point gives the value of b which indicates the slope of the relationship between egg number (Y) and length (X). The value of Y can be estimated from the equation $Y-\bar{y} = b(X-\bar{x})$, where $\bar{y}$ is the mean egg number and $\bar{x}$ is the mean length. ●, 1961; ○, 1963.

Sida crystallina also shows wide fluctuations in the regression of egg number on length (Fig. 16). These fluctuations can be related to the fluctuations in the chlorophyll content of the water. The high peaks of chlorophyll are usually followed by an increase in the regression of egg number on length. When the regression coefficients are used to correct the egg number to a grand mean size the high peak in the spring of 1961 is reduced

but not eliminated (Fig. 17). During the summer of 1961 the corrected mean egg number remained low, but increased during September and October when the chlorophyll content of the water increased. The corrected egg numbers for 1963 remained above those of 1961 throughout June, July and August, but fell below those of 1961 during September and October when the chlorophyll content of the water fell below the values found in the same months of 1961.

LATITUDINAL VARIATION IN EGG PRODUCTION

In Fig. 18 the mean egg numbers of all the samples of *Simocephalus vetulus* from the Long Water are plotted against the mean length of the females carrying the eggs. It is obvious that the mean egg number increases with increasing size of the females. When data from two populations of the same species from Greenland are plotted on the same figure it is seen that both produce fewer eggs for the same length of female than the population from the Long Water.

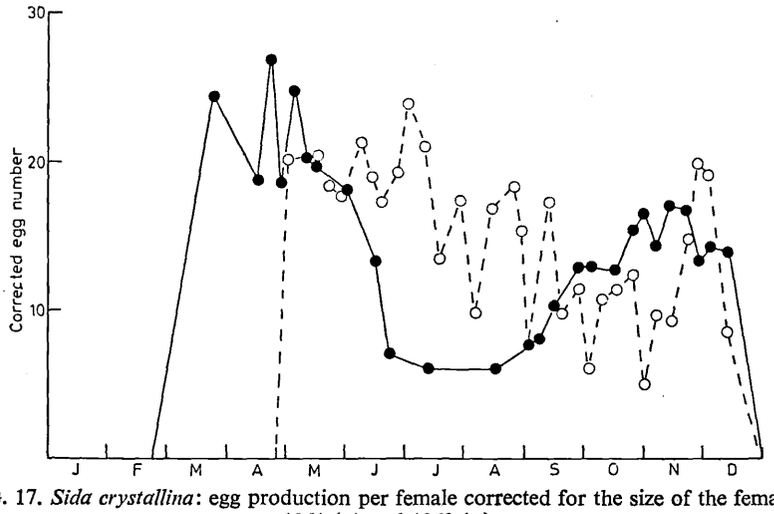

FIG. 17. *Sida crystallina*: egg production per female corrected for the size of the females, 1961 (●) and 1963 (○).

Similarly, in Fig. 19, the mean egg numbers of *Scapholeberis mucronata* from several British and Danish localities all show a similar relationship with body length. Four samples from separate populations in Greenland all produce fewer eggs at a given body length than the British and Danish specimens. A sample from Northern Italy (Fondo Toce, Lago Maggiore) where the summer temperatures are appreciably higher than those in Britain and Denmark, shows a larger number of eggs at a given body length.

This latitudinal difference in the relation between body length and egg production in *Simocephalus vetulus* and *Scapholeberis mucronata* is correlated with a latitudinal variation in the size of their eggs (see later section).

SEASONAL VARIATION IN THE SIZE OF EGGS

Agar (1913) demonstrated that the eggs laid by females of *Simocephalus vetulus* kept at 15·5–18·5° C gave rise to larger young than eggs laid by females kept at 28·5–31·5° C.

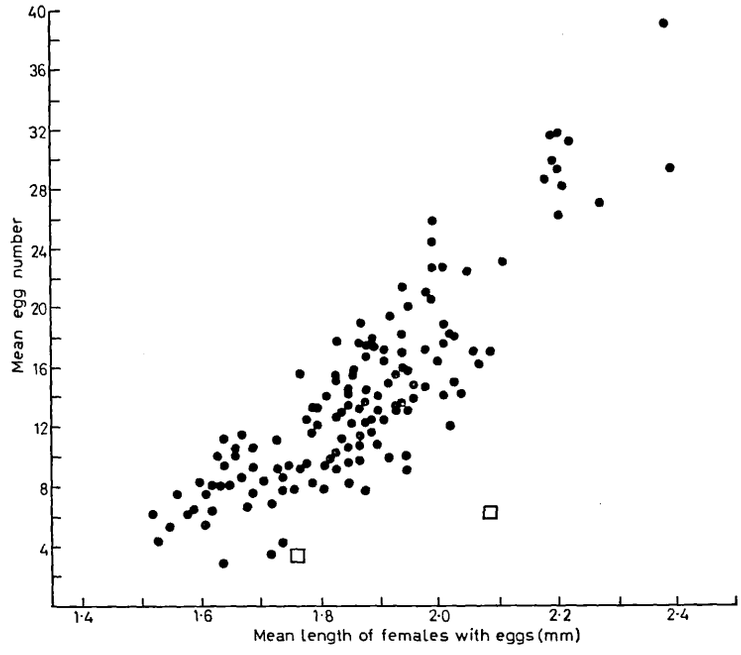

FIG. 18. *Simocephalus vetulus*: relationship between length and egg number per female. ●, From Hampton Court Long Water; □, from Greenland. Each point is based on a sample of ten or twenty females.

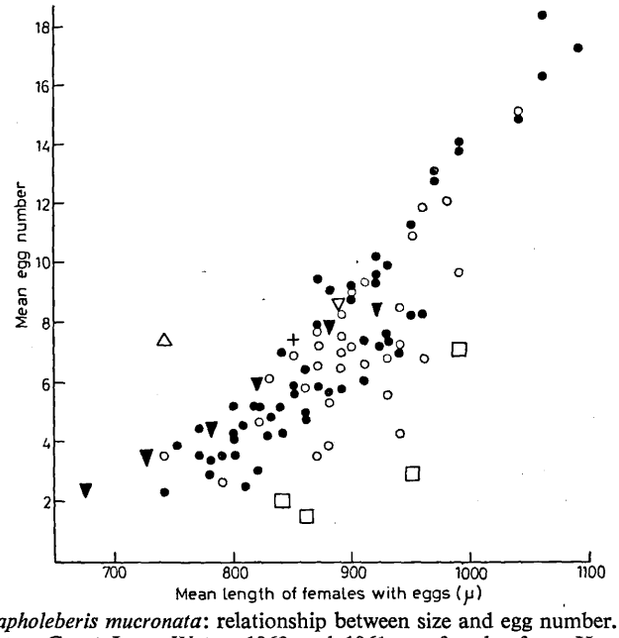

FIG. 19. *Scapholeberis mucronata*: relationship between size and egg number. ●, Females from Hampton Court Long Water, 1960 and 1961; ○, females from Hampton Court Long Water 1963; +, females from Esthwaite; □, females from Greenland; ▽, females from Ager Sø, Denmark; ▼, females from Løg Sø, Denmark; △, females from Fondo Toce, Lago Maggiore, Italy. Each point is based on a sample of ten or twenty females, except the data relating to Løg Sø where the six points are based on a total of fifty females.

In Cladocera which do not secrete a nutritive fluid into the brood pouch (i.e. excluding genera such as *Moina* and *Polyphemus*) the size of the eggs and the size of the young may be considered together, the size of the young being proportional to the size of the egg (Green 1956). The question arises whether there is in natural populations any seasonal

Table 3. Simocephalus vetulus: *diameters and volumes of parthenogenetic eggs, Long Water*, 1963, *and Greenland*, 1961

Date	Water temperature at 09.00 hours (°C)	No. of eggs measured	Mean egg diameter (μ) Greatest	Least	Egg volume (millions μ^3)
30 April	15	60	246	218	6·1
14 May	14	40	248	228	6·7
10 June	21	70	245	216	6·0
13 Aug.	18	50	252	223	6·6
28 Aug.	16	40	255	233	7·3
25 Sept.	14·5	12	262	235	7·6
16 Oct.	12	20	272	247	8·6
6 Nov.	10	30	270	243	8·3
27 Nov.	7·5	55	270	250	8·8
4 Dec.	6	20	271	244	8·4
11 Dec.	3·5	30	287	259	10·1
20 Dec.	0·5	50	280	254	9·5
GREENLAND					
28 July	11	22	308	273	12·0

variation in the size of the eggs, and whether this can be related to temperatures lower than those studied by Agar. It is possible that other factors might obscure the effect of temperature. Agar (1914) showed that in a monoclonal population of *Simocephalus exspinosus* at a given temperature, in females of the same age, when the size of the

Table 4. Sida crystallina: *diameters and volumes of eggs, Long Water*, 1963

Date	Water temperature at 09.00 hours (°C)	Type of egg	No. measured	Mean egg diameter (μ) Greatest	Least	Egg volume (millions μ^3)
30 April	15	Partheno.	50	366	209	8·4
14 May	14	Partheno.	25	360	229	9·8
20 May	15	Partheno.	15	385	241	11·7
7 June	21	Partheno.	10	349	215	8·4
12 June	22	Partheno.	25	360	223	9·4
17 June	18	Partheno.	30	376	214	9·0
25 Sept.	14·5	Partheno.	5	410	230	11·4
30 Oct.	9	Resting	50	342	264	12·5
27 Nov.	7·5	Resting	50	340	259	11·9
4 Dec.	6	Resting	50	319	266	11·8

parent is constant, the size of the eggs, as estimated by the size of the young developing from them, varies inversely as their number. Given the number of eggs to be the same, their size varies as the size of the animal which laid them. These relationships apply only to eggs laid by females of the same age, since it has been shown that the size of eggs and young varies with the age of the mother (Green 1954a, 1956).

During 1963, measurements were made of samples of eggs dissected from the brood pouches of *S. vetulus*, *Scapholeberis mucronata* and *Sida crystallina*. The resting eggs of *Polyphemus pediculus* were also measured; the parthenogenetic eggs of this species were not measured because they receive nutrient material from the mother during the course of development in the brood pouch, so that their size is not relevant in the present context. The results of these measurements are given in Tables 3–6.

Table 5. Polyphemus pediculus: *diameters and volumes of resting eggs, Long Water*, 1963, *and Greenland*, 1961

Date	Water temperature at 09.00 hours (°C)	Mean egg diameter (μ) Greatest	Least	Egg volume (millions μ^3)
13 June	21	303	270	11·5
30 Oct.	9	288	265	10·6
GREENLAND				
29 July	12	293	263	10·6

When the egg volumes were plotted against the sizes of the females carrying them no relation was found, similarly no relation between egg volume and egg number was found. In *Simocephalus*, there is a clear inverse relationship with temperature (Fig. 20), and in the other Daphniid, *Scapholeberis*, there is a suggestion of a similar trend (Fig. 21). In these two species the seasonal change in temperature seems to be the most important factor governing the size of eggs, overruling other intrinsic and extrinsic factors. The

Table 6. *Egg diameters and volumes in the genus* Eurycercus

Locality and date	Species	No. of eggs measured	Mean egg diameter (μ) Greatest	Least	Egg volume (millions μ^3)
GREENLAND					
Godhavn 29 July 1961	*glacialis*	10	479	476	56·8
Kangarssuk 28 July 1961	*glacialis*	11	552	550	87·4
ESTHWAITE 28 July 1953	*lamellatus*	23	316	249	10·3
MOSS ECCLES TARN 28 July 1953	*lamellatus*	8	329	259	11·6

eggs of *Sida crystallina* do not show the same clear relationship (Table 4), and the variation is complicated by the fact that when the temperature falls below 10° C most of the females change to producing resting eggs. The size of the resting eggs of *Polyphemus* does not seem to be influenced by variations in temperature (Table 5).

LATITUDINAL VARIATION IN EGG SIZE

Table 3 shows that the eggs of *Simocephalus vetulus* from Greenland are much larger than those of British specimens, even when the British specimens are reproducing at temperatures lower than the summer temperatures in Greenland. The significance of this presumably genetic difference will be considered in the Discussion. Similarly the

eggs of *Scapholeberis mucronata* in Greenland are larger than those of British specimens, and when specimens from Northern Italy are compared they are found to be smaller than both the British and the Greenland eggs (Fig. 21), though in this case the temperature was higher than that at which this species was found in Britain.

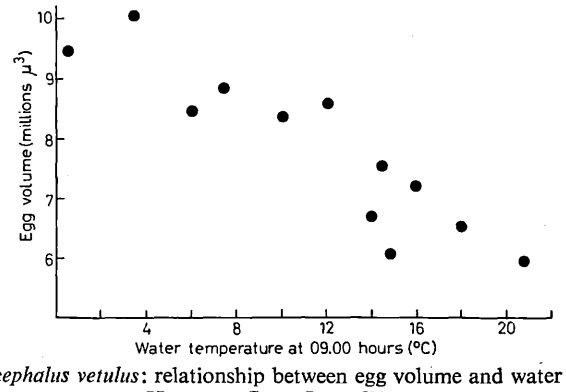

FIG. 20. *Simocephalus vetulus*: relationship between egg volume and water temperature in Hampton Court Long Water.

When different (but closely allied) species from different latitudes are examined there may be even greater differences. I have measured eggs of *Eurycercus glacialis* from Greenland and eggs of *E. lamellatus* from Britain. The eggs of *E. glacialis* are much larger (Table 6).

The resting eggs of *Polyphemus pediculus* from Greenland are similar in size to those of British specimens (Table 5). This is in agreement with the lack of variation in size with temperature in the British specimens.

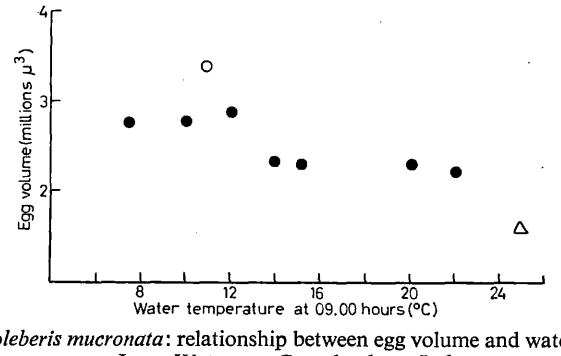

FIG. 21. *Scapholeberis mucronata*: relationship between egg volume and water temperature. ●, Long Water; ○, Greenland; △, Italy.

DISCUSSION

Many species of Cladocera in the Long Water cease reproducing during the winter. This seems to be due largely to changes in temperature (or day length which varies in the same general cycle). The amount of food available, as estimated by the chlorophyll content of the water, does not seem to have much influence at the onset of winter. For instance, the number of reproducing species declined as usual in December 1961 even though the chlorophyll content of the water was very high at that time. This diminution in reproductive activity during the winter reduces the possibility of competition for food at this time

when the rate of replacement of the phytoplankton is low. In the spring and summer the production of phytoplankton increases and the total population of Cladocera which can be supported is correspondingly increased. The improvement in food supply enables the species which have been reproducing throughout the winter to increase their rate of reproduction and enables additional species to become active.

The species which were studied in detail show some clear differences in the nature of their seasonal cycles in the Long Water. They may be divided into three types:

1. In the first group are the species which reproduce through the winter, and show only short periods of bisexual reproduction resulting in the production of few resting eggs. *Simocephalus vetulus* and *Chydorus sphaericus* belong to this group. In some years with mild winters *Simocephalus vetulus* did not produce any resting eggs. *Eurycercus lamellatus* also belongs to this group, although it differs from the other two species in becoming very sparse in the shallow water during the summer.

2. The second group contains those species which overwinter as resting eggs and emerge in March or April. They have a distinct period of bisexual reproduction before the onset of winter. An important point is that the autumn peak of resting egg production dies down well before the species cease reproduction in December, so that there is a short period following the bisexual period when most of the adult females revert to parthenogenesis. The two species studied in detail in this group are *Scapholeberis mucronata* and *Polyphemus pediculus*. The former species was found to be producing small numbers of resting eggs at the end of May or at the beginning of June in four different years, but the percentage of females with resting eggs never rose much above 1 per cent. This early summer bisexual period of *Scapholeberis* was quickly over and all the females reverted to parthenogenesis until September (cf. Fig. 4 in Green 1963). *Polyphemus pediculus* also started to produce resting eggs in June but continued to do so for the rest of the breeding season showing a maximum percentage of females with resting eggs in October (Fig. 6).

3. *Sida crystallina* also overwinters as resting eggs, but differs from the previous groups in that the change-over to bisexual reproduction in the autumn is much more complete and there is no drop in the percentage of females carrying resting eggs before the species becomes inactive. With the almost complete change to production of resting eggs this species persists as large individuals into December and even to the beginning of January.

The results of these studies on the Long Water are difficult to compare with observations from other areas. Most workers have been concerned with the numbers of individuals and have not studied their egg production. Yet egg production is more accurately determined than the size of the population. Throughout this study I have been particularly conscious of the patchy distribution of even the commonest species and the fact that it would be possible to take quantitative samples within a few feet of each other which would give widely different ideas of the density of the population. An important point of interest in the results from the Long Water is that the spring peaks of egg production coincide with the peaks of chlorophyll content of the water. When other workers have studied numbers they find a lag between the maximum phytoplankton and the maximum numbers. There is no disagreement between this and the coincidence of the peaks in the Long Water; clearly the maximum production of eggs will give rise later to a large population.

The data on seasonal occurrence can be compared with those of Berg (1929) from Denmark and Poulsen (1940a, b) from East Greenland. In general it may be said that

fewer species overwinter in an active state in Denmark than in the Long Water. No Cladocera seem to remain active through the winter in East Greenland. In Denmark *Simocephalus vetulus* survives until January, but it was not found in Berg's samples taken in March. In East Greenland *S. vetulus* emerges from its resting eggs in June, and some of the females begin to produce resting eggs while others produce the normal quickly developing eggs. Towards the end of August all the females produce resting eggs and the species then disappears from the samples.

Sida crystallina first becomes active in April or May in Denmark and disappears at the end of November, so that the period of activity is somewhat shorter than that observed in the Long Water. This species does not occur in East Greenland, or even in West Greenland, where conditions are not quite so severe (Røen 1962).

Chydorus sphaericus overwinters in an active state in Denmark, but the production of resting eggs is more pronounced than in the Long Water. Berg found one population which was engaged exclusively in bisexual reproduction. The life cycle of *C. sphaericus* in East Greenland has been described by Poulsen (1940b). The resting eggs hatch in June and July, there then follows a short period of parthenogenesis, but before the end of the month some males appear and the females begin to produce resting eggs. The species becomes scarce at the end of August, but a few individuals may survive until the beginning of November. A further shortening of the annual cycle of *C. sphaericus* is found in Spitzbergen (Olofsson 1918) where the females which emerge from the resting eggs produce a generation of males and females which produce resting eggs, the whole cycle being completed in under 2 months.

If one accepts that the number and size of the eggs of a cladoceran have been subjected to natural selection, then there must be some adaptive significance in the latitudinal differences in egg size described above. A large egg gives rise to a large neonate, and there is a tendency within a species for the larger neonatae to become mature at an earlier instar than smaller neonatae (Green 1954a, 1956). Larger eggs give rise to individuals which become mature after fewer moults. At 22° C neonates of *Daphnia magna* with an intial length of 0·98–1·06 mm became mature in the fifth instar, while those with an initial length of 0·78–0·84 mm became mature in the sixth instar (Green 1954a). Those which matured earlier gained 2 or even 3 days over the later maturers. At lower temperatures the gain in time would be even greater. This may give a selective advantage in situations where a premium is set on rapid maturation. A clear example of this is found in arctic populations which enjoy only a short summer. The eggs of *Simocephalus* are very much larger than those of British specimens (Table 3), and the eggs of *Scapholeberis mucronata* from Greenland are also larger than those from Britain, though not to the same extent as those of *Simocephalus vetulus*. A smaller number of moults before maturity might enable these species to pass through an extra generation during the summer. The same principle applies to the eggs of *Eurycercus glacialis* when compared with those of *E. lamellatus*.

When the population of *Scapholeberis mucronata* from the warm water of the Lago Maggiore is compared with that from the Long Water it seems that natural selection operates in favour of the production of large numbers in warm water. The eggs of the Italian specimens are smaller and the number produced by a female is larger than that produced by a female of the same size from the Long Water. In warm water the time taken to reach maturity is short and the advantage of becoming mature an instar earlier seems to be outweighed by the advantage of producing large numbers. At high temperatures a cladoceran becomes mature at a smaller size than at lower temperatures

(MacArthur & Baillie 1929); this combined with the rapid rate of moulting at high temperatures would compensate for the extra moult which might be necessitated by the small size of the neonatae.

The same arguments can be applied to the seasonal variation in egg size found in the Long Water. In cold water large eggs will ensure the maximum speed of maturation, but in warm water the greater number of smaller eggs will ensure the maximum rate of increase of the population. The actual size of the eggs produced by a cladoceran at a given time will be influenced by the differing advantages of large and small size.

These arguments may not apply to the resting eggs of Cladocera. The temperature at which they are laid does not have any direct relationship with the temperature at which they will develop and hatch. There must still be some balance between the advantages of few large eggs and more numerous smaller eggs, but it is of direct interest in this respect that the resting eggs of *Polyphemus pediculus* did not show any seasonal variation in size in the Long Water, and the eggs measured in Greenland were similar in size to the British eggs. The resting eggs of larger Crustacea, which take more time to reach maturity, may be subjected to selection for large size in Arctic regions. The eggs of the notostracan *Lepidurus arcticus* (Pallas) are much larger than eggs of Notostraca from warmer climates, and the larva emerges at a much later stage of development (Longhurst 1955).

Low temperature is not the only factor which may favour the production of large eggs. It is noticeable that lake dwelling species of *Daphnia* produce fewer but larger eggs than pond dwelling species of the same size (Green 1956). In Lake Ohrid *Daphnia pulex* produces much larger eggs than pond dwelling specimens of the same species in Britain (Green 1964). This tendency to produce large eggs in lakes may have evolved in association with the relatively lower availability of food in lakes compared with ponds and with the greater stability of conditions in lakes, where many Cladocera continue to produce parthenogenetic eggs throughout the year even though they are potentially capable of producing resting eggs (Berg 1931). In Lake Sevan it has been found that when food is scarce the number of eggs produced by *Daphnia* falls to a very low level, but each egg is large. The largest eggs are produced when a single egg is laid in each brood, and the ovaries function alternately (Pyatakov 1956). A young cladoceran will presumably stand a better chance of survival in poor food conditions if it is well supplied with nutriment at the beginning of its life.

SUMMARY

1. The occurrence of thirty-six species of Cladocera in Hampton Court Long Water during the course of four years is recorded. Two of the species, *Scapholeberis aurita* and *Pleuroxus denticulatus*, have been recorded only once previously in Britain.

2. The period of maximum abundance of the oligochaete *Chaetogaster diaphanus* coincided with the maximum abundance of *Chydorus sphaericus* which formed the main food of the oligochaete.

3. *Simocephalus vetulus* normally remained active and reproduced parthenogenetically throughout the winter. This species produced very few resting eggs in the Long Water.

4. *Chydorus sphaericus* also reproduced parthenogenetically throughout the winter and reached a maximum of abundance in the early spring.

5. *Scapholeberis mucronata* and *Polyphemus pediculus* overwintered as resting eggs and emerged to begin parthenogenetic reproduction in March and April. Both species produced large numbers of resting eggs in October and November, but then reverted to parthenogenesis for a short period before dying.

6. *Sida crystallina* overwintered as resting eggs, and emerged in March or April. During October the females changed from parthenogenetic reproduction to the production of resting eggs and might persist until January before the active population died. The period of production of resting eggs was not followed by any reversion to parthenogenesis.

7. Low temperatures (2–7° C) restricted the production of eggs by *Simocephalus vetulus*, even when food was abundant.

8. The low numbers of eggs per female of *S. vetulus* in summer were caused by a combination of factors, but the dominant factor was the small size of adult females at temperatures above 18° C.

9. In *S. vetulus* and *Scapholeberis mucronata* the number of eggs produced by a female was directly proportional to her size. This relation was modified in populations from different latitudes. Arctic populations, from Greenland, produced fewer and larger eggs at a given body size. A population of *S. mucronata* from Italy produced more but smaller eggs than British and Danish specimens of the same size. *Eurycercus lamellatus* in Britain produced smaller eggs than *E. glacialis* in Greenland.

10. The sizes of the parthenogenetic eggs of *Simocephalus vetulus* and *Scapholeberis mucronata* varied seasonally. This variation was governed largely by an inverse relationship with the temperature of the water.

11. The size of a cladoceran egg is partially determined by the differing advantages given at different temperatures. In cold water a large egg ensures that the neonate has a maximum chance of reaching maturity and reaches this state after fewer instars. In warm water the greater number of smaller eggs ensured a greater rate of population growth.

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Variation in carotenoid pigmentation of *Simocephalus vetulus* (Crustacea: Cladocera)

J. GREEN

Westfield College, London

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(With 11 figures in the text)

Simocephalus vetulus (O. F. Müller) is coloured by carotenoids, which may be free in globules in the gut wall and fat body, or associated with proteins in the cytoplasm of the fat cells, carapace epidermis, ovary and eggs. A green carotenoprotein is found in the blood. The transfer of carotenoids from one tissue to another is described; these pigments can be excreted *via* the gut in fully developed embryos. An intermoult cycle, related to egg laying, is shown to cause variation in the pigmentation of the fat cells, carapace and blood.

A seasonal cycle of pigmentation of the fat cells can be related to the reduction of egg laying caused by low temperatures in winter. A female *Simocephalus* may pass half her total carotenoids into her eggs. The total carotenoid content is determined by the amount of pigment obtained from food, and is modified by variations in light and temperature.

Contents

Introduction

A considerable body of information is available concerning the location, formation and functions of haemoglobin in *Daphnia* (Fox, 1948; Fox, Hardcastle & Dresel, 1949; Fox, Gilchrist & Phear, 1951; Fox & Phear, 1953; Fox, 1955; Chandler, 1954; Green, 1955*a*,*b*; Green, 1956*a*; Smaridge, 1956). The haemoglobin of *Simocephalus* has also been studied in some detail (Hoshi, 1957, 1959, 1963*a*,*b*). A smaller body of information is available concerning carotenoids in *Daphnia* (Teissier, 1932; Green, 1957; Thommen & Wackernagel, 1964). The carotenoids of Cladocera other than *Daphnia* have received little or no attention.

The purpose of the present paper is to give an account of the variation in carotenoid pigmentation of a natural population of *Simocephalus vetulus* (O. F. Müller), and to describe a field study of the environmental factors which cause this variation. The complexity of the natural environment is such that the results obtained can only be regarded as indications which will require confirmation by controlled laboratory experiments.

Material and methods

The material used in this study was collected from the Long Water at Hampton Court. A general description of this habitat and a detailed account of the reproductive biology of the Cladocera dwelling therein has been published elsewhere (Green, 1966). Collections were made at approximately weekly intervals during the course of 4 years. All the collections were made at 09.00 h (BST or GMT according to season) and the temperature of the water was measured at the same time.

Samples of 20 or more adult females were taken. The length of each female, the number of eggs or embryos in the brood pouch, the stage of development of these embryos (cf. Green, 1956*b*, 1965, for descriptions of these stages), and the colours of the various tissues were noted. In the figures the results have been expressed as monthly means, so that each point represents 80 or 100 females. The specimens were always examined alive on the day of collection.

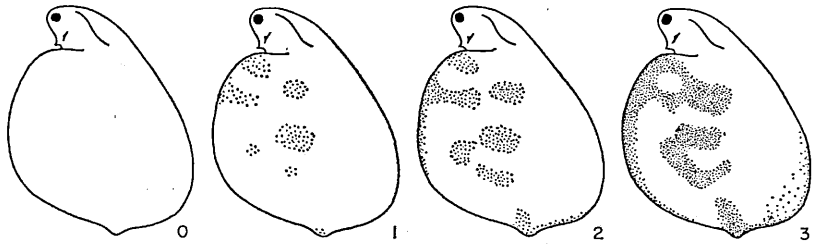

FIG. 1. Lateral views of head and carapace of *Simocephalus vetulus* to show the four categories of extent and intensity of the markings on the carapace.

During 1961 and 1963 a separate sample of water was taken and 100 ml passed through a Millipore DA filter (pore diameter 0·65 μ). The chlorophyll content of the phytoplankton retained by the filter was estimated after extraction with acetone. Absorption of light by the acetone extract was measured at a wavelength of 665 μ in a Unicam SP500 spectrophotometer. Residual absorption and scattering were measured at 750 mμ and deducted from the measurement at 665 mμ. The chlorophyll content was used as an estimate of the food available to *Simocephalus*. This usage gives rise to certain inaccuracies. A low concentration of chlorophyll in summer, when the turnover of phytoplankton may be high, might represent a greater amount of food available than a similar or even slightly greater concentration of chlorophyll in winter, when the rate of turnover would be lower. In spite of such errors the range of chlorophyll concentrations was found to be very great, and the fluctuations do give a rough guide to the amount of food available to *Simocephalus*. A full discussion of the interaction between food, as estimated by chlorophyll, and temperature in determining the reproductive rate of Cladocera is given in Green, (1966).

Changes in the distribution of carotenoids in the tissues of the body were studied by taking random samples of mature females, measuring the diameter of the largest globule in the fat body, and assigning a numerical value to the colour of the cytoplasm of the fat cells and to the globules in these cells. This was done in the simplest possible manner. Cytoplasm which appeared colourless under standard lighting was scored 0; if a pale shade of purple was discernible it was scored as 1; if distinctly purple it was scored 2; and if the purple colour was intense it was scored 3. These estimates are of necessity subjective, but the range of colour is great, and few specimens caused any doubt as to which category they belonged. The same region of the fat body, at the posterior end of the trunk on the ventral side, was examined in each specimen. This was the region in which the fat cells were always conspicuous and fairly constant in number. Similar methods

were used to estimate the orange colour of the globules and the green colour of the blood. This last feature was most clearly visible in the region of the heart, and the colour of the blood was checked by piercing the carapace in this region and noting the colour of the blood as it flowed out. The pigmentation of the carapace was studied by using a similar scale. Four categories based on the extent and intensity of pigmentation were used; these are illustrated in Fig. 1. For all these estimations the maximum possible score for a sample of 20 females is 60. The scale of pigmentation for a sample will thus range from 0 to 60, with the possibility of all intermediates. The monthly means are calculated from several samples of 20 females.

The total carotenoid content of *Simocephalus* was estimated by taking samples of mature females, measuring the length of each, drying the sample on filter paper and then homogenizing in methanol. The volume of methanol was made up to 4·5 ml and then centrifuged to clarify it before examination in the spectrophotometer. Absorption of light by the methanol solution was measured at a wavelength of 470 mμ, and residual absorption and scattering were measured at 600 mμ. The latter reading was deducted from the former to give an estimate of the total carotenoid in the sample. The total was divided by the cube of the mean length of the females in the sample to give an estimate of concentration per unit volume. The concentrations are given in terms of a sample of 30 females.

Results

Distribution of carotenoids in tissues

Free carotenoids are found dissolved in small fat globules in the wall of the gut, and in larger globules in the fat cells which lie on either side of the gut and in the bases of the thoracic limbs. Carotenoproteins are found as brown, green or purple patches in the epidermis of the carapace. The pigment is present in the epidermal cells in the form of granules with a diameter of about 0·5 μ. In the fat cells, the cytoplasm is often coloured homogeneously by a purple carotenoprotein. The ovaries and eggs contain a green carotenoprotein, and a similar colour can be found in the blood. The intensity of the colour in the blood varies during the course of an intermoult in a manner similar to that described for *Daphnia magna* (Green, 1957), indicating that the pigment passes from the blood into the ovary.

In this paper the term carotenoprotein is used in a wide sense, implying an association between a carotenoid and a protein which causes a change in the colour of the carotenoid. No particular type of chemical linkage is implied, although some such form of linkage may be involved. The identification of the colours in *Simocephalus* as carotenoproteins is based on the following observations:

(1) all the colours change to orange when treated with denaturing agents, such as dilute acids, acetone, methanol and ethanol;

(2) concentrated nitric acid produces a strong xanthoproteic reaction when applied to the pigmented areas;

(3) aqueous extracts can be prepared which when cleared by filtration and centrifugation retain the predominant green colour of the eggs and ovary. When such extracts are treated with acetone they change to an orange colour and yield carotenoid pigments with single peaked spectral absorption curves similar to those of the diketo-carotenoids. The centrifuged residue yields an extract which has an absorption curve typical of a mixture of carotenoids;

(4) observation of embryos in the later stages of development shows that the green colour of the fat cells diminishes and the orange colour of the fat globules intensifies. This is in agreement with the breakdown of a water soluble green carotenoprotein liberating a fat soluble carotenoid.

A question which naturally arises is how are these various sites of carotenoid deposition interrelated, and can pigments pass from one to the other?

It is convenient to start investigating this problem with the developing embryo, where the green carotenoprotein and the free carotenoid in the fat globules become restricted to the embryonic fat cells. At the end of embryonic development the link with the protein breaks.

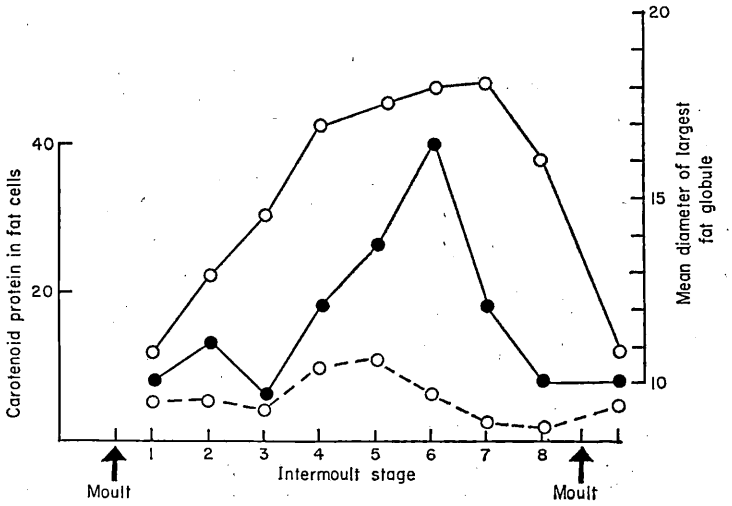

FIG. 2. *Simocephalus vetulus*. Intermoult cycle of pigmentation in fat cells.

—●—●—, Purple pigment in cytoplasm, November 1958; —○— —○—, purple pigment in cytoplasm, July 1960; —○——○—, diameters of largest fat globules, November 1958.

The cytoplasm becomes paler and the colour of the globules becomes intensified. Further, in some embryos when the concentration of carotenoprotein is high, carotenoids are excreted through the gut. The pigment disappears from the fat cells and appears in the gut lumen. Sometimes the concentration in the gut lumen is so high that distinct crystals are formed and their progress down the gut can be followed. In one brood of fully developed embryos at 11.00 h the guts were found to be evenly coloured by orange crystals of carotenoid. At 14.00 h the crystals had moved posteriorly, and at 17.30 h all the gut lumens were clear and without colour. Thus carotenoids can be moved from the fat cells and excreted by the gut.

The early post embryonic stages often have colourless cytoplasm in the fat cells, but as the animals approach maturity this cytoplasm generally becomes purple with carotenoprotein. After eggs have been laid the colour of the fat cell cytoplasm is paler, and the number and size of the globules in the fat cells are both reduced. The intermoult cycle of pigmentation in the fat cells is shown in Fig. 2, where the stages of development of the embryos in the brood pouch are used as markers for stages of the intermoult cycle. There

is a gradual increase in pigmentation after moulting, and a rapid decrease before the next moult and egg laying. This indicates that pigments are transferred from the fat cells to the ovary. The cycle of moulting and egg laying is repeated at intervals of a few days throughout the adult life of *Simocephalus*.

The pigmentation of the carapace also shows variation. When a large sample is examined, a clear effect of size is found. Figure 3(a) shows that the pigmentation of the carapace

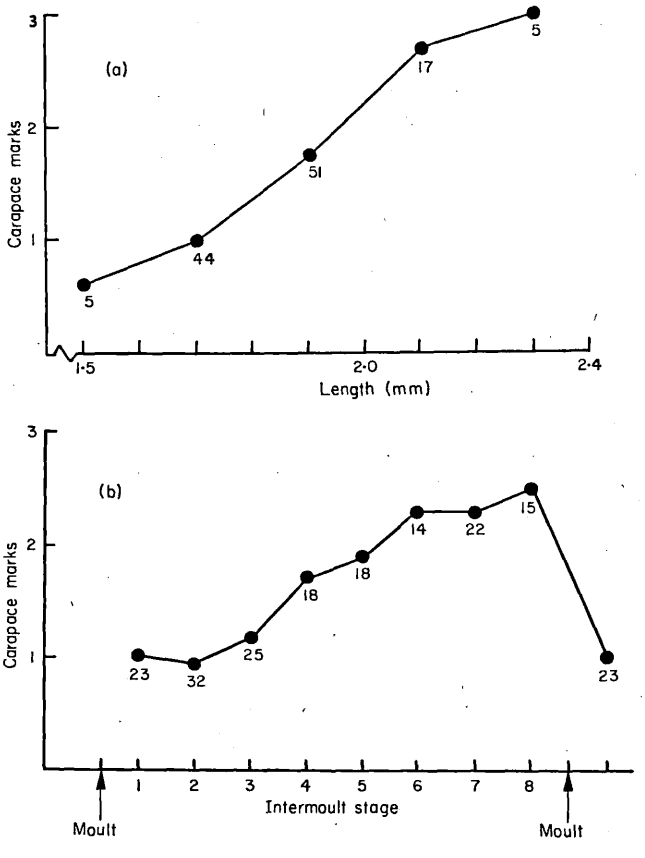

FIG. 3. (a) *Simocephalus vetulus*. Relation between length and intensity of carapace markings, November 1958. The small numbers give the number of females examined for each point.

(b) Intermoult cycle of pigmentation of the carapace. All the females were between 1·6 and 2·0 mm in length. The small numbers give the numbers of females examined for each point.

increases as the animal grows. There is also an intermoult cycle which draws upon the pigment deposited in the carapace in the same way that carotenoproteins are withdrawn from the fat body. This cycle was found when females of a restricted size range were examined. Figure 3(b) shows that the markings on the carapace are faintest when the female has just laid her eggs, and they gradually intensify during the course of the intermoult.

