AN ECOLOGICAL STUDY OF THE MEIOFAUNA OF SLOW SAND FILTERS, WITH PARTICULAR REFERENCE TO THE OLIGOCHAETES

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LODGE, D.V. AN ECOLOGICAL STUDY OF THE MEIOFAUNA OF SLOW SAND FILTERS, WITH PARTICULAR REFERENCE TO THE OLIGOCHAETES

ABSTRACT

In this thesis an examination is made of the ecology of the meiofauna in a slow sand filter used for the purification of river derived water for industrial and domestic supply. The physical and operational features of the filter are described in relation to the biology of the organisms inhabiting it. Problems encountered with sampling the meiofauna of such a system are discussed and a criticism of the sampling methods employed is presented.

A discussion is made of the uses of total particulate organic carbon as a parameter for characterising the biomass changes in the system. An evaluation of the horizontal and vertical distributions of the meiofauna in the filter is presented. Similar observations are made regarding the distributional heterogeneity of the organic carbon in the system.

Special attention is directed to the oligochaetes present. A relationship is determined between length and dry weight and also segment number and dry weight of Enchytraeus buchholzi, the only actively sexually reproducing oligochaete worm present. The segment number - dry weight relationship is exploited to obtain quick estimates of the dry weight of the worm for biomass estimates. Seasonal changes in population abundance of the meiofauna are presented.

The need to obtain estimates of parameters not easily measured in an operational filter is discussed. The design and functioning of an experimental small-scale filter is described and criticised. A method is detailed for obtaining interstitial water samples without them coming into contact with air. An account of a micro-Winkler oxygen analysis is given. The results of redox potential measurements and dissolved oxygen analyses, made in the pilot filter, are presented together with a discussion on the insight which these measurements give as far as bacterial activity in the filter is concerned.

A discussion is made of the ecology of the meiofauna in slow sand filters and further suggestions for research investigations are made.

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CHAPTER 1 INTRODUCTION

This investigation is part of a continuing study of the biology of water treatment systems, involving collaboration between Dr. A. Duncan and other workers of the Zoology Department of Royal Holloway College and the staff of the Metropolitan Water Division of the Thames Water Authority, formerly the Metropolitan Water Board. The work of this department has been mainly involved with planktonic and benthic energetics and fish population studies of the various reservoirs of the Metropolitan Water Division. The Senior Biologist of the Metropolitan Water Division, Mr. Alan Steel, has been concerned with research involving factors affecting phytoplankton production in these reservoirs. Interest in the biological and chemical processes involved in the slow sand filter stages of the water treatment process has been generated partly because of the relative paucity of biological information available and partly due to their high cost of operation caused by high cleaning costs. Staff of the Metropolitan Water Division have for some time been collecting physio-chemical data from some of the slow sand filter beds used in the water treatment process. In 1974 this study was initiated in order to gain some basic information concerning the biology of these filter beds.

The main aims of the investigation were

1. To devise a sampling method which would allow the collection of representative samples from the sand during the course of a filter run
2. To describe the spatial and temporal variations of the meiofaunal organisms inhabiting the interstices of the filter
3. To assess the role of the oligochaetes in any fluctuations in the organic content of these filters.

The first two aims were largely fulfilled; however, it became apparent during the course of this investigation that, in order to more fully understand some of the spatial and temporal fluctuations of the meiofauna, information was needed concerning the dissolved oxygen and redox regime of the sand filter. As information of this kind was difficult to obtain from an operational filter, a major decision was made to design and build a small scale filter which could be operated in a laboratory or in the field, which allowed detailed depth examinations of these parameters and would be of use for energetic and

respiratory studies by subsequent workers. Because of the time involved in the design, building and testing of this equipment, it was not possible to complete the oligochaete energetics studies which had been originated earlier.

The thesis consists of various sections. The remainder of this chapter consists of an account of relevant slow sand filter studies. Chapter 2 describes the structure and physical environment of slow sand filters and chapters 3 and 4 describe field and laboratory methods of analysis. Results of field analyses are discussed in chapters 5 and 6. Methods and results of the oxygen and redox investigations of the pilot filter are discussed in chapters 7 and 8. The final chapter interrelates the results and discussions of previous chapters.