The colour of the blood also varies during the course of an intermoult. Figure 4 shows

that the blood is pale immediately after the eggs have been laid; it then becomes progressively greener, but becomes paler again just before the next eggs are laid. The green carotenoprotein in the blood is presumably in transit between the various tissues. For instance,

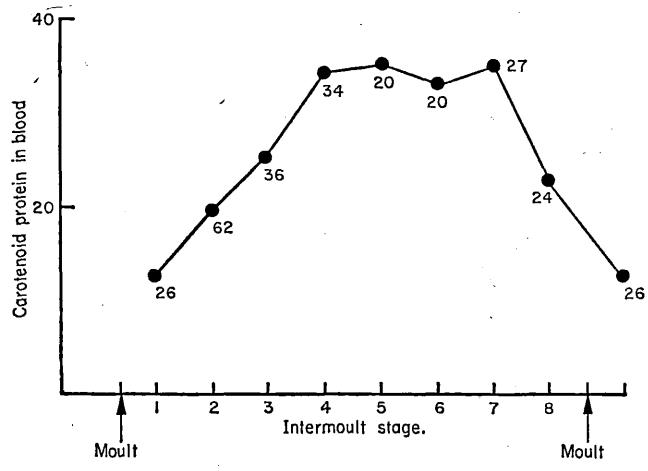

FIG. 4. *Simocephalus vetulus*. Intermoult cycle of pigmentation of the blood. The small numbers give the numbers of females examined for each point.

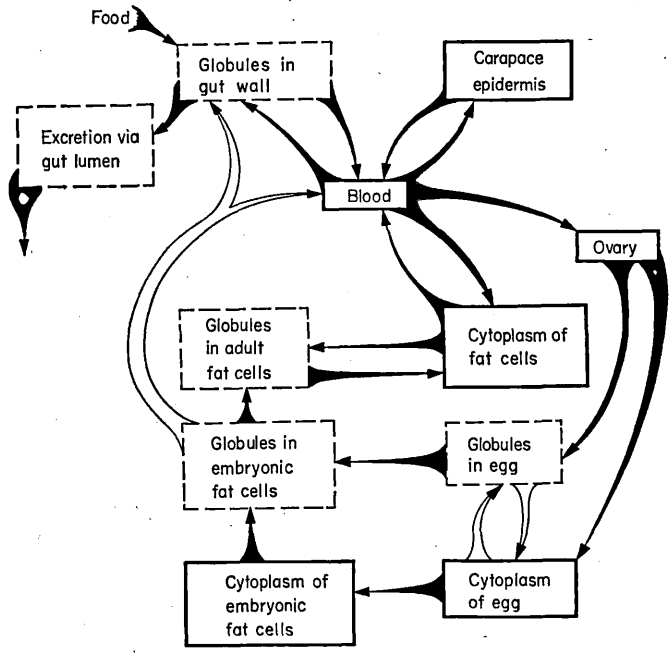

FIG. 5. Pathways of carotenoid transfer in *Simocephalus vetulus*. The black arrows indicate transfer established by direct observation or by analysis of intermoult cycles. The white arrows indicate probable routes of transfer, lacking direct observational data. The broken lines indicate situations in which the carotenoid is free; the whole lines enclose situations in which the carotenoid differs in colour from free carotenoid, and is presumed to be linked to a protein.

the cells of the carapace epidermis have no direct contact with the ovary, and yet the pigmentation of the epidermis shows a cycle which is related to egg laying. Similarly, most of the fat cells have no contact with the ovary, and must use the blood as a means of transporting their pigment to the ovary. In Fig. 5 the routes of transfer from one tissue to another are shown. The blood occupies a central position as an intermediary. There is a possibility that some direct transfer of carotenoid from the fat cells to the wall of the gut may take place. This has been shown in Fig. 5 as a white arrow from the embryonic fat cells because the disappearance of carotenoids from the fat cells coincides with the appearance of carotenoids in the gut wall and lumen, without any noticeable increase in the pigmentation of the blood.

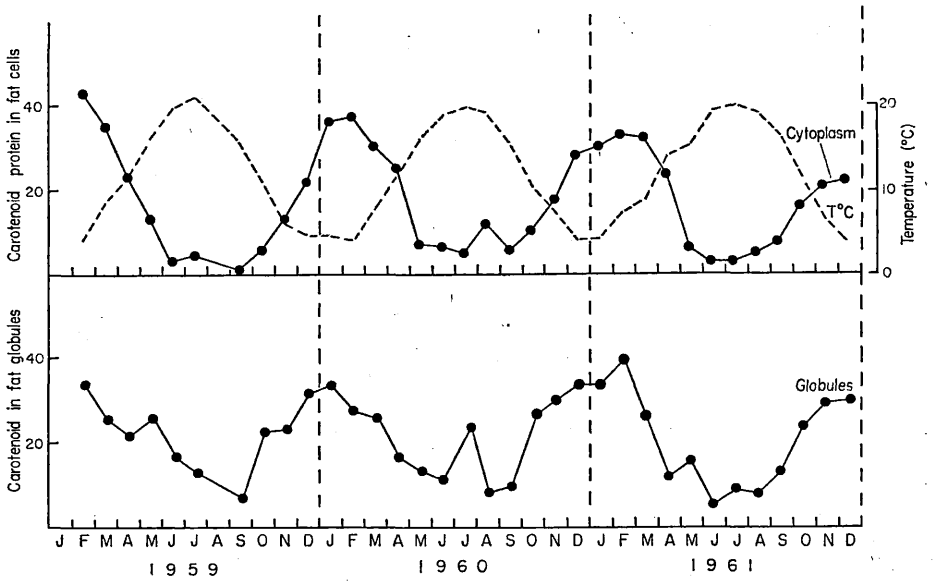

FIG. 6. *Simocephalus vetulus*. Seasonal variation in pigmentation of the fat cells in relation to the temperature of Hampton Court Long Water. Each point is based on several samples of 20 adult females. The vertical scale represents the total score achieved by a sample of 20 females. Means have been calculated on a monthly basis.

Seasonal variation in pigmentation

In addition to the intermoult cycle there is a seasonal cycle of pigmentation. Figure 6 shows the variation in pigmentation of the fat cells in the population of *Simocephalus vetulus* at Hampton Court during the course of three years. There was a general inverse relationship with temperature, but occasional irregularities, such as the increase in pigmentation in July and August 1960, indicate that other factors were also operating.

The seasonal cycle is analysed in more detail in Figs 7 to 14, where the two years 1961 and 1963 are compared. It was a fortunate chance that there was a marked contrast in the cycle of environmental factors in the two years. In 1961 there was a mild spring, and the greater part of the summer was warmer than in 1963 (Fig. 7(a)). The early part of 1963 was very cold, and a thick layer of ice covered the Long Water until the beginning of March. The

chlorophyll content of the phytoplankton showed a different cycle in the two years (Fig. 7(b)). In 1961 there was a peak in the spring, a low level through the summer, and a strong steady rise to a very high level in the autumn. In contrast 1963 showed a high chlorophyll content during the spring, with a fluctuating but generally high level through the summer, falling to a low level at the end of September and remaining low for the last three months of the year.

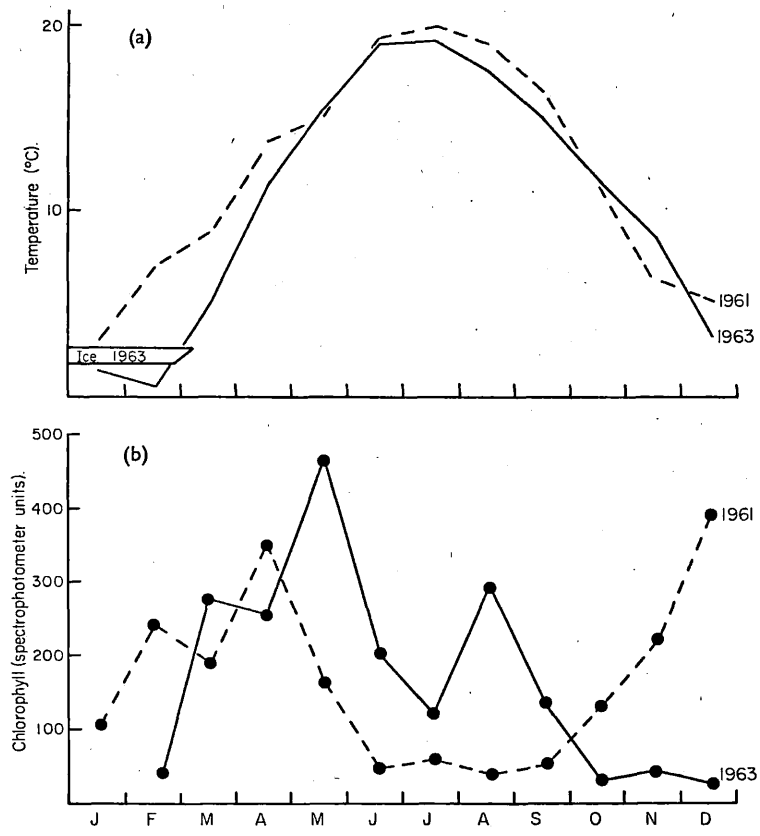

FIG. 7. (a) Seasonal variation in the water temperature at 09.00 h in Hampton Court Long Water during 1961 and 1963.

(b) Seasonal variation in the chlorophyll content of the phytoplankton in Hampton Court Long Water during 1961 and 1963.

The population of *Simocephalus vetulus* became inactive and overwintered as resting eggs during the very cold weather in 1963, and did not become active again until April. The first samples adequate for complete analysis were taken in May. Figure 8 shows that the purple pigment in the fat cells declined as usual during the summer, and increased during the autumn. The important feature of 1963 is this increase in pigmentation during the autumn, even though the chlorophyll content of the phytoplankton was very much lower than in 1961. This indicates that the dominant factor governing the autumn increase

in pigmentation of the fat cells is not the amount of food available, as might be thought if the data for 1961 were the only ones available. The increase in pigmentation of the fat cells can be related in 1963, as in previous years, to decreasing temperatures during the autumn.

The green carotenoprotein in the blood varied in different ways in the two years (Fig. 8(b)). In 1963 there was a general decline in the colour of the blood from May to December,

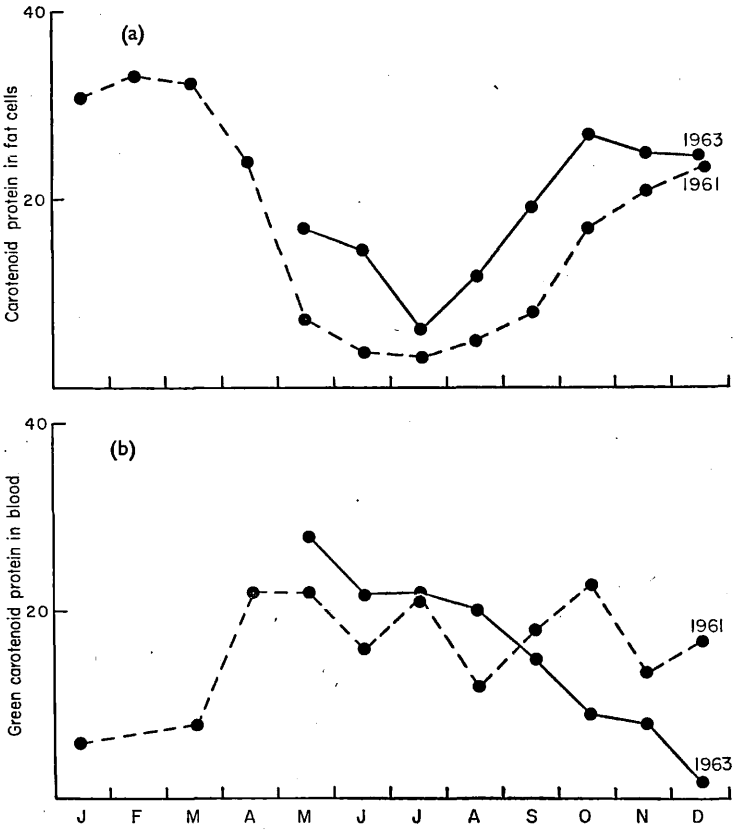

FIG. 8. (a) *Simocephalus vetulus*. Variation in the pigmentation of the fat cell cytoplasm during 1961 and 1963. (b) Variation in pigmentation of the blood during 1961 and 1963.

with a check in the decline in July. In 1961 the blood was pale in January, but became greener in April and May. Thereafter the colour fluctuated for the rest of the year, but did not fall to the low level found at the end of 1963. The higher level of pigmentation of the blood during the period from October to December 1961 compared with 1963 can be related to the higher chlorophyll content of the phytoplankton. Similarly, the high level of pigmentation in May 1963 can be related to the peak of chlorophyll in that month. But the causes of the fluctuations during 1961 are not known.

The size and intensity of the carapace markings also varied during the course of the year (Fig. 9). The general trend was for the markings to be large in the spring, smaller and paler

in the summer, to increase again in the autumn, and finally to become paler and smaller in the winter. Now the intensity of pigmentation of the carapace of *S. vetulus* has been shown to have a strong relation with the size of the animal (Fig. 3), so that the observed cycle of pigmentation may be merely a reflection of a cycle of variation in size. When the sizes of the females are plotted as monthly means (Fig. 10(a)) the general form of the graph shows some similarities to that of the carapace markings. This indicates that fluctuation in size is one of the factors producing the observed fluctuations in pigmentation of the carapace. But there are discrepancies which indicate that other factors also play a part.

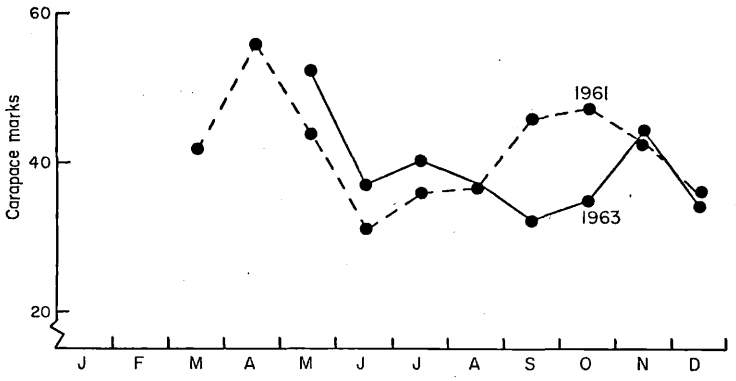

FIG. 9. *Simocephalus vetulus*. Variation in pigmentation of the carapace during 1961 and 1963.

In many members of the Daphniidae the size of the females and the number of eggs produced by them show similar fluctuations (Green, 1955*b*, 1956*b*, 1963, 1966), so that if one uses egg production as an index of nutrition a correction must be made for the size of the females. In Fig. 10(b) a correction has been made by calculating the regression of egg number on size for each sample. It was then possible to calculate egg production at a grand mean size for all the samples. In September and October 1963 the mean size of the females was larger than in the corresponding months of 1961, and yet the corrected mean egg numbers for September and October 1961 were higher. The pigmentation of the carapace followed the corrected egg number, so that in this instance the effect of size was overruled by some other factor, probably related to the quality of food available. The difference in the chlorophyll content of the phytoplankton in the two years indicates that more food was available in the autumn of 1961, but it is not clear why the females did not grow to a larger size.

Estimates of the total carotenoid content of adult females of *Simocephalus vetulus* from the Long Water were made at intervals throughout 1960. Females were selected with recently laid eggs, so that any variation due to different stages of the intermoult cycle were avoided. The results are shown in Fig. 11, together with a series of measurements made in 1963. The mean value in May 1960 was about half the January estimate. The high value in May 1963 can be related to the high peak of chlorophyll at this time, but it is noteworthy that the total carotenoid content did not fall to a very low level in October and November 1963 when there was a great reduction in the chlorophyll content of the phytoplankton.

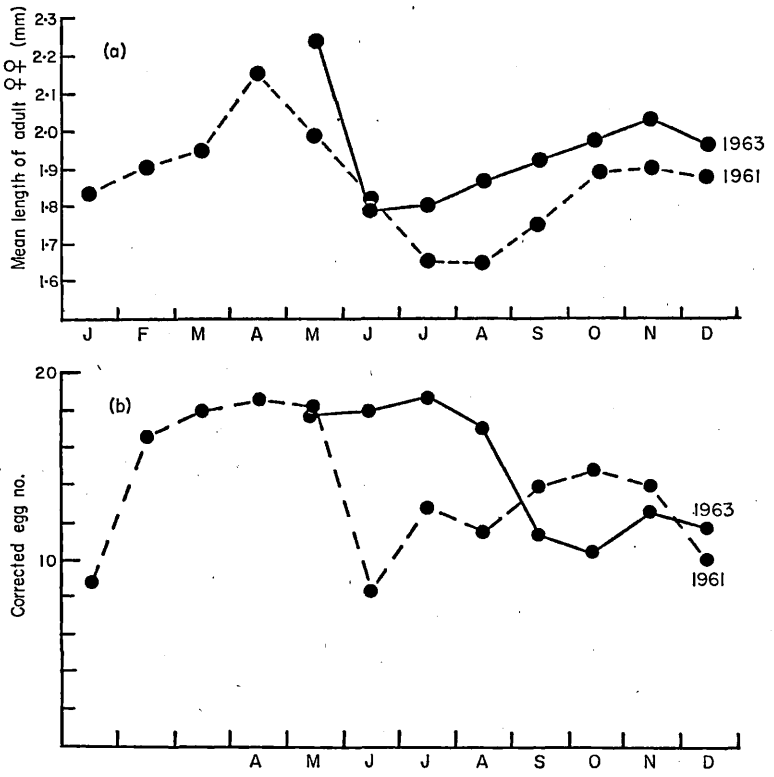

FIG. 10. (a) *Simocephalus vetulus*. Variation in the mean length of adult females during 1961 and 1963. (b) Variation in egg production during 1961 and 1963. The mean egg number per female has been corrected for size, using regression coefficients of egg number on size for each sample.

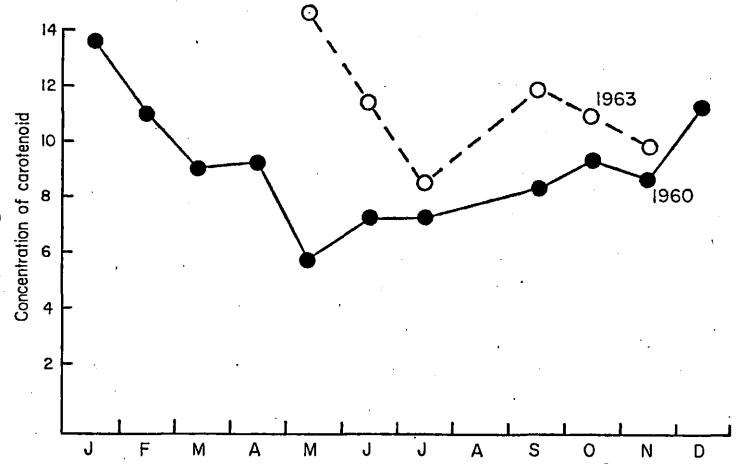

FIG. 11. *Simocephalus vetulus*. Variation in the total carotenoid content per unit volume during 1960 and 1963. See text for method of estimating concentration.

Several estimates were also made to assess the amount of carotenoid passed into the eggs. This was done by comparing the carotenoid content of females with recently laid eggs with the carotenoids in females with fully developed embryos in their brood pouches. The latter females had large ovaries, which would have a carotenoid content equivalent to that of the recently laid eggs in the first group. Any difference between the two groups will give an estimate of the carotenoids in the embryos. Table I shows that females with well developed embryos always have a higher concentration of carotenoids then females with

TABLE I

Total carotenoid content of females of ***Simocephalus vetulus****: concentration estimated as optical density † of a methanol extract from 30 females divided by the cube of the mean length*

Date	Carotenoids in ♀♀ with new eggs	Carotenoids in ♀♀ with fully developed embryos	Percentage difference
5 January	10·1	15·3	52
26 January	15·6	29·2	87
3 March	8·8	12·2	39
31 March	9·0	13·3	48
20 April	8·9	11·0	24
27 April	9·7	12·9	33
3 May	7·1	12·3	73
10 May	6·4	8·1	27
25 May	5·6	9·8	75
14 June	8·3	13·1	58
1 July	7·7	12·2	58
13 July	6·7	10·4	55
13 October	8·9	15·1	71
4 November	8·4	11·3	35

† An optical density of 0·1 represents 100 units

recently laid eggs. The difference, representing the pigment in the embryos, varies between 24 and 87%, with a mean of 52·6% for the 14 comparisons. Thus a female will, at the beginning of each instar, pass about half her total carotenoids into her eggs. The exact amount will vary with factors such as the number of eggs produced and the amount of light that the female receives (cf. Green, 1957).

A detail requiring attention in making a comparison between two groups of females at different stages of the intermoult cycle is the possible presence of epibiotic algae. These become more abundant towards the end of an instar, so that it is always necessary to check that they are not present in any significant quantity before any comparison can be regarded as valid. Throughout the work on *Simocephalus* this factor has been under constant surveillance.

Discussion

The results of the present study indicate that food and temperature can influence the carotenoid content of various tissues in *Simocephalus*. Previous work has shown that light

also exerts an important effect on the carotenoid content of *Daphnia* (Green, 1957). The simple experiment of starving a cladoceran illustrates the basic importance of a supply of carotenoids in the food. When *Simocephalus* is starved all the tissues gradually become depleted of carotenoids. When a cladoceran has an adequate supply of carotenoids in its food the amount found in various tissues will depend on the influence of light and temperature. Different tissues respond differently. The purple carotenoprotein in the fat cells of *Simocephalus vetulus* increased in the autumn both in 1961 and 1963, when the supply of food was quite different in the two years. In contrast the green carotenoprotein in the blood decreased during the autumn of 1963, but not during 1961.

Table I shows that the total carotenoid content of females of *Simocephalus vetulus* varied about three- or fourfold during the course of a year. Now the purple carotenoprotein in the fat cells showed a variation which was at least 20-fold (Fig. 6). This difference between the range of total pigmentation and the range of pigmentation of a particular group of cells indicates that there is a seasonal differential distribution of the pigments. A comparison of Figs 8(a) and 11 shows that the increase of pigmentation of the fat cells in winter is much greater than one would expect from the comparatively small increase in total carotenoid. This increase in pigmentation of the fat cells can be related to decreasing temperatures. The low temperatures in winter reduce the production of eggs, even in the presence of abundant food (Green, 1966), so that less carotenoprotein is likely to pass from the fat cells to the ovary. It is also known that light promotes the deposition of carotenoprotein in the eggs of *Daphnia* (Green, 1957) so that during the autumn it is probable that decreasing length of day will also play a part in reducing the stimulus to pass carotenoprotein into the eggs, and more will be available for retention in the fat cells. If this occurs over several intermoults the result will be an increase in the pigmentation of the fat cells and a gradual increase in the total carotenoids during the winter. A decline in the pigmentation of the fat cells will result when egg production increases in the spring, making greater demands on the reserves held in the fat cells.

Summary

(1) The carotenoid pigmentation of a population of *Simocephalus vetulus* has been studied during the course of four years.

(2) Carotenoids are found either free in fat globules in the gut wall and fat cells, or linked to proteins in the cytoplasm of the fat cells, carapace epidermis, ovary and eggs. The blood contains a green carotenoprotein.

(3) The amount of pigment in the fat cells, blood and carapace varies during the course of an intermoult cycle, and is related to the deposition of pigment in the eggs.

(4) Carotenoids can be excreted *via* the gut in fully developed embryos.

(5) Pigmentation of the carapace epidermis increases with increasing size.

(6) The fat cells show an increase in pigmentation during the winter, when egg production is reduced by low temperatures. A female may pass about half her total carotenoids into her eggs.

(7) Pigmentation of the blood by a green carotenoprotein appears to be promoted by high food levels.

(8) Starvation causes depletion of carotenoids in all tissues.

(9) The total carotenoid content of *S. vetulus* varies during the course of a year. The cycle in each year varies according to the food available and is influenced by the annual cycle of temperature.

Most of this work was done in the Zoology Department of Bedford College, where Professor N. Millott provided me with every facility I required. I am also grateful to Dr B. M. Gilchrist for her critical reading of my first draft of this paper.

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The distribution and variation of *Daphnia lumholtzi* (Crustacea: Cladocera) in relation to fish predation in Lake Albert, East Africa

J. GREEN

Westfield College, Hampstead, London, N.W.3

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(With 4 figures in the text)

In Lake Albert *Daphnia lumholtzi* is found in two forms. One has a pointed anterior prolongation, or helmet, on the head. The other has a shorter rounded head, and was originally described as a separate species, *D. monacha*. The latter form dominates the zooplankton in the middle of the lake where planktivorous fish are rare or absent. The helmeted form becomes commoner near the margins of the lake and reaches its greatest abundance in Ndaiga Lagoon, where planktivorous fish are common. The possession of a helmet is associated with a reduction in the size of the carapace compared to the round headed form. The carapace with its contained eggs is the most conspicuous part of a cladoceran, so that the helmeted forms are at an advantage in the presence of planktivorous fish which locate their prey by sight. The mid-lake *monacha* forms are larger than specimens of the same form in Ndaiga Lagoon, where it is shown that *Alestes baremose* feeds selectively on the larger specimens of the *monacha* form.

The helmeted form produces more, but smaller eggs than the *monacha* form. The total brood volume (= mean egg volume × mean number eggs per female) is greatest in the midlake *monacha* forms. The selective advantages of variations in egg size and the possession of a helmet are discussed. It is concluded that the data from Lake Albert support the hypothesis of Brooks (1965) concerning the adaptive significance of helmet development in *Daphnia*.

Contents

Introduction: Brooks's hypothesis

In the temperate regions there are many populations of lake-dwelling species of *Daphnia* which undergo seasonal variation in body form, known as cyclomorphosis, or seasonal polymorphism. The most striking feature of this variation is the elongation of the head

during the summer. The crest of the head extends forwards as a thin transparent helmet. During the winter the helmet is reduced, and the anterior border of the head assumes a rounded form. These changes occur in populations; no individual of any species of *Daphnia* survives in the plankton for a complete year. The young born in summer have longer heads than those born in winter, and the helmets grow relatively faster in summer.

Research on this phenomenon has centred on two main aspects: the environmental factors determining the change in form, and its adaptive significance. The work of Brooks (1947, 1964), Hrbáček (1959) and Jacobs (1961, 1962) has led to some degree of understanding of the main environmental determinants of cyclomorphosis in temperate lakes. High temperatures and turbulence promote an increase in helmet size. For turbulence to be effective the animals must be exposed to light. The summer environment, with long days and warm turbulent water provides the ideal conditions for helmet growth in species of *Daphnia* inhabiting the epilimnion. During the winter the short days, cold water and reduction in turbulence, particularly when there is ice cover, inhibit the production of helmets.

At the beginning of this century the development of helmets was considered as an adaptation to increase the floating capacity of *Dapnia*. This was thought to compensate for the decreased supporting capacity of the water in summer. Wesenberg-Lund (1900) thought in terms of the decreased specific gravity of the water in summer, and Ostwald (1902) thought in terms of decreased viscosity. Support for these ideas has declined in recent years, and Brooks (1965) has put forward a hypothesis which relates the seasonal variation in helmet size to seasonal variation in predation by planktivorous fish. Briefly, the cornerstones of Brooks's hypothesis are as follows. Predation by planktivorous fish in temperate lakes is known to decrease markedly, or even stop, in winter, and to increase in spring and summer. *Daphnia* is particularly liable to predation by fish, and large forms are more susceptible than small forms (Hrbáček, 1962). The development of a helmet is accompanied by a restriction in the size of the carapace. The helmet is transparent and not easily visible, but the carapace with its contained eggs, is relatively conspicuous. Restriction of the size of the carapace will increase the chances of survival in the face of predation by a predator locating its prey by sight. A small carapace can be achieved in two ways. Dwarfed, slow growing forms without helmets, such as the races of *D. cucullata* Sars described by Hrbáček & Hrbáčková-Esslová (1960), can survive in the presence of planktivorous fish, but suffer the disadvantage of a reduction in food-gathering and reproductive capacities related to their small size. The development of a helmet allows the immature female to assimilate food at a high rate and to grow quickly, but at the same time reduces the liability to predation by being associated with restricted growth of the conspicuous carapace.

The purpose of the present paper is to consider Brooks's hypothesis in relation to the populations of *Daphnia* in Lake Albert, Uganda. This lake has no significant seasonal variation in its high water temperature, and no known seasonal variation in predation by planktivorous fish. But there is variation in the intensity of predation in different parts of the lake.

Material and methods

Plankton samples were taken by means of vertical hauls from the bottom of the lake to the surface, using standard zooplankton and phytoplankton nets supplied by the Freshwater Biological Association. The samples were preserved in 5% formalin. Counts were made using methods already described (Green, 1960). The numbers of the larger species were estimated from the coarse

net samples, and the small forms such as rotifiers and cyclopoids were estimated from the fine net samples. The percentage composition of the zooplankton was estimated from the combined counts of the coarse and fine nets.

Samples from the main stations were taken by Mr M. J. Holden at approximately monthly intervals from November 1961 to October 1962. During September and October 1962 several

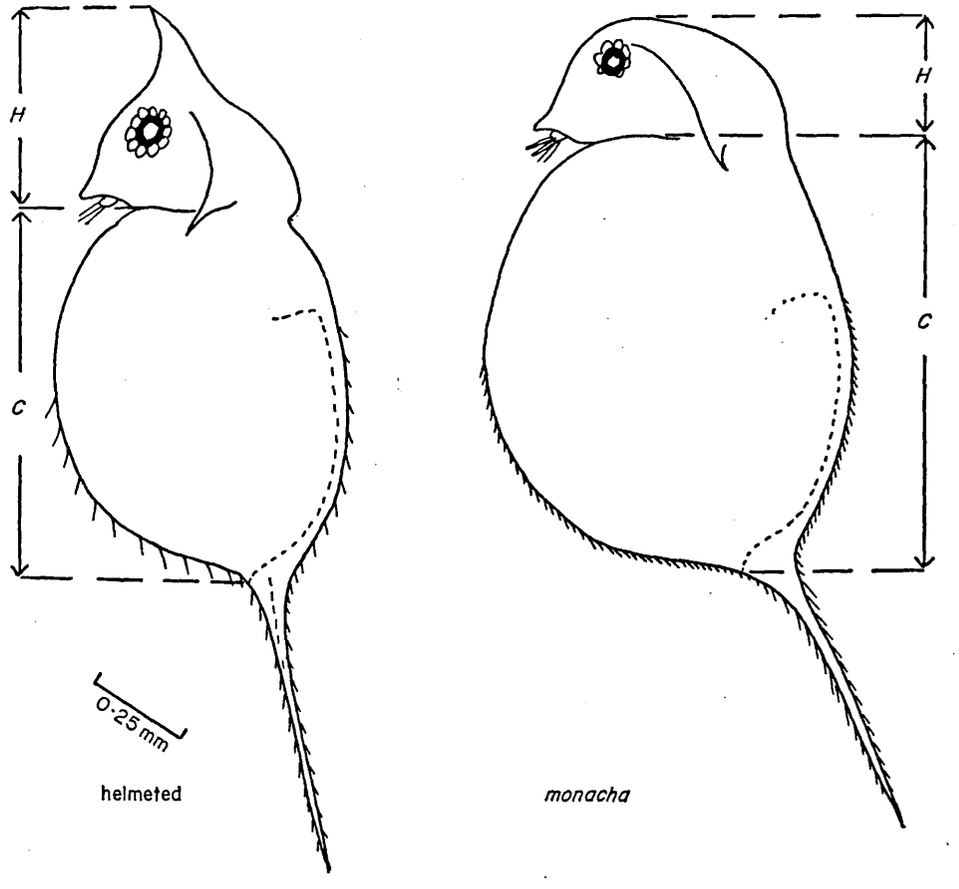

FIG. 1 *Daphnia lumholtzi:* adult females of the helmeted and *monacha* forms to show the limits used in making measurements. Both are drawn to the same scale. *H*, head length; *C*, carapace length.

transects were made of different parts of the lake to study the horizontal distribution of the zooplankton. The samples taken during these transects were taken with coarse nets only. The coarse nets retain all stages of *Daphnia*, but do not retain the smaller cyclopoids and rotifers.

Measurements under the microscope were made with a calibrated eyepiece micrometer, using the limits shown in Fig. 1. When eggs were measured they were covered with water. This supported the eggs and reduced the danger of compression due to their own weight. Two diameters were measured: the greatest and the least. The depth of the egg was assumed to be equal to the least diameter. That this was reasonable assumption was shown by rolling eggs about on the slide and

measuring the least diameter in different positions. The mean egg volumes were calculated from the mean diameters, so that they represent volumes of eggs of mean greatest and least diameters. This method gives a slightly smaller figure than the true mean egg volume, which would be found if the volume of each egg was calculated separately and the mean taken.

The contents of fish stomachs were collected for me by Mr M. J. Holden, who was making a fisheries survey of the lake.

Daphnia lumholtzi Sars and *D. monacha* Brehm

Daphnia lumholtzi was first described by Sars (1885) using individuals hatched from dried mud sent to him from Australia. The species has subsequently been found to be widespread in Asia and Africa.

In 1912 Brehm described *Daphnia monacha* from specimens collected from Lake Albert by the Duke of Mecklenberg's first expedition to Central Africa (1907–8). Further specimens were collected by Dr. R. Lieper in 1907 and were recorded by Cunnington (1915). In a later survey of the recorded fauna of the African Lakes Cunnington (1920) regarded *D. monacha* as endemic to Lake Albert. My own unpublished observations show that the *monacha* form is present in Lake Edward.

Wagler (1936), with remarkable insight, regarded *D. monacha* as being merely a form of *D. lumholtzi*, and Brehm (1959) has accepted this view.

The two extreme forms could easily be taken for separate species (Fig. 1). *Daphnia lumholtzi* typically has a pointed helmet, a long spine at the end of the carapace, long pointed fornices, and spines along the ventral edge of the carapace widely spaced and projecting at a wide angle. The *monacha* form has a rounded head, a relatively shorter tail spine, shorter fornices, and the spines on the ventral margin of the carapace are closer together and do not project at such a wide angle. Intermediates between these two forms have been found at Pakwach and Ndaiga, but in the main body of Lake Albert intermediates were not found. For the sake of convenience in the account which follows the round headed form will be referred to as the *monacha* form, and the form with a pointed helmet will be called the helmeted form.

The distribution of *Daphnia* in Lake Albert

Lake Albert lies in the western branch of the Great Rift Valley. The length of the lake is about 145 km, and the width is a fairly even 40 km, except at the northern end where it narrows to the outflow of the Albert Nile. The main inflow to the body of the lake is the River Semliki, which enters at the southern end. The Victoria Nile, which is a much larger river, enters at the northern end of the lake, but almost immediately turns northwards and flows out again as the Albert Nile. The greatest depth of the lake is about 58 m, and a large area of the lake is over 20 m deep (Fig. 2).

The chemistry of the lake water has been summarized by Talling (1963). The temperature of the water is remarkably constant, between 25 and 29°C throughout the year. Higher temperatures (up to 31°C) have been recorded at the surface on very hot days, but the temperature of the deep water differs very little from that of the water at 5 m below the surface. Thermal and chemical stratifications of short duration occur, and the oxygen content of the lower layers is sometimes seriously depleted, but the water is also mixed frequently by violent storms with strong winds which blow, particularly from the south.

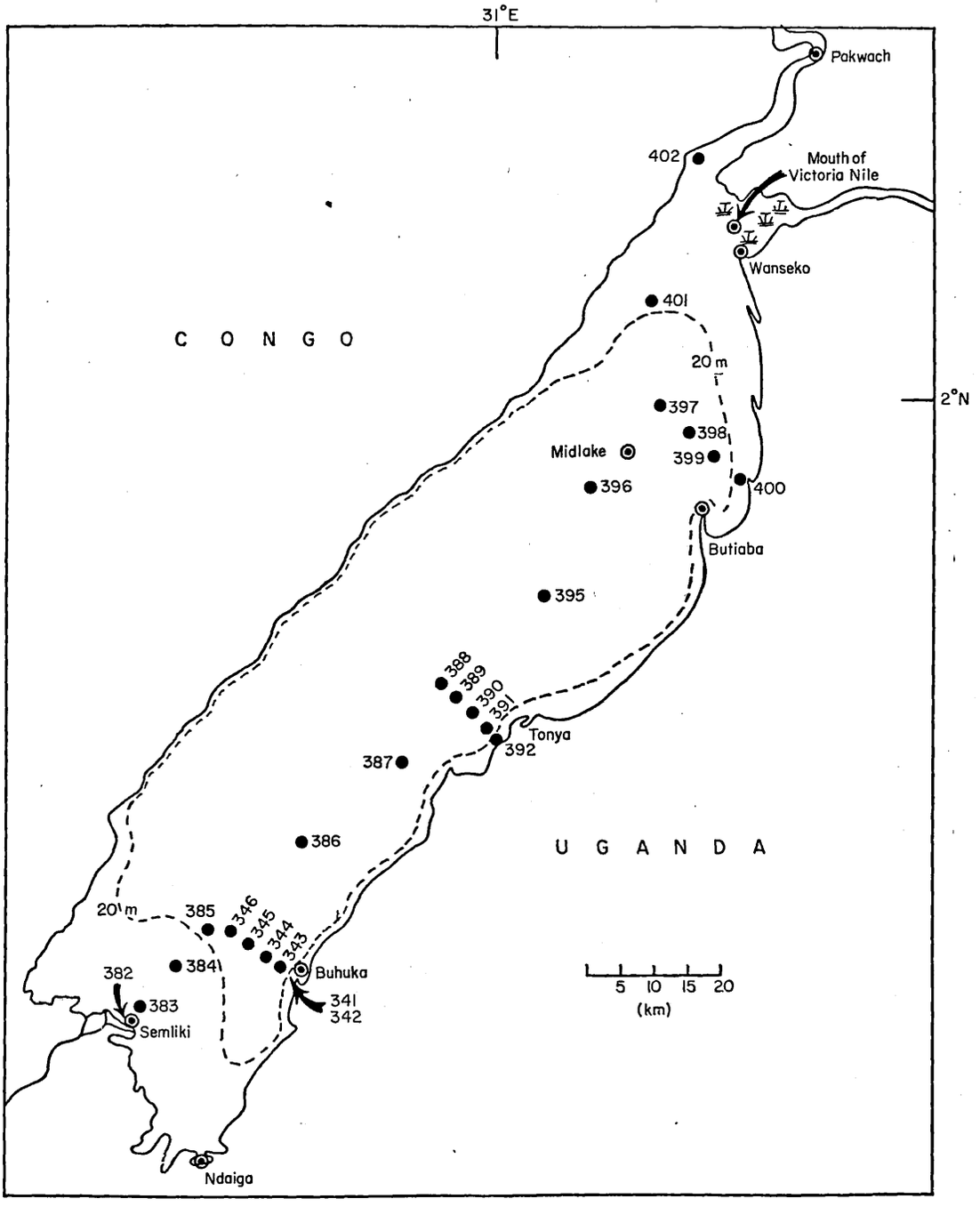

FIG. 2. Lake Albert to show sampling stations. The main stations are shown thus ⊙, and the transect stations are shown by a black dot and a number.

The sampling stations are shown in Fig. 2. In the following account they are dealt with in sequence from north to south. The percentage of the total zooplankton formed by *Daphnia lumholtzi* is given in Table I.

(1) Pakwach. This station lies midstream in the Albert Nile, about 25 km from the region where the river leaves Lake Albert. The depth varied between six and 11 m during the year. Samples were taken with the launch adrift so that it was possible to compensate for the flow of the river and to make the hauls approximately vertical. By the time the

TABLE I

Percentage of the total zooplankton formed by the helmeted and ***monacha*** *forms of* ***Daphnia lumholtzi*** *at six stations on Lake Albert*

Station	Nov.	Dec.	Jan.	Feb.	Mar.	Apr.	May	June	July	Aug.	Sept.	Oct.
Pakwach												
helmeted	0	0	4·8	1·3	3·2	—	5·6	2·8	12·9	4·2	8·7	25·1
monacha	0·01	0	2·2	0·4	1·6	—	1·2	1·4	41·5	5·5	13·1	23·3
Midlake												
helmeted	0	0	0	0·16	0·03	0·05	0	0	0	0·1	0·1	0
monacha	17·1	31·5	24·8	33·6	20·2	6·2	13·3	59·1	38·7	43·3	25·2	37·2
Butiaba												
helmeted	—	0·09	0·11	0·14	0·03	0	0	0·09	0·1	0	0·14	0
monacha	—	2·4	1·6	0	1·1	0·05	2·7	2·4	13·8	3·2	4·7	3·2
Buhuka												
helmeted	0·2	—	0	—	—	—	—	0·4	0·1	0·2	0·1	1·2
monacha	1·2	—	0·1	—	—	—	—	0·1	0·1	1·3	2·4	6·0
Ndaiga												
helmeted	3·1	—	—	—	—	—	2·7	3·1	4·4	23·8	9·2	15·3
monacha	2·9	—	—	—	—	—	0·4	0·4	0·4	0·1	0	2·1
Semliki												
helmeted	0	0	0	0·9	5·1	—	1·8	0·2	0·5	1·9	0·3	0·4
monacha	0	0	0	0·02	0·3	—	0·6	0·1	0·2	0·3	0·1	0

Note. 0 indicates that a sample was taken and no specimens were found, — indicates that no sample was taken.

outflow has reached Pakwach the waters from the Victoria Nile and the main body of the lake have become mixed. The plankton can be expected to be a mixture of shallow water species with deeper water species. Helmeted *Daphnia lumholtzi* varied in abundance, forming between 0 and 25% of the total zooplankton, with a mean of 6·2% for the year. The *monacha* form was slightly more abundant, forming between 0 and 41·5% of the zooplankton, with an annual mean of 8·2%.

(2) Mouth of the Victoria Nile. This station lies in one of the channels between papyrus swamp. The depth varied between 3 and 7 m during the year. The water was usually heavily laden with plant debris, and the strength of the current was such that truly vertical hauls were difficult to make. This combination of factors reduces the counts to little more than rough approximations, but they make an interesting comparison with the other stations. No specimens of *Daphnia* were found in any sample from this station.

(3) Wanseko Lagoon. This lagoon lies just south of the mouth of the Victoria Nile. The depth varied between 3 and 4 m, and samples were taken only during the last six months of the year. A few specimens of the helmeted *Daphnia lumholtzi* were found in the samples taken in May and June, and a single specimen of the *monacha* form was found in the sample taken in June. Otherwise the samples from this station were lacking in *Daphnia*.

(4) Midlake. This station lay approximately 16 km northwest from Butiaba. The depth varied from 45 to 49 m. The helmeted *D. lumholtzi* was absent from seven of the samples, and in the other samples its maximum density was only 0·16% of the zooplankton. In contrast the *monacha* form was present in all the samples, and formed between 6 and 59% of the zooplankton, with an annual mean of 29·2%.

TABLE II

Mean numbers of helmeted and monacha forms of Daphnia lumholtzi at six stations on Lake Albert

Station	No. of monthly samples taken	Mean no. of helmeted forms under 1 sq m	Mean no. of *monacha forms* under 1 sq m
Pakwach	11	7187	7462
Midlake	12	244	127,068
Butiaba	11	15	842
Buhuka	7	277	1303
Ndaiga	7	11,965	665
Semliki	13	2531	373

(5) Butiaba. Worthington (1929) gives a sketch plan of Butiaba Spit which shows a long narrow spit extending nearly 2 km northwards into the lake. During the period when the present series of samples were taken this spit was submerged, but a small part of the northern end projected above the water at the beginning of the year. Samples were taken from the shallow water just beyond the northern end of the spit. The bottom of the lake shelved very steeply, but most of the samples were hauled from a depth of 3 or 4 m. The general characteristics of the plankton from this station are those of a rather poor midlake sample. The helmeted form was much less abundant than the *monacha* form. This resemblance between the midlake samples and the Butiaba samples may be due to the action of the currents which are responsible for the formation of the sand spit.

(6) Buhuka Lagoon. This station lies about 35 km north of the southeastern corner of the lake. The depth varied between 3·5 and 4 m. Samples were taken in January and in each month from June to October 1962. During this period the lagoon was in wide communication with the main body of the lake, and the samples had some of the characteristics of more open water. The helmeted form of *D. lumholtzi* formed between 0 and 1·2% of the zooplankton, while the *monacha* form varied in abundance from 0·1 to 6% of the zooplankton.

(7) Ndaiga Lagoon. This lagoon lies in the southeastern corner of the lake. The depth varied from 5 to 7 m. Samples were taken in November and in each month from May to October 1962. During this period the communication of the lagoon with the lake was more restricted than that of Buhuka Lagoon. Ndaiga was the station where the helmeted form was most consistently present. Between 2·7 and 23·8% of the zooplankton was formed by the helmeted form, and the annual mean number under one square metre was nearly 12,000. The *monacha* form was much less abundant, varying from 0 to 2·7% of the zooplankton.

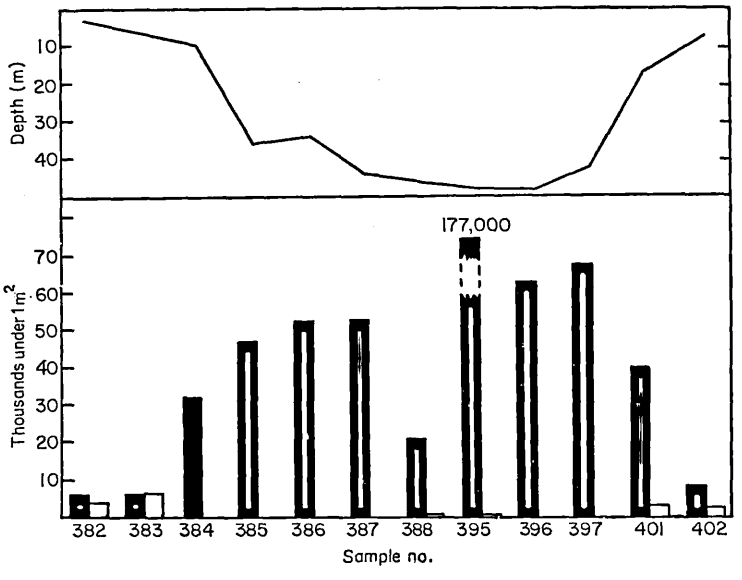

FIG. 3. Distribution of *Daphnia lumholtzi* along the length of Lake Albert. Transect made 2 to 4 October 1962. □, Helmeted; ■, *monachi.*

(8) Mouth of the River Semliki. This station lies at the southern end of the lake where the Semliki flows through a large area of reed swamp. Samples were taken there the water was between 2·5 and 4 m deep. The helmeted form had an annual mean abundance of 0·9% and the *monacha* form was much less abundant (Table II), forming only 0·17% of the annual mean zooplankton.

Distribution of *Daphnia* in transects of the lake

Longitudinal transect

During the period from 2 to 4 October 1962 the whole length of Lake Albert was traversed, and vertical hauls were taken at intervals along the length. The positions of the sampling stations are shown in Fig. 2, and the abundance of the two forms is shown in Fig. 3.

The helmeted form of *D. lumholtzi* was present at the mouth of the Semliki, forming up to 9% of the coarse net zooplankton at the second station which lay a mile away from the first. At the third station, which lay four miles further into the lake, the helmeted form

had disappeared from the samples and did not reappear in any numbers until the depth of the water was less than 20 m at the northern end of the lake.

The *monacha* form was also present at the mouth of the Semliki, but in contrast to the helmeted form it increased greatly in abundance in the deeper water, and in some regions formed over 70% of the zooplankton retained by the coarse nets.

Transects from the shore to midlake (Table III)

Transect out from Buhuka Lagoon, 18 September 1962. The helmeted form was present in small numbers in the samples from the three inshore stations, and was not present in the samples from the three stations nearer the middle of the lake. The *monacha* form was abundant along the whole length of the transect.

TABLE III

*Distribution of helmeted and **monacha** forms of **Daphnia lumholtzi** in transects from the shore to the middle of Lake Albert*

	From Buhuka, 18 September 1962					
Station no.	341	342	343	344	345	346
Depth (m)	6·5	27·0	34·0	33·0	34·5	36·5
Helmeted no.	170	141	778	0	0	0
%	0·6	0·2	1·7	0	0	0
monacha no.	13,857	32,734	16,404	15,555	16,827	17,533
%	51·8	47·5	36·5	56·1	56·6	49·2

	From shore S. of Tonya, 2 October 1962				
Station no.	392	391	390	389	388
Depth (m)	15·0	33·0	41·0	44·0	46·5
Helmeted no.	85	0	283	283	283
%	0·5	0	0·4	0·6	0·7
monacha no.	4412	22,412	29,411	26,725	21,493
%	23·8	36·0	37·7	52·0	50·1

	From shore N. of Butiaba 3 October 1962			
Station no.	400	399	398	397
Depth (m)	7·0	30·0	35·5	42·5
Helmeted no.	566	1980	0	0
%	1·2	0·6	0	0
monacha no.	8484	204,182	108,312	68,720
%	18·6	65·4	55·0	52·6

Note. Numbers under 1 sq m. The percentages are calculated from the zooplankton retained by the coarse nets.

Transect from the shore south of Tonya, 2 October 1962. The helmeted form was present in small numbers in all the samples except the second from the shore. The *monacha* form was abundant at the four outer stations which were all more than 30 m deep, but diminished in abundance at the shallowest station which was 15 m deep.

Transect from the shore north of Butiaba, 3 October 1962. The helmeted form was not present at the two outer stations, but appeared in small numbers in the samples from the

two inner stations. The *monacha* form was very abundant at the three outer stations which were 30 m deep or more, but decreased greatly in abundance at the inshore station which was 7 m deep.

Variation in size of the two forms of *Daphnia lumholtzi*

The *monacha* form in the middle of the lake shows little variation in the length of the carapace of adult females during the course of a year (Fig. 4). The helmeted *D. lumholtzi*

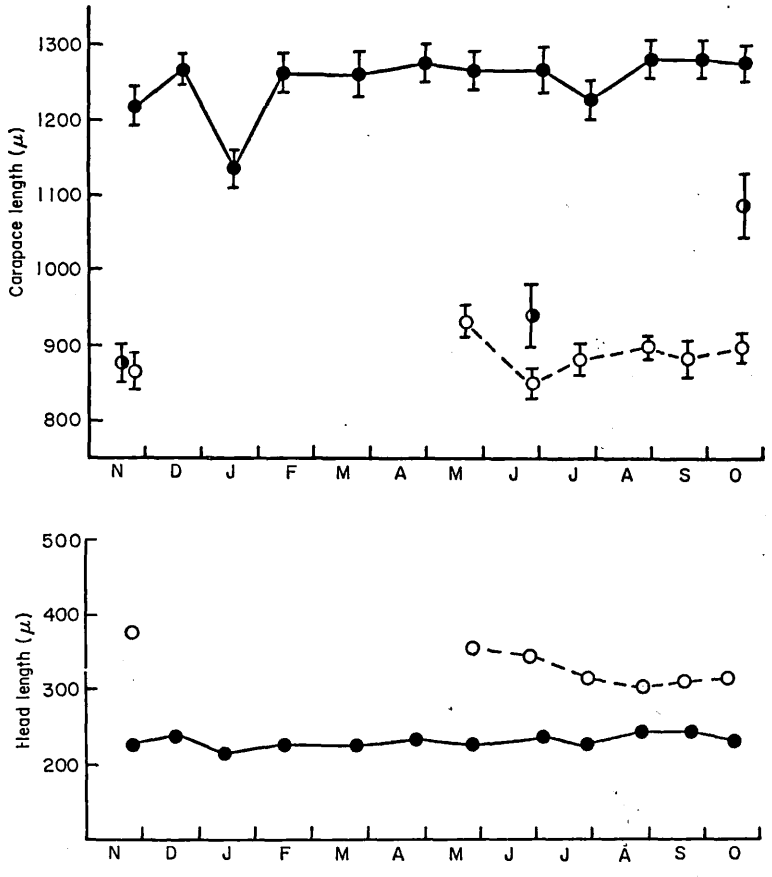

FIG. 4. *Daphnia lumholtzi:* variation in length of the carapace and head. Each point is based on a random sample of 20 adult females. The short vertical lines give one standard error above and below the mean length of the carapace. The standard errors of the head lengths are too small to show in the figure, and the head lengths of the *monacha* form in Ndaiga lie very close to those of the midlake specimens. ●, Midlake *monacha*; ◑, *monacha*—Ndaiga; ○, helmeted—Ndaiga.

from Ndaiga Lagoon had a larger head and a smaller carapace than the midlake *monacha*. The combined head and carapace of the helmeted form was shorter than the combined head and carapace of the midlake *monacha* form.

In Ndaiga Lagoon the *monacha* form was much scarcer than the helmeted form, and the

specimens of *monacha* were often immature. On three occasions it was possible to find sufficient adult females of *monacha* to enable a comparison to be made. In November and June there was no significant difference in the carapace length of the two forms; in October there was a significant difference, but the lagoon specimens of *monacha* were also significantly smaller than the midlake *monacha*.

These results indicate that in Ndaiga Lagoon the adult females of the *monacha* form are smaller than those in midlake, and approach the carapace size of the helmeted forms in the lagoon.

TABLE IV

Size and number of eggs produced by ***Daphnia lumholtzi*** *in Ndaiga Lagoon and midlake Albert. Based on samples taken in June*

Locality	Form	No. of eggs measured	Mean no. eggs per ♀	Volumes in million cu. μ smallest egg	mean egg	largest egg	total brood (mean)
Ndaiga	helmeted	16	3·95	1·59	2·18	2·72	8·61
Ndaiga	*monacha*	10	2·10	3·10	3·94	4·86	8·27
Midlake	*monacha*	20	1·60	3·76	5·64	8·14	9·02

The sample taken from Ndaiga in June provided the opportunity to compare the number and size of the eggs produced by the two forms. The results of this comparison are shown in Table IV. In this table the mean number of eggs per female is based on a sample of 30 females, but only eggs in stage 2 were measured (cf. Green, 1956*b*).

The *monacha* form was producing fewer, but larger eggs than the helmeted form. Each egg of the helmeted form was a little over half the volume of the average egg produced by the *monacha* form in the lagoon. There was no overlap in the sizes of the eggs produced by the two forms. When the total volume of all the eggs in the brood pouch (i.e. the brood volume) was calculated the helmeted form was found to have a slightly greater total than the *monacha* form.

The size of the eggs produced by the helmeted form in June was found to be similar to the size of the eggs produced in other months. The mean volume in June was 2·18 million cubic microns per egg, and the mean volume of a sample which included eggs from several other months was 2·14 million cubic microns.

The eggs produced by the *monacha* form in Ndaiga Lagoon were smaller than those produced by the same form in the middle of the lake. In this case there was some overlap in the sizes of the eggs, but the largest eggs from Ndaiga did not equal the mean size of the eggs from midlake (Table IV).

Distribution of planktivorous fish in Lake Albert

During the period of the present survey the most important planktivorous fish in the lake was *Alestes baremose* (Joannis). This species formed the basis of the main fisheries in shallow water at the two ends of the lake. Verbecke (1959) has shown that this species

feeds mainly on zooplankton, but will also take other small animals such as insects and occasional small molluscs, as noted by Worthington (1929). The distribution of *A. baremose* is curiously restricted to the comparatively shallow areas of the lake; Worthington noted this, and Holden (1963) confirms that this species is rarely found in water deeper than 20 m.

Alestes nurse (Rüppell) is also found in Lake Albert, but it is much less common than its congener. Worthington & Ricardo (1936) found that the stomachs of three specimens from Lake Rudolf contained only zooplankton. The stomachs of four specimens collected from the south end of Lake Albert in November 1961 were found to be packed with the *monacha* form, together with a few specimens of *Caridina nilotica, Ceriodaphnia reticulata*

TABLE V

Stomach contents of ***Engraulicypris bredoi****, from Butiaba and the mouth of the River Semliki*

Food	Butiaba		Semliki	
	total no. in 10 stomachs	percentage of total food	total no. in 10 stomachs	percentage of total food
Crustacea				
Cyclopoid copepodids	353	64·9	203	29·7
Cyclopoid nauplii	161	29·6	223	32·7
Harpacticoida	1	0·2	0	0
Diaphanosoma	7	1·3	2	0·3
Moina dubia	1	0·2	1	0·2
Ceriodaphnia	5	0·9	24	3·5
Daphnia lumholtzi	0	0	1	0·2
Insecta				
Chaoborus larva	2	0·4	0	0
Rotifera				
Brachionus calyciflorus	2	0·4	10	1·5
Brachionus caudatus	7	1·3	2	0·3
Keratella tropica	4	0·8	3	0·5
Pedalia mira	0	0	2	0·3
Lecane bulla	1	0·2	0	0
Algal colonies	0	0	212	31·0

and cyclopoid copepods. These other components formed less than 1% of the stomach contents, while *Daphnia* was the outstandingly abundant food. This species of *Alestes* is also restricted to water less than 20 m in depth (Greenwood, 1958).

Distichodus niloticus (Linn.) is the only other of the 22 species examined from Lake Albert by Verbecke (1959) which he records as being predominantly "zooplanktonophage". Worthington (1929) found that this species fed largely on bottom growing plants, but also noted that four of the 20 specimens examined had eaten considerable quantities of Cladocera. A stomach of this species sent to me by Mr M. J. Holden was found to contain thousands of the *monacha* form compressed into a solid mass together with a few cyclopoid copepods. Although it may be a considerable consumer of *Daphnia* on some occasions, *Distichodus niloticus* does not seem to extend into water more than 10 m deep, and usually remains in shallower water.

Engraulicypris bredoi Poll is a small fish which lives in the surface water close to the shore of Lake Albert. The food of this species has not previously been investigated. I have examined two samples, one from the mouth of the Semliki and the other from Butiaba. As shown in Table V the main food in both localities consisted of cyclopoid copepods and their nauplii, with a colonial alga forming an important component at the mouth of the Semliki. Only a single juvenile *Daphnia lumholtzi* was found in the 20 fish examined. In the main this species seems to exert its greatest effect on the inshore populations of cyclopoids, and to have a negligible effect on the populations of *Daphnia*.

Some of the other fish in Lake Albert, such as *Citharinus citharus* (Geoffr.), also take *Daphnia* as part of their diet, but they are also inshore species and during the period of this survey were much less abundant than *Alestes baremose*. The small tiger fish, *Hydrocyon forskali* Cuvier, also takes *Daphnia*, but only the young fish have this habit; the larger specimens feed on *Caridina* and small fish.

TABLE VI

Stomach contents of ***Alestes baremose*** *captured in Buhuka lagoon, compared to plankton sample from the same region*

Organism	Percentage of total in *Alestes*	Percent ofage total in plankton
Daphnia lumholtzi f. *monacha*	89·0	0·1
Daphnia lumholtzi—helmeted	0	0·4
Ceriodaphnia (2 spp.)	2·5	2·0
Diaphanosoma excisum	0·8	3·7
Moina dubia	0	5·3
Cyclopoid copepodids	7·5	59·9
Cyclopoid nauplii	0	26·3
Caridina nilotica	0·2	0·1
Rotifera	0	2·0

Size of *Daphnia* eaten by *Alestes*

The specimens of *Daphnia* found in the stomachs of *Alestes baremose* and *A. nurse* during this study were all of the *monacha* form. In a sample taken from *A. baremose* in Buhuka Lagoon in June the mean carapace length was $1{\cdot}281 \pm 0{\cdot}026$ mm. A plankton sample was also taken from Buhuka Lagoon in June, and the carapaces of the helmeted and *monacha* forms were measured. The mean carapace length of the adult females of the *monacha* form was $1{\cdot}06 \pm 0{\cdot}024$ mm, and of the helmeted form $0{\cdot}915 \pm 0{\cdot}022$ mm. These means are considerably smaller than the mean length of the specimens actually eaten by *Alestes*, and indicate that predation falls most heavily on the larger specimens.

The selection of the *monacha* form by *Alestes baremose* is illustrated by Table VI, which compares the percentage composition of the stomach contents with the percentage composition of the zooplankton in Buhuka Lagoon.

Discussion

For Brooks's hypothesis to hold the following conditions must be met.

(1) The development of a helmet requires more metabolic effort than the development of a smaller rounded head.

(2) If a helmet is not developed the metabolic effort is transferable to the production of eggs.

According to Brooks's hypothesis the roundheaded forms should be the more successful in the absence of predation. The population in the middle of Lake Albert supports this view. The helmeted form is rare or absent, and the individuals of the *monacha* form are more abundant, and larger than those found near the shore of the lake (Fig. 4 and Table III). The total brood volume per female in midlake is also larger than that produced in Ndaiga Lagoon (Table IV). The scarcity of the helmeted form in midlake is probably due to competitive elimination by the *monacha* form, which does not expend metabolic effort in making a helmet.

Around the margins of the lake, and in the lagoons the *monacha* form is constantly subjected to predation. The helmeted form is more frequent than in the middle of the lake, and is most frequent in lagoons with limited communication with the open waters of the lake. In Ndaiga Lagoon the helmeted form was found to be more abundant than the *monacha* form. In areas where the water is less than 20 m deep the populations of both forms are exposed to the danger of being eaten by planktivorous fish. The examination of fish stomachs revealed that *monacha* was taken as food, whereas the helmeted form was rarely found in fish stomachs. The numbers of the *monacha* form around the edge of the lake have the potentiality of being continually replaced from the thriving population in the middle of the lake. This process of replacement would be aided by the frequent storms and high winds which mix up the waters of the lake. Where this potential replacement is reduced by limited access, as in Ndaiga Lagoon, the helmeted individuals form a higher percentage of the zooplankton.

When the two forms are found together in an area where they are subject to fish predation, as in Ndaiga Lagoon, the size of the carapace tends to be similar (Fig. 4), the carapace of the *monacha* form being smaller than that of the midlake specimens. There is a difference in the sizes of the eggs produced by the two forms, but if the total volume of a brood is calculated it is found that the volume of egg material per female is slightly greater in the helmeted form than in the *monacha* form of a similar carapace length taken from the same lagoon sample. This difference in total brood volume is very small, but it may indicate that the helmeted forms are assimilating more food than the *monacha* form. If the growth rate of the *monacha* form in Ndaiga Lagoon is less than the growth rate of the midlake *monacha*, as seems probable from the smaller size of the adult females, it is likely that egg production would also be reduced. Table IV shows that the total brood volume produced by the midlake females is distinctly higher than that produced by the females in Ndaiga Lagoon. Clonal differences have been found in other species of *Daphnia*. In a comparison of two clones of *Daphnia magna* Straus it was found that the slower growing clone became mature at a smaller size and produced fewer and smaller eggs than the faster growing clone (Green, 1956*a*,*b*).

As predation falls most heavily on the larger individuals there will be selection towards slow growing dwarf forms when planktivorous fish are present (Hrbáček & Hrbáčková-

Esslová, 1960). The *monacha* form in Ndaiga Lagoon can be regarded as dwarfed in comparison with the midlake specimens, and although the total brood volume approaches that of the helmeted form the number of eggs produced by each female is lower.

The difference in the number and size of the eggs produced by the two forms in Ndaiga Lagoon may be explained as follows. A large egg gives rise to a large neonate which becomes mature after fewer moults than a small neonate (Green, 1956*b*). The *monacha* form, by producing larger eggs, may reduce the number of moults before maturity. The helmeted form produces more numerous smaller eggs. Now if the possession of a helmet gives the advantage of enabling a rapid growth rate in the immature phase, as postulated by Brooks, without making the carapace conspicuous, this may compensate for the initial small size of the neonate. The larger number of eggs will then be advantageous in terms of population growth. The balance of advantage will clearly be delicate. That both large and small eggs have advantages is shown by the coexistence of the two forms in certain parts of the lake.

The midlake *monacha* forms produce large eggs in small numbers, but the total brood volume is greater than that produced by lagoon dwelling specimens of both forms. It is probable that the amount of food available to each individual in the dense midlake population is relatively small. In good feeding conditions one would expect a *Daphnia* the size of the midlake *monacha* to produce about 20 eggs in each brood. A similar production of few large eggs in poor feeding conditions has been recorded from Lake Sevan (Pyatakov, 1956).

The overall impression of the intensity of predation on *Daphnia lumholtzi* in Lake Albert is one of moderation. In the face of intense predation *Daphnia* species can be completely eliminated. Brooks & Dodson (1965) have given good examples from lakes near the Connecticut coast, where some of the lakes contain a planktivorous herring-like fish, *Alosa pseudoharengus* (Wilson), which eliminates *Daphnia* from the plankton. Lakes which have contained this fish for a number of years lack *Daphnia* in the plankton, and if the fish is introduced into a lake containing *Daphnia* this cladoceran is rapidly eliminated and replaced by smaller Crustacea.

Intensive predation may well be the reason for the lack of any species of *Daphnia* in the plankton of Lake Tanganyika. This lake is unique in Africa in having two endemic clupeoid fish, *Stolothrissa tanganicae* Boulenger, and *Limnothrissa miodon* Boulenger, which between them occupy the open water of the lake and feed on zooplankton. That the intensity of predation on the midlake zooplankton of Lake Tanganyika is exceptional is indicated by the large size of the shoals of these two clupeoids, which occur in sufficient numbers to form the basis of a large fishery (Collart, 1956; Marlier, 1957).

The results from Lake Albert support Brooks's view that the development of a helmet has survival value in the presence of planktivorous fish, but the picture is complicated by the large midlake population of round headed forms which can replace those removed by planktivores near the edge of the lake.

The existence of round-headed and helmeted forms in a tropical lake raises the problem of the environmental factors which control the development of a helmet. Lake Albert is warm and turbulent throughout the year and perhaps throughout its depth, and it is dfficult to see how the factors which operate in temperate lakes can operate on the two forms in Lake Albert. A possible difference between inshore and offshore water may be caused by drainage from the land, but the nature of the factors which stimulate helmet growth in *Daphnia lumholtzi* are quite unknown.

Summary

(1) *Daphnia lumholtzi* occurs in Lake Albert in two forms. One bears a pointed anterior prolongation of the head and is here referred to as the helmeted form. The other has a rounded head and is here called the *monacha* form.

(2) The *monacha* form dominates the zooplankton in the centre of the lake where planktivorous fish are rare or absent.

(3) In Ndaiga Lagoon, which has limited communication with the open water of the lake, the helmeted form is much more numerous than the *monacha* form. Planktivorous fish are common in the lagoon.

(4) Adult females of the helmeted form have smaller carapaces than the midlake *monacha* forms. The carapace is the most conspicuous part of a cladoceran, so that the helmeted forms are at an advantage in the presence of a predator locating its prey by sight. The *monacha* forms are smaller in Ndaiga Lagoon than in midlake.

(5) The characin fish *Alestes baremose* feeds selectively on the larger specimens of the *monacha* form, so that smaller specimens are at an advantage in the presence of this fish.

(6) The small planktivorous fish *Engraulicypris bredoi* is not an important predator of *Daphnia*, but feeds mainly on cyclopoid copepods and their nauplii.

(7) The eggs produced by the helmeted form are smaller and more numerous than those produced by the *monacha* form. The total brood volume (= mean egg volume × mean number eggs per female) is greatest in the midlake *monacha* and least in the same form in Ndaiga Lagoon.

(8) The selective advantages of variation in egg size and the possession of a helmet are discussed. It is concluded that the data from Lake Albert support the hypothesis of Brooks (1965) concerning the adaptive significance of helmet development in *Daphnia*.

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Associations of Rotifera in the zooplankton of the lake sources of the White Nile

J. GREEN

Zoology Department, Westfield College, University of London

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Plankton samples for rotifers were collected by means of vertical hauls with phytoplankton nets at eight stations on Lake Albert during the course of a year. Similar hauls were taken from Lakes Kyoga, Victoria, George and Edward in October and November. The rotifers found in the samples are listed and some estimates of their abundance and seasonal occurrence in Lake Albert are given. The most important planktonic rotifers are *Keratella tropica* and several species of *Brachionus*. Other species may also become locally or temporarily abundant; *Lecane bulla* becomes numerous in samples taken near vegetation after disturbance by rough weather, or where blue-green algae are abundant.

The associations of rotifers at the stations on Lake Albert and in the other lakes have been compared by means of the Sorensen Index and the index of diversity. The highest diversity is found in situations with a high rate of flow, as at the mouth of the Victoria Nile, where the extra species are non-planktonic forms swept into suspension by the current. This high diversity is associated with low numbers of individuals per unit volume. In Lake Albert rotifers are most consistently present and abundant at the mouth of the River Semliki, where the rate of flow is moderated by a large reed swamp. The middle of Lake Albert is poor in rotifers, and this may be related to the sparseness of planktivorous fish coupled with competitive elimination of the rotifers by larger crustacean zooplankters.

The associations of rotifers in Lakes Kyoga and George are similar, and resemble one another more than they resemble the associations in their neighbouring deeper lakes. The association in Lake Kyoga also resembles the associations found in water of a similar depth at the northern end of Lake Albert.

Contents

Introduction

The White Nile has its origins in five major lakes in Uganda and neighbouring territories. A major outflow leaves Lake Victoria at the Owen Falls and flows into the western side of Lake Kyoga. From Lake Kyoga the river, here called the Victoria Nile, flows westwards

over the Murchison Falls and into the northern end of Lake Albert. This inflow immediately turns northwards and flows out as the Albert Nile. At its southern end Lake Albert receives its major inflow from the Semliki River, which flows out of Lake Edward. The Kazinga channel connects Lake Edward with Lake George. Because of these various interconnections each of the five lakes may lay claim to being a source of the White Nile. A description of the general ecology of Lake Victoria is given by Graham (1929) and the other lakes are described by Worthington (1929, 1932).

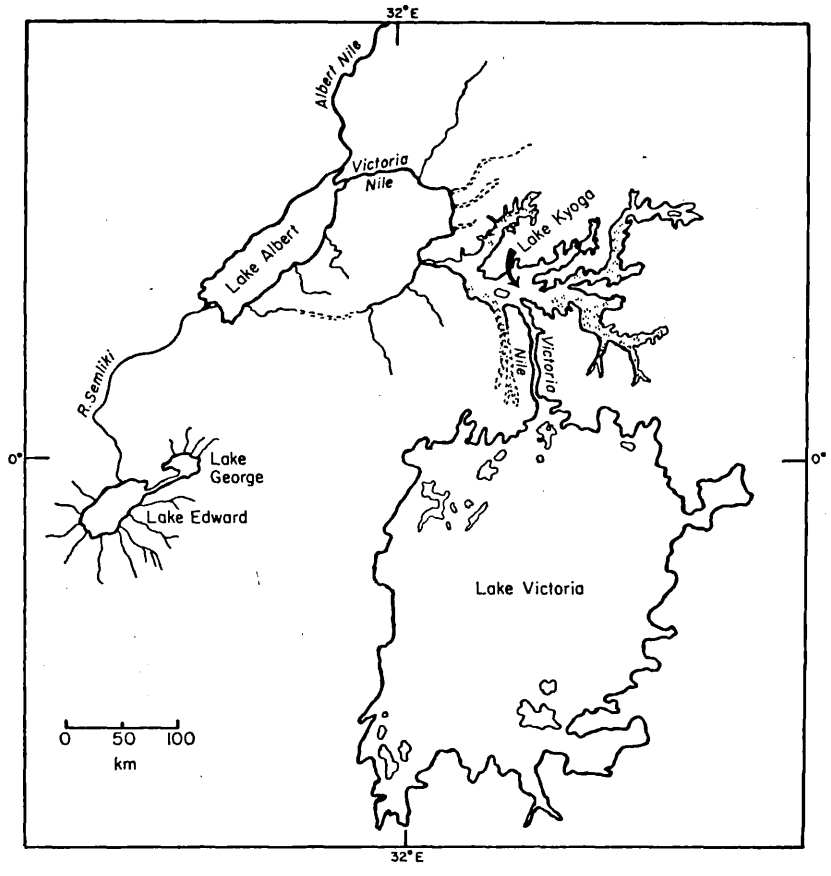

FIG. 1. Map of the lake sources of the White Nile.

In the autumn of 1962 I visited all these lakes and took samples of the zooplankton. In addition I have had access to collections made by Mr M. J. Holden during the course of a year on Lake Albert.

The present paper has three aims. The first is to present a systematic account of the species of Rotifera found in the plankton of these lakes. The second is to describe the seasonal variation in abundance of the rotifers in the plankton of Lake Albert, and the third is to make a quantitative comparison of the associations of rotifers at various stations in Lake Albert and the other four lakes.

Material and methods

Plankton samples for rotifers were taken with standard phytoplankton nets supplied by the Windermere laboratory of the Freshwater Biological Association. Vertical hauls from close to the bottom to the surface were made after the depth had been measured with a weight attached to a wire cable running over a Bergen Nautic metre wheel. On Lake Albert the cable winch was operated from the fisheries launch St Claire. On Lake Kyoga samples were collected using the launch St Peter, on Lake Victoria the launch Darter was used, and on Lake Edward a small police patrol boat was made available. On Lake George a fishing launch was placed at my disposal by TUFMAC Ltd.

The samples were preserved in 5% formalin. The numbers of each species in the samples were estimated by the technique used in the study of the Rotifera of the River Sokoto (Green, 1960). This involved an estimation of the total number of rotifers in each sample, followed by a critical identification of 100 specimens from each to establish the relative abundance of each species. No corrections were made for variations in filtering rate, any such variations were probably very small because the nets never showed signs of clogging except on Lake George where the surface layers contained a dense bloom of blue-green algae.

The vertical hauls were used to estimate the numbers per unit volume and per unit surface area at each station. In the tables the depth given is that measured with the weight on the cable; the depth of the haul was usually 0·5 m less. The numbers under 1 m^2 have been calculated from the total numbers in each haul. The numbers per cu m have been calculated by dividing the numbers under 1 m^2 by the depth of the haul, not by the total depth.

In addition to the vertical quantitative hauls some horizontal and oblique hauls were taken. The purpose of these hauls was to provide larger numbers of rotifers so that the composition of associations could be analysed in greater detail.

Sampling stations

Lake Albert

(1) *Pakwach.* This station lies midstream in the Albert Nile approximately 25 km from the region where the river leaves Lake Albert. The depth varied between 6 and 11 m during the year. Samples were taken with the launch adrift so that it was possible to compensate for the flow of the river and make the hauls approximately vertical.

(2) *Mouth of the Victoria Nile.* This station lies in one of the channels between papyrus swamp. The depth varied between 3 and 7 m during the year. The water was usually heavily laden with plant detritus, and the strength of the current was such that truly vertical hauls were difficult to make. This combination of factors reduces the counts to little more than rough approximations, but they nevertheless make an interesting comparison with the other stations.

(3) *Midlake.* This station lay approximately 16 km NW from Butiaba. The depth varied between 45 and 49 m.

(4) *Butiaba Island.* Worthington (1929) gives a sketch plan of Butiaba Spit which shows a narrow spit extending nearly 2 km northwards into the lake. During the period when the present series of samples were taken this spit was submerged, but a small part of the northern end projected above the water at the beginning of the year. Samples were taken in shallow water just beyond the northern end of the spit. The bottom of the lake shelved very steeply, but most of the samples were hauled from a depth of 3 or 4 m.

(5) *Mouth of the River Semliki.* This station lies at the southern end of the lake where the Semliki flows very slowly through a large area of reed swamp. Samples were taken where the water was between 2·5 and 4 m deep.