Although no known work has been carried out on slow sand filters concerning their physical mechanisms of filtration, much of the information gathered from rapid filters is applicable to the slow sand filter. The relevant literature concerning these aspects of filtration is discussed in chapter 2. As well as treating water used for supply, slow sand filters are sometimes used to treat water prior to ground water recharge and this is also discussed in chapter 2. Another minor use of the slow sand filter is in the tertiary treatment of sewage, although the intermittent nature of these filters means that they are not strictly comparable with slow sand filters used to treat water for supply. Work by Brink (1967) largely centred around the bacteriological and biochemical processes involved in reducing the organic content of the sewage entering the filters.

Before this study was initiated very little information was available on the biology of interstitial meiofaunal organisms inhabiting slow sand filters. Most work has centred on bacteriological and microbiological aspects of their biology. Very early work on slow sand filters was carried out by Kemna (1899) who was mainly concerned with listing the organisms found in the surface waters and on the surface skin of the filter. Van de Vloed (1955) considered some of the physical effects of the filter in removing particles from the water. More recent work has involved bacteriological investigation of the sand. Burman and Lewin (1961) showed that most removal of bacteria originally in suspension in the water was achieved in the top 3 inches of the filter. They also found that whereas coliform organisms were capable of multiplication in the filter skin, Escherichia coli organisms were

not; thus suggesting that increases in the numbers of the former type of organism were due to multiplication and accumulation, whereas increases in E. coli were just the result of accumulation. They considered that reduction in the numbers of these organisms in the sand may well have been due to the predatory effects of Protozoa and detritus feeding organisms. Problems encountered with aerobic sporing bacilli and high E. coli counts in "blanket weed" following cleaning of these filters have been reported in Metropolitan Water Board reports (M.W.B. 1971) and discussed in chapter 2.

The effect of sand filter bacteria in removing dissolved organic compounds from interstitial water was investigated by the Metropolitan Water Board (1974) by the addition of phenols to the inflow water. It was demonstrated that the bacteria of the filters could remove these phenols quite effectively from the water even when added at concentrations far higher than would normally reach a slow sand filter. However, it was found that a period of acclimatisation was necessary to the particular phenol before effective removal would be achieved. Schmidt (1977) has demonstrated 70 - 90% elimination of detergents entering slow sand filters used for ground water recharge, again a period of "acclimatisation" was required before these maximal values were achieved. Similar results were found with the removal of indol, produced during anaerobic digestion of algae. Halogenated hydrocarbons were almost totally eliminated during passage through this slow sand filter. The presence of large amounts of organic material, especially algae and bacteria, in the upper layers of the slow sand filter caused the removal of most of the mercury and other heavy metals which were experimentally introduced into the system. No information was available, however, on the effect which these pollutants had on the larger biota of the filter. Predatory bacteria, bdellovibrios, were demonstrated (M.W.B. 1974) to have a predatory effect on sand filter bacteria grown in culture. However, their overall effect on a sand bed was difficult to assess because of the difficulty in eliminating the effect of predatory Protozoa from the experiment and also because a variety of plaques were produced by bacteria other than bdellovibrios.

Species lists of the attached and unattached algal flora of slow sand filter beds are given in Brooks (1954 and 1955). He found that the surface of the filters were dominated by filamentous diatoms for most of the year. Two well-defined phases were recognised - in the spring and in the autumn, when different species were found to

dominate. Marked differences were found in the algal composition of recently cleaned beds and those which had been operating for long periods and he considered that the predatory effects of Protozoa and surface dwelling chironomid, ephemeropteran and trichopteran larvae may have been important. Ridley (1967) recognised three stages of algal succession in slow sand filter beds

1. Marked by a proliferation of members of the Chlorophyceae in the supernatant water during the first seven or so days of the filter run
2. From the fifth day onwards the surface was seeded with diatoms which he considered were either derived from influent water or from the sand bed during backcharging procedures.
3. A proliferation of the larger sized filamentous algae from the tenth day onwards.

Bellinger (1968) divided the algal flora of slow sand filters into two categories

1. Those which were present in large numbers simply due to accumulation and were planktonic in origin
2. Those which entered the beds, often in small numbers and produced large populations simply by active division, and would compare with Ridley's 1 and 3 stage algae. Bellinger also investigated the distribution of algae on the sand surface and found very heterogenous distributions which he related to the direction of the prevailing wind and position of the inlet water source. Algal cells of most species were demonstrated to penetrate into the sand to depths of up to 7cm, although the majority of living cells were found in the top 2·5cm.