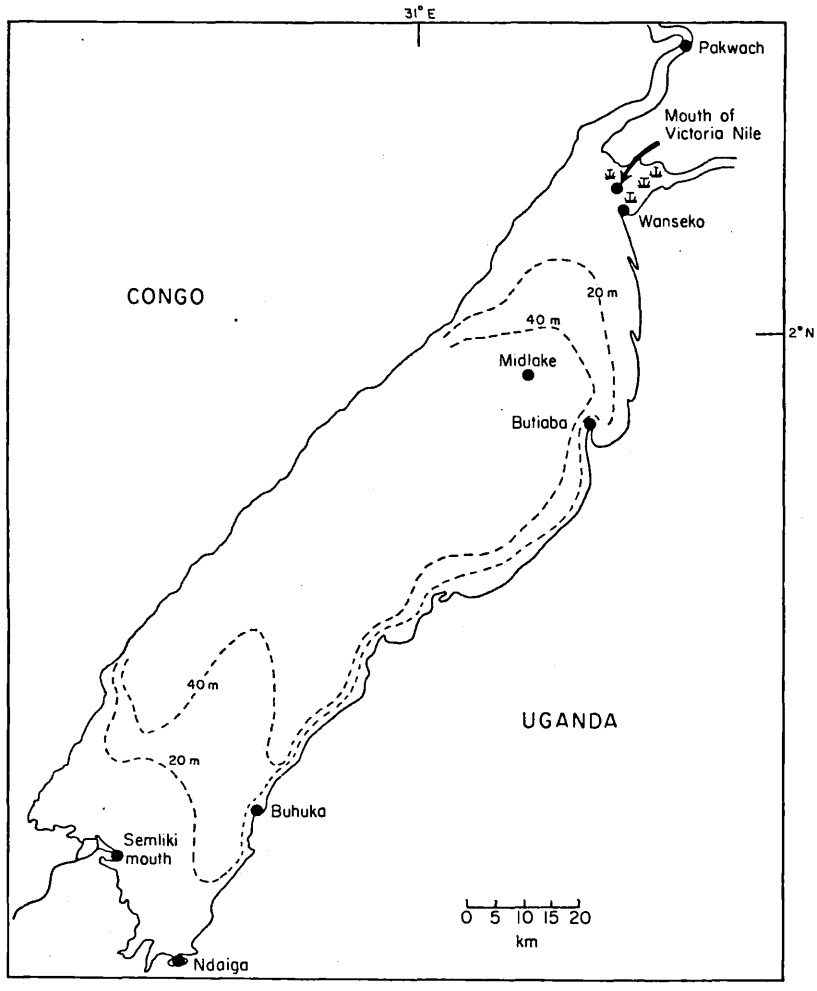

FIG. 2. Map of Lake Albert to show sampling stations. The isobaths at 20 and 40 m are based partly on the map given by Verbeke (1957) and partly on information provided by Mr M. J. Holden.

(6) *Wanseko Lagoon.* This lagoon lies just south of the mouth of the Victoria Nile. The depth varied between 3 and 4 m, and samples were only taken during the last six months of the year.

(7) *Buhuka Lagoon.* This station lies about 35 km north of the southeastern corner of the lake. The depth varied between 3·5 and 4 m. Samples were taken in January and in each month from June to October 1962.

(8) *Ndaiga Lagoon.* This lagoon lies in the southeastern corner of the lake. The depth varied between 5 and 7 m. Samples were taken in each month from May to October 1962.

Lake Kyoga

This lake lies in a large area of swamp to the north of Lake Victoria. The area of open water is subject to variation as floating islands of papyrus move across the lake. Worthington (1929) found that the greater part of the open water did not exceed 3·5 m in depth, and the deepest sounding he made was 5·7 m. During the present survey the depths were generally somewhat greater than those recorded by Worthington, and the deepest open water encountered was 6·25 m at stations 3 and 5 (Fig. 3). Prolonged heavy rain during

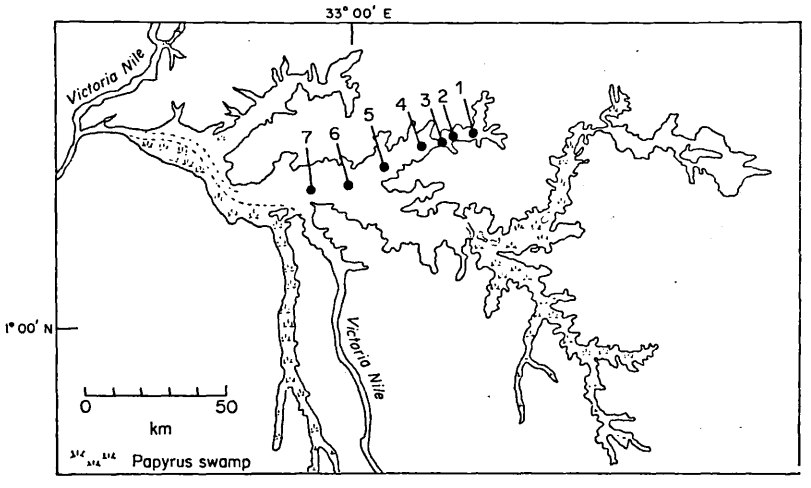

FIG. 3. Map of Lake Kyoga showing the sampling stations.

1961 and 1962 accounts for the raised level of the lake. Seven samples were taken along a line extending from near the inflow of the Victoria Nile (stn 7) past the Bugondo Ferry (stn 3) into the region known as Lake Koweri (stns 1 and 2).

Lake Victoria

The samples taken during the present survey can only be considered as representative of the plankton of a small part of the northern edge of the lake. Three stations, with depths of 15, 19 and 27 m, along a line approximately SE from Entebbe were sampled.

Lake George

This shallow lake is characterized by a dense bloom of blue-green algae lying very close to the surface. The depth is only 2 to 3 m over the greater part of the open water. The northern shore is formed by papyrus swamp which occasionally liberates islands to float

around the lake. A sample was taken in July by Mr M. J. Holden near the mouth of the Kazinga Channel. In November I took three samples along a line from the gap between Kakuranga and Akika islands NE towards the centre of the lake (Fig. 4).

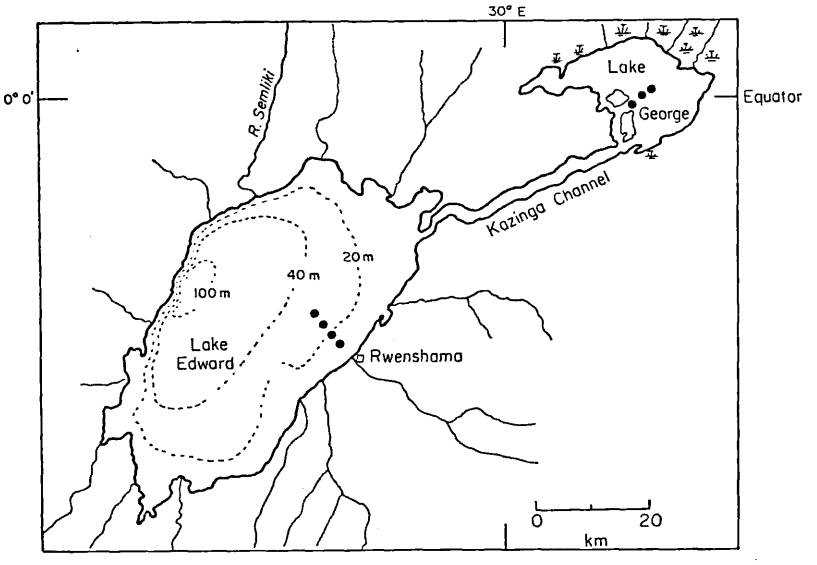

FIG. 4. Map of Lakes Edward and George showing the sampling stations. The isobaths in Lake Edward are based on the map given by Verbeke (1957).

Lake Edward

This lake reaches a greater depth than the others, but it was not possible to sample the deepest water. Four stations with depths of 8·5, 19, 24·5 and 29·5 m were sampled. These stations lay along a line running approximately NW from Rwenshama.

Systematic survey and seasonal occurrence

Order MONOGONONTA
Suborder Ploima
Family Brachionidae

Anuraeopsis fissa Gosse

Gillard (1948) regards *A. congolensis* Evens as a synonym of this species, and this view is adopted here. This is a small species which probably is not retained quantitatively by the net.

Lake Albert

The greatest numbers (1450 under one square metre) were found in the sample from the mouth of the Semliki in February (Fig. 5). There was only one period of relative abundance at the mouth of the Semliki, but at Pakwach low peaks were found in March and September.

The September peak was also reflected in the samples from Wanseko. This species was also present in the sample taken from the mouth of the Victoria Nile in May. No specimens were found in the Midlake samples, or in the samples from Butiaba, Buhuka and Ndaiga.

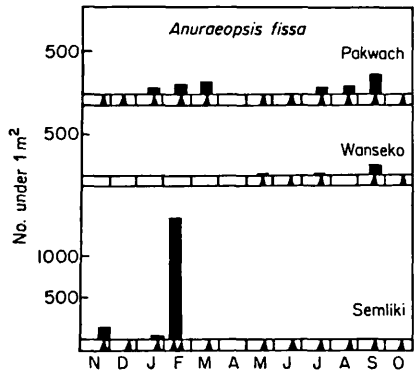

FIG. 5. Seasonal occurrence of *Anuraeopsis fissa* at three stations in Lake Albert. The black triangles indicate when samples were taken.

Lake Kyoga

All the samples along the transect in Lake Kyoga contained some specimens of *A. fissa*, which formed between 1 and 3% of the rotifers in the samples. The highest numbers were found at station 1 (286 under one square metre).

Lake George

A few specimens were found in one of the samples taken from Lake George in November.

Anuraeopsis navicula Rousselet

This is another small species which was much less frequent in the samples than *A. fissa*.

Lake Albert

The specimens from Lake Albert are similar to those from the River Sokoto (Green, 1960). This species was not found in the Midlake samples, or in the samples from Butiaba, Buhuka and Ndaiga, but was fairly common in the July sample from Wanseko and slightly more abundant at the mouth of the Semliki at the end of August, when a population of 144 under one square metre was found.

Brachionus bennini Leissling

Lake Victoria

This species was found in small numbers at two of the three stations on Lake Victoria, but was not found in the samples from the other lakes.

Brachionus budapestinensis Daday

Lake George

A few specimens were found in one of the November samples from Lake George, but none was found in the samples from the other lakes.

Brachionus calyciflorus Pallas

Lake Albert

The seasonal occurrence of this species is shown in Fig. 6. It was present through most of the year at the mouth of the Semliki, but absent in February and July. It was also absent

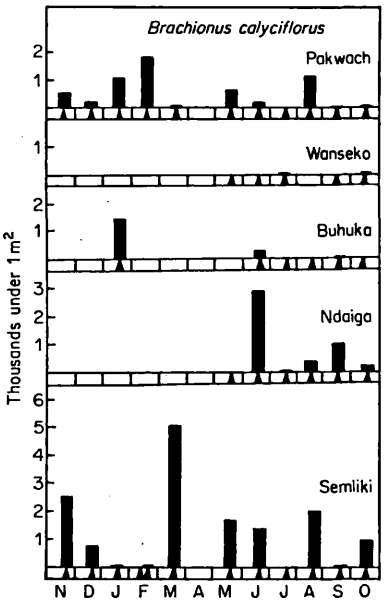

FIG. 6. Seasonal occurrence of *Brachionus calyciflorus* at five stations in Lake Albert. The black triangles indicate when samples were taken.

from the samples taken in July at Pakwach, Wanseko and Buhuka, but present in the sample taken from Ndaiga in that month. The greatest numbers were found at the mouth of the Semliki in March (5110 under one square metre).

Lake Kyoga

This species was present in small numbers in the samples taken from stations 4, 5 and 6; it formed only 1% of the total rotifers.

Lake Victoria

No specimens were found in the present study, but the species has been recorded from Lake Victoria by Daday (1910) under the name *Brachionus pala* Ehrenberg.

Lake George

This species was present in all the samples from Lake George, but did not exceed 4% of the total rotifers. The highest number found was 293 under one square metre.

Lake Edward

No specimens were found in the present study, but the species has been recorded from Lake Edward by de Beauchamp (1939) under the name *B. pala.*

Brachionus caudatus Barrois & Daday

Many of the specimens in this study were small, lacking posterior spines, but as in the study of this species in the River Sokoto (Green, 1960) intermediates were found showing all stages in the formation of posterior spines.

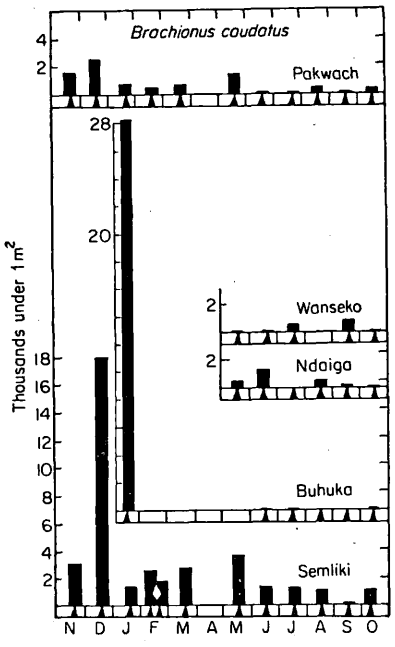

FIG. 7. Seasonal occurrence of *Brachionus caudatus* at five stations in Lake Albert. The black triangles indicate when samples were taken.

Lake Albert

This species formed an important part of the total rotifer population at the mouth of the Semliki, where it formed up to 67% of the total rotifers in January. The numbers found in the samples from the other stations (Fig. 7) were relatively low, apart from the remarkably high number found in the sample from Buhuka in January.

Lake Kyoga

This species formed between 6 and 32% of the total rotifers along the transect of Lake Kyoga. The highest number under one square metre (2498) was found at station 3.

Lake Victoria

This species was very sparse in the samples from Lake Victoria. The highest number under one square metre was 150, and the species did not form more than 4% of the total rotifers.

Lake George

In the samples from Lake George this species formed between 11 and 35% of the total rotifers. It was most abundant (8611 under one square metre) in the sample taken near the mouth of the Kazinga Channel in July.

Lake Edward

The numbers of this species in Lake Edward were very low (74 under one square metre), and it formed only 5% of the total rotifers.

Brachionus falcatus Zacharius

Lake Albert

The seasonal occurrence of this typical warm water species is shown in Fig. 8. There was a distinct peak of abundance at the mouth of the Semliki in December, otherwise the numbers were relatively low. The percentage of the total rotifers formed by this species was usually under 12.

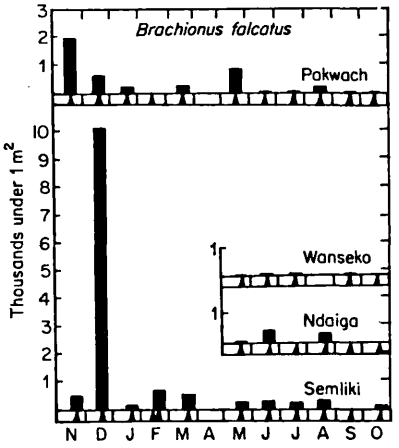

FIG. 8. Seasonal occurrence of *Brachionus falcatus* at four stations in Lake Albert. The black triangles indicate when samples were taken.

Lake Kyoga

This species was present in all the samples. The highest number (1738 under one square metre) was recorded at station 3, where this species formed 16% of the total rotifers.

Lake Victoria

A few specimens were found in one of the samples.

Lake Edward

This species was present in small numbers, forming about 2% of the rotifers in the samples.

Brachionus forficula Wierzejski

Lake Victoria

This species was recorded from Lake Victoria by Daday (1910). In the present series of samples it formed about 3% of the total rotifers. It was not found in the samples from the other lakes.

Brachionus patulus O. F. Müller

In placing this species in the genus *Brachionus* I am following the recent work by Wulfert (1965). This species is not completely planktonic, but spends much of its time among vegetation. This habit accounts for its general scarcity in the samples. In addition to the records given below this species was very abundant in a small roadside pool near Kampala. The specimens from this locality had bright purple ovaries.

Lake Albert

All the samples from Wanseko contained this species. At Pakwach small numbers were found in December, February, March, May and September. At the mouth of the Victoria Nile in May this species formed about 2% of the total rotifers. Only a single specimen was found in the samples from the mouth of the Semliki, in October, and none was found in the samples taken from Ndaiga and Buhuka.

Lake Kyoga

This species was found at five of the seven stations on Lake Kyoga. The highest numbers were found at station 2 (428 under one square metre), where it formed about 5% of the rotifers in the sample.

Lake Victoria

Although not found in the present study this species has been recorded from Lake Victoria by Daday (1910) under the name *Noteus militaris* (Ehrenberg).

Brachionus quadridentatus Hermann

Lake Albert

Small numbers of this species were found in all the samples from Pakwach except that taken in December. Similar numbers were taken from Wanseko, but none were found in samples from other parts of the lake.

Lake Kyoga

This species was found at six of the seven stations on Lake Kyoga. It was most abundant at station 6 where it reached a density of 1974 under one square metre, and formed 21% of the rotifers in the sample.

Lake Victoria

This species was present in the samples from two of the three stations on Lake Victoria. The highest number was 238 under one square metre.

Lake George

The sample taken in July did not yield any specimens, but all the samples taken in November contained this species. The highest number under one square metre was 515, when this species formed 7% of the rotifers.

Lake Edward

A few specimens were found in one of the samples. de Beauchamp (1939) recorded this species from Lake Edward under the name *B. bakeri* O. F. Müller.

Brachionus rubens Ehrenberg

Lake Albert

This species was not common in the samples, and never formed more than 2% of the total rotifers. Small numbers were found at Pakwach in September, at Wanseko in July, and at the mouth of the Semliki in March and October. The last occurrence was the most abundant, but even this only reached a density of 228 under one square metre.

Brachionus rubens is normally epibiotic on Cladocera, and during the present study it was found attached to *Moina dubia* De Guerne & Richard. There are also records in the literature of this species attaching itself to aquatic Hemiptera (Bartoš, 1947) and to mosquito larvae (Russell, 1957).

This species was not found in the samples from the other lakes.

Brachionus urceolaris O. F. Müller

Lake Kyoga

A few specimens were found at station 2, but not elsewhere.

Colurella uncinata O. F. Müller

Lake Albert

Small numbers were found in the samples from Wanseko in May, June and July.

Dipleuchlanis propatula Gosse

Lake Albert

All the samples from Wanseko contained small numbers of this species, but it was not found at the other stations. The maximum number under one square metre was 53 in July.

Diplois daviesiae Gosse

Lake Albert

This species formed about 7% of the rotifers in the sample taken from Wanseko in October, but it was not found in any other sample.

Epiphanes macroura Barrios & Daday

The specimens agree with the figures given by de Beauchamp (1932) of *Notops mollis* Hempel, which is a synonym of *E. macroura*.

Lake Albert

This species was never abundant in the samples, but it was most frequent at the mouth of the Semliki, where the maximum number under one square metre was 468 in June. At Pakwach and Wanseko the numbers did not exceed 84 under one square metre. No specimens were found in samples from Midlake, Butiaba, Buhuka and Ndaiga.

Lake Kyoga

Specimens were found in the samples from stations 5 and 6. At the former station the population reached a density of 376 under one square metre and formed 4% of the rotifers in the sample.

Lake Victoria

This species was recorded from Lake Victoria by Daday (1910), but was not found in the present series of samples.

Lake George

No specimens were found in the samples taken in July, but in November this species was present in the samples taken from two of the stations. This highest number under one square metre was 573 when this species formed 6% of the total rotifers.

Lake Edward

de Beauchamp (1932) recorded this species from Lake Edward under the name *Notops mollis*, but no specimens were found in the present samples.

Euchlanis dilatata Ehrenberg

Lake Victoria

In the sample taken from the deepest station this species formed 14% of the total rotifers, and numbered 416 under one square metre. It was slightly less abundant at the other two stations.

Keratella tropica Apstein

Lake Albert

This is the most regularly occurring of all the rotifer species in the plankton of Lake Albert. It is the only species which becomes moderately abundant in the Midlake samples, and it forms a high percentage of the total rotifers at all the stations except Wanseko. Even at Wanseko it formed 20% of the rotifers in the September samples. At the mouth of the Semliki it sometimes formed over 70% of the rotifers, and in Buhuka lagoon it formed 91% of the rotifers in the samples taken in August. The seasonal occurrence of this species is shown in Fig. 9.

Lake Kyoga

This species formed between 8 and 24% of the rotifers along the transect of Lake Kyoga. The highest number under one square metre was 2142 at station 1.

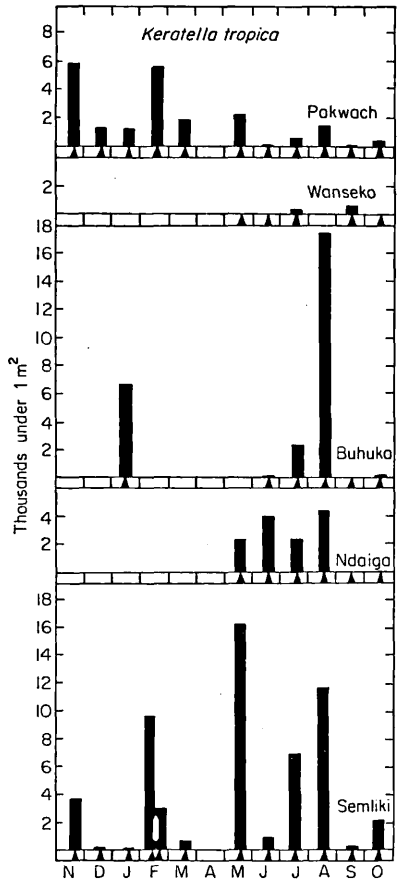

FIG. 9. Seasonal occurrence of *Keratella tropica* at five stations in Lake Albert. The black triangles indicate when samples were taken.

Lake Victoria

This species was not abundant in the samples from Lake Victoria; the highest number under one square metre (602) was recorded where the depth was 19 m.

Lake George

In the sample taken from the mouth of the Kazinga Channel in July this species reached a density of 7382 under one square metre, and formed 30% of the rotifers. In the November samples it was not so abundant: the numbers varied from 543 to 2444 under one square metre, and the species formed between 8 and 27% of the total rotifers.

Lake Edward

Although not abundant (only 441 under one square metre) this species formed about 30% of the rotifers in the samples from Lake Edward.

Keratella lenzi Hauer

This species has been recorded once previously in Africa, from the Transvaal, under the name *Keratella valga* f. *aspina* by Edmondson & Hutchinson (1934). This identification was corrected by Berzins (1955) whose diagnosis is followed here.

Lake Albert

A few specimens were found in the samples from Wanseko in September and from Pakwach in January and February.

Lake Kyoga

A few specimens were found in the sample taken from station 1.

Lake Victoria

Small numbers were found in the samples from the shallowest and the deepest stations.

Keratella cochlearis Gosse

Lake Albert

This species was very rare in the samples from Pakwach, and was found only in November and June. The length of the lorica, excluding anterior and posterior spines, varied between 93 and 97 μ and the posterior spine varied between 18 and 25 μ in length. No specimens were found in the samples from the other stations.

Lake Victoria

This species was less abundant than *K. tropica*, the highest number under one square metre was 475 at the shallowest station. The length of the lorica varied from 90 to 94 μ and the length of the posterior spine varied from 19 to 35 μ.

Lepadella patella O. F. Müller

Lake Albert

Single specimens were found in the samples from Pakwach in December, Wanseko in May, and the mouth of the Semliki in September.

Lepadella sp.

Lake George

Small numbers of this species were found in the samples taken from Lake George in July and November. The total length of the lorica was 81 μ and the toes were equal in length (16 μ) and inserted side by side. The foot had three segments, each slightly longer than wide. This is very similar in general form to *L. patella*, and it may be a small form of that species.

Manfredium eudactylotum Gosse

In using this name I am following Gallagher (1957) and Edmondson (1959).

Lake Albert

This species was found only in the samples from Wanseko and the mouth of the Victoria Nile. It never formed more than 2% of the rotifers in the samples.

Mytilina ventralis Ehrenberg

Lake Albert

A single specimen was found in the sample from the mouth of the Semliki in October, and another was found in the sample from Wanseko in July.

Lake Victoria

A single specimen was found in the sample from the shallowest station.

Platyias leloupi Gillard

In the original description this species was regarded as a variety of *P. quadricornis*, but Wulfert (1965) has given it full specific rank.

Lake Albert

This species was present in all the samples from Wanseko, where it was about twice as abundant as *P. quadricornis*. It was not found in the samples from the other stations.

Platyias quadricornis Ehrenberg

Lake Albert

This species was also present in all the samples from Wanseko, but was found in only one sample from Pakwach (in May). It was also present in the same month in the samples from the mouth of the Victoria Nile, but was not found in the samples from the other stations.

Trichotria tetractis Ehrenberg

Lake Albert

A single specimen was found in the sample from Wanseko in June.

Family Lecanidae

Lecane bulla Gosse

Lake Albert

This is the most abundant species of the genus in Lake Albert. It is only partly a planktonic species, and its occurrence in the samples is often associated with disturbance of water near vegetation. This aspect of its occurrence is most strikingly shown in the samples taken from the mouth of the Semliki in February. Samples were taken on the 19 and 20 February. On the first day there was a strong northerly wind blowing throughout the day so that the water at the mouth of the Semliki was subjected to considerable agitation. In

the samples collected on the 19 *Lecane bulla* was the most abundant rotifer (14,993 under one square metre), forming 31% of the total rotifers. In the samples collected on the following day, which was calm, *L. bulla* formed only 2% of the total rotifers, and five other species were more abundant. This species was consistently present in the samples collected at Wanseko, and was frequently the dominant rotifer, forming up to 35% of the total rotifers. At Pakwach its occurrence was erratic, and it never formed more than 5% of the rotifers in the samples. At Buhuka and Ndaiga the occurrence of this species was rare, and it never formed more than 2% of the rotifers. A few specimens were found in the samples from Butiaba, but none was found in the samples from Midlake.

Lake Kyoga

This was one of the most abundant rotifers at all the stations on Lake Kyoga. The highest number under one square metre was 4713 at station 1, where this species formed 33% of the rotifers in the samples.

Lake Victoria

The highest number under one square metre was 1188 at station 3 where this species formed 40% of the rotifers in the sample. The number under one square metre was similar at station 1, but here this species formed 18% of the total rotifers. This species has previously been recorded from Lake Victoria by Daday (1910) under the name *Monostyla bulla.*

Lake George

The species was abundant in all the samples from Lake George, and was most abundant (5905 under one square metre) in the sample taken near the mouth of the Kazinga channel in July.

Lake Edward

This species was not found in the present series of samples from Lake Edward, but has been recorded in this lake by de Beauchamp (1939).

Lecane cf. acronycha Harring & Myers

Lake Albert

A single specimen was found in the sample taken from the mouth of the Victoria Nile in May. The specimen was smaller than those described by Harring & Myers (1926), but was otherwise very similar. The total length of the lorica was 130 μ, and each toe was 62 μ long, including a claw with a length of 7 μ.

Lecane aculeata Jakubski

Lake Kyoga

A single specimen was found in the sample taken from station 5.

Lecane cornuta O. F. Müller

Lake Albert

A few specimens were found in the samples taken from the mouth of the Victoria Nile in May and from Ndaiga lagoon in September.

Lecane curvicornis Murray

Lake Albert

This species was most frequently found at the mouth of the Semliki, where the annual mean per cubic metre was 454, and on 19 February 1962 it formed 27% of the rotifers in the sample. On the following day the percentage had fallen to five, so that the great abundance of this species was related to the disturbed conditions associated with a strong wind blowing from the North (cf. *L. bulla*).

Lecane decipiens Murray

Lake Albert

This species formed 2% of the total rotifers in the sample taken from Wanseko in May, and a single specimen was found in the sample taken from the mouth of the Victoria Nile in the same month. It was not found in any other sample.

Lecane hamata Stokes

Lake Albert

A few specimens were found in the sample taken from the mouth of the Semliki on 19 February 1962.

Lecane leontina Turner

Lake Albert

The samples taken from Wanseko in the period from June to October contained small numbers of this species, which never formed more than 4% of the total rotifers. It was not found in the samples from the other stations.

Lecane ludwigi Eckstein

Lake Albert

This species was found only in the samples from Wanseko, taken in June and July, when it formed 1% of the total rotifers.

Lecane luna O. F. Müller

Lake Albert

The sample from Wanseko in May yielded a few specimens of this species, otherwise the mouth of the Semliki was the only station where it was found. The maximum number of 387 per cubic metre was found in the sample taken on 19 February when the mouth of the Semliki was disturbed by strong winds.

Lecane papuana Murray

Lake Albert

A single specimen was found in the sample taken from the mouth of the Semliki in February.

Lecane pyriformis Daday

Lake Albert

Small numbers of this species were found at the mouth of the Victoria Nile and in Wanseko Lagoon. In September this species formed about 6% of the total rotifers in the sample from Wanseko.

Lake Kyoga

This species was found in the samples taken from stations 1 to 4, but never formed more than 2% of the total rotifers.

Lake Victoria

About 2% of the rotifers taken in the sample from the shallowest station belonged to this species, but it was not found in the other samples.

Lake George

This species was somewhat more abundant in Lake George than in the other lakes, and formed 6% of the rotifers in one of the samples taken in November.

Lecane quadridentata Ehrenberg

Lake Albert

A single specimen was found in the sample taken from Wanseko in October.

Lecane unguitata Fadeew

Lake Albert

This species formed 1% of the total rotifers in the samples taken from Wanseko in May and June, but was not found in any other sample.

Lecane ungulata Gosse

Lake Albert

This characteristic and cosmopolitan species formed about 1% of the total rotifers in the samples from Wanseko during May, June and July. In the samples from Pakwach it was found only in December, when it formed 1% of the rotifers, and at the mouth of the Semliki it was found in the October sample. It was not found in the samples from the other stations.

Lecane spp. indet.

Eight additional forms have been found during the course of the present study, and while it seems certain that they belong to distinct species it has not been possible to assign them to known species. It would be necessary to have much more material if an attempt was made to describe them as new species, and this procedure would be of doubtful value in the present state of the taxonomy of the genus, which is in need of a careful revision taking cognisance of variation in living and preserved specimens. None of the eight forms was of any numerical significance in the samples, and several were represented by single specimens.

Family Trichocercidae

Trichocerca cylindrica Imhof

Lake Albert

A single specimen was found in the sample taken from Wanseko in October.

Trichocerca rattus O. F. Müller

Lake Albert

A single specimen was found in the sample taken from Wanseko in October.

Lake Kyoga

Single specimens were found in three of the seven samples from Lake Kyoga.

Lake George

This species was present in all the samples from Lake George. The highest number under one square metre was 543.

Trichocerca similis Wierzejski

Lake Kyoga

Single specimens were found in the samples from stations 1 and 3.

Lake Victoria

Two of the samples yielded single specimens.

Trichocerca sp. a.

Lake Albert

This is a small form, varying in length from 72 to 93 μ, with a single posterior spine varying in length from 57 to 65 μ. Single specimens were found in seven of the samples from Pakwach and in single samples from Ndaiga, Wanseko and the mouth of the Semliki.

Trichocerca sp. b.

Lake Albert

A single specimen of what may be a small form of *T. chattoni* (de Beauchamp) was taken from Pakwach in March.

Trichocerca sp. c.

Lake Albert

Two specimens of an unidentified species were found in the sample taken from Pakwach in October. The length of the body was 140 μ, and the longest posterior spine was 72 μ long. It was not possible to decide if any small spines or teeth were present on the anterior border of the lorica.

Family Testudinellidae

Filinia longiseta Ehrenberg

I am following Pejler (1957) in regarding *F. limnetica* (Zacharias) as a synonym of *F. longiseta.*

Lake Albert

This species was found in all the samples taken from the mouth of the Semliki, and reached its maximum abundance (7273 under one square metre) in March. It was also present in all the samples from Ndaiga, but was less abundant, the maximum number under one square metre being 933 in May. At the other stations its occurrence was more erratic and the numbers were generally low.

Lake Kyoga

This species was present in the samples taken from all but one of the stations on Lake Kyoga. It was most abundant at station 6, where 735 were found under one square metre, and it formed 13% of the rotifers in the sample.

Lake George

This species formed 1 or 2% of the rotifers in the samples taken in November; the highest number under one square metre was 181.

Filinia terminalis Plate

Pejler (1957) regards *F. maior* (Colditz) as a synonym of *F. terminalis*, and this view is adopted here.

Lake Albert

A few specimens were found in the sample taken from the mouth of the Semliki on 19 February.

Lake Victoria

At station 3 this species formed 4% of the rotifers in the sample, and the population density was 119 under one square metre.

Lake George

In the sample taken in July this species formed 1% of the rotifers; the number under one square metre was 246.

Hexarthra mira Hudson

Lake Albert

The highest numbers of this species (up to 748 under one square metre) were recorded at the mouth of the Semliki, but it was not present at this station throughout the year. It was present in all the samples from Ndaiga, where the maximum recorded under one square metre was 333. At Pakwach it formed a small proportion of the total rotifers, and on one occasion the numbers under one square metre reached 540. No specimens were found in the samples from Buhuka, but it was recorded twice, in June and July, at Wanseko.

Lake Kyoga

This species was present in six of the seven samples from Lake Kyoga, but it never formed more than 8% of the total rotifers. The highest number under one square metre (1142) was found at the first station.

Lake Victoria

A few specimens were found in the samples from stations 1 and 2.

Lake Edward

A few specimens were found in two of the samples. All the specimens that I have examined were referable to *H. mira*, but another species has also been recorded from Lake Edward by de Beauchamp (1932), who found *H. intermedia* Wiszniewski in the samples collected by the Cambridge Expedition to the East African Lakes.

Pompholyx sulcata Hudson

Lake Albert

A few specimens were found at the mouth of the Victoria Nile in May.

Testudinella emarginula Stenroos

Lake Albert

A few specimens were found in the sample taken from the mouth of the Victoria Nile in May. The length of the lorica was 100 μ.

Testudinella patina Hermann

Lake Albert

This species was present in all but one of the samples from Wanseko, but was never abundant. At Pakwach it was recorded twice, in December and March; on each occasion it formed 2% of the total rotifers. At the mouth of the Semliki it was found in the sample taken on 19 February, when it formed 1% of the rotifers in the sample. It was not found in the Midlake samples, or in the samples from Butiaba, Buhuka and Ndaiga.

Lake Kyoga

Small numbers were found at stations 1 and 2, but not at the other stations.

Testudinella trilobata Shephard

This is a larger and rarer species than *T. patina*. The original brief description was given by Shephard (1892); a more detailed description of specimens collected in Natal by Kirkman (1901) was given by Rousselet (1901). Specimens from the Ivory Coast described by de Beauchamp (1955) under the name *T. dendradraena* also belong to this species.

Lake Albert

Small numbers were found at Wanseko in June and October, and at Pakwach in May.

Lake Kyoga

A single specimen was found in the sample from station 3.

Tetramastix opoliensis Zacharias

Lake Albert

This species was present in most of the samples taken from the mouth of the Semliki, where it formed up to 14% of the rotifers in the November sample. The highest numbers were also recorded in November (1770 under one square metre). At Pakwach it was less

abundant (maximum recorded—363 under one square metre) and never formed more than 5% of the rotifers. At Ndaiga it reached a density of 777 under one square metre in June, being in general more abundant than at Pakwach, but less abundant than at the mouth of the Semliki. At Wanseko and Buhuka its occurrence was restricted to small numbers in September and June respectively.

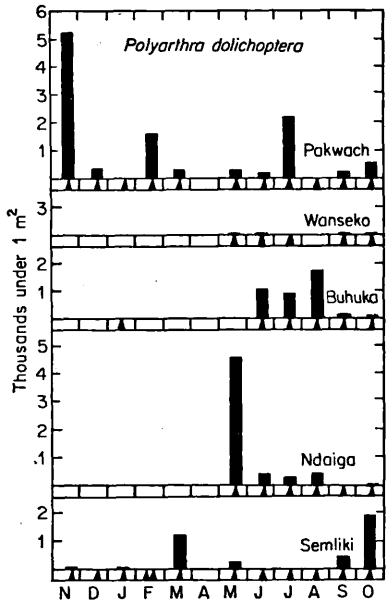

FIG. 10. Seasonal occurrence of *Polyarthra dolichoptera* at five stations in Lake Albert. The black triangles indicate when samples were taken.

Lake Kyoga

This species was present in all the samples from Lake Kyoga, forming between 2 and 8% of the rotifers. The maximum number under one square metre was 434 at station 3.

Lake Victoria

All the samples from Lake Victoria contained this species, which formed up to 3% of the rotifers. The highest number under one square metre was 113.

Lake Edward

This species was sparse in Lake Edward, the highest number found was 76 under one square metre.

Family Synchaetidae

Polyarthra dolichoptera Idelson

Lake Albert

The seasonal occurrence of this species is shown in Fig. 10. It was much less abundant at Wanseko than at the other localities. The relative importance of this species varied greatly

from month to month and from station to station. In Buhuka Lagoon in September this species formed two-thirds of all the rotifers in the samples, and a similar proportion was found at Pakwach in July. The highest number under one square metre was 5238 at Pakwach in November.

Lake Kyoga

This species was present in four of the seven samples from Lake Kyoga, forming between 5 and 11% of the total rotifers. The highest number under one square metre was 543 at station 3.

Lake George

A few specimens were found in the samples taken in November.

Lake Edward

This species was not found in the present series of samples, but de Beauchamp (1939) has recorded *P. platyptera* from Lake Edward. This record might possibly refer to *P. dolichoptera* because it was made before Carlin (1943) had made his detailed revision of the genus.

Polyarthra vulgaris Carlin

Lake Albert

This species is not always easy to separate from *P. dolichoptera*, and introgressive populations occur (Pejler, 1957). In the samples from the mouth of the Semliki in March some of the specimens of *Polyarthra* were referable to typical *P. vulgaris* while the majority were *P. dolichoptera*. It is possible that the two forms are extremes of a "Rassenkreis" in the sense of Rensch (1929), as suggested by Ruttner-Kolisko (1963). Forms attributable to *P. vulgaris* were not found at the other stations.

Synchaeta sp. cf. pectinata Ehrenberg

Specific identification of preserved specimens of *Synchaeta* spp. is difficult and often impossible. The specimens which were well preserved resembled *S. pectinata*, but it was not possible to be certain of the identity of all the specimens counted in the samples.

Lake Albert

A few specimens were found in the sample from Pakwach in December, and in the sample from the mouth of the Victoria Nile in May.

Lake Kyoga

Five of the seven stations on Lake Kyoga yielded specimens of *Synchaeta*. The highest number under one square metre (1222) was found at station 5.

Lake Victoria

All the samples from Lake Victoria yielded this species, which formed between 20 and 46% of the total rotifers. The highest number under one square metre was 2732 at station 1.

Lake Edward

About 50% of the rotifers in the samples from Lake Edward belonged to this species, but the total numbers under one square metre were low and the maximum recorded was only 764 at station 1.

Family Asplanchnidae

Asplanchna brightwelli Gosse

Lake Albert

This species reached its highest numbers at the mouth of the Semliki in December, when 2620 were found under one square metre. At Pakwach its occurrence was limited to November and December, and at Wanseko a few specimens were found in July and October.

Lake Kyoga

Stations 3 and 7 yielded a few specimens, but the total under one square metre did not exceed 113.

Order BDELLOIDEA

Family Philodinidae

Rotaria neptunia (Ehrenberg)

Lake Albert

A single specimen was found in the sample taken from the mouth of the Semliki on 20 February.

Comparison of the sampling stations

Lake Albert

Variations in the total number of rotifers under one square metre at each of eight stations are shown in Fig. 11, and Table I compares the mean numbers under one square

TABLE I

Rotifera in Lake Albert

Station	Mean depth (m)	Mean no. of rotifers under 1 m^2	Mean no. of rotifers in 1 m^3	Total no. of species	Index of diversity
Pakwach	8·7	5686	851	30	6
Victoria Nile	5·1	1042	305	27	12
Wanseko	3·8	1763	549	42	10
Butiaba	3·5	301	117	6	1
Midlake	47·2	371	8	5	1
Buhuka	4·1	10,416	3273	8	1
Ndaiga	5·8	5556	1064	11	1
Semliki	3·0	15,812	6455	30	6

metre, the mean numbers per cubic metre and the total number of species found at each station. It is clear that rotifers are most consistently present and abundant at the mouth of the River Semliki, and consistently present, but not so abundant at Pakwach. The samples from the Midlake station and Butiaba are similar in having few rotifers and showing only a minor increase in May, when *Keratella tropica* was the commonest species at both stations. In Wanseko Lagoon the total number of rotifers was generally lower than at Buhuka and Ndaiga, but at Wanseko there were many more species in the samples.

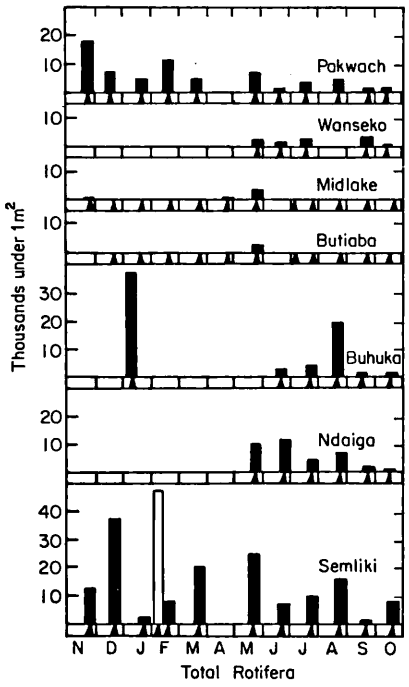

FIG. 11. Seasonal variation in the total Rotifera at seven stations in Lake Albert. The black triangles indicate when samples were taken. The clear column in February at the mouth of the Semliki represents a day when the water had been disturbed by a strong wind blowing from the North.

The index of diversity has been estimated to the nearest whole number from the graph given by Williams (1964, p. 311). In making this estimation the total number of specimens critically identified was used, and not the total per square metre. The indices of diversity fall into three distinct groups (Table I). The first group, with low diversity, includes Butiaba, Buhuka, Midlake and Ndaiga. The second group includes Pakwach and the mouth of the Semliki, and the third group, with a much higher diversity contains the mouth of the Victoria Nile and Wanseko Lagoon. The high diversity at these two stations is caused by non-planktonic species, particularly of the genus *Lecane*. These rotifers are frequently encountered in shallow water near vegetation.

The sampling stations have also been compared by means of the Sørensen index of similarity. This was calculated for each combination of stations according to the following equation

$$S = \frac{2c}{a+b} \times 100$$

where c = number of species common to both associations, a = number of species in one association, and b = number of species in the other association. The index was first used to compare plant associations (Sørensen, 1948), but it has also been used to compare associations of leaf hoppers (Kontkanen, 1950), and the Steinhaus-Marczewski modification, which gives parallel results, has been applied to rotifer associations in fish ponds (Hillbricht-Ilkowska, 1964). The results of calculating the Sørensen indices for 28 pairs of stations are given in Table II, and the highest indices for each station are indicated,

TABLE II

Sørensen indices for rotifer samples from eight stations on Lake Albert

	Pakwach	Victoria Nile	Wanseko	Butiaba	Midlake	Buhuka	Ndaiga	Semliki
Pakwach	+	49	61	33	29	42	49	73 h
Victoria Nile	49 h	+	49 h	36	31	40	47	49 h
Wanseko	61 h	49	+	25	21	32	38	56
Butiaba	33	36	25	+	73	86 h	71	33
Midlake	29	31	21	73	+	77 h	63	29
Buhuka	42	40	32	86 h	77	+	84	42
Ndaiga	49	47	38	71	63	84 h	+	49
Semliki	73 h	49	56	33	29	42	49	+

h, Indicates the highest value in each horizontal row.

so that it is possible to see which stations have the greatest similarity in species composition. There is considerable agreement with the grouping based on the index of diversity. The samples from Pakwach show the greatest similarity to those from the mouth of the Semliki. Butiaba, Buhuka, Ndaiga and Midlake yield high interrelated indices of similarity, and all show least similarity to the samples from Wanseko. The association of rotifers at the mouth of the Victoria Nile is similar to those at the two other river stations, and to the nearby Wanseko Lagoon.

The Sørensen index gives equal weight to every species, whether rare of abundant, and so may give undue emphasis to the rare species and to the occasional non-planktonic species in the samples. Another method of comparison is to use the "percentage difference" between pairs of stations. This method has been used successfully by Odum (1950) in a study of bird populations. The sum of the differences in numbers of individuals of each species at two stations is calculated as a percentage of the total individuals in the combined samples from both stations. The greatest emphasis is thus placed on the most abundant species, and when two stations differ greatly in total numbers, as when comparing the mouth of the Semliki with Midlake, the difference will not be influenced much by the

specific composition of the samples. The percentage differences between the samples from the stations on Lake Albert have been calculated from the mean number per cubic metre. Table III shows that the greatest differences are found between stations which differ most in total numbers of rotifers, and the smallest differences are found when the total numbers are similar. In these samples the main information gained from this type of analysis is related more to similarity in total production of rotifers than to similarity in specific

TABLE III

Percentage differences of rotifer samples from eight stations on Lake Albert, based on annual mean numbers of each species

	Pakwach	Victoria Nile	Wanseko	Butiaba	Midlake	Buhuka	Ndaiga	Semliki
Pakwach	+	70	63	88	99	70	31 s	78
Victoria Nile	70	+	49 s	83	93	91	84	95
Wanseko	63	49 s	+	68	96	89	79	88
Butiaba	88	83	68 s	+	82	90	72	95
Midlake	99	93	96	82 s	+	99	98	99
Buhuka	70	91	89	90	99	+	63	41 s
Ndaiga	31 s	84	79	72	98	63	+	71
Semliki	78	95	88	95	99	41 s	71	+

s, Indicates the smallest percentage difference in each horizontal row.

composition of the samples. In this case the calculation of percentage difference does not give any more significant information than a simple numerical comparison. The percentage difference method is probably more valuable in discerning differences in samples which yield similar numbers of specimens.

Koch (1957) has devised an index of biotal dispersity (IBD) which can be used to assess how widely dispersed species are between a number of stations

$$\text{IBD} = \frac{T-S}{S(n-1)} \times 100,$$

where T is the arithmetical sum of species living in each of n compared associations, and S is the total list of species in n compared associations. If each station had a completely different set of species S would equal T and the IBD would be 0. If each station had an identical set of species T would equal $n \times S$ and the IBD would be 100.

When the Koch index for all eight stations is calculated the resulting IBD is 22, but if separate indices are calculated for the four lake stations with low diversity (Butiaba, Buhuka, Ndaiga and Midlake) and for the four stations associated with rivers (Pakwach, Wanseko, mouth of the Victoria Nile and Semliki) there is an increase in the IBD in both cases. For the lake stations the figure is 58, and for the riverine stations the figure is 35. These large increases show that the rotifer assemblages in the lake stations resemble each other more than the riverine stations, and support the conclusions reached using the Sørensen index and the index of diversity.

Lake Kyoga

The seven stations in this lake are compared in Table IV. Rotifers were most abundant at station 1 and least abundant at stations 4 and 7. Viewing the lake as a unit (Table IX) the most important species are *Lecane bulla*, *Keratella tropica* and *Brachionus caudatus*.

TABLE IV

Numbers of Rotifera in Lake Kyoga

Station	Depth (m)	No. Rotifera under 1 m²	Mean no. Rotifera in 1 m³	No. of species
1	5	14,281	2856	15
2	5·25	8555	1629	17
3	6·25	10,860	1810	14
4	4	2545	727	14
5	6·25	9403	1567	15
6	5	5656	1257	14
7	4	2828	808	9

Samples taken 13 and 14 November 1962

Lake Victoria

In Table V the three stations are compared in terms of total numbers of rotifers. The total is lowest at the deepest station, and the number of species is also lowest at this station. Table IX shows that *Synchaeta pectinata*, *Lecane bulla*, and *Keratella tropica*, are the most

TABLE V

Numbers of Rotifera in Lake Victoria

Station	Depth (m)	No. Rotifera under 1 m²	Mean no. Rotifera in 1 m³	No. of species
1	15	5939	397	14
2	19	3762	198	15
3	27·5	2969	107	9

Samples taken 24 October 1962.

important species, while *Keratella cochlearis* and *Euchlanis dilatata* occupy an intermediate position between the dominant species and the species which were found in very small numbers.

Lake Edward

The main characteristic of the rotifer fauna in the samples from this lake is sparseness, both in terms of individuals per unit volume and in numbers of species (Table VI). It

should be noted that in addition to the seven species recorded in the present survey de Beauchamp (1939) has recorded 13 other species from the lake. The rotifer fauna of Lake Edward is probably more similar to that of Lake Victoria than is indicated by the present series of samples. *Synchaeta pectinata* and *Keratella tropica* were the most important species in the samples.

TABLE VI

Numbers of Rotifera in Lake Edward

Station	Depth (m)	No. Rotifera under 1 m²	Mean no. Rotifera in 1 m³	No. of species
1	8·5	1471	173	6
2	19	141	7	3
3	24·5	622	25	5
4	29·5	1273	43	5

Samples taken 3 November 1962.

Lake George

The sample taken in July was much richer than the samples taken in November (Table VII). *Lecane bulla* was most numerous in the samples, but *Brachionus caudatus* and *Keratella tropica* were also relatively important.

TABLE VII

Numbers of Rotifera in Lake George

Station	Depth (m)	No. Rotifera under 1 m²	Mean no. Rotifera in 1 m³	No. of species
1 (July)	3·0	24,604	12,302	8
2 (Nov.)	2·5	6787	3394	12
3 (Nov.)	2·75	9050	4019	11
4 (Nov.)	3·5	7353	2451	10

Samples collected 11 July and 1 November 1962.

Inter-lake comparisons

Table X shows the results of calculating the Sørensen indices for Lakes Victoria, Kyoga, George and Edward. The samples from the two shallow lakes, Kyoga and George, are more similar to each other than they are to their neighbouring deeper lakes. The main features which these two lakes have in common are their depths and the abundance of floating papyrus. The two deeper lakes, Victoria and Edward, show a less striking similarity, but it is surprisingly high in view of the considerable difference in the numbers of species in the samples and the corresponding difference in the index of diversity.

TABLE VIII

Rotifera in Lakes Victoria, Kyoga, George and Edward, based on plankton samples taken in October and November 1962

Lake	Mean depth of haul (m)	Mean no. of Rotifera under 1 m²	Mean no. of Rotifera in 1 m³	Total no. of species	Index of diversity
Victoria	20·5	4223	234	19	5
Kyoga	4·8	7732	1524	24	5
George	2·5	7730	3287	15	3
Edward	19·0	1372	110	7	1

TABLE IX

Mean numbers of Rotifera per cubic metre based on samples taken in October and November 1962

Species	Lake Victoria	Lake Kyoga	Lake George	Lake Edward
Anuraeopsis fissa	—	34	11	—
Brachionus bennini	4	—	—	—
budapestinensis	—	—	11	—
calyciflorus	—	5	84	—
caudatus	4	219	375	5
falcatus	1	126	—	3
forficula	2	—	—	—
patula	—	32	—	—
quadridentatus	7	97	107	5
urceolaris	—	2	—	—
Keratella tropica	26	255	542	32
lenzi	5	4	—	—
cochlearis	15	—	—	—
Lepadella	—	—	35	—
Mytilina ventralis	1	—	—	—
Epiphanes macroura	—	12	92	—
Euchlanis dilatata	13	—	—	—
Lecane bulla	55	384	1,644	—
pyriformis	3	17	101	—
aculeata	—	2	—	—
sp. A	—	—	27	—
sp. B	5	—	—	—
sp. C	1	—	—	—
Trichocerca similis	2	7	—	—
rattus	—	5	145	—
Filinia longiseta	—	43	46	—
terminalis	1	—	—	—
Tetramastix opoliensis	4	56	—	2
Testudinella patina	—	9	—	—
trilobata	—	1	—	—
Polyarthra dolichoptera	—	49	11	—
Synchaeta sp.	83	79	—	58
Hexarthra mira	2	61	56	5
Asplanchna brightwelli	—	7	—	—
Bdelloid sp. indet.	—	19	—	—

There are of course severe limitations to the present data. One would like the data from all the lakes to be comparable in number to those obtained from Lake Albert. But even with the present limited data the Sørensen index gives results which lend themselves to fairly simple ecological explanations.

TABLE X

Sørensen indices for rotifer samples from Lakes Victoria, Kyoga, George and Edward

	Victoria	Kyoga	George	Edward
Victoria	+	48	35	54 h
Kyoga	48	+	63 h	47
George	35	63 h	+	36
Edward	54 h	47	36	+

h, Indicates the highest value in each horizontal row.

When the four lakes are compared with the eight stations in Lake Albert (Table XI) the Sørensen indices show that the rotifers in Lake Kyoga are most similar to the associations at the northern end of Lake Albert. There are geographical and ecological reasons for this resemblance. The outflow from Lake Kyoga reaches Pakwach *via* the mouth of the

TABLE XI

Comparison of rotifer samples from four lakes and eight stations in Lake Albert, using Sørensen indices

Station in Lake Albert	Lake Victoria	Lake Kyoga	Lake George	Lake Edward
Pakwach	41	70 h	44	38
Victoria Nile	35	56	38	24
Wanseko	20	58	42	24
Butiaba	32	41	38	46
Midlake	17	36	50	33
Buhuka	44 h	52	52	53
Ndaiga	40	53	54 h	55 h
Semliki	29	57	40	27

h, Is used to indicate the highest index for each lake.

Victoria Nile. The depths of the two regions are similar, and both are subjected to the influence of floating islands. The samples from Lake Kyoga also show considerable similarity to the samples from the mouth of the River Semliki. This resemblance is to be expected on ecological grounds. As noted in the previous section the samples from the riverine stations of Lake Albert and more similar to each other than they are to the samples from the stations on the main body of the lake. The overall similarity of the samples from

Lake Kyoga is with the riverine stations of Lake Albert. In contrast, the association of rotifers in the other shallow lake, Lake George, shows its greatest similarity to the rotifers in Ndaiga and Buhuka lagoons. The Sørensen indices are not very high, being about the same as those of Lake Kyoga when compared with these lagoons, but they do indicate that Ndaiga and Buhuka have associations of rotifers which show more resemblance to the association in Lake George than is shown by the rotifers in other parts of Lake Albert. Ndaiga and Buhuka also show greater resemblance in their rotifers to Lake Edward than do the other stations in Lake Albert.

The Sørensen indices comparing Lake Victoria to the eight stations in Lake Albert are all rather low. Not one index reaches 50. The association of rotifers in Lake Victoria shows the greatest resemblance to that in Buhuka lagoon, but even in this comparison the index is only 44. This may indicate that the association of rotifers in Lake Victoria is somewhat divergent from that in the other lakes. It is noteworthy that Lake Victoria is the only lake from which the samples taken in this study contained *Branchionus forficula*, *Brachionus bennini*, and *Euchlanis dilatata* although it is possible that these species may be found in the other lakes when more detailed studies have been made. They were nevertheless absent from the numerous samples taken during the course of a year on Lake Albert.

Discussion

It is clear from Table I that the mouth of the Semliki is the area of Lake Albert most favourable to the development of rotifer populations. The flow of water through this area is relatively slow, but is sufficient to cause the plankton to develop the characteristics of a river plankton, with a relative increase in the proportion of Rotifera. There is a marked similarity between the samples from the mouth of the Semliki and the rotifer fauna of the River Sokoto in Nigeria (Green, 1960). The dominance of *Keratella tropica* and *Brachionus caudatus* (Fig. 12) is a particular point of similarity.

At Pakwach the Rotifera are less abundant than at the mouth of the Semliki. This is probably due to the greater rate of flow at Pakwach. It was found in the River Sokoto that when the river level was raised the total rotifer population decreased, although the diversity of species in the plankton increased. This type of phenomenon is seen particularly clearly at the mouth of the Victoria Nile. Here there is a strong current through relatively narrow channels. The total number of rotifers per unit area is low, but the index of diversity is high. In only one sample from the mouth of the Victoria Nile were rotifers numerous enough to enable an estimate to be made of the species composition. This was the sample taken in May. The dominant rotifers were *Lecane bulla*, which formed 21 % of the rotifers, and *Keratella tropica*, which formed 15 % of the total. These two species were accompanied by 24 other species. This high diversity and the relative importance of *Lecane bulla* are characteristic of the rotifer populations in situations with a high rate of flow.

The Midlake samples are particularly poor in rotifers, and share this feature with the samples from Butiaba. This seems strange at first because the Butiaba station is close inshore and might be expected to yield samples similar to those from Buhuka and Ndaiga. Although close inshore the Butiaba station lay outside the sand spit, and the bottom of the lake fell away steeply in this region. The close resemblance between the Midlake and the Butiaba samples may be due to the action of the currents which are responsible for the

formation of the sand spit. These currents probably sweep water from the middle of the lake along the outer edge of the sand spit.

The sparseness of rotifers in the Midlake samples is not due to a general sparseness of zooplankton. In fact there is a considerable population of planktonic Crustacea in the middle of the lake. It may be that this is a case of competitive elimination of the rotifers by larger zooplankters. Brooks & Dodson (1965) have found that there is a general decrease in the mean size of zooplankters when plankton-eating fish are introduced into a lake,

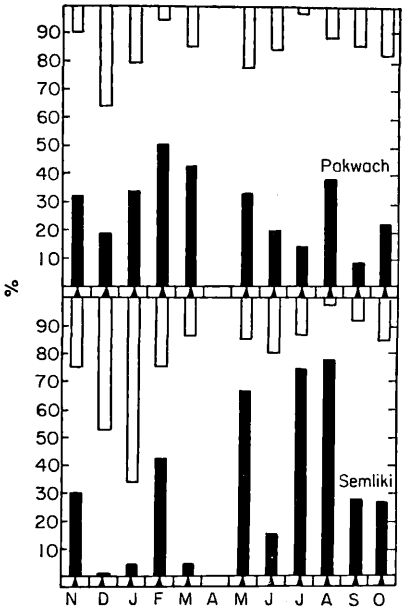

FIG. 12. Seasonal variation in the percentages of the total Rotifera formed by *Brachionus caudatus* and *Keratella tropica* at Pakwach and at the mouth of the Semliki. □, *B. caudatus;* ■, *K. tropica.*

and Hillbricht-Ilkowska (1964) has found marked increases in the rotifer populations in heavily stocked fish ponds. Now the main planktivorous fish in Lake Albert live in water less than 20 m deep, so that the Midlake zooplankton is not subjected to much predation. (Green, 1967, for a discussion of the distribution of planktivorous fish in Lake Albert). In the absence of predators the larger zooplankters are at an advantage because they are more efficient at gathering food, but where predators are present their large size is disadvantageous because the larger animals are more easily seen and are selectively eaten (Brooks & Dodson, 1965; Green, 1967). The sparseness of rotifers in the middle of Lake Albert may thus be partly explained by the sparseness of planktivorous fish.

Lakes George and Kyoga both yield samples with a high proportion of rotifers. *Brachionus caudatus* and *Keratella tropica* are again important, but both lakes also show a high proportion of *Lecane bulla* (Table IX). The occurrence of this species in plankton samples is often associated with the disturbance of water near vegetation. The floating islands in these lakes may be associated in some way with the frequent occurrence of *L. bulla.* The

numbers of *Lecane* in Lake George may also be influenced by the abundance of blue-green algae. *Lecane bulla* can attach itself to such algae, so that although not strictly a planktonic species this rotifer can be found in considerable numbers in plankton samples which contain an abundance of blue-green algae. A parallel to this is found in the highly eutrophic Fredriksborg Castle Lake in Denmark, where the benthic cladoceran, *Chydorus sphaericus* becomes abundant in the plankton at the time when the blue-green algae are forming blooms (Berg & Nygaard, 1929). The phytoplankton in the eastern arm of Lake Kyoga is sometimes dominated by the filamentous blue-green alga *Lyngbya limnetica* Lemmerman (Evans, 1962), which may also serve as a substratum for *Lecane bulla.*

This work was made possible by the generous co-operation of other people. Mr M. J. Holden collected most of the samples from Lake Albert and made arrangements for my work on this lake. The Fisheries Department at Entebbe made it possible for me to visit the other lakes and arranged local contacts for me. I am also indebted to Messrs Tufmac Ltd., who made a launch available on Lake George. My thanks are also due to the Leverhulme Trust for a grant which made possible the journey to Uganda.

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J. 1

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Biliverdin in the Eggs of *Nereis fucata*

Nereis fucata (Savigny) is a polychæte worm living in the gastropod shells carried by the hermit-crab *Eupagurus bernhardus* (L.) at Plymouth. The heteronereis or breeding stage found with hermit-crabs during late February was found to contain apparently ripe, blue eggs in the cœlom. The eggs were removed by slitting the body-wall of fresh worms, rinsed in sea water to remove hæmoglobin, and separated from cœlomic cells by repeated centrifuging. The pigments in the eggs were extracted into a methanol–sulphuric acid mixture (19 : 1), when a turquoise-blue solution was produced. A purple pigment remained in the eggs; the nature of this pigment is being investigated. The original blue colour of the eggs was apparently due to the mixture of the purple pigment and the turquoise extract. The turquoise solution was filtered through glass wool, diluted with an equal volume of water and shaken with chloroform. The chloroform layer became bluish-green. This was evaporated to dryness *in vacuo*, and the pigment redissolved in methanol. The absorption spectrum of this solution showed a sharp peak at 392 mμ and a broader peak at 650 mμ, with minimum absorption at 500 mμ. The addition of zinc acetate and iodine produced a brilliant pink fluorescence in ultra-violet light. When more iodine was added the fluorescence became green. The presence of biliverdin was confirmed by the production of a purple colour, with a spectral absorption maximum near 490 mμ, by the addition of concentrated nitric acid, and by the destruction of the blue-green pigment with concentrated sulphuric acid.

This appears to be the first time that biliverdin has been recorded in an egg, although it is known from the egg shells of certain birds[1], and a green pigment giving the Gmelin reaction is found in the eggs of the parasitic cirripede *Septosaccus*[3]. Biliverdin has been found in the body-wall of *Nereis diversicolor* O. F. Müller[2], but a re-examination of this species by us has revealed no biliverdin in the eggs. The pale yellow colour of *N. diversicolor* eggs may be due to minute amounts of carotenoids, but acetone extracts did not yield sufficient pigment for characterization. Similar quantities of *N. fucata* eggs yielded sufficient acetone-soluble pigment to be purified and identified as β-carotene. A further contrast is provided by the cœlomic cells, which are yellow in *N. fucata* and green in *N. diversicolor*, contrasting

markedly with the colours of the eggs. Further work on the pigments present in the cœlomic cells and eggs of these and various other polychætes is in progress.

J. GREEN
R. PHILLIPS DALES

Zoology Department,
Bedford College,
Regent's Park,
London, N.W.1.

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The pigments of *Artemia*

By Barbara M. Gilchrist and J. Green

The pigments of *Artemia*

By Barbara M. Gilchrist and J. Green
Bedford College, University of London

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The eggs of the brine shrimp, *Artemia salina*, vary in colour from pale cream to dark brown. This variation is due to different amounts of haematin in the egg shells.

Nauplii of *Artemia* are bright orange in colour owing to a carotenoid pigment, esterified astaxanthin. The same carotenoid is present in the eggs.

Adult *Artemia* which has been reared on bakers' yeast, in which we found no carotenoids, contains only a small amount of astaxanthin ester, presumably derived from that present in the egg.

The carotenoids of the unicellular algae *Dunaliella tertiolecta* and *Phaeodactylum tricornutum* have been examined as a preliminary to feeding experiments with *Artemia*. The carotenoids identified from *Dunaliella* were β-carotene, γ-carotene, a carotene oxide, lutein, violaxanthin, and neoxanthin; those from *Phaeodactylum* were β-carotene, diadinoxanthin, fucoxanthin and neofucoxanthin.

Adult *Artemia* reared on *Dunaliella* retains varying amounts of all the algal carotenoids, and in addition changes some of them to astaxanthin which becomes esterified and is quantitatively the most abundant carotenoid in the animal. A keto-carotenoid has been found in *Artemia* examined soon after being fed on *Dunaliella*.

Artemia fed on *Phaeodactylum* retains all the algal xanthophylls to some extent. No β-carotene was found in the animals; a large amount of a keto-carotenoid was found, as well as astaxanthin.

There is evidence that β-carotene in the algal food is the precursor of astaxanthin found in the adult *Artemia* and that the transformation proceeds through the keto-carotenoids.

Introduction

In his original description of the brine shrimp, *Artemia salina* (L.), Schlosser (1756) remarked that the red colour of the animals tinted the water in the tanks in which they were found. Many authors since have referred to the colours of *Artemia*, in particular to the red colour of the adults. The general coloration has been briefly described by Ermakow (1928); he says the females are salmon pink with green heads and limbs, while the males have green bodies with reddish limbs. There is, however, considerable variation in colour; Bond (1933) believed that this variation was due to differences in the food of the animals. This is not always so: there are, for instance, two distinct causes of reddish coloration in *Artemia*. The nauplii are pink or orange because of carotenoid pigment (Needham & Needham 1930). The red or orange colours of adults may also be due to carotenoids (Lochhead & Lochhead 1941), but another frequent cause of redness in adult brine shrimps is the presence of haemoglobin.

The occurrence and function of haemoglobin in *Artemia* has already been analyzed in detail (Gilchrist 1954). It was shown that the concentration of haemoglobin in the blood of *Artemia* varies inversely as the oxygen content of the water in which the animal swims. In natural populations an increase in redness of the animals with increase in salt content of the environment has been noted several times (Payen 1836; Joly 1840; Schmankewitsch 1877; Kellogg 1906; Artom 1905). The observations of the last author are of particular interest because he noted that the

adults were red at high salinities, but the nauplii were red at all salinities of the medium. The increased redness of the adults at high salinities is not a direct effect of the salinity, but is due to the low oxygen content of strong brines stimulating the synthesis of haemoglobin (Gilchrist 1954).

Some information is also available concerning other haem pigments in *Artemia*. Phear (1955) detected a haemochromogen in the gut, and Needham & Needham (1930) have shown that a high concentration of haematin is present in the shells of the resting eggs.

We thus have a fairly detailed picture of the haem compounds present in *Artemia*, but there are no comparable data available for the other pigments. The presence of an ommochrome pigment in the eye has been demonstrated by Becker (1942), but beyond this record of its presence no further information is available. The references to carotenoids (Needham & Needham 1930; Lochhead & Lochhead 1941) are very general, with no critical identification of the pigments.

There is a considerable body of information available concerning the carotenoids present in the decapod Crustacea, but our knowledge of these pigments in the lower Crustacea is meagre. This is particularly true of the subclass Branchiopoda, which is generally acknowledged to include the most primitive of the living Crustacea. The only detailed account of carotenoids in a branchiopod is for the cladoceran *Daphnia* (Green 1957). We know of no account dealing with an anostracan, conchostracan or notostracan. In the present paper we have attempted to answer the following questions in relation to an anostracan, namely *Artemia*. First, what pigments are available at the onset of development within the enclosed system of the resting egg? Secondly, which carotenoids are found in the adult, and to what extent are they derived from those available in the food of the animal?

Artemia has certain advantages for this type of study. The resting eggs are obtainable in large amounts, so that the carotenoids available at the very onset of life can be determined. The adults are fairly easily reared in the laboratory and will reproduce at a rate sufficient to produce the large numbers required for the application of chromatographic and spectrophotometric methods to the identification of the pigments.

Material and methods

Nauplii and adults of *Artemia salina* (L.) were reared from a large stock of resting eggs obtained from California; these eggs are known to be not less than 8 years old. All cultures were derived from the same stock of eggs. The animals were reared in sea water at room temperature (18 to 20 °C) in large glass containers kept in a window facing north. Some cultures were fed only on a suspension of bakers' yeast, others only on *Dunaliella tertiolecta* Butcher 1959 and a third group were fed only on the diatom *Phaeodactylum tricornutum* Bohlin emend. Lewin 1958. Both *Dunaliella* and *Phaeodactylum* were cultured in an 'Erd-Schreiber' solution recommended by Dr Mary Parke of the Plymouth Laboratory.

All the chemical reagents used in the extraction and identification of pigments were of B.D.H. Analar quality. The whole carotenoid extract was passed through columns of adsorbents in order to separate the different carotenoids present.

Columns of alumina (B.D.H. for chromatographic adsorption analysis) and icing sugar (Tate & Lyle, which contained up to 1·5 % calcium phosphate) were found to be most satisfactory. Sometimes the alumina was activated by heating overnight in an oven at 70 °C, at other times unactivated adsorbents were used. Chromatographic columns were made in glass tubes of diameter 2 cm and about 20 cm long.

When extracts from algae were made, the plant material was first separated from its culture medium by centrifuging. The algae were ground with fine acid-washed sand in a mortar and acetone was used to extract the pigments. The crude acetone extract was filtered, then diluted with water and the pigments taken into light petroleum, various boiling ranges of which were used as available. The light petroleum solution was then washed thoroughly with water to remove traces of acetone. This was found to be essential because incomplete washing led to poor chromatographic separation of the pigments. After washing, the light petroleum solution was dried over anhydrous sodium sulphate. Two different procedures were then followed to isolate the various carotenoids. In the first method the light petroleum solution was washed with several changes of 90 % methanol to remove hypophasic pigments. The epiphasic pigments in the light petroleum were then chromatographed on alumina using increasing amounts of acetone in light petroleum for separation and elution. The hypophasic pigments were taken back from 90 % methanol into light petroleum by diluting the methanol with water and chromatographed separately on alumina or icing sugar. In the second method the dried light petroleum solution was poured directly on to a column of icing sugar. The column was developed by washing with light petroleum and the first fraction to pass through was found to contain all the epiphasic pigments, which were subsequently rechromatographed on alumina. Thus essentially the same basic separation of pigments was achieved as in the first method. The pigments remaining on the sugar column were separated and eluted by adding increasing amounts of *n*-propanol to the light petroleum with which the column was washed. It was found that icing sugar was much more satisfactory than alumina for the separation of hypophasic pigments.

The absorption spectra of the separated pigments were measured in a Unicam S.P. 500 spectrophotometer. The accuracy of the instrument was periodically checked using the hydrogen lamp emission lines at 4861 and 6563 Å.

Quantitative estimates of the relative amounts of carotenoids present were made by adjusting the volumes of each fraction to a known quantity and measuring the maximum light absorption in the visible range. In calculating the relative amounts of each carotenoid it was assumed that all the carotenoids had the same extinction coefficient. This assumption leads to certain inaccuracies, so that the percentages given in the tables can only be used as a general indication of the relative amount of each pigment present.

When extracts of pigment were made from adult *Artemia*, males and females were used separately. After separating the sexes the animals were washed with distilled water and then dried by gently rolling them on filter-paper. The pigments were extracted with acetone and the subsequent treatment was essentially the

same as that for the algal pigments so that direct comparison was possible. The animals were starved for about 24 h before the extraction of pigments. This was a precaution against including algal pigments derived from gut contents in the animal extracts.

Pigments in the eggs of *Artemia*

Haematin

The presence of haematin in the shell of the eggs of *Artemia* has been demonstrated by Needham & Needham (1930). Several authors have remarked upon the differences in the colours of the eggs, which may vary from pale cream to very dark brown (Joly 1840; Boone & Baas Becking 1931; Mathias 1932). We have confirmed the presence of haematin and attempted to determine to what extent the colour of the eggs is due to different amounts of this pigment in the shell.

Haematin was extracted from whole eggs by soaking them overnight in 1 % aqueous sodium hydroxide. A dark green solution was obtained which became red on the addition of sodium dithionite and, on shaking vigorously in air, became dark green again. The absorption spectrum of the alkaline haematin was measured in 1 % sodium hydroxide; an excess of sodium dithionite was then added and the absorption spectrum again determined.

The absorption curve of alkaline haematin was diffuse, with an ill-defined maximum about 582 mμ. On the addition of sodium dithionite, however, the characteristic absorption spectrum of a haemochromogen was obtained; this has a sharp peak at 558 mμ and a weaker one at 527 mμ.

In order to compare the amount of haematin in pale and dark coloured eggs of *Artemia* the total haem content of the eggs was measured. This includes not only the haem compounds in the shell but also the cytochromes in the tissues of the enclosed nauplius. We have never detected haemoglobin in the eggs or nauplii of *Artemia*. Since the absorption spectrum of haematin is diffuse, a pyridine haemochromogen was prepared from the eggs; it has a clearly defined absorption maximum at 557 mμ.

The method used to measure total haem has been described by one of us (Green 1956). The intensity of the α-band of a standard solution of pyridine haemochromogen prepared from human blood is compared with that of a solution of pyridine haemochromogen prepared from the eggs of *Artemia*. The intensity of the α-bands of the two solutions is matched using the comparator microspectroscope method of Elliot & Keilin (1934). The results obtained by this method were verified by measuring with the spectrophotometer the intensity of absorption of the two solutions at a wavelength of 557 mμ. Since the intensity of absorption of the haemochromogen solutions was found to decrease markedly the longer they were kept, measurements on both the comparator microspectroscope and on the spectrophotometer were made at the same time interval after the preparation of the solutions.

The standard solution of haemochromogen was made from a haemoglobin solution containing 0·1 ml. of blood in 75 ml. distilled water. To 40 ml. of this diluted solution was added a pinch of sodium dithionite and 10 ml. pyridine. This

high proportion of pyridine ensures that all the haem is converted to the haemochromogen.

A pyridine haemochromogen solution was prepared from the eggs of *Artemia* as follows. One thousand eggs were counted and homogenized with a pinch of sodium dithionite and 2 ml. of pyridine. This homogenate was put into a small glass tube and the volume made up to a fixed mark with pyridine. The tube was then left undisturbed for exactly 3 min to allow fragments of shell and particles of sodium dithionite to settle. In this way a clear supernatant solution of pyridine haemochromogen was obtained; for measurements made with the comparator microspectroscope 1 ml. of this solution was used.

Under normal culture conditions in the laboratory, *Artemia* reared in sea water reproduces viviparously, actively swimming nauplii being liberated from the mother's brood pouch. Occasionally, for reasons not yet understood, females cultured in sea water produce shelled eggs, the so-called 'resting eggs'. These are liberated from the brood pouch and are found in the culture vessels. In the experiments to be described estimations of total haem were made on two different samples of eggs. One sample was obtained from a sea-water culture of parthenogenetic females of *Artemia* derived from salt works at Sète, in southern France. These eggs were pale cream in colour. They were removed from the culture vessels and left to dry. The total haem content of one thousand of these eggs was compared with that of the same number of eggs collected in salt works at Sète from a region where the brine was moderately concentrated (salinity 180 to 200‰); these eggs were much darker in colour than those laid in sea water.

The results of these experiments are given in table 1. The comparator microspectroscope method is referred to as method 1 and the determinations made with the spectrophotometer as method 2. The actual measurements obtained by these two methods are not comparable since different scales of measurement are used, but the ratio of total haem in pale and dark eggs is comparable.

TABLE 1. TOTAL HAEM CONTENT OF PALE AND DARK COLOURED EGGS OF *ARTEMIA*; INTENSITY OF ABSORPTION AT 557 mμ IN ARBITRARY UNITS

	total haem content of eggs		ratio	
	pale	dark	pale	: dark
expt. 1				
method 1	7·3	28·7	1	3·9
method 2	56·0	200·0	1	3·6
expt. 2				
method 1	7·5	29·7	1	4·0
method 2	50·0	176·0	1	3·5

It is clear from these results that dark brown eggs of *Artemia* contain from three and a half to four times as much haem as pale cream coloured eggs. Since the egg shell contains haematin it is probable that differences in the colour of the eggs are due to different concentrations of haematin in the shell. In order to confirm this the total haem content of one thousand nauplii newly hatched from dark-coloured eggs was measured using the same methods as described for eggs. The amount of

haem in the nauplii was found to be negligible. The difference in the total haem content of pale and dark eggs is due, therefore, to different amounts of this pigment in the shell.

Carotenoids

Dried eggs of *Artemia* were homogenized with acetone; the extract was filtered and the pigments taken into light petroleum by the addition of water. On partition between light petroleum and 90 % methanol the pigments were entirely epiphasic.

The light petroleum extract was passed through a column of activated alumina and a single broad pink band of pigment appeared near the top of the column. In some experiments this washed slowly through the column and was eluted with light petroleum but in others it was necessary to use light petroleum containing 5 % of acetone in order to elute the pigment.

The absorption spectrum of the pigment in light petroleum (b.r. 60 to 80 °C) showed a single broad maximum at about 466 mμ. In carbon disulphide the maximum was at 502 mμ, in hexane at 466 to 468 mμ and in pyridine at 488 mμ. These figures agree well with those given by Goodwin & Srisukh (1949) and by Vevers (1952) for esterified astaxanthin.