The Protozoa of slow sand filters have been studied by Lloyd (1973 and 1974), whose sampling equipment has been discussed and criticised in chapter 3. He found that the only group to feed upon bacteria in suspension were the Peritrichia and of these Vorticella spp was dominant. Lloyd also found a very rapid colonisation of Vorticella spp within the first 10 or so days of a filter run and that this was inversely related to the number of bacterial colonies in the filtrate water. He concluded that Vorticella spp could have a significant effect on the number of suspended bacteria in the interstitial water. However, he found that bacteria in suspension did tend to adhere to the surface of the sand grain and could thus be passively removed from suspension in this way. Lloyd also compared the effect of filtration rate on depth penetration of the Protozoa into the sand

and found that increasing the filtration rates from 15 to 45cm.h^{-1} (normal works filtration rate is approximately 20cm.h^{-1}) caused larger populations of Vorticella spp to occur at deeper depths and also found that, whereas extinction of the population at 15cm.h^{-1} was at 18cm., at 45cm.h^{-1} it was calculated to be at 35cm. He considered that this effect was likely to increase the depth at which the Vorticella spp population would actively remove bacteria from the interstitial water and thus assist in purification. The M.W.B. (1974) have reported that experimental filters could be run at rates of up to 50cm.h^{-1} without any deterioration in the chemical and bacteriological parameters.

The results of relevant investigations into the biology of slow sand filters by Rittersbusch (1976) and Husmann (1968) have been fully discussed in the text. In summary, Husmann's (1968) work centred around descriptions of the meiofauna of a slow sand filter, whereas Rittersbusch (1976) found that rotifers and nematodes did not play a great part in the purification of a slow sand filter, as far as nitrate removal is concerned, although the results were not very conclusive.

CHAPTER 2 THE CONSTRUCTION AND OPERATION OF A SLOW SAND FILTER

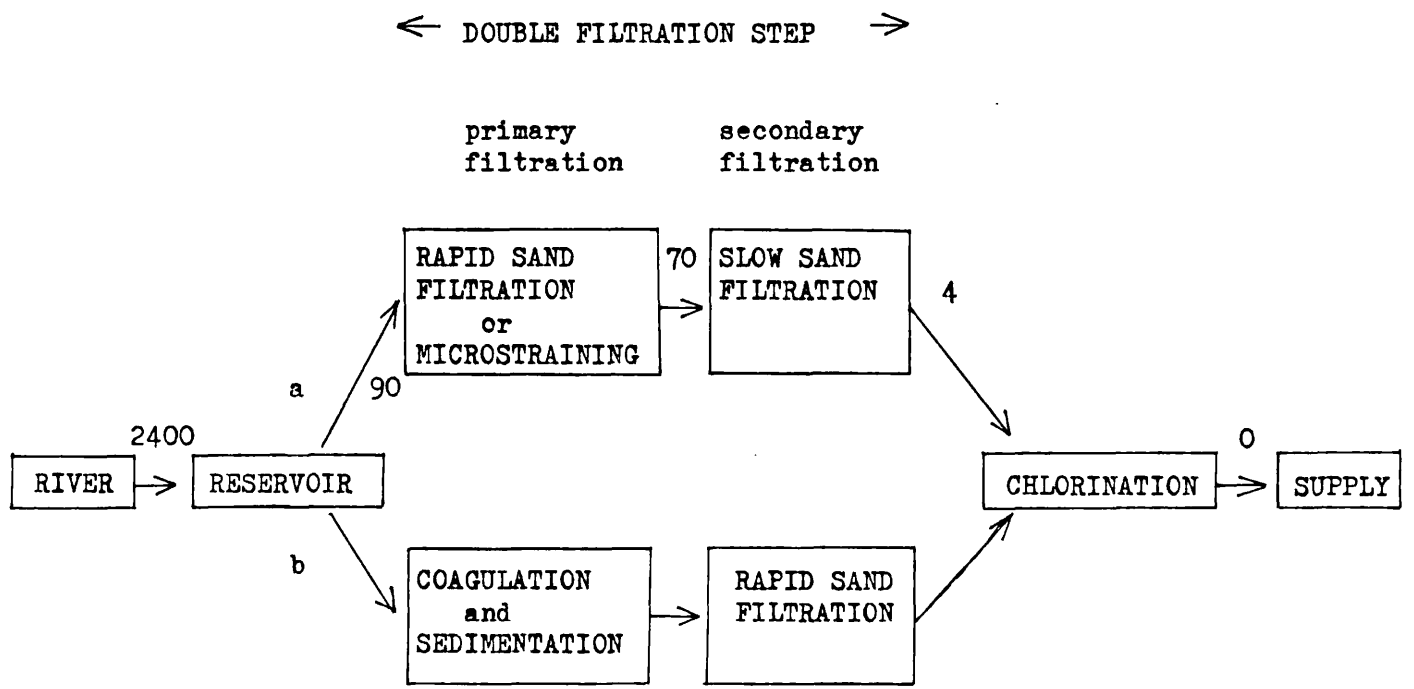
2.1 Requirements for raw water treatment prior to domestic and industrial supply