The pigment was saponified by boiling vigorously for a few minutes in a concentrated solution of potassium hydroxide in ethanol. After saponification the pigment was entirely hypophasic on partition between light petroleum and 90 % methanol; on the addition of a few drops of glacial acetic acid it became epiphasic. This characteristic behaviour confirms that the astaxanthin in the eggs of *Artemia* is esterified.

PIGMENTS IN THE NAUPLII OF *ARTEMIA*

The nauplii of *Artemia* are intensely orange in colour; the colour resides mainly in the gut wall and in fat cells. No haemoglobin has been detected in nauplii although they have been described as 'blood red' in colour (Packard 1883; Jensen 1918). The orange colour is due to carotenoid pigment.

It has been shown above that the only carotenoid present in the eggs of *Artemia* is esterified astaxanthin. It is likely that the pigment is located in the enclosed nauplius and not in the egg shell. In order to verify this and also to determine whether any changes in the nature of the pigment present occur at the time of hatching, carotenoids were extracted from newly hatched nauplii. The nauplii were concentrated on bolting silk, rinsed in distilled water and the excess water drawn off with filter-paper. The extraction and subsequent treatment of the extract was the same as described for eggs.

On partition between light petroleum and 90 % methanol the extracted pigments were epiphasic. When chromatographed on activated alumina in light petroleum two bands of pigment separated on the column. A lower pink band washed slowly through the column with light petroleum and an upper pink band moved down the column with 80 % (v/v) ether in light petroleum and was finally eluted with 100 % ether.

The absorption maxima of these two fractions were found to be identical, at 468 mμ in light petroleum, 500 mμ in carbon disulphide and 466 mμ in hexane.

It seems therefore that only esterified astaxanthin is present in the newly hatched nauplii. On each of several occasions that we have extracted and chromatographed carotenoids from nauplii of *Artemia* two bands of pigment have separated on the column. This suggests that there are two esters of astaxanthin in the nauplius.

It is stated by Needham & Needham (1930), with reference to the carotenoid present in the eggs and nauplii of *Artemia*, that 'it is certain that the orange pigment (crustaceorubin?) is synthesised during development'. Their evidence for this is the stronger colour of the alcohol extracts obtained from nauplii than from eggs and that more fatty material is found in nauplii.

Clearly, it would be of interest to confirm this statement, for it would be the only known case of the synthesis of a carotenoid by an animal not supplied with carotenoid precursors. In view of this an attempt has been made to obtain a quantitative estimate of the carotenoid content of eggs and of newly hatched nauplii of *Artemia* and thus to get a measure of the suggested synthesis of carotenoid within the enclosed system of the egg.

During preliminary experiments on the quantitative extraction of carotenoids from eggs and nauplii, the material was ground with acid-washed sand in a mortar for $\frac{3}{4}$ h and the pigment extracted with acetone. When acetone extracts from equal quantities of eggs and nauplii were filtered and the pigment taken into equal volumes of light petroleum the extracts from newly hatched nauplii were much stronger in colour than those from dried eggs. This could mean that carotenoids are synthesized within the egg during the 36 to 48 h before the nauplius hatches. When the residue on the filter-paper was examined, however, many intact eggs were found in spite of the prolonged grinding with sand; all the carotenoid had not been extracted from the eggs. If the total carotenoid content of eggs and nauplii is to be compared quantitatively a more efficient method of extracting the pigment from the eggs must be used.

Improved extraction was obtained by grinding the material in a Griffiths thimble homogenizer. No difference could then be seen in the intensity of the colour of carotenoid extracts from equal quantities of eggs and of nauplii. This indicated improved extraction from the eggs; this type of homogenizer was therefore used in the quantitative extractions to be described below.

Equal quantities of eggs and of newly hatched nauplii were obtained by weighing two batches of 100 mg of dried eggs and allowing one batch to hatch in sea water. Not all the eggs gave rise to active nauplii, but it was observed that even among the small percentage which did not liberate nauplii most of the shells had split and a well-developed embryo could be seen inside.

A quantitative extract of the carotenoids in the eggs and newly hatched nauplii was made by homogenizing the material in about 2 ml. methanol for 6 min. The extract was then filtered and the volume of filtrate made up to 10 ml. with methanol. Exactly 20 min after extraction was begun the intensity of absorption of this solution of the pigment was measured at 470 mμ, the absorption maximum of esterified astaxanthin in methanol. Since carotenoid pigments are readily destroyed by light, the intensity of absorption must be measured in all samples after exactly the same time interval from the beginning of pigment extraction.

The results obtained from a number of samples in which the carotenoid content of eggs and newly hatched nauplii was compared are given in table 2. These show that the carotenoid content of newly hatched nauplii is about 8 % higher than that of dried eggs. The difference is statistically significant. These results might mean that the carotenoid content of the nauplius increases within the egg prior to or at the time of hatching. We consider, however, that this difference in the carotenoid extracts could still be accounted for in terms of incomplete extraction of the pigment from the eggs. This may be due partly to adsorption of carotenoids on the calcium salts in the fragmented shells and partly to different degrees of adsorption on to tissues which differ in composition in eggs and nauplii (Needham & Needham 1930).

TABLE 2. COMPARISON OF THE CAROTENOID CONTENT OF EQUAL QUANTITIES OF DRIED EGGS AND NEWLY HATCHED NAUPLII OF *ARTEMIA*. THE OPTICAL DENSITY IS GIVEN OF EQUAL VOLUMES OF PIGMENT MEASURED AT 470 mμ IN METHANOL

	eggs	nauplii
	411	365
	402	403
	388	400
	374	402
	375	461
	420	472
	442	474
	418	455
	417	500
	464	470
	410	490
	441	486
	454	480
	445	488
	448	510
	440	
mean and S.E.	422 ± 6·9	457 ± 11·2
ratio	1·00 :	1·08

The extinction coefficients of the astaxanthin esters in the nauplii and eggs may also differ slightly. It is clear from the chromatographic results that there are certain differences; one band only is formed with extracts from eggs, while two bands appear when extracts from nauplii are chromatographed.

PIGMENTS AVAILABLE IN THE FOOD OF *ARTEMIA*

Phaeodactylum tricornutum Bohlin emend. Lewin

Chromatography on icing sugar of the crude light petroleum extract resulted in the separation of five fractions which were eluted from the column in the following sequence.

(1) A yellow fraction passed through the sugar column with light petroleum. When rechromatographed on alumina there was one dense orange band which was eluted with 0·5 % acetone in light petroleum. A faint pink band remained at the top of the column, but we have not been able to elute this successfully as the

pigment disappears, apparently owing to oxidation on the column. The strong orange fraction is epiphasic and has absorption maxima at 450 and 477 mμ in hexane, showing that it is β-carotene.

The epiphase left after washing the crude light petroleum solution with 90% methanol was also chromatographed on alumina and gave the same result: thus β-carotene is the main epiphasic pigment in *Phaeodactylum*. There was, however, a faint yellow band on the column below the β-carotene but it was too faint to permit any attempt at identification.

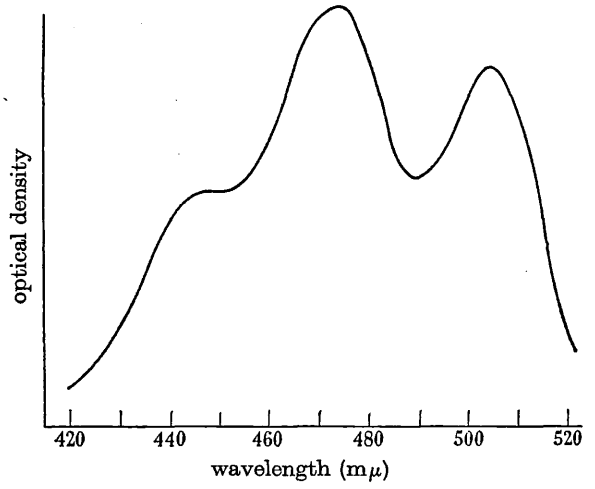

FIGURE 1. Spectral absorption curve of diadinoxanthin in carbon disulphide.

(2) A second fraction, forming a yellow band of pigment, was eluted from the column with 0·25% *n*-propanol in light petroleum. The pigment was hypophasic. Absorption maxima were found at 426 to 428, 447 to 448 and 477 mμ in ethanol, which are in good agreement with those given by Strain, Manning & Hardin (1944) for diadinoxanthin. The absorption spectrum of this pigment in carbon disulphide was also measured. The result is shown in figure 1; maxima were found at 450 to 452, 475 and 506 mμ. We do not know of any published figures with which to compare these, but they are very close to the absorption maxima of lutein, so that it was necessary to verify that the pigment was diadinoxanthin and not lutein. This was done by making a mixed chromatogram with some authentic lutein obtained from *Dunaliella*. The two pigments were easily separated on icing sugar and were eluted with 0·25% *n*-propanol in light petroleum. The lutein came off the column below the diadinoxanthin.

We have carefully rechromatographed the diadinoxanthin to see if we could separate any diatoxanthin from it. Strain *et al.* (1944) found diatoxanthin in all the diatoms which they examined, but we were unable to detect any in our extracts from *Phaeodactylum*. Even the first few drops of diadinoxanthin eluted from the sugar showed a normal absorption spectrum, which would not have been the case if any of the less strongly adsorbed diatoxanthin had been present.

(3) The next fraction was quite the most abundant of the pigments; it formed a broad orange band on the column and was eluted with 0·5% *n*-propanol in light

petroleum. The pigment was hypophasic. In ethanol the absorption spectrum had a single broad peak at 451 to 452 mμ; in hexane absorption maxima were found at 449 and 477 mμ, and in carbon disulphide maximum absorption was at 477 mμ. These data, taken in conjunction with the position of the band on the chromatogram, indicate that the pigment is fucoxanthin.

(4) The fourth fraction formed a narrow yellow band on the column and was eluted with 1 % *n*-propanol in light petroleum. The pigment was hypophasic and showed a single broad absorption maximum at 444 to 446 mμ in ethanol. The position of the pigment on the chromatogram coupled with its simple absorption spectrum indicate that it is an isomer of fucoxanthin, neofucoxanthin B (Strain *et al.* 1944).

(5) A narrow orange band remained at the top of the column and was not eluted by high concentrations of *n*-propanol. The column was extruded and attempts were made to elute the pigment from the icing sugar. Ethanol, water, di-ethyl ether and glacial acetic acid were used, but the pigment would not leave the sugar. After treatment with glacial acetic acid the pigment was no longer visible and its nature remains unknown.

In addition to the pigments considered in detail above, two green fractions were eluted from the sugar column. These were chlorophyll *a*, which came off the column below diadinoxanthin but above β-carotene, and chlorophyll *c* which came off immediately after the neofucoxanthin B.

Dunaliella tertiolecta Butcher

The carotenoids which occur in *Dunaliella tertiolecta* can be considered in two groups: those which are epiphasic and pass through an icing sugar column in light petroleum, and those which are hypophasic and are retained by sugar so that the addition of small amounts of *n*-propanol to the light petroleum is necessary to ensure elution.

Once separated from the hypophasic pigments the epiphase, in light petroleum, was chromatographed on alumina. This produced three fractions which were eluted from the column in the following order.

(*a*) The first fraction, orange in colour, washed through the column with light petroleum to which 0·5 % acetone had been added. The acetone was removed by washing with water and the solution then dried over anhydrous sodium sulphate. The absorption spectrum in light petroleum (b.r. 60 to 80 °C) showed maxima at 477 to 478, at 451 and a shoulder at 425 to 427 mμ. In hexane absorption maxima were found at 450 and 477 mμ. These figures are typical of β-carotene.

A point of interest about this fraction is that when the solvent was evaporated by heating on a water-bath and the pigment taken into carbon disulphide the resulting solution had absorption maxima at 482 to 483 and 508 mμ. These are not typical figures for β-carotene, which has absorption maxima at 485 and 520 mμ in carbon disulphide. In some of our early extractions the pigment was taken into carbon disulphide without first measuring the absorption maximum in light petroleum. The wavelength at which maximum absorption occurred in carbon disulphide led us to suspect a mixture of α- and β-carotenes. When the first few

drops of the fraction were collected from the column after rechromatographing and the absorption spectrum in carbon disulphide determined maxima were found at 477 and 509 mμ. These figures are identical with those given by Karrer & Jucker (1950) for α-carotene. The absorption spectrum in light petroleum does not, however, indicate the presence of α-carotene, and the conclusion is reached that evaporation to dryness before taking the pigment into carbon disulphide causes partial isomerization of the β-carotene. The absorption spectrum of the first part of the fraction when in carbon disulphide is in good agreement with the β-carotene isomer described as pseudo-α-carotene by Gillam & El Ridi (1936) which is probably identical with neo-β-carotene B (Polgar & Zechmeister 1942).

(*b*) The second fraction was eluted with the same concentration of acetone as the first, but was pinker in colour and was clearly separated on the column. The absorption spectrum in light petroleum (b.r. 60 to 80 °C), after washing out the acetone and drying over anhydrous sodium sulphate, showed maxima at 461, 491 and a shoulder at 433 to 435 mμ. In hexane absorption maxima were found at 460, 491 and a shoulder at 435 to 436 mμ, and in carbon disulphide peaks were found at 469, 495 and 527 mμ. The positions of the main peaks (461 mμ in light petroleum, 460 mμ in hexane and 495 mμ in carbon disulphide) are in good agreement with those of γ-carotene, but there are small discrepancies between the minor peaks as recorded by us and those published in the literature (see Karrer & Jucker 1950). These can probably be explained by partial isomerization of the pigment; Zechmeister & Polgar (1945) have produced a series of γ-carotene isomers by treatment with light and heat, and some of these isomers have absorption maxima removed from those of the naturally occurring substance by 8 or 9 mμ.

(*c*) The third band of pigment was eluted from alumina with 5% acetone in light petroleum, but if the alumina was activated it required much higher concentrations, up to 80 or 100% acetone, to elute the pigment successfully. This band was pale yellow in colour and when the solution in light petroleum was examined in the spectrophotometer absorption maxima were found at 445 and 470·5 mμ. In carbon disulphide the maxima were at 472 to 473 and 499 to 500 mμ. The identity of this pigment is uncertain, but the absorption spectrum strongly suggests a carotene oxide; the di-epoxide of β-carotene, for instance, has maxima at 472 and 502 mμ in carbon disulphide and 443 and 470·5 mμ in light petroleum (data from Karrer & Jucker 1950).

After the epiphasic pigments had been separated from the hypophasic the latter were chromatographed on icing sugar, and the column was developed with small amounts of *n*-propanol in light petroleum. In addition to the two chlorophylls (*a* and *b*) which were eluted separately from the column, four carotenoid fractions were collected in the following sequence.

(i) The first fraction, which was yellow in colour, was eluted from the column with 0·5% *n*-propanol. The absorption spectrum in ethanol had peaks at 421, 446 and 475 mμ; in carbon disulphide the peaks were at 448, 474 and 506 mμ. These figures indicate that the pigment is lutein.

(ii and iii) Both these fractions were eluted with 2% *n*-propanol. The absorption spectra are very similar (table 3) and it seems probable that they are both isomers

of violaxanthin. The lower position and clear separation of fraction ii would be in agreement with an identification as taraxanthin, which is isomeric with violaxanthin. We did not always get a clear separation of these two pigments, however, and in the quantitative estimates have included both under violaxanthin.

TABLE 3. ABSORPTION MAXIMA (mμ) OF HYPOPHASIC FRACTIONS ii AND iii FROM *DUNALIELLA TERTIOLECTA*

	ethanol	carbon disulphide
fraction ii	442, 470	443, 469 to 470, 501
fraction iii	417, 441, 470	441, 467 to 468, 499

(iv) The last carotenoid to be eluted from the sugar column formed a narrow bright yellow band which washed through the column with 5 % *n*-propanol. The absorption spectrum had maxima at 414 to 415, 437 and 466 mμ in ethanol and at 437, 462 and 494 mμ in carbon disulphide, indicating that the pigment is neoxanthin (Strain 1938).

Bakers' yeast

No carotenoid pigments were detected in bakers' yeast. On grinding yeast with sand in acetone a pale yellow solution was obtained. The pigment was insoluble in light petroleum, chloroform and di-ethyl ether.

CAROTENOIDS IN MALE AND FEMALE *ARTEMIA*

In cultures of *Artemia*, well fed on *Dunaliella* or *Phaeodactylum*, it is noticeable that females are more orange in colour than males; the orange colour is due to carotenoids. The pigments are located mainly in fat cells (phagocytic storage cells of Lochhead & Lochhead (1941)) of the trunk, limbs, labrum and antennae. The cells of the gut wall are also frequently orange; here the pigment is in the form of granules. Occasionally, particularly in males, the exopodites of the limbs are reddish-orange; there appear to be small granules of pigment in the cells of the exopodites.

The ovary of *Artemia* may be whitish, bluish-green or orange in colour. On treating the whitish and bluish-green ovaries with acetone or acid, the eggs in the ovaries become bright orange. There is, therefore, a carotenoprotein in the eggs while they are in the ovary. This is also true of the bluish-green eggs which sometimes appear in the lateral and median brood pouches of the females. Frequently, however, the eggs in the ovary are bright orange as are the eggs and nauplii in the lateral and median brood pouches, respectively.

Most of our early extracts of carotenoids from adults of *Artemia* were made from animals reared on *Dunaliella*, but our chromatographic results using columns of alumina were confusing and inconsistent. It was only when icing sugar was used to separate the hypophasic pigments that we were able to obtain consistent results with clean separation of the pigments.

Artemia *fed on* Phaeodactylum

Phaeodactylum is the least efficaceous of the three foods we have used in culturing *Artemia*. The animals grew slowly and reached a smaller final size than those fed

on *Dunaliella* or yeast. Egg production by the females was also greatly reduced compared with that of females reared on other foods. Although *Phaeodactylum* is rich in carotenoids the animals did not accumulate these pigments to the extent that they did when fed on *Dunaliella*.

The results obtained from one sample of females are given in table 4. Males produced a similar pattern on the chromatogram but the bands were very faint and faded rapidly on the column. The first fraction to pass through the sugar column was rechromatographed on alumina. A single pink band of pigment appeared near the top of the column and was eluted slowly with 5 % acetone in light petroleum; it showed no signs of separating into two fractions. The pigment was epiphasic on partition between light petroleum and 90 % methanol and had a single absorption maximum at 456 to 458 mμ in hexane and 460 mμ in light petroleum (b.r. 80 to 100 °C); it is probably a keto-carotenoid (see p. 134).

TABLE 4. CAROTENOID PIGMENTS IDENTIFIED IN *ARTEMIA* FED ON *PHAEODACTYLUM*, AFTER CHROMATOGRAPHIC SEPARATION ON ICING SUGAR IN LIGHT PETROLEUM (B.R. 80 TO 100 °C) CONTAINING VARYING AMOUNTS OF N-PROPANOL

			absorption maxima (mμ)		
fraction	colour	% *n*-propanol for elution	light petroleum	ethanol	identification
1	pale orange	0	460	—	keto-carotenoid
2	orange	0·25	468	—	esterified astaxanthin
3	yellow	1·00	—	447, 477	diadinoxanthin
4	pale orange	2·00	448, 476	—	fucoxanthin
5	yellow	5·00	—	446 to 450	neofucoxanthin

Artemia *fed on* Dunaliella

Figure 2 shows the results obtained from a sample of females of *Artemia*. No trace of chlorophyll was found in the extract and so it is unlikely that any undigested plant material was included. Males gave a similar pattern but the relative proportions of the various carotenoids were different (table 5).

When animals containing traces of chlorophyll were examined some additional fractions were found; these were generally present only as faint bands on the column and were not usually identifiable. One such band, however, appeared with some consistency above β-carotene on the alumina. This fraction was epiphasic and showed a single absorption maximum at 453 mμ in hexane. It is probably a keto-carotenoid of the echinenone type. These additional bands may represent intermediates in the formation of astaxanthin from the algal pigments. The fact that they were found only when traces of chlorophyll were present in the extracts indicates that they are formed very soon after the digestion of the algae has begun.

Artemia *fed on yeast*

Both males and females of *Artemia* fed on yeast yielded only small amounts of astaxanthin esters, and no other carotenoids were identified. A faint yellow band, however, appeared on one of our columns but faded before it could be eluted. The

small amount of astaxanthin is probably that which the animals received from the maternal ovary.

A characteristic feature of yeast-fed cultures of *Artemia* is the green colour of the animals, particularly the males. This green colour resides in the blood, as already suggested by Lochhead & Lochhead (1941). We have been unable to

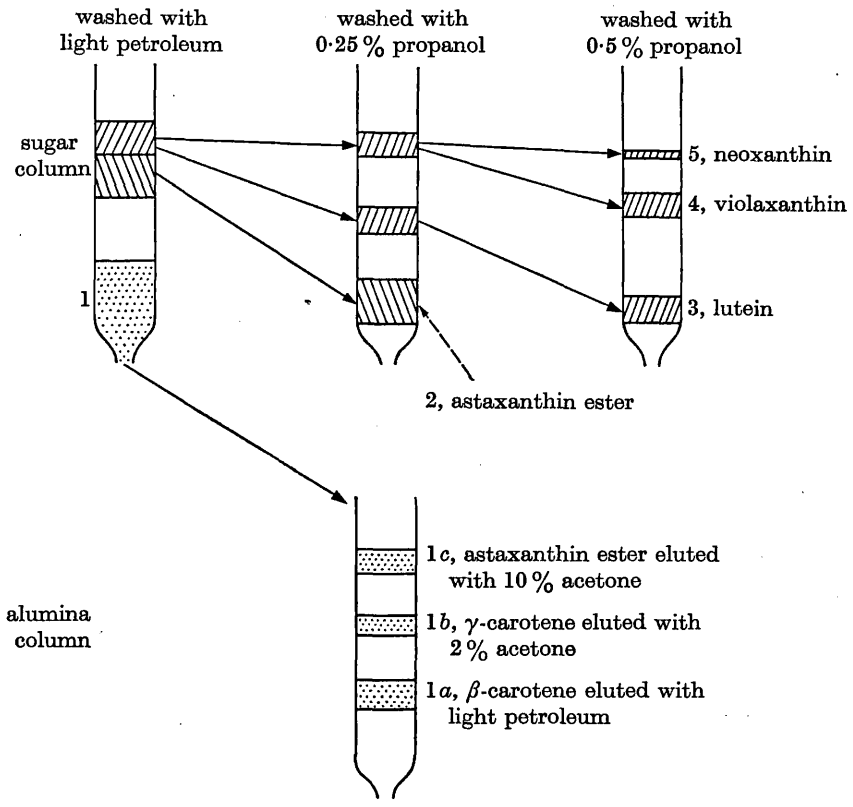

FIGURE 2. Diagram illustrating the chromatographic separation of carotenoid pigments from a light petroleum extract of females of *Artemia* fed on *Dunaliella*.

identify this pigment. A green carotenoprotein is known to occur in the blood of various cladocerans (H. M. Fox 1955; Green 1957). Attempts to identify a carotenoprotein in the blood of green individuals of *Artemia* have been inconclusive, likewise have tests for bile pigments and verdohaemochromes. The identity of this green pigment remains unknown.*

* *Footnote added in proof* 27 *January* 1960. Since this paper went to press our attention has been drawn to the work of Janine Dutrieu (*C.R. Acad. Sci., Paris*, **248**, 1959, p. 2522). She presents her results briefly and we are unable to discuss their significance without further data concerning the way in which they were obtained. One point, however, deserves comment. This is the statement that the green colour in the blood of *Artemia* is probably due to a mixture of chlorophylls *a* and *b*. We cannot accept this on the data she supplies. It is intrinsically improbable that the green colour could be due to chlorophyll, because it is very marked when *Artemia* is reared on yeast, and as yeast lacks chlorophyll, it would imply synthesis of the pigment by *Artemia*. Such a unique phenomenon would be worthy of detailed examination, rather than the brief statement that absorption bands were found 'à 645 et 690 mμ'; the solvent and method of measuring the spectra were not specified.

Discussion

The presence of haematin in the egg shells of *Artemia* has a parallel in the presence of haemoglobin derivatives, namely, bilins and a porphyrin, in the egg shells of birds. In both the pigment is associated with calcification of the shell (Needham & Needham 1930). It is interesting to speculate upon the source of the haematin in the egg shell of *Artemia*. The pale cream-coloured eggs were found in a sea-water culture; we have never seen dark-coloured eggs in sea-water cultures, those eggs which are occasionally laid in this medium are always pale in colour. By contrast, the majority of eggs collected in the field from the edges of salt pans containing concentrated brine are dark brown. This suggests that female brine shrimps in a concentrated medium have more haem available to put into their eggs than females in sea water. It is known that females in concentrated brine have more haemoglobin in solution in the blood than females in more dilute media (Gilchrist 1954). It may be, therefore, that the haem found in the egg shell is derived from the haemoglobin in the blood. In the cladoceran *Daphnia* haemoglobin passes from the blood of the females into the parthenogenetic eggs (Dresel 1948). No haemoglobin has been detected in the eggs or nauplii of *Artemia*, but perhaps haem is passed from blood to the shell glands in the maternal brood pouch and so to the egg shell. Thus the conditions which promote the synthesis of haemoglobin seem also to result in the production of dark brown eggs rich in haematin. In sea-water cultures *Artemia* reproduces viviparously, resting eggs are rarely produced and haemoglobin is rarely synthesized; those eggs which are occasionally produced contain little haematin. There may therefore, be a parallel between the passage of haemoglobin from blood to parthenogenetic eggs of *Daphnia* and the passage of haem from blood to egg shells of *Artemia*.

Artemia is not the only branchiopod crustacean to have a haem pigment in its egg shell. The eggs of *Triops* spp. are surrounded, when in the maternal oviduct, by a bright red liquid containing a protohaemochromogen with its α-band at 563 mμ; this pigment passes into the egg shells but not into the ova (H. M. Fox 1955).

Our results on carotenoids are summarized in table 5. The relative amounts of the different carotenoids identified in *Artemia* are given, together with the carotenoids identified in the food available to the animals.

The carotenoids mobilized by *Artemia* are clearly derived from the food; we have no conclusive evidence of the synthesis of carotenoids by the animal. It has been shown by Lwoff (1927) that most of the carotenoids in the harpacticoid copepod *Idya furcata* (Baird) are derived from these pigments in the animal's food. He suggested at the time, however, that small amounts of carotenoid were synthesized by the copepod, but has since agreed that a biosynthesis has not conclusively been demonstrated (D. L. Fox 1953). Likewise, the cladoceran *Daphnia* is unable to synthesize carotenoids (Teissier 1932); when fed on a carotenoid-free diet the green carotenoprotein characteristic of the parthenogenetic eggs disappeared, but the addition of carotene to the diet resulted in the appearance of the carotenoprotein in the eggs.

Our identification of the pigments of *Phaeodactylum* are in general agreement with the findings of Strain *et al.* (1944) in their studies on diatoms. We did not, however, find any diatoxanthin and only found one neo-isomer of fucoxanthin. This may be due to the fact that *Phaeodactylum tricornutum* is a very atypical diatom (Lewin 1958; Lewin, Lewin & Philpot 1958).

TABLE 5. QUANTITATIVE ESTIMATES OF THE DIFFERENT CAROTENOIDS IDENTIFIED IN *ARTEMIA* AND ITS FOOD, AS % TOTAL CAROTENOIDS

	% total carotenoids		
	females	males	food
(*a*) *Artemia* fed on *Phaeodactylum* pigments identified			
β-carotene	0	—	8
keto-carotenoid	22	—	0
astaxanthin esters	64	—	0
diadinoxanthin	8	—	27
fucoxanthin	3	—	52
neofucoxanthin	3	—	13
(*b*) *Artemia* fed on *Dunaliella*			
β-carotene	8	15	27
γ-carotene	7	12	7
carotene oxide	0	0	1
astaxanthin esters	50	27	0
lutein	13	28	32
violaxanthin	15	8	21
neoxanthin	6	11	12
(*c*) *Artemia* fed on yeast very small amounts of astaxanthin esters			

The carotenoids identified in *Artemia* are not entirely the same either qualitatively or quantitatively as those available in the food; *Artemia* is able to select and alter the pigments taken into the body (table 5). This is known to occur in *Daphnia magna*: when only *β*-carotene and lutein were identified in its food both *γ*-carotene and astaxanthin were present in the animal in addition to those carotenoids found in the food (Green 1957). More recently it has been found that the ostracod *Heterocypris incongruens*, when fed on a diet containing over 80 % of carotenoids in the form of hypophasic xanthophylls, does not accumulate any of these pigments but only epiphasic pigments, namely, *β*-carotene and esterified astaxanthin (Green 1959).

More information is available on the mobilization of carotenoids derived from food in Malacostraca. In the shore crab, *Carcinus maenas*, fed on a carotenoid-free diet both the liver and ovary were lacking in carotenoids; fed on a carotenoid-rich diet, however, first the liver then the blood and finally the ovary acquired a yellow-orange colour due to carotenoids (Abeloos & Fischer 1926). The work on *C. maenas* has been extended by Lenel (1953*a*, *b*, 1955) who showed that the most consistent and well-defined carotenoids in the shore crab are *β*-carotene derived from the food and esters of astaxanthin formed by the animal from precursors in the food, probably *β*-carotene. Feeding experiments (quoted by D. L. Fox (1953))

on the 'spiny-lobster', *Panulirus interruptus*, whose chief carotenoid is astaxanthin esterified in the tissues and free in the shell, have failed to reveal the carotenoid precursor of astaxanthin in *Panulirus*.

In *Artemia* esterified astaxanthin is the only carotenoid in the eggs and nauplii and the most abundant one in the adults. This pigment is absent from *Phaeodactylum*, *Dunaliella* and yeast, the foods of *Artemia* in our experiments. It is seen in table 5 that the proportions of β-carotene and xanthophylls in the animals, particularly females, is much lower than in the food. This suggests either lack of accumulation of these pigments by *Artemia* or that they are taken into the body and used in the formation of astaxanthin.

We have no evidence from our experiments of any intermediate stages in the formation of astaxanthin from xanthophylls. The absence of β-carotene, however, together with the presence of a high proportion of a keto-carotenoid in females of *Artemia* fed on *Phaeodactylum* strongly suggests that the β-carotene of the food is being used in the formation of astaxanthin. That β-carotene is the precursor of astaxanthin has been suggested by Goodwin (1949) as the result of his observations on the distribution of carotenoids in locusts. Further, the role of keto-carotenoids as intermediates in the metabolism of astaxanthin from β-carotene is suggested by de Nicola (1954). She identified the carotenoids in the integument of the echinoderm *Ophidiaster ophidianus*; astaxanthin formed over 50 % of the total carotenoids identified while the other main pigments present were neo-β-carotene B, β-carotene, cryptoxanthin and two keto-carotenoids. One of the latter pigments had an absorption maximum at 460 mμ in light petroleum which is identical with the figure obtained by us for the keto-carotenoid in the females of *Artemia*. More recently Vevers & Millott (1957) have separated a pigment, probably a keto-carotenoid, from the integument of the starfish *Marthasterias glacialis*, which also contains β-carotene and astaxanthin.

We have only identified a keto-carotenoid from females of *Artemia* fed on *Phaeodactylum* and rarely from those fed on *Dunaliella* (see p. 130). A possible explanation of this may be found by considering the reproductive capacity of these animals. Females fed on *Phaeodactylum* produce very few eggs or nauplii whilst those fed on *Dunaliella* produce large numbers of young. The only carotenoid in the eggs and nauplii is esterified astaxanthin. It is likely, therefore, that in the animals fed on *Dunaliella* there is a continuous loss of astaxanthin from the females through the eggs or nauplii liberated from the maternal brood pouch. It has been shown by Goodwin & Srisukh (1949) that the red colour of the copepod *Tigriopus fulvus* Fisch, is due to astaxanthin, both free and esterified. They found that males and females contained about the same amount of the pigment, but that gravid females lost about 50 % of their store of astaxanthin to their eggs. Since, then, in *Artemia* fed on *Dunaliella* there is a rapid turn-over of astaxanthin it is unlikely that any of the intermediate stages in the formation of astaxanthin from β-carotene could accumulate in sufficient amounts to allow of their separation and identification. This probability is strengthened by the fact that we have been able to identify a keto-carotenoid from *Artemia* fed on *Dunaliella* when the extracts were made soon after feeding.

The low proportion of xanthophylls in females of *Artemia* is probably due to lack of accumulation of these pigments. Teissier (1932) found that the addition of carotene and also an extract of lobster carapace to the diet of *Daphnia* resulted in the appearance of a green carotenoprotein in the parthenogenetic eggs; the addition of the xanthophyll lutein to the diet did not produce this result. The ostracod *Heterocypris incongruens* when fed on a diet rich in xanthophylls does not accumulate any of these pigments (Green 1959).

We wish to thank Professor H. Munro Fox, F.R.S. for his critical and helpful reading of our manuscript. Our thanks are also due to Professor N. Millott for the working facilities placed at our disposal. Dr Mary Parke kindly provided the original cultures of algae from which our own stocks were grown and advised us concerning their taxonomy.

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THE HYDROLOGY AND PLANKTON OF THE RIVER SOKOTO

By M. J. HOLDEN

*West African Fisheries Research Institute Field Station, Birnin Kebbi, Sokoto Province, Nigeria**

AND

J. GREEN

Department of Zoology, Bedford College, University of London

(With 12 Figures in the Text)

During the course of work on fish populations in the River Sokoto and its surrounding flood plain it was decided to investigate the hydrology of the river and seasonal abundance of plankton because it was considered that a knowledge of these would permit a better understanding of the populations and life cycles of the fish. The field work on which this paper is based was carried out by one author (M. J. H.) who is responsible for the part of the paper dealing with the collection of data, the physical climate, hydrology and phytoplankton. The second author (J. G.) was responsible for working up the zooplankton collection and writing the part of the paper dealing with it.

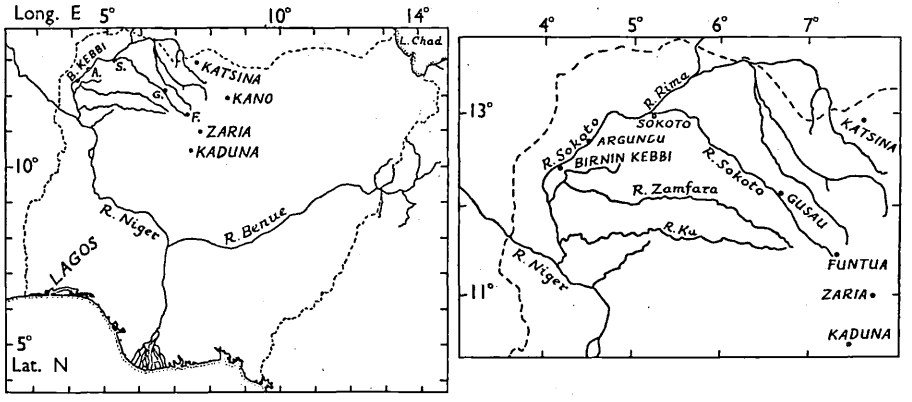

Fig. 1. Nigeria showing the Rivers Niger, Benue and Sokoto, and north-western Nigeria showing the River Sokoto system.

THE AREA AND STATIONS

The River Sokoto is a tributary of the River Niger rising near Funtua in northern Nigeria (Fig. 1). From just upstream of the town of Sokoto the river bed is narrow with steep, rocky sides and is dry, except during the rainy season. To the west of Sokoto the river is joined by the River Rima and from there until it reaches the

* Present address: Fisheries Laboratory, Lowestoft.

River Niger there is permanent water, although the flow in the northern reaches may be slight at the end of the dry season. From Birnin Kebbi at least, and southwards, the river flows throughout the year, reaching a maximum rate of flow in the main channels of approximately 7 km/h during the floods. From approximately the confluence of the Rivers Sokoto and Rima the banks flatten out and there is a large plain of varying width that is inundated when the river floods. A flood plain of this type is called 'fadama' by the Hausa natives. At Birnin Kebbi,

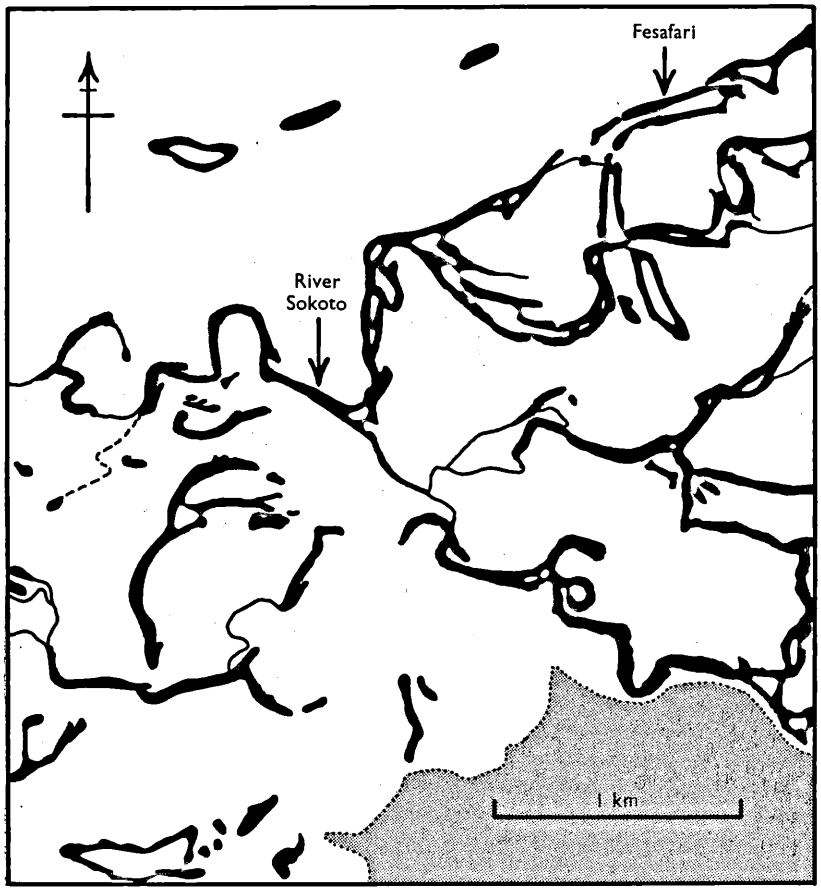

Fig. 2. Flood plain of the River Sokoto to the west of Birnin Kebbi with the two sampling stations shown.

where the present work was carried out, the flood plain is 8 km wide and bounded by an escarpment. At the height of the floods it is entirely covered with water and large areas are cultivated with deep water rice. As the river subsides to its dry season bed, which may be as narrow as 30 m, it leaves a series of pools that are permanent throughout the dry season. Fig. 2 shows that part of the flood plain to the west of Birnin Kebbi which is situated on the escarpment bordering the flood plain.