In Great Britain the main sources of raw water are rivers, lakes and underground aquifers and in the London area water for drinking comes mainly from the River Thames and River Lee. This water is not immediately available for human consumption as it contains a variety of particulate material including zooplankton, algae, bacteria and detritus (the dead and decaying remains of organic matter). The former two components are not dangerous to man in themselves, but can impart unpleasant tastes to the water when present in sufficient quantities. The bacteria, however, which are often associated with the detritus are potentially dangerous. Plate counts from the River Thames at Walton in 1972 gave a total bacterial count of 700 - 17,000.ml^{-1} and of these Escherichia coli formed 500 - 4,000.$100ml^{-1}$, with total coliforms of the order 3,000 - 31,000.$100ml^{-1}$. The World Health Organisation (1970) recommended for drinking water that 95% of the 100ml samples taken throughout the year should contain no coliforms (which corresponds to an average density of 1 coliform in 2 litres of water). Thus normal river levels are clearly too high. The significance of E. coli is not that it is pathogenic in itself, but that it is the most frequent type of coliform organism present in human and animal intestines. Thus its presence is indicative of faecal pollution and can point to possible contamination by more potentially dangerous pathogens.

2.2 Methods of treatment of raw water

There are many different forms of water purification, but all of them conform to a similar basic plan (see figure 2.1). Water is pumped from the primary sources into a reservoir where it stays for some time. At this stage there may be a passive form of treatment in that the environmental conditions have changed from a fast-flowing, relatively shallow river, to a deep, relatively static body of water. The result is that there is a decrease in riverine species and an increase in lacustrine species. The main effects of significance are that there is a decrease in the number of bacteria at this stage, plus sedimentation of organic particles. Alternatively the reservoir may be stirred by the use of large inlet jets, a device used to control algal blooms in order to reduce the algal biomass passed on to the

Figure 2.1 SUMMARY OF VARIOUS SYSTEMS OF WATER TREATMENT USED TO PRODUCE POTABLE WATER



a = eg Metropolitan Water Division of the Thames Water Authority.
b = eg Grafham Water, and Bough Beech Reservoir.

N.B. Figures indicate number of E. coli. $100ml^{-1}$ present in water after the appropriate treatment, based on a 10 years average (1961-70) from Thames Water Authority.

treatment works.

This water is then passed on to the treatment works, which is where the main differences in the type of treatment are encountered. Basically the water may be treated by varying combinations of rapid sand filtration, slow sand filtration, microstraining and chemical coagulation and sedimentation. As far as this study is concerned, the methods which will be discussed will be those used by the Metropolitan Water Division of the Thames Water Authority.

The Metropolitan Water Division employs a system of double filtration, consisting of a primary filtration step followed by one of secondary filtration. The primary filtration step may be composed of microstrainers and/or rapid sand filters.

The microstrainers consist of large cylinders of diameter 1·5 - 3m, composed of mesh of 23, 35 or 65 μm aperture. This is continuously rotated, raw water flows axially into the cylinder through an open end and passes out through the mesh. A series of water jets arranged above the drum washes away the debris accumulated on the inner surface of the mesh.

Rapid sand filters operate at a higher flow rate than slow sand filters - generally at 600cm.hr^{-1}. They consist of bodies of sand 0·6 to 0·9m deep and of a coarser nature than that found in slow sand filters, supported on a bed of gravel. The depth of water overlying the sand may be from 1 to 1·5m. Some of the material in suspension in the filter water remains behind in the sand and is removed after a period by a process of reversing the direction of flow and increasing its velocity. The organic matter floats to the surface of the water and is washed off into overflow sills. Treatment at this stage is basically mechanical, although rapid sand filters play a valuable role in oxidising free ammonia. This is brought about by the activities of nitrifying bacteria which are present in the bacterial film surrounding the sand particles. Removal of ammonia is important where free residual chlorination is required later on in the treatment processes.

The procedure at Hampton Treatment Works is to use rapid sand filters only in the primary filtration step.

The secondary filtration step consists of passing the water through a slow sand filter, which will be discussed later. The overall effect of the slow sand filter is to remove the majority of the bacteria (see figure 2.1) which remains in suspension after the primary

filtration step.

The final process at the treatment works is for the water to be dosed with chlorine in order to remove any remaining bacteria and to leave a residual chlorine concentration which will be effective in killing any bacteria which may enter the water via cracks in the distributive system. This water is then held in covered service reservoirs before being distributed to domestic homes and industry.

2.3 Brief history of the slow sand filtration method of treating water

A comprehensive summary of the historical development of water filtration is given in Lloyd (1974) and so this review is intended to emphasise only the important points.

A type of sand filtration for treating water for human consumption was documented in Sanskrit texts as long ago as 2,000 B.C. Various forms of filtration using granular material have been found in the literature since then and up to more recent times (review in Lloyd 1974).

The first record of sand filtration in this country was that used by John Gibb in 1804 at Paisley in Scotland, using a lateral flow system. Although the filtrate was intended for use in a bleaching works, surplus water was sold for domestic consumption (Sinclair 1808).

A self cleaning sand filter was put into use in 1827 by Robert Thom at Greenock in Scotland and used 5ft of fine sand through which water was allowed to percolate downwards. When the water yield decreased, due to silting up of the pores by organic material, the filters were cleaned by reversing the direction of water flow.

In 1829 the method of slow sand filtration was adopted for the first public supply when James Simpson used a design to treat water supplied by the Chelsea Water Co. in London, using water taken from the River Thames (Shadwell 1899). This was used following the reporting of a Royal Commission in 1828 which spoke favourably of filtration as a method of providing clean water from a polluted supply.

In 1852 the Metropolis Water Act was passed which required that all water derived from the River Thames within 5 miles of St. Paul's Cathedral should be filtered, or drawn from wells, before being supplied for human consumption.

Simpson's filter was very similar in concept and design to

those slow sand filters in use today and was cleaned by scraping the surface film off and also using reverse flow techniques.

The classical convincing proof of the effectiveness of water filtration was given in 1892 and concerned the fates of the populations of two cities, Hamburg and Altona, both of which abstracted their drinking water from the River Elbe. Hamburg treated its supply merely by settlement, whereas Altona filtered the whole of its drinking water. When the river became infected with the cholera-causing bacterium, many of the inhabitants of Hamburg died as a result of the disease, whereas the people of Altona escaped almost unscathed.

The rapid sand filter was first developed in the U.S.A. and at first was operated under pressure rather than gravity. In 1899 the first patented rapid sand filters were used at Reading in England. They were so effective in removing a considerable amount of the suspended organic matter load of the water that, as a result, it was possible to almost double the amount of water treated by the slow sand filter before it required cleaning itself. This system of double sand filtration was later used by the Metropolitan Water Board of London in 1923.

Today the slow sand filter is used in many European cities and also in America and Japan. Its high quality of product, simplicity and economy of construction and operation in regions of the world where land and manpower are cheap, make it a good proposition for developing countries who need treated water for human consumption (World Health Organisation, 1970).

2.4 Construction and physical environment of a slow sand filter

Sand filters vary in surface shape and area; bed 45 at Hampton Treatment Works is approximately 60m square and between 2 and 3m. deep. The basic design of a slow sand filter can be seen in figure 2.2. Concrete underdrains support a layer of sand varying from 0·3 to 0·75m in depth. Above the sand layer lies a layer of surface water 1 to 1·5m deep which has received primary treatment - in this case rapid sand filtration. The surface water is kept at a constant depth during the course of a filtration run, in order to maintain a constant head of water which provides the pressure to drive the water through the filter.