Two main stations were regularly sampled. One of these was a pool called Fesafari that became cut off during the dry season, and the other was in the main river (Fig. 2). Fesafari was 450 m long at the end of the dry season with a minimum recorded depth of 0·75 m on 5 April 1955 and a maximum depth of 4·5 m on 23 September 1954. The bottom was hard sand-mud mixture in 1954 but changed to soft mud, probably in the flood period of 1955. There was a fringing belt of water lilies that increased with the change over to a muddy bottom but the centre of the pool, where samples were taken, was always free of vegetation. It is linked to the river at the beginning of the wet season when the river reaches 0·7 m depth approximately. It is not possible to state a definite level because there is not a clear division between the pool and swamps that run into the river. The station in the river was selected because it was near an experimental set of gill nets and afforded a long reach in which to make zooplankton tows. The minimum depth was 2·5 m on 5 May 1954, and the maximum depth 7·5 m on 23 September 1954. There was a soft mud bottom and the area was free of vegetation. From time to time samples were taken at other points on the flood plain. These observations were not systematic and will only be referred to in discussion as required.

Sampling was first commenced at Fesafari and only later in the river. At first weekly observations were made, and pH, oxygen and temperature readings taken at the surface and 0·25 m from the bottom, or at 2 m intervals when the depth of the water was 4 m or more. These samples showed that there were little or no differences between depths and that weekly fluctuations were not marked. Thereafter sampling was done monthly as far as possible and at 1 m below the surface only unless the depth of water was less than 1·25 m, when it was done at 0·25 m from the bottom. No distinction has been made in the results between samples from different depths.

It was not possible to take samples always at the same hour and some of the fluctuations shown must be due to daily fluctuations in the values observed. River temperatures were recorded at three-hourly intervals on one occasion but it is possible that other values varied throughout the day; *e.g.* Blanc, Daget & d'Aubenton (1955) found that aquatic vegetation could cause a fluctuation of 2 pH units in 24 h. Therefore no account has been taken of minor fluctuations in considering the overall trends shown by the observations, the repetition in whose cycles from year to year confirms that the results are a true representation of hydrological and planktonic cycles in the river despite the imprecision of some of the data. It was not always possible to maintain the continuity of sampling. From May to October 1955 the writer was on leave. Accidents to equipment that could not be replaced quickly meant that sampling had to be discontinued, at least temporarily, or that observations could not be made as accurately as desired.

ENVIRONMENTAL CONDITIONS

(*a*) *Rainfall*

Rainfall figures were copied from those kept at the Government Hospital, Birnin Kebbi. Readings were taken at 0900 h each day to the nearest 0·01 in.

and have been converted to the nearest 0·1 cm. There is one wet season each year (see Fig. 3). The rains normally start in April but may not begin until May. There is no set pattern to the rains as can be seen by comparison of the histograms for the $3\frac{1}{2}$ years. The earliest rains especially do not fall steadily but are torrential and are accompanied by violent thunderstorms. The heaviest rainfall occurring in any 24-hour period during the time of these observations was 11·4 cm. This caused a rise in river level of 0·18 m.

(*b*) *Air shade temperatures*

Those from February to November 1954 were read daily on a maximum and minimum thermometer by the Malaria Control Project personnel to the nearest degree F. Monthly means have been calculated and converted to the nearest 0·5° C below. From March 1956 to July 1957 daily temperatures were read at the Field Station on a maximum and minimum thermometer to the nearest 0·5° C.

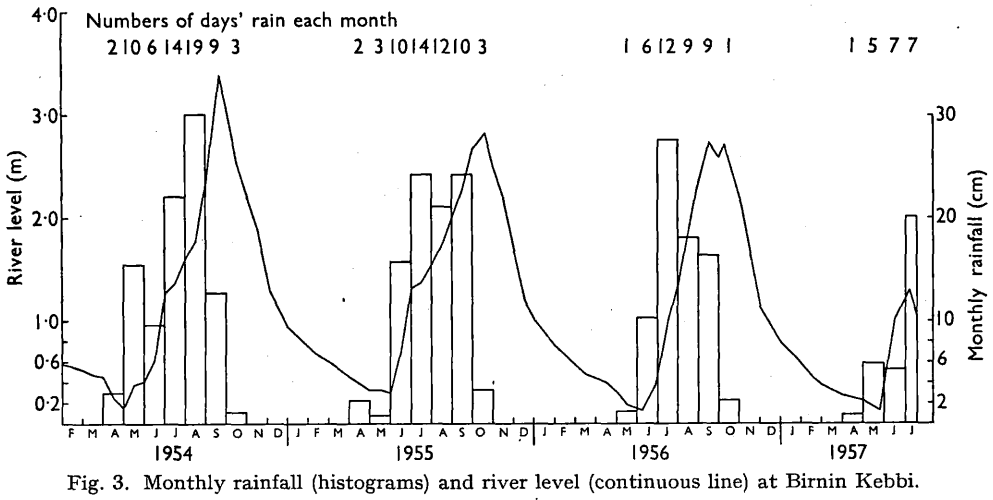

Fig. 3. Monthly rainfall (histograms) and river level (continuous line) at Birnin Kebbi.

A Stevenson screen was not used at either recording point, a thatch cover being used. The results are given in Fig. 4. There is a hot season preceding the rains, which cause a rapid fall of temperature at their onset. With the cessation of the rains maximum temperatures start to rise until the beginning of the harmattan, a dry, dust-laden wind from the Sahara. Minimum temperatures fall rapidly, 9·5° C being the lowest recorded in January 1957. Maximum temperatures fall owing to the wind and dust obscuring the sun. The harmattan is followed by the hot season.

(*c*) *Water level*

Levels were measured on a depth gauge situated on the edge of the flood plain. Readings were taken to the nearest inch and converted to the nearest 0·01 m. For periods when the author was absent from Birnin Kebbi the records kept by the Native Authority were copied. The results are shown in the graph in Fig. 3. The

first rains do not affect the river level, which continues to fall, because they are absorbed by the parched soil or evaporated by the sun. The maximum height of the floods is not reached at the peak of the rains but later. This is due to rain in the

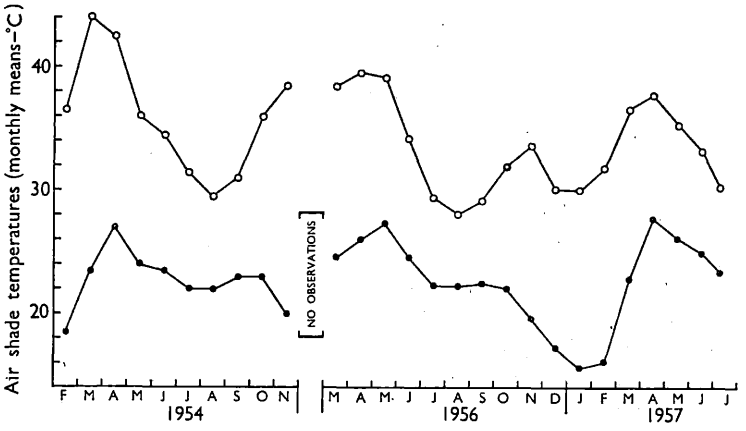

Fig. 4. Minimum and maximum air shade temperatures at Birnin Kebbi.

upper reaches of the River Sokoto near Funtua where the rains finish later than at Birnin Kebbi, and also to the flooding of the Niger which hinders the escape of water from the River Sokoto.

(*d*) *Water temperature*

Temperatures were read to the nearest 0·1° C holding the thermometer in a

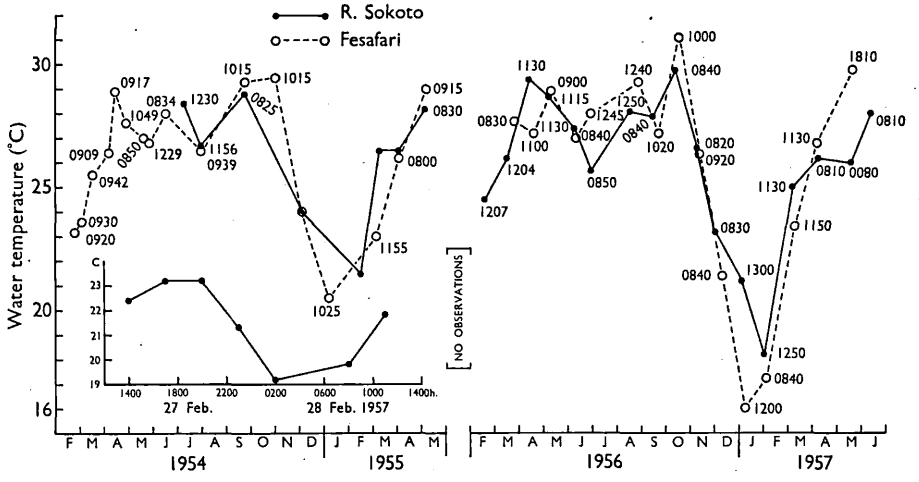

Fig. 5. Water temperatures in the River Sokoto and in Fesafari. Where available, the time of sampling is shown by each point. Inset shows daily fluctuation in river temperature for 27-28 February 1957.

bottle containing the water sample until a steady temperature was reached. The results are given in Fig. 5 with an inset showing the daily fluctuations over one 24-hour period, 27-28 February 1957. So that a comparison between the two

stations can be made the times of sampling have been given. In only one instance is there any large difference that is inexplicable on a time difference basis and that is in January 1957 and this was due to a rapid fall in air temperature between the two dates. The river was sampled on 3 January 1957 and the minimum air temperature for that day was 17·5° C and the maximum 31·0° C. Corresponding temperatures for 8 January 1957 when Fesafari was sampled were 10·5° C and 23·0° C. Most months did not show such large daily fluctuations in maximum and minimum air temperatures and the observed water temperatures are therefore a fairly true record of annual fluctuations.

For most months the water temperature follows the air minima. This is due to the fact that temperatures were taken usually before 1200 h, so that observations were nearer minimal rather than maximal temperatures. During the rainy season they approached maximal air temperatures although there was no difference in the times of taking temperatures. Macan (1958) explains this effect as being due to evaporation which reduces water temperature in dry weather.

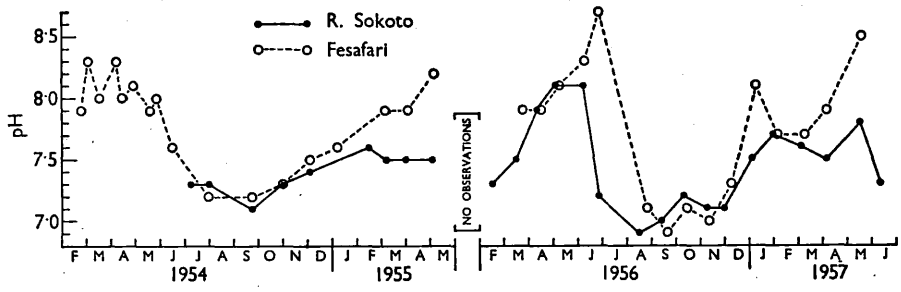

Fig. 6. Seasonal fluctuations in pH in the River Sokoto and Fesafari.

(*e*) *Hydrogen ion concentration and alkalinity*

During 1954 and 1955 a B.D.H. Capillator set was used in the field. During 1956 and 1957 a Lovibond Nessleriser was used and read in the laboratory. Comparisons showed that there was no change in pH during the time it took to transport samples back to the laboratory. Results are given in Fig. 6. The pH rarely fell below 7·0 owing to the calcareous soil through which the river flows at Sokoto. Alkaline waters are not typical of Africa. The high pH level at the end of the dry season is due to evaporation of the water and concentration of calcium salts (see Table 1). It rises higher in Fesafari because this is an isolated body of water, and this higher value is maintained longer until the pool is relinked with the river, when both values fall together owing to the influx of stagnant water of lower pH from the swampy areas and the dilution of calcium salts by rain water.

Alkalinity was measured with N/10 HCl prepared from B.D.H. standard solutions using methyl orange as an indicator. The results are given in Fig. 7 and agree with those for pH.

(*f*) *Oxygen concentration*

In 1954 this was determined by the standard Winkler titration. In 1956 and 1957 a Lovibond Nessleriser was used. The results are given in Fig. 7. The main factor affecting oxygen concentration appears to be wind. The rains are preceded by strong winds and the harmattan is accompanied by them and their wave action causes the May-June and January peaks. The concentration rises higher in Fesafari owing to its lesser depth. The low levels in July and August are due to the washing out of stagnant swamp water by the rains. The August-September peak is probably caused by an outburst of filamentous algae that occurred in all three years and was noted by its presence in the zooplankton samples and by its deposition on gill nets. The times of these observations are marked on Fig. 7. There is a delay between the first appearance of these algae and the rise in oxygen concentration due to a masking effect by the swamp water. Calculation of oxygen concentration as percentage saturation showed that temperature has very little effect on observed oxygen concentration.

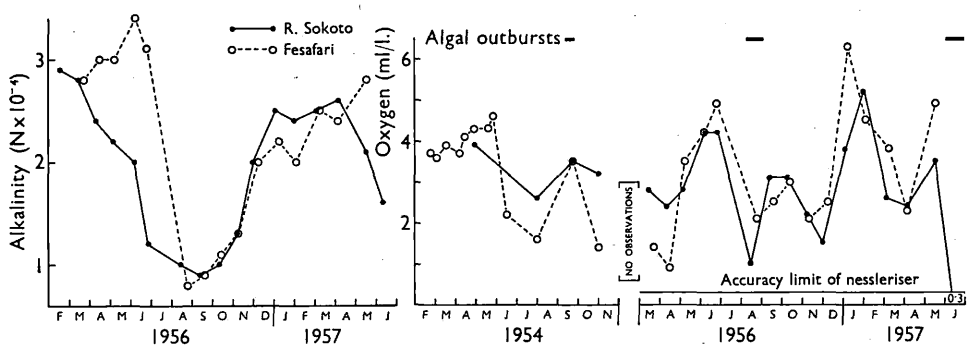

Fig. 7. Seasonal fluctuations in alkalinity and oxygen concentration in the River Sokoto and Fesafari. Outbursts of filamentous algae are indicated by horizontal bars.

(*g*) *Transparency*

This was measured with a 10 cm Secchi disc. The results are shown in Fig. 8. The main factor affecting water transparency is the zooplankton concentration which shows an incomplete inverse correlation with transparency. The transparency of Fesafari is for the most part greater than in the river although the zooplankton concentration is higher in the former. Fine silt held in suspension in the flowing waters of the river may be the cause of this.

The very high transparency of the water in October-November is due to the cessation of the rains when no further silt is being carried into the river off the land and before the plankton has started to increase. Daget (1957) found a similar increase in the Middle Niger.

(*h*) *Chemical composition*

Water analyses were done by the Government chemist, Kaduna. There was a delay of at least 48 hours between the time of collecting the samples and their

Table 1. *Chemical analyses (parts per million)*

	River Sokoto							Fesafari			
	13 Feb. 1956	2 Apr. 1956	4 June 1956	7 Aug. 1956	1 Oct. 1956	10 Dec. 1956	4 Feb. 1957	13 Feb. 1956	2 Apr. 1956	4 June 1956	4 Feb. 1957
NH_4	<0·004	0·004	0·004	0·102	0·05	0·12	0·012	0·093	0·14	0·299	0·012
Mg	7·0	10·0	9·0	2·0	3·5	4·0	9·0	7·0	10·0	9·0	7·0
Ca	32·0	42·0	32·0	12·0	10·0	28·0	32·0	28·0	30·0	32·0	28·0
Fe^{++}	0	0·2	<0·2	–	–	–	–	0	0·2	<0·2	–
Fe (total)	0·5	1·4	0·8	3·0	0·24	0·2	0·50	0·28	0·36	0·6	0·08
Mn	<0·025	0·075	<0·025	<0·025	<0·025	<0·025	<0·025	<0·025	0·025	<0·025	<0·025
Na	8·7	5·7	7·5	7·8	3·1	11·0	10·8	3·2	8·8	16·6	12·4
K	2·8	3·0	13·2	4·0	5·2	6·3	6·8	5·2	5·2	13·2	7·4
F	0·4	0·5	0·7	0·2	0·1	0·3	0·5	0·4	0·5	0·7	0·5
Cl	6·0	15·0	8·0	0·5	1·0	4·0	6·0	6·0	15·0	12·0	5·0
NO_2	0·046	0·07	0·007	0·01	<0·003	<0·003	0·003	0·231	0·03	0·03	<0·003
NO_3	0·44	0·132	<0·132	0·22	<0·11	<0·11	<0·025	<0·132	0·132	<0·132	<0·025
PO_4	<0·1	0·1	<0·1	<0·1	0·1	<0·1	<0·1	<0·1	0·1	<0·1	0·1
SO_4	0	tr	tr	0	0	0	tr	0	tr	tr	0
HCO_3	77·8	74·7	58·9	25·9	29·0	65·6	74·7	70·2	83·9	102·2	68·6
CO_3	0	0	0	0	0	0	0	0	0	0	0
SiO_2	20·0	16·0	10·0	10·0	14·0	12·0	11·0	16·0	12·0	12·0	14·0
Alkalinity, as $CaCO_3$	127·5	122·5	96·5	42·5	47·5	107·5	122·5	115·0	137·5	167·5	112·5
Hardness ($CaCO_3$)	75·0	85·0	95·0	27·5	34·0	79·0	103·0	60·0	70·0	95·0	85·0

tr, trace; –, no analysis.

arrival at Kaduna. The results must therefore be treated with caution because of possible bacterial and other changes, but the overall results agree with observations made at the Field Station. No separate samples were taken from Fesafari during the floods because other observations show that during this period there are no significant differences between the two stations.

The results are given in Table 1. The water is generally poor in nutrient salts. Sulphate is almost completely absent. This is typical of African inland waters (Beauchamp 1953). Phosphate is practically absent, while nitrate, nitrite and ammonia are all at low levels. Iron is present, the local soil having abundant laterite deposits. The rise in iron concentration in the rains is probably due to the

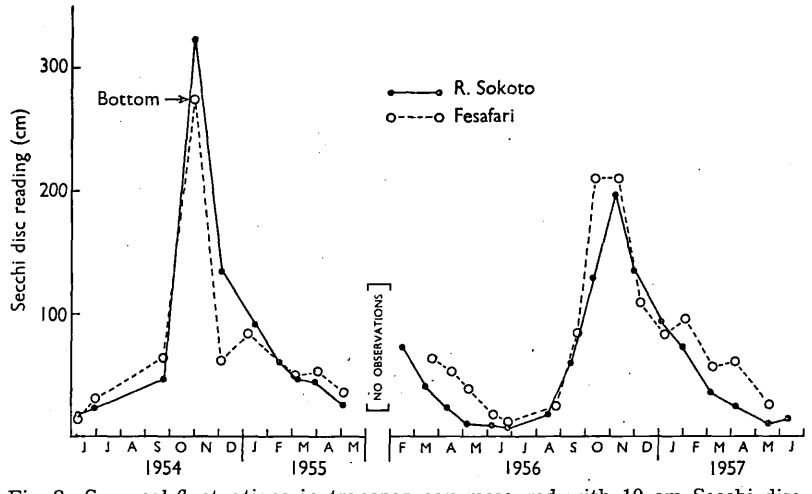

Fig. 8. Seasonal fluctuations in transparency measured with 10 cm Secchi disc in the River Sokoto and Fesafari.

release of iron at low oxygen tensions (Mortimer 1949). Calcium is abundant and total alkalinity correlates with pH observations.

(*j*) *Phytoplankton*

During 1954 and 1955 a few observations were made using a phytoplankton net. During 1956 and 1957 100 ml samples were filtered through a membrane filter using a Steffi-Cole's filter funnel and the phytoplankton preserved in 70% alcohol. The samples were examined by Dr E. Lind of Makerere College. It is difficult to draw conclusions from the results (Table 2) but it would appear that Fesafari has a higher productivity than the river, and that at both stations the productivity was greater in 1956 than in 1957. This can be partly associated with the concentration of nutrient salts. In February 1956 the concentrations of ammonia, nitrite and nitrate at both stations were higher than those in February 1957, and they were higher in Fesafari than in the river. This association does not extend into 1957 when there was little or no difference between the concentrations of these ions at the two stations despite the higher productivity in Fesafari. The low phyto-

plankton concentrations during the floods are due to the diluting effect of the rains both on the phytoplankton itself and on nutrient salts.

Records of a fairly superficial examination of phytoplankton samples from Fesafari on 6 January 1954, 6 January 1955 and 1 April 1955 and from the river on 1 April 1955 indicate that the same pattern of production is followed each year. The observation of 'Nothing' on 7 June 1956 suggests rapid fluctuations in the phytoplankton. There is a similar observation on 12 September 1956 whereas on 3 September 1956 filamentous algae had been recorded as abundant on gill nets. In 1954 this outburst occurred in the river and Fesafari at the same time, so that

Table 2. *Phytoplankton analyses*

	RIVER SOKOTO
16 Feb. 1956	Diatoms, *Cosmarium* occ.
6 Jun. 1956	Scattered cells of Myxophyceae
12 Sep. 1956	Nothing
7 Nov. 1956	Nothing
1 Dec. 1956	Nothing
3 Jan. 1957	Nothing
1 Feb. 1957	Nothing
4 Mar. 1957	Scattered cells of Myxophyceae
6 Apr. 1957	Scattered cells of Myxophyceae
	FESAFARI
23 Mar. 1956	*Synedra* c., *Melosira* occ., *Pediastrum* occ.
10 Apr. 1956	*Aphanocapsa, Merismopedia,* diatoms, *Pediastrum, Scenedesmus, Oscillatoria* fragments, all frequent
7 May 1956	*Aphanocapsa, Chroococcus, Melosira* (not common)
7 Jun. 1956	Nothing
25 Jun. 1956	*Melosira* and other diatoms, *Euglena, Phacus, Trachelomonas, Scenedesmus*
24 Aug. 1956	Nothing
19 Sep. 1956	Nothing
13 Oct. 1956	Nothing
10 Nov. 1956	Nothing
8 Jan. 1957	Nothing
3 Feb. 1957	Nothing
7 Mar. 1957	Occ. diatoms and *Euglena*
5 Apr. 1957	*Melosira, Euglena,* diatoms, *Aphanocapsa, Dictyosphaerium,* all occ.
17 May 1957	*Melosira, Euglena, Coelastrum, Oscillatoria,* all occ.

c. = common
occ. = occasional

their presence would have been expected in the samples of 24 August 1956 and 19 September 1956 both of which record 'Nothing'. While the coincidence of the samples from year to year and their similarity to the zooplankton results indicate that the picture of phytoplankton production is probably correct, the results cannot be considered a very detailed picture of this production.

ZOOPLANKTON

Timed tows were made using a net of 5 meshes per mm with mouth 30·5 cm diameter over an approximately fixed distance of 300 m. It was not always possible to make the tow over the same distance owing to obstructions such as long lines in

the water, nor at the same speed because high winds blew the boat along rapidly, but as far as possible tows were made comparable. They were made with the net just below the surface of the water; no meter was used. In Fesafari the tow was made from the north-east end of the pool in a westerly direction down the middle line into the southerly arm. In the river the tow was made from the westerly end of the reach marked (Fig. 2) upstream in a south-easterly direction.

(*a*) *Method of counting*

The sample in each tube was made up to 30 ml with 5% formalin, and the whole sample was thoroughly mixed. A sub-sample of 0·5 ml was quickly taken with a wide-mouthed graduated pipette and ejected into a petri dish containing a thin film of water. The water in the petri dish was agitated so that the animals were evenly distributed. The base of the petri dish had lines ruled with a diamond at intervals just a little under the field diameter of a binocular microscope with a magnification of 30 diameters, to aid in systematic searching and counting. All the animals in the sub-sample were counted.

The volume of animal material in each sample was estimated by allowing the sample to settle for 5 days and then measuring the height of material in the tube to the nearest millimetre. All the tubes were of the same diameter so that the volume of settled material could easily be calculated. When the sub-sample was taken for the count, an estimate was made of the amount of detritus and plant material in the sample. This was estimated after examination of at least ten fields of the microscope. The estimate was subjective, and attempted to assess the amount of non-animal material to the nearest tenth by volume, but because of the subjective method no claim to accuracy greater than one-fifth by volume is made. The estimate of non-animal material was always made before the count to avoid any bias due to previous knowledge of the number of animals in the sample. The estimated percentage of detritus was deducted from the volume of material in the tube to give an estimate of the volume of zooplankton.

In the results which follow counts have been presented in terms of major groups, and not divided into species. The systematics of some of the species are complicated and since they are not relevant to the purpose of the present paper they have been left for more detailed treatment elsewhere.

(*b*) *Rhizopoda*

The most striking feature of the seasonal occurrence of the rhizopods in the Sokoto is that they appear most abundantly at the times when other groups are relatively sparse. They are generally more abundant in the river than in Fesafari. According to Kofoid (1908) the appearance of testaceous rhizopods in river plankton can be due to two different causes. The first of these is the action of the river as it rises and flushes out rhizopods from swampy, heavily vegetated areas. The second cause is the production of light-shelled planktonic forms by species which are not normally planktonic. In the Sokoto the level of the river appears to be of overriding importance. Fig. 9 shows that in the river the abundance of rhizopods

in the plankton is greatest when the river is rising to its maximum height, and the lowest numbers occur when the river is very low. The relatively high numbers in the plankton of the river in March and April of 1956 may be due to the production of plankton forms by non-planktonic species; this question will be examined in more detail when the systematics of the Sokoto species have been worked out. When the relative importance of the rhizopods in relation to the rest of the zooplankton is considered there is a very good agreement with river level, as can be seen from the two lines in Fig. 9, where the rhizopods are plotted as a percentage of the total zooplankton. From September to December rhizopods formed between 40 and 50% of all the zooplankters.

Most of the species in the Sokoto belong to the genera *Arcella* (*A. vulgaris* Ehrenberg, *A. macrostoma* Penard, *A. dentata* Ehrenberg) and *Difflugia* (*D. corona* Wallich, *D. scalpellum* Penard, *D. acuminata* Ehrenberg, *D. limnetica* Levander). These species form the greater part of the rhizopods in all the samples, but various

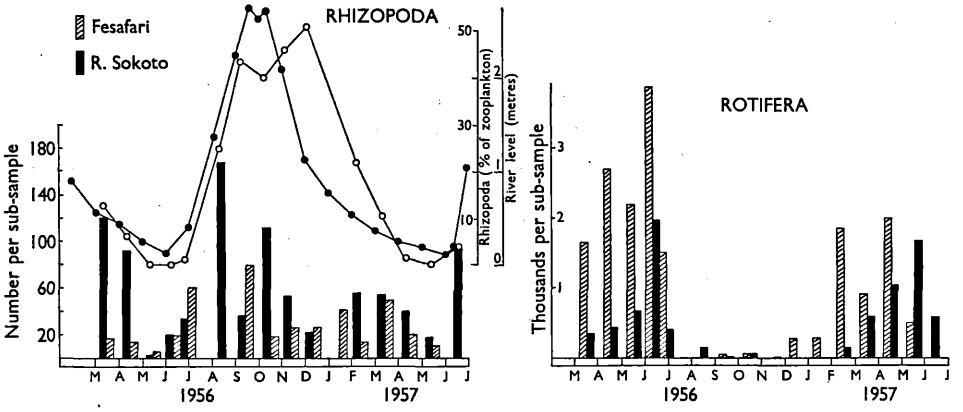

Fig. 9. Seasonal variation in abundance of Rhizopoda and Rotifera in the River Sokoto and Fesafari. Histograms give numbers per sub-sample. —o—o—, Rhizopoda as a percentage of the total zooplankton. —•—•—, height of the river.

other species are also found, such as *Centropyxis aculeata* Stein, *Euglypha acanthophora* (Ehrenberg) and *Lesquereusia spiralis* (Ehrenberg).

(*c*) *Rotifera*

The seasonal fluctuations in total rotifers are shown in Fig. 9. In both 1956 and 1957 the maximum production of rotifers was earlier and greater in Fesafari than in the river. The numbers in Fesafari in 1957 did not reach the same height as in 1956, but the numbers in the river did not differ significantly in the two years. The population peaks are produced by the following four species: *Brachionus caudatus* Barrois & Daday, *B. falcatus* Zacharias, *Keratella tropica* (Apstein), and *Tetramastix opoliensis* Zacharias. Of these the first is generally the most numerous, and *Brachionus falcatus* is more important in Fesafari than in the river. *Asplanchna brightwelli* Gosse forms up to 17% of the rotifer population in the river in April.

When the river is in flood the importance of these species is greatly reduced, and numerous other species appear, though the total number of rotifers is very much lower than in the dry season. The species appearing with the floods are dominated by several species of *Lecane*, including the following: *L. bulla* (Gosse), *L. ungulata* (Gosse), *L. leontina* (Turner) and *L. curvicornis* (Murray). Other species which occur frequently at this time include *Trichocerca bicristata* (Gosse) and *Macrochaetus collinsi* (Gosse).

(*d*) *Cladocera*

The dominant species in this group are *Diaphanosoma excisum* Sars, *Moina dubia* Guerne & Richard, and *Ceriodaphnia cornuta* Sars. These species are responsible for the population peaks shown in Fig. 10. In the River Sokoto the cladoceran population built up to high peaks in June in both 1956 and 1957, while in Fesafari 1956 produced a high peak, but the population failed to develop in 1957. In the period from August to March, when Cladocera are scarce, there is a change in the species which are present. The species listed above as responsible for the population peaks are rare or absent, while various others, which generally live on or near the bottom, are found. *Grimaldina brazzai* Richard, *Macrothrix triserialis* Daday, *Leydigia ciliata* Gauthier, *Alona verrucosa* Sars and *Chydorus barroisi* (Richard) have been recorded. Even the mud dwelling *Iliocryptus spinifer* Herrick appeared in some of the samples. The occurrence of such bottom-dwelling species in the plankton is associated with the swollen state of the river; the increased flow swirls such creatures up from the bottom into the plankton.

(*e*) *Copepoda*

The fluctuations in numbers of copepods throughout the year are shown in Fig. 10. The Calanoida were most abundant in Fesafari in June 1956, when the pool had five times as many as the river. In 1957 the calanoid population remained sparse, and the numbers in the river were a little higher than in Fesafari. Calanoids disappear completely from the samples during the period from August to November in Fesafari and for even longer in the river, where they are not found again until April. The Cyclopoida contrast with the Calanoida in being more abundant in the river than in Fesafari in June 1956. This group of copepods persists throughout the floods, but in greatly reduced numbers. The maximum numbers in both the river and the pool were lower in 1957 than in 1956. A few copepodid stages of a lernaeid were found in the samples (Table 3). The adults of this family are parasitic on the gills of fish, but the larvae are actively swimming members of the plankton, and seek out new hosts. The number of copepodid stages found was too small to enable any conclusions to be drawn about seasonal occurrence.

All copepod nauplii were counted together. They were found to persist throughout the year in Fesafari, though the numbers were very low during the floods. In the samples from the river no nauplii were found between the August sample and the December sample. The time of maximum abundance of nauplii coincides

with the maximum of the later stages. This is in agreement with the findings of Kofoid (1908) in the River Illinois, but disagrees in some respects with the work of Cohn (1903) who found that the maxima of nauplii preceded the maxima of later stages in two cases out of three in certain German lakes. Steur (1901), working on the Danube, found the maxima of nauplii coinciding with those of later stages on two occasions out of three. There is no real conflict in such results because an abundance of nauplii may be due to two causes. The first is a large hatch from resting eggs which may have been laid in the previous year. Nauplii may then be abundant without any adults being present. An example of this type has been described by Comita (1956) who studied a population of *Limnocalanus johanseni* in an Arctic lake. The second source of abundant nauplii is from eggs hatching from the sacs of mature females; in this case one would expect nauplii to be most

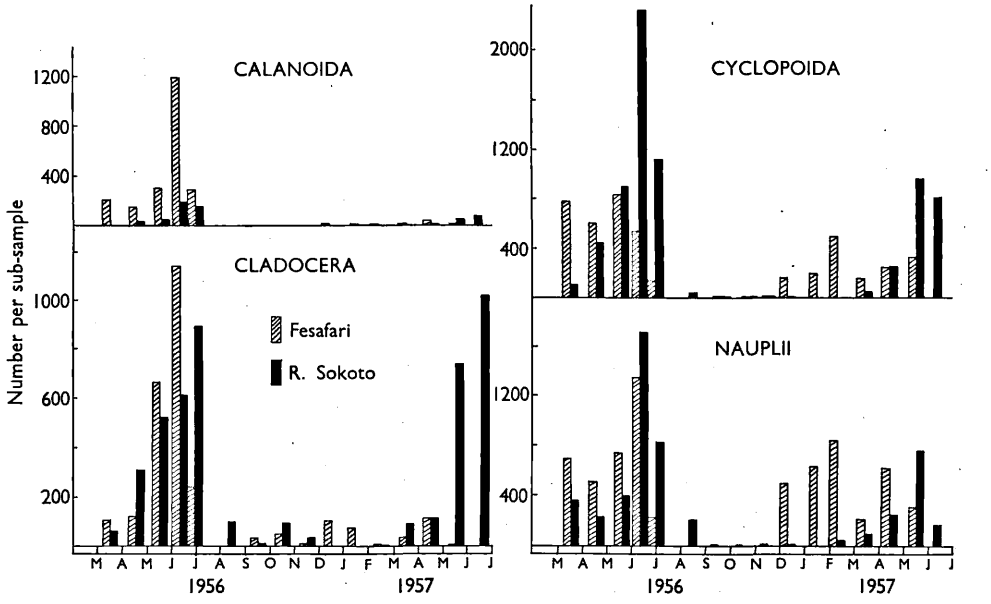

Fig. 10. Seasonal variation in abundance of Cladocera and Copepoda in the River Sokoto and Fesafari.

abundant when the adult population was most abundant. This is the situation found in the Sokoto.

(*f*) *Ostracoda*

The freshwater ostracods are mainly bottom dwellers, and their appearance in the zooplankton is adventitious. They are commonest in the Sokoto, as one would expect, when the river is in flood (Table 3).

(*g*) *Other groups*

The only truly planktonic species included under this heading is the freshwater medusa *Limnocnida tanganyikae* which appeared in Fesafari in January, February, March and April 1957.

The rhabdocoele turbellarians are mainly benthic in habit, but their occurrence

in the Sokoto does not show any marked correspondence with the floods. Some species are known to swim fairly well, and Kofoid records one occasion when 19 250 per cubic metre were found in the River Illinois. No such outbreaks were found in the Sokoto (Table 3).

Table 3. *Numbers of zooplankters in sub-samples from net hauls*

	Rhizopoda	Rhabdocoela	Rotifera	Cladocera	Cyclopoida	Calanoida	Nauplii	*Lernaea* sp.	Ostracoda	Insect larvae	Hydracarina
R. SOKOTO											
1956											
14 Mar.	121	–	335	62	103	–	360	–	1	–	1
10 Apr.	93	1	438	310	441	25	234	–	–	1	3
5 May	3	–	659	521	897	54	392	–	1	1	–
6 June	19	–	1982	608	2316	191	1714	–	–	–	1
26 June	34	–	417	886	1111	145	830	1	1	–	–
14 Aug.	168	–	159	96	40	–	201	–	4	2	5
12 Sept.	36	2	22	10	6	–	–	–	1	2	3
9 Oct.	112	–	56	87	15	–	–	–	10	–	3
7 Nov.	53	–	19	34	5	–	–	–	–	1	3
1 Dec.	21	–	4	–	8	–	6	–	–	1	1
1957											
1 Feb.	56	–	140	5	9	–	41	–	–	1	–
4 Mar.	54	–	578	86	52	–	99	–	1	–	–
6 Apr.	40	–	1050	112	243	1	255	–	–	–	–
16 May	18	1	1695	737	965	43	768	2	–	–	1
10 June	98	2	580	1013	825	84	171	1	–	2	3
FESAFARI											
1956											
23 Mar.	15	–	1649	104	781	198	702	–	–	–	–
18 Apr.	14	–	2692	719	599	142	517	–	–	1	–
7 May	6	–	2212	663	829	298	743	–	–	–	2
7 June	20	–	3890	1130	538	1190	1354	–	–	–	2
25 June	61	–	1507	273	127	285	299	1	2	–	–
19 Sept.	80	–	42	31	6	–	1	–	–	3	3
13 Oct.	18	3	50	46	4	–	1	1	2	2	4
10 Nov.	26	–	4	11	3	–	4	1	–	–	1
8 Dec.	25	1	276	99	164	9	476	–	–	–	2
1957											
8 Jan.	42	–	277	72	187	4	643	1	–	–	–
3 Feb.*	14	–	1838	11	494	8	856	–	–	1	1
7 Mar.†	50	1	889	33	149	4	216	–	–	1	–
5 Apr.	21	1	1996	112	238	30	635	1	–	–	1
17 May	11	–	486	4	323	2	312	–	–	–	–

* Also 2 *Limnocnida* medusae. † Also 1 *Limnocnida* medusa.

Insect larvae and Hydracarina occur sporadically in the samples throughout the year, but are most abundant during the floods.

(*h*) *Relative importance of zooplankton groups*

The figures showing seasonal distribution indicate that all the plankton groups except the rhizopods are most abundant in May and June, and it can be seen in Table 3 how the numbers of each group compare with each other. Another feature of importance is the composition of the plankton throughout the year; this is

shown in Fig. 11, where each group is plotted as a percentage of the total. It is evident from these figures that there are two distinct seasonal plankton assemblages. From January to July the Copepoda and Rotifera form the greater part of the assemblage, while from July to December the Rhizopoda increase in importance. The Cladocera also show a relative increase in importance, centred about October. We have already shown that there is a change in the cladoceran species present during the floods, and that the species present at this time are mainly bottom-dwelling species which as a rule are much smaller than true planktonic Cladocera. This means that in spite of an increased relative numerical importance the decrease in Cladocera at the end of June is even more marked, in terms of food material available for fish, than is shown in the histograms.