The grain size ranges from less than 63 μm diameter to greater

Figure 2.2 SIMPLIFIED DIAGRAM OF A SECTION THROUGH A SLOW SAND FILTER
(Not to scale)

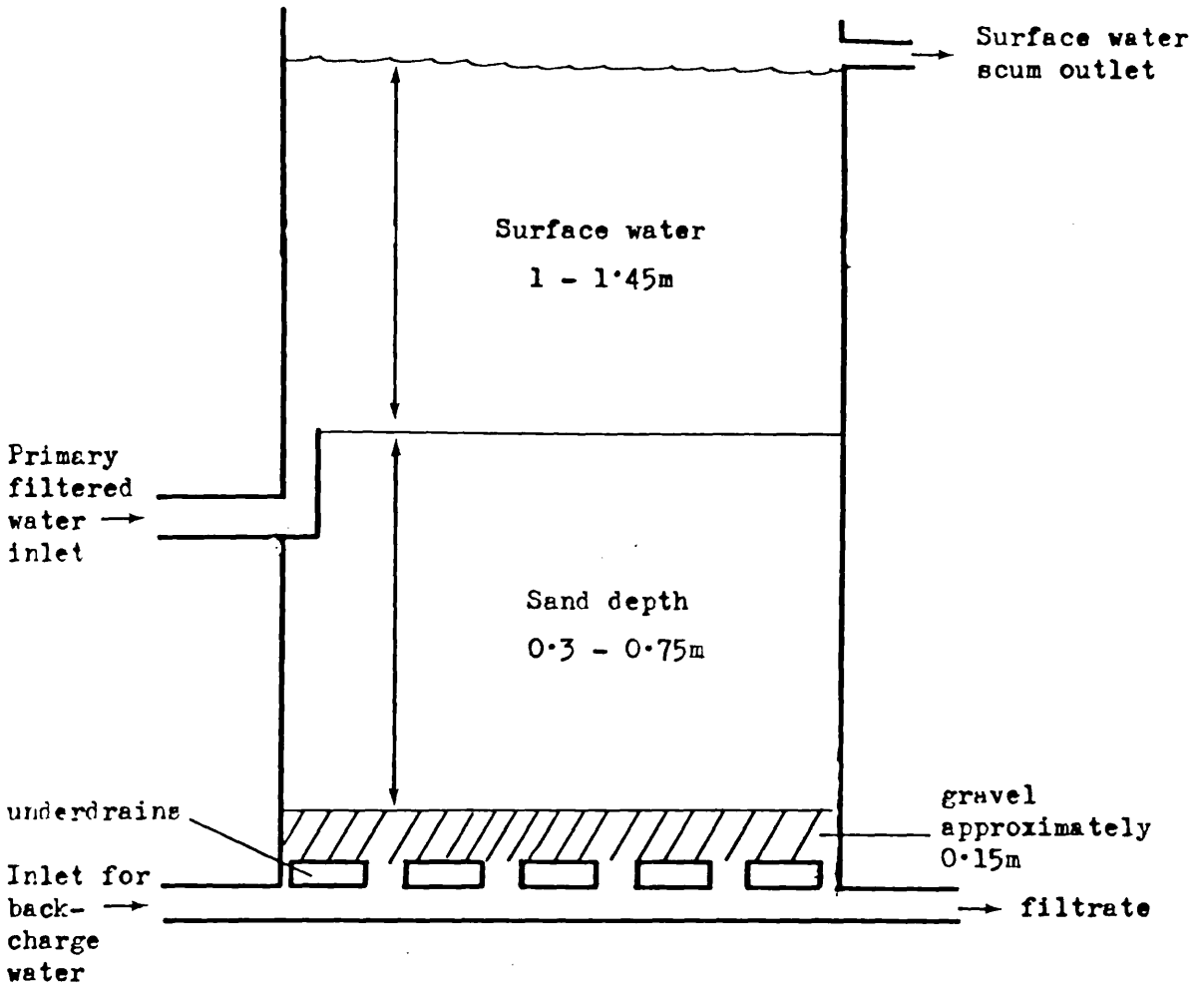