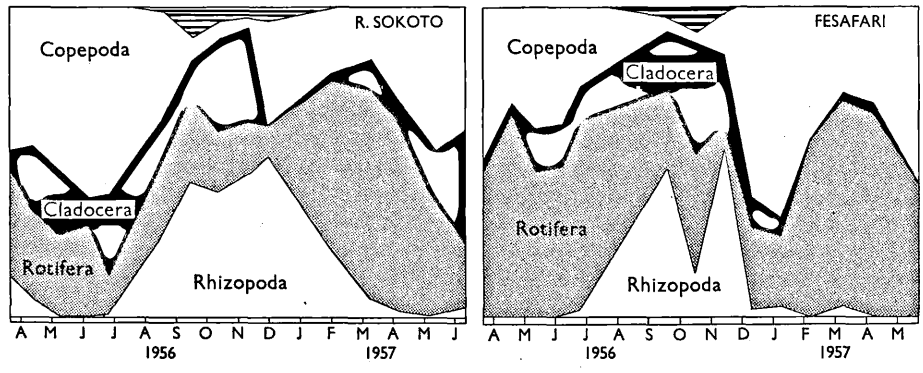

Fig. 11. Seasonal variation in the composition of the zooplankton of the River Sokoto and Fesafari. Each group is plotted as a percentage of the total zooplankton.

The occurrence and relative importance of non-planktonic groups in the samples coincide with the period of flooding. The duration of occurrence of these forms is longer in the river than in Fesafari, due no doubt to an earlier reduction in the rate of flow through Fesafari as the floods decline.

(*j*) *Total zooplankton*

Consideration of the total zooplankton has been left until the end so that the seasonal fluctuation of the whole can be viewed against the background of the fluctuations of the constituent groups. Seasonal fluctuations are shown in Fig. 12. The most obvious and striking feature is the great drop in both number and volume with the onset of the floods. The increase in zooplankton begins much earlier in the pool than in the river so that in the early part of the year, from January to May, there is about twice as much zooplankton in the pool as in the river. In both the pool and the river there was a much greater production of zooplankton in 1956 than in 1957. The reduction in production in the latter year was more severe in Fesafari than in the river, and the failure to develop can be attributed in particular to the Cladocera and Calanoida. It is noteworthy that both these groups feed mainly on small planktonic algae, while the Cyclopoida have a much greater range of diet (Fryer 1957). The amount of phytoplankton in Fesafari in 1957 was

noticeably smaller than in 1956, and this may be attributed to the lower concentrations of ammonia, nitrates and nitrites. In the river the reduction in numbers in 1957 cannot be attributed to the failure of the Cladocera; they were just as abundant as in 1956. There is also only an insignificant reduction in the Rotifera in the river. The copepod population is mainly responsible for the lower figures in the river in 1957. It is difficult to find any reason why this group, and particularly the Cyclopoida, should fail when the Cladocera increased to the level of the previous year.

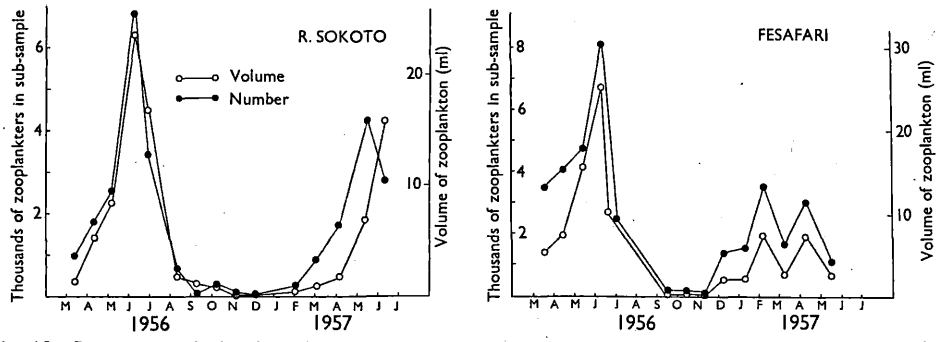

Fig. 12. Seasonal variation in volume and numbers of zooplankton in the River Sokoto and Fesafari.

The two methods of estimating total zooplankton agree very closely. On only one occasion did the total number in the sample fall while the volume of the sample continued to rise. This was in the river from May to June 1957. The explanation for this is seen when the frequency histograms for the various groups are examined. The rotifers and nauplii decrease markedly at this time, while the Cladocera increase. Each cladoceran is as large as 200 rotifers, so that a small increase in Cladocera could more than compensate, in terms of volume, for a considerable reduction in the rotifer population.

Table 4. *Maximum numbers recorded per m³*

	Cladocera	Copepoda	Rotifera
River Sokoto	2700	7500	6000
Fesafari	3400	5100	11 700
Blue Nile (Rzóska *et al.* 1955)	108 000 (all Crustacea together)		202 000
Illinois (Kofoid 1908)	440 000	1 000 000	5 000 000

In the main part of this paper no attempt has been made to express the abundance of the various zooplankters in absolute quantitative terms. The sampling method was too crude to allow this. A rough calculation can, however, be made to get an idea of plankton production in the Sokoto to compare with other localities. In this calculation there have been no corrections for the net filtering rate, or for variation in the time taken to complete a tow, which varied somewhat with the prevailing wind. We have calculated that each sub-sample represents about $\frac{1}{3}$ m³ of water. When the figures for rotifers, Cladocera and copepods are converted

to numbers per cubic metre they can be compared with the figures published by Kofoid (1908) for the River Illinois and those of Rzóska, Brook & Prowse (1955) for the Nile. In Table 4 maximum figures are given since these probably provide as good a basis for comparison of production as any other. The table shows that the temperate, polluted Illinois is very much more productive than either the Nile or the Sokoto. Even allowing for the crudity of sampling of the Sokoto there can be no doubting the paucity of production in this river. It is relevant to note that the values for nitrate from waters of the Illinois are sometimes ten or twenty times as high as those recorded from the Sokoto.

DISCUSSION

The hydrology and plankton of the River Sokoto are dominated by the water level. During the flood period the volume of water entering the river is so great that it dilutes the plankton and important chemical contents to a considerable degree so that the plankton crop, while possibly as large overall as in the dry season, is small per unit volume of water. The great dilution of nutrient substances inhibits the development of the phytoplankton. It is only when the rains cease that the level of nutrient salts can start to rise, and even at the end of the dry season this leaves certain ions limiting. Probably the chief limiting ion is sulphate. The amount of this ion in the water of the Sokoto is extremely small, and there is some indication from the work of Fish (1956a) and Beauchamp (1953) that sulphate acts as a limiting ion in other African waters. Phosphates are also scarce in the Sokoto, but Komarovsky (1953) found that minute traces were sufficient for diatom production, although it was increased by their addition. In Fesafari, where concentration due to evaporation starts earlier and can go on longer there is a greater plankton production than in the river, but, as the zooplankton figures in Table 4 show, production is very poor compared with other similar waters.

While the concentration of nutrient salts is generally low, there is a high calcium concentration in the river due to its flowing through calcareous soil at Sokoto. Daget (1957) gives a calcium concentration of 3 mg/l. for the River Niger at Diafarabé; this is well below the lowest concentration (10 mg/l) found in the Sokoto. The alkalinity in the Sokoto is also high, the highest value being 0·0034 N, compared with 0·00115 N recorded by Daget in the Middle Niger. The alkalinity of the Niger is usually much lower than this maximum figure; Daget gives a figure of 0·0004 N, while a similar figure of 0·0006 N is given by Johnels (1954) for the River Gambia. Only at the beginning of the rains was a comparable figure observed in the Sokoto. The alkalinity of the Sokoto is correlated with a high pH, which rarely fell below 7. Blanc, Daget & d'Aubenton (1955) obtained pH values of 6 to 7 in the River Niger, but observed rapid fluctuations which would be expected with a low alkalinity. Johnels (1954) found the pH varying between 7·0 and 7·5 in the River Gambia, but in the associated swamps it fell to 4·3.

Temperature appears to be of little importance in the nutrient and plankton cycles in the Sokoto, but the low values in December and January probably delay the plankton outburst by slowing down rates of growth and reproduction.

The oxygen concentration had no observed effect, although the work of Fish (1956b) shows that the level to which the oxygen concentration falls at the beginning of the rains is below the 100% saturation level of the blood of *Lates albertianus* Worthington and *Tilapia esculenta* Graham. The Sokoto species of these two genera might be expected to have similar blood-loading capacities. The local *Tilapia* species are abundant in the river near the end of the dry season, but disappear at the start of the rains, being found to some extent in the shallows. This may be caused by lack of oxygen, but it could also be a breeding movement. The fisheries department of the Gold Coast (Anon 1952) has reported Nile perch floating helplessly down the River Kamba after heavy rain; deoxygenation of the water was put forward as a possible cause.

Johnels (1954) advances the theory that the flood permits rapid feeding and growth by fish because it brings various organisms with the rain water into the river and because 'the river is possibly fertilized by substances from land which will indirectly add to the production of the river'. This is probably true as far as predatory fish are concerned, but the rains dilute the plankton of the Sokoto to such an extent that for plankton-feeding fish the floods must be regarded as a time of impoverishment. This may be the cause of the slow growth of *Tilapia* species in the area, a subject to be discussed in a later paper. Le Roux (1956) showed the preference of small *Tilapia* for zooplankton. The species he worked with are not the same as those in the River Sokoto at Birnin Kebbi, but are closely related in feeding habits. Daget (1956) observed *T. zillii* (Gerv.) of 25-30 mm length with their stomachs full of copepods on 23 June in the River Niger. During the floods he found similar fish filled only with filamentous algae; he gives no date for this latter observation. The data from the Sokoto do not contradict Daget's observations. Copepods are abundant in June, and there may be a similar outburst of filamentous algae in the Middle Niger as there is in the Sokoto. *Tilapia* breed at the beginning of the rains so that the fry are subjected to a period when the plankton is greatly diluted and their preferred food item is at a very low concentration. The data for the whole year suggest that the river is not very productive. This means that for fish that are entirely plankton feeders growth will be severely limited throughout their life cycle, and this appears to be so in *T. galilaea* (Artédi), a phytoplankton feeder, which is the commonest species in the area.

ACKNOWLEDGMENTS

We wish to thank the Medical Officers in charge of the Government Hospital at Birnin Kebbi during 1954 to 1957 and Dr J. Haworth, the Officer-in-Charge of the Western Sokoto Malarial Control Project, for permission to use their records of rainfall and air temperatures, and Dr E. Lind of Makerere College, Kampala, Uganda, for examining the phytoplankton samples.

One author (M. J. H.) owes much to the unpaid assistance of his wife without whose help many of the observations could not have been made, and we both express our appreciation to Dr B. M. Gilchrist for her helpful criticisms of the manuscript.

SUMMARY

1. Two stations were sampled in the River Sokoto, Nigeria, near Birnin Kebbi, one in the main stream the other in a pool isolated during the dry season.

2. Birnin Kebbi has one wet season each year.

3. Approximately monthly data over $3\frac{1}{2}$ years are given for water level, temperature, pH, alkalinity, oxygen and transparency. Samples were also taken for chemical analysis, phytoplankton and zooplankton.

4. Water temperatures tend to follow air shade minima. Nutrients, especially sulphate, are low, but calcium is abundant and pH and alkalinity usually high. Oxygen rarely falls to levels lethal to fish. Transparency is high at the end of the rains and falls to a minimum due to plankton at the end of the dry season. Differences between river and pool are attributed to evaporation.

5. Phytoplankton is scarce except from March to June and more abundant in the pool than the river.

6. Rotifera, Cladocera and Copepoda are most abundant in the dry season, Rhizopoda during floods. Differences between years are attributed to differences in nutrients and phytoplankton.

7. The zooplankton is sparse compared with that of the Illinois River and the Blue Nile.

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Carotenoid Pigments in Rotifera

THE Rotifera are remarkable among the Metazoa for their small size, often smaller than the largest ciliated Protozoa. Many rotifers are colourless or at most show some slight colour in the gut wall, clearly due to pigment taken in with food. A few species may have pigment spread throughout the body, others may have red eyespots and the fat globules of resting eggs may be bright orange in colour. This combination of small size and general lack of colour makes investigation of the pigments of this group difficult and so far as we are aware no rotifer pigments have previously been identified.

We have been fortunate in obtaining, more or less by accident, two species of rotifer in sufficient numbers to extract pigments from and apply chromatographic and spectrophotometric techniques.

A dense population of *Philodina roseola* Ehrb. was found in a dish in which the anostracan crustacean *Chirocephalus diaphanus* was being cultured. The rotifers formed dense pink patches on the sides of the dish, and when these patches were scraped off with a clean scalpel they were found to consist of thousands of individuals of *P. roseola*, together with even greater numbers of pink eggs intermingled with a few yeast cells which had been added as food for *Chirocephalus*.

The food available to the rotifers was almost entirely yeast, but as the culture dish was placed near a window a few algal cells may also have been present. There were no other species of rotifer in the material scraped from the dish and a few yeast cells would not interfere with the analysis since no carotenoid pigment is detectable in the bakers' yeast which we used[1].

Carotenoid pigments were extracted from *Philodina* in acetone and transferred to light petroleum (b.p. 40°–60° C) after dilution of the aceone with water. The solution in light petroleum was washed with water to remove traces of acetone and dried over anhydrous sodium sulphate. The absorption spectrum of this solution showed that a mixture of carotenoids was present. The solution was poured on to an alumina column and the chromatogram developed by adding increasing amounts of acetone to the light petroleum used to wash the column. Four fractions were separated.

(1) A yellow fraction passed through the column with light petroleum and showed a maximum absorp-

tion at 449 mμ. The position of this absorption maximum together with the chromatographic behaviour of the fraction is in agreement with the presence of β-carotene. This fraction did not form more than 10 per cent of the total carotenoid pigments.

(2) This fraction was eluted with 30 per cent acetone and showed a single absorption maximum in light petroleum at 468–470 mμ. It formed about 35 per cent of the total pigment and appears to be an ester of astaxanthin.

(3) A third fraction was eluted with 30 per cent acetone and came off the column after the astaxanthin ester. The amount of pigment was too small to examine spectroscopically and formed less than 5 per cent of the total carotenoids.

(4) The final fraction formed a deep orange band at the top of the column. It was resistant to elution with acetone but moved rapidly down the column when a trace of glacial acetic acid was added to the acetone. The pigment was taken into light petroleum, washed well to remove the acetic acid and acetone and dried over anhydrous sodium sulphate. The pigment was hypophasic when partitioned between light petroleum and 90 per cent methanol even when the hypophase was further diluted, but on the addition of a drop of glacial acetic acid the pigment moved to the epiphase. The absorption spectrum in light petroleum showed a single maximum at 468–470 mμ, which, coupled with the characteristic phase behaviour, indicates that the pigment is astacene derived from astaxanthin. This pigment formed about 50 per cent of the total, so together with the esterified fraction, astaxanthin formed about 85 per cent of the total carotenoids in *Philodina*.

These results suggest that *Philodina* is able to accumulate carotenoids from food and convert these into astaxanthin. Observations on another rotifer, *Brachionus calyciflorus* Pallas, further support this suggestion. This rotifer was collected in large numbers from the overflow water at the east end of the Long Water in Hampton Court Park. The plankton sample consisted mainly of *B. calyciflorus* together with a very few other rotifers and small crustaceans. The rotifers were separated from the crustaceans by making use of their reactions to light. The rotifer sample was then placed in a Petri dish and warmed until the animals became immobile. When the sample was examined under a binocular microscope it was found that only a very few other rotifers were present and counts showed the sample to contain 99·8 per cent *B. calyciflorus*.

The carotenoid pigments were extracted from *Brachionus* in acetone. The extract was much more

dilute than from *Philodina* and so the absorption spectrum was measured in acetone before trying any further treatment. Since the gut of these rotifers probably contained algal cells taken in as food, it was decided to compare the absorption curve of an acetone extract of *Brachionus* with that of a sample of phytoplankton taken from the Long Water on the same day as the rotifers were collected. This would show whether the rotifers were accumulating carotenoids or whether the carotenoids in the rotifer extract were

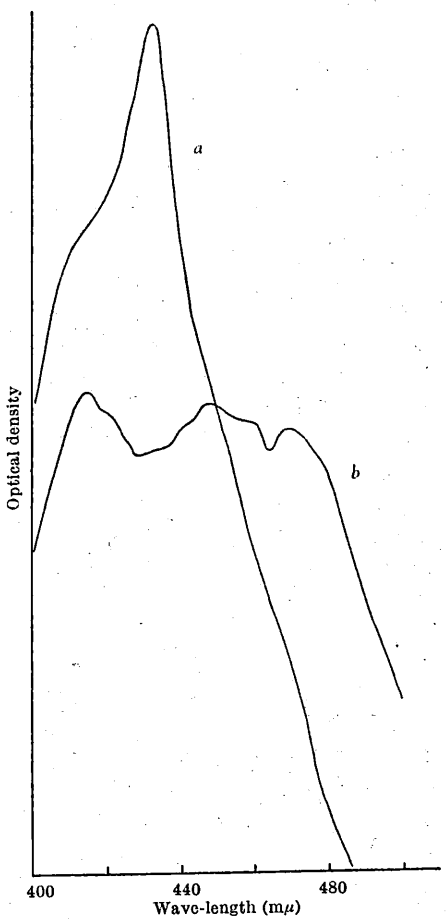

Fig. 1. Spectral absorption curves of acetone extracts of: (*a*) phytoplankton and (*b*) *Brachionus calyciflorus* from the Long Water, Hampton Court

from algal cells in the gut. These curves are shown in Fig. 1 and it can be seen that they differ markedly in shape. The curve from phytoplankton (Fig. 1*a*) shows the absorption maximum of chlorophyll *a* at 432 mμ but in the curve from *Brachionus* (Fig. 1*b*) this maximum has shifted to 414 mμ, suggesting that the chlorophyll has been converted to phæophytin *a* in the gut of the rotifers. The curve from *Brachionus* also shows a region of maximal absorption over the range 450–480 mμ not present in the phytoplankton curve. This is the result of the accumulation of carotenoids by the rotifers. The shape of the curve from *Brachionus* suggests a mixture of carotenoids and this was confirmed by chromatography of the extract in light petroleum on a very small alumina column. Two bands of carotenoid separated on the column ; a lower yellow band was eluted with 60 per cent acetone and an upper pink band was eluted with acetone to which a few drops of glacial acetic acid were added. Too little of the pigments was obtained to examine their spectral properties but the behaviour of the upper band suggests the presence of astaxanthin.

An additional feature of interest in *Brachionus calyciflorus* is that the eggs are coloured pale blue and on treatment with acetone or dilute mineral acids they become pale orange. This change in colour, coupled with the suggestion of astaxanthin on the alumina chromatogram, indicates that the eggs contain a carotenoprotein, thus showing remarkable similarity to the eggs of many Crustacea.

These results add another phylum to those which include animals known to accumulate astaxanthin and since *Philodina* belongs to the Bdelloidea and *Brachionus* to the Ploima, show that the ability to accumulate carotenoids is present in the two major subgroups of the Rotifera.

B. M. Gilchrist
J. Green

Zoology Department,
Bedford College,
Regent's Park,
London, N.W.1.

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BILE PIGMENT IN *CHIROCEPHALUS DIAPHANUS* PREVOST (CRUSTACEA: ANOSTRACA)

BARBARA M. GILCHRIST and J. GREEN

Bedford College, University of London

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Abstract—1. *Chirocephalus diaphanus* is frequently greenish-blue in colour. This may be due both to carotenoprotein in solution in the blood and to granular deposits of blue pigment in various tissues.

2. This blue pigment is identified as a bilatriene and is more abundant in females than in males. When cultured in water deficient in dissolved oxygen, females contain more bilin than if cultured in well-aerated water and more haem is also synthesized.

3. It is suggested that during haemoglobin synthesis in females there is a production of excess haem relative to the available protein. This haem may be broken down and deposited in various tissues as a bilin.

INTRODUCTION

MANY references to the colours of the freshwater fairy shrimp, *Chirocephalus diaphanus* Prévost, are to be found in the literature. Shaw (1791) commented on the red colour of the caudal furca of males and females, the bluish-green of the head and legs of males, and the "spine of the back" of females which is of a deep dull blue. Similar references to the red tail and blue-green body of females were made by Prévost (1803), and the colour plates of Jurine (1820) give an indication of the general pigmentation of the animals.

The colours of *Chirocephalus* seem in part to be related to the available food. Animals collected in the field, whose gut contents show that they have fed mainly on algae, are predominantly orange in colour while those cultured in the laboratory and fed on yeast are predominantly greenish-blue.

It has recently been shown by Lenel & Nourisson (1961) that the red colour of the caudal furca in males of *Chirocephalus* is due to deposits of a keto-carotenoid, while β-carotene and the same keto-carotenoid were found in the rest of the body. No information is available, however, on the nature of the greenish-blue colour of *Chirocephalus*. Another anostracan, the brine shrimp *Artemia salina* (L.), is known to be greenish-blue both when fed on algae and on yeast. This colour resides in the blood and seems to be due to different pigments in the two cases. In green yeast-fed animals, tests for carotenoprotein, bile pigments and verdohaemochromes were all inconclusive (Gilchrist & Green, 1960) and the nature of this green pigment remains unknown. The greenish-blue blood and the ovaries of algal-fed animals contain a carotenoprotein, readily identified by the bright orange colour on treatment with acetone or acid. This green pigment in the blood of algal-fed *Artemia* has been identified as chlorophyll by Dutrieu (1960). The interpretation of her results is

confused by the fact that the wavelength readings of the spectrophotometer were not correctly aligned, thus the absorption maxima of oxyhaemoglobin are given at 560 and 595 mμ instead of the more usual 544 and 576 mμ. This makes the identification of chlorophyll, by means of an absorption curve with a shoulder at 640 mμ and a peak at 700 mμ, very uncertain. No mention is made of the red fluorescence in ultraviolet light so characteristic of chlorophyll.

There also appear to be two separate causes of green and blue colours in *Chirocephalus*. Algal-fed animals in well-aerated water may be greenish-blue as the result of carotenoprotein in solution in the blood, whilst yeast-fed animals are bright green as the result of the deposition of blue pigment granules in various parts of the body. This pigment is also present in algal-fed animals but the colour may be masked by large amounts of carotenoid and carotenoprotein in the fat cells and in the blood respectively. Lack of any information as to the nature of the blue pigment led us to investigate this pigment in *Chirocephalus* and to consider its possible origin.

MATERIAL

The animals used in the present study were derived from eggs laid by a stock of *Chirocephalus diaphanus* collected from a temporary pool in the New Forest, Hampshire. The animals were reared at room temperature (18–20°C) in large vessels containing tap water and fed on a suspension of bakers' yeast. In all experiments involving the quantitative extraction of pigment, the concentration of animals was 100 individuals in 2 l. of tap water and the volume of yeast suspension added was in proportion to the number of animals in the culture.

RESULTS

Distribution of the blue pigment

In both males and females of *Chirocephalus* the blue pigment is deposited in rhomboidal granules which are so regular in shape as to have the appearance of crystals. When, however, they are seen from the end they have the appearance of two cones applied together at their bases. The granules appear to be extracellular and variable in size up to a maximum length of about 24 μ. In both males and females the granules are found in the limbs, particularly in the endites and endopods, the epipods of the last pair of thoracic limbs and also in the caudal furca. Small amounts of pigment are also deposited in the epidermis beneath the cuticle in the ventral wall of the abdomen and in the labrum.

The remaining distribution is strikingly different in the two sexes. In females the gut wall is intensely blue, the colour being most pronounced towards the dorsal aspect of the gut. The pigment granules appear to be embedded in the epimysium covering the thin layer of muscle outside the gut. The granules are deposited in an annular pattern around the gut as indicated in the colour plates of Baird (1850), and are more concentrated dorsally and therefore appear darker in colour. These annular patterns of blue pigment are found in both the thoracic and abdominal regions of the gut, but we have never observed any blue granules in the gut wall of the two genital segments. Large deposits of pigment are also found in the epimysium

of the main diagonal trunk muscles which go from an antero-dorsal attachment in one segment to a postero-ventral attachment in the segment behind. Finally, in females these pigment granules are found in the epithelium covering the foregut caeca and the median group of shell glands, never the anterior or posterior groups.

In males no blue pigment granules have been observed in the epimysium of either the gut wall or the trunk muscles. The large antennal muscles, however, are distinctly greenish-blue in colour; this is due to very small blue granules in the epimysium, the muscles themselves being colourless.

Extraction and identification of the blue pigment

A preliminary test for the presence of bilins (bile pigments) was made by treating the blue gut wall of *Chirocephalus* with yellow concentrated nitric acid and observing the results under a binocular microscope. On contact with the acid there was an immediate change of colour; the whole gut wall rapidly became yellowish-orange and no further colour change took place for several seconds. Subsequently, however, that part of the gut nearest to the acid front became green, then purple, red and finally yellow. This positive Gmelin reaction, although a little atypical, clearly indicates the presence of a bilin.

Extracts of the blue pigment were made by soaking several hundred animals overnight either in methanol or in acid methanol (5 per cent by volume concentrated hydrochloric acid in methanol). The yellowish-orange methanol extract had an absorption maximum at 640 mμ measured in a Unicam S.P. 500 spectrophotometer, as well as maxima in the 450–470 mμ range. These latter are due to traces of carotenoids present in the extract, while the peak at 640 mμ indicates the presence of a bilatriene. This was confirmed by the appearance of an intense red fluorescence in ultraviolet light on the addition of zinc acetate and iodine. The red fluorescent solution had an absorption maximum at 635 mμ, which is characteristic of biliverdin, with proto- rather than meso- side chains.

The acid methanol extracts had absorption maxima at 637·5–640 mμ and at 680–685 mμ, which suggests that there was only partial conversion of the pigment to the hydrochloride. This was further indicated by our repeated failure to take the pigment into chloroform on dilution of the acid methanol with water. When the acidity of the acid methanol extract was increased, the pigment moved rapidly into chloroform.

When the blue pigment was extracted overnight in strong acid methanol (13 per cent by volume concentrated hydrochloric acid in methanol) the resulting solution had a single absorption maximum at 685 mμ, the peak at 637·5–640 mμ having been eliminated. This indicates complete conversion to the hydrochloride and the pigment was then readily taken into chloroform. On the addition of a drop of aqueous ammonia followed by zinc acetate and iodine an intense red fluorescence was obtained in ultraviolet, confirming the presence of a bilatriene.

It is known that in acid solution biladienes may be oxidized to bilatrienes (Gray, Lichtarowicz-Zulczycka, Nicholson and, in part, Petryka, 1961; O' hEocha & Lambe, 1961). Thus any biladiene present in *Chirocephalus* could have been

oxidized to a bilatriene as the result of overnight extraction in acid methanol. Since, however, we got no red fluorescence on the addition of zinc acetate alone to the methanol extract but only on the subsequent addition of iodine, it seems that the bilin is present in *Chirocephalus* as a bilatriene.

Relation between total haem and bilin

Chirocephalus, like other Anostraca, always has traces of haemoglobin in solution in the blood, but when there is little dissolved oxygen in the medium the animal synthesizes much more of the pigment. It may be that the bilin is derived from the breakdown of the haemoglobin in the blood. In an attempt to determine the possible origin of the bilin, experiments were made to show the relation between the concentration of total haem and bilin in animals cultured in waters of different dissolved oxygen content.

The dissolved oxygen content of the water was controlled by bubbling either oxygen or air through the medium. To obtain a low dissolved oxygen content of the water, the initial content was reduced by bubbling nitrogen through the medium until the oxygen concentration was about 2 ml/l. This level was then maintained by bubbling air very slowly; without this slight aeration the medium became depleted of oxygen as the result of the respiration of *Chirocephalus*, yeast cells and bacteria. The dissolved oxygen content was measured every second day, using the syringe pipette modification of the Winkler technique of Fox & Wingfield (1938).

A few hours before making quantitative extracts of pigment the animals were removed from the culture medium and kept in several changes of tap water in order to empty their guts of yeast cells. To make extracts of total haem, five animals were dried on filter paper, weighed and homogenized with a little sodium dithionite and 3 ml of pyridine. The extract was made up to a total volume of 5 ml with pyridine and centrifuged. The optical density of this pyridine haemochromogen was then measured at 557 mμ in a spectrophotometer.

To obtain a quantitative extract of bilin, twenty-five animals were dried on filter paper, weighed and soaked overnight in 4 ml acid methanol (13 per cent by volume concentrated hydrochloric acid in methanol). The optical density of the resulting clear greenish-blue extract was measured at 685 mμ. In order to avoid any difference in pH of the extracts which might affect the concentration of the pigment, sufficient acid methanol was made up initially for all extractions in these quantitative experiments. The concentration of total haem and of bilin was expressed in arbitrary units per 500 mg wet weight of *Chirocephalus* tissue.

In one series of experiments a large stock of young *Chirocephalus* was cultured for 3 weeks as described above in water through which air was gently bubbled. At the end of this period the total haem and bilin concentration of the animals was measured; the stock was then divided into two batches, each of 100 animals. One batch was then cultured for a further 10 days in well-aerated water and the other in water of a low dissolved oxygen content. At the end of this period the total haem and bilin concentration was again measured. The results of two such experiments are given in Table 1.

Another experiment was made to try to culture *Chirocephalus* free from bilin. A large stock of newly hatched nauplii was divided into three batches each of

TABLE 1—GAIN AND LOSS OF TOTAL HAEM AND BILIN IN *Chirocephalus* AFTER 10 DAYS IN AERATED AND OXYGEN-DEFICIENT WATER. OPTICAL DENSITY OF PYRIDINE HAEMOCHROMOGEN MEASURED AT 557 mμ AND BILIN IN ACID METHANOL AT 685 mμ IN ARBITRARY UNITS PER 500 mg WET WEIGHT OF TISSUE

	Mean oxygen conc. ml/l.	Mean wet wt./animal mg		Total Haem		Total Bilin	
		♀♀	♂♂	♀♀	♂♂	♀♀	♂♂
Exp. 1							
Initial	4·8	33·6	31·7	290	115	117	60
Final	5·6	63·5	60·4	263	100	108	51
	1·4	52·5	51·8	528	366	141	53
Exp. 2							
Initial	4·2	39·1	39·1	370	216	130	64
Final	5·0	73·5	75·0	324	154	100	47
	1·8	63·6	63·7	479	290	166	56

100 nauplii and these were reared in tap water which was oxygenated or aerated or which had a low dissolved oxygen content. After 2 to 3 weeks the concentration of total haem and of bilin was measured. Two experiments were made, the results of which are shown in Table 2. The results of all four experiments are combined

TABLE 2—COMPARISON OF TOTAL HAEM AND BILIN IN *Chirocephalus* CULTURED FOR 2 TO 3 WEEKS IN WATERS OF DIFFERENT DISSOLVED OXYGEN CONTENT. OPTICAL DENSITY OF PYRIDINE HAEMOCHROMOGEN MEASURED AT 557 mμ AND BILIN IN ACID METHANOL AT 685 mμ IN ARBITRARY UNITS PER 500 mg WET WEIGHT OF TISSUE

	Mean oxygen conc. ml/l.	Mean wet wt./animal mg		Total Haem		Total Bilin	
		♀♀	♂♂	♀♀	♂♂	♀♀	♂♂
Exp. 1	10·60	36·4	34·9	173	94	83	52
	5·80	42·1	37·9	299	167	88	52
	1·28	31·6	31·6	599	462	167	53
Exp. 2	12·40	28·1	26·3	144	77	81	51
	5·85	32·2	31·8	241	142	88	57
	1·24	25·0	26·7	619	435	126	53

in Fig. 1 to show the relation between total haem and bilin in males and females of *Chirocephalus*.

It is clear from these results that females always have a higher total haem content than males under the same experimental conditions. Further, the increase in

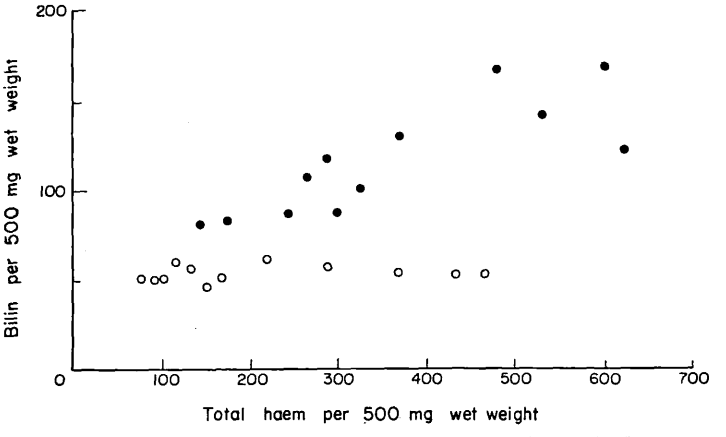

FIG. 1. Relation between total haem and bilin in *Chirocephalus diaphanus* measured in arbitrary units. ● females; ○ males.

concentration of bilin with increasing concentration of total haem in females contrasts with the steady level of bilin in males irrespective of their total haem concentration.

DISCUSSION

The occurrence of bilins among the entomostracan Crustacea is rare. They have been recorded only from parasitic cirripedes (Fox, 1953), from some freshwater ostracods (Green, 1959) and from the eyes of the cladoceran *Polyphemus pediculus* (L.) (Green, 1961). Among Anostraca no bilins have been seen in representatives of the families Artemiidae (*Artemia salina*), Streptocephalidae (*Streptocephalus dregei*) and Branchinectidae (*Branchinecta paludosa* and *B. lindahli*) which we have been able to examine. Unfortunately it has not been possible for us to observe living specimens of other species of *Chirocephalus* or other genera in the family Chirocephalidae. From descriptions in the literature, however, the occurrence of blue and green colours is not infrequent within the family. Hsü (1933), with reference to females of *Chirocephalus nankinensis* (Shen), describes "the deep blue colour of the muscles of the back and of the alimentary canal" while Borcea (1913) refers to the greenish-black of the intestines of *C. spinicaudatus* Simon. Blue pigment granules have been observed in the ovisac wall and lateral shell glands of females of *Chirocephalopsis bundyi* Forbes (Linder, 1959). These granules are rhomboidal in shape and Linder believes the colour to be due to a carotenoprotein.

Of particular interest are references to the colours of *Eubranchipus serratus* Forbes (Pearse, 1913; Dexter & Ferguson, 1943) and of *E. vernalis* (Dexter, 1943)

belonging to another genus in the family Chirocephalidae. These authors observed that early in the season in natural populations the animals were orange-pink in colour while later in the season, when they became mature and were larger, the females were bluish-grey and the males light green in colour. In a later publication Dexter (1946) points out that if orange-pink immature individuals of *E. vernalis* are collected in the field and then kept in the laboratory they change colour as they mature and the females become bluish-grey. He states that the pigment is distributed throughout the body and particularly in the head and tips of the appendages. As has already been mentioned, when *Chirocephalus diaphanus* is collected in the field it is usually bright orange in colour owing to carotenoid pigments. When kept in the laboratory and fed on yeast, the animals gradually become greenish-blue. This is largely the result of the gradual loss of carotenoid pigments which mask any other colour; it is particularly marked in females where there is a continuous loss of carotenoid into the ovary. With loss of carotenoid pigments the blue colour of the bilin becomes more apparent. These various references to the colours of *Chirocephalus* species and *Eubranchipus* suggest that *Chirocephalus diaphanus* may not be unique among Chirocephalidae in its ability to accumulate bilin.

It is clear from Tables 1 and 2 that females accumulate more bilin than males and that they also synthesize more haem under the same experimental conditions. This greater synthesis of haem by females of *Chirocephalus* is in agreement with what is known for *Artemia salina* (Gilchrist, 1954). The relation between total haem and bilin is clearly shown in Fig. 1. In females the concentration of bilin increases with increase in total haem. In males, however, there is no such relation and the amount of bilin remains at the same low level irrespective of changes in the dissolved oxygen content of the medium and of the total haem concentration in the animals.

In studies of the origin of bilins in man it has been shown that there are two main sources of the pigments. The first is from the breakdown of red cells at the end of their life, but the second source arises in association with haemopoiesis. It seems unlikely that in *Chirocephalus* the increase in bilin when females are in poorly aerated conditions could be due to breakdown of haemoglobin at the end of its functional life. Our experiments only lasted 2 to 3 weeks and this would necessitate a very short life for the haemoglobin of *Chirocephalus*. In man the production of bilin from the second source, which Gray & Scott (1959) have termed the haemopoietic fraction, is greatly enhanced in conditions such as congenital porphyria, sickle-celled anaemia and pernicious anaemia, which provoke increased haemopoiesis. This is a striking parallel with females of *Chirocephalus* when placed in poorly aerated conditions. The difference between males and females may be related to the egg production of the females which will impose a strain on the protein metabolism, far greater than the demands made by the production of spermatozoa in males. This may cause a relative deficiency of proteins available to females for other purposes such as making haemoglobin. When females are in poorly aerated water, the haemopoietic stimulus may result in the production of more haem than the females can convert into haemoglobin. This excess haem

could be the source of the additional bilin made by females in water containing little dissolved oxygen.

It is of interest to note that in *Artemia salina*, where we have been unable to detect bilins, haematin is deposited in the shell of the eggs and that more haematin is deposited in eggs laid in concentrated brine than in more dilute media (Gilchrist & Green, 1960). It is also known that females of *Artemia* in concentrated brine synthesize more haemoglobin than do those in dilute media (Gilchrist, 1954). Thus, the conditions which promote haemoglobin synthesis result in the deposition of more haematin in the eggs. The eggs of *Chirocephalus* are pale cream in colour, both in well-aerated and in oxygen-deficient water, and extracts of these eggs have revealed only traces of haem such as could be accounted for by the tissue haems of the enclosed nauplius.

There is an indication, therefore, that during haemoglobin synthesis in females of *Artemia salina* and *Chirocephalus diaphanus* there is an excess haem in relation to the available protein. This haem may either be deposited in the shell of the eggs of *Artemia* as haematin or broken down and deposited in various tissues of *Chirocephalus* as a bilin.

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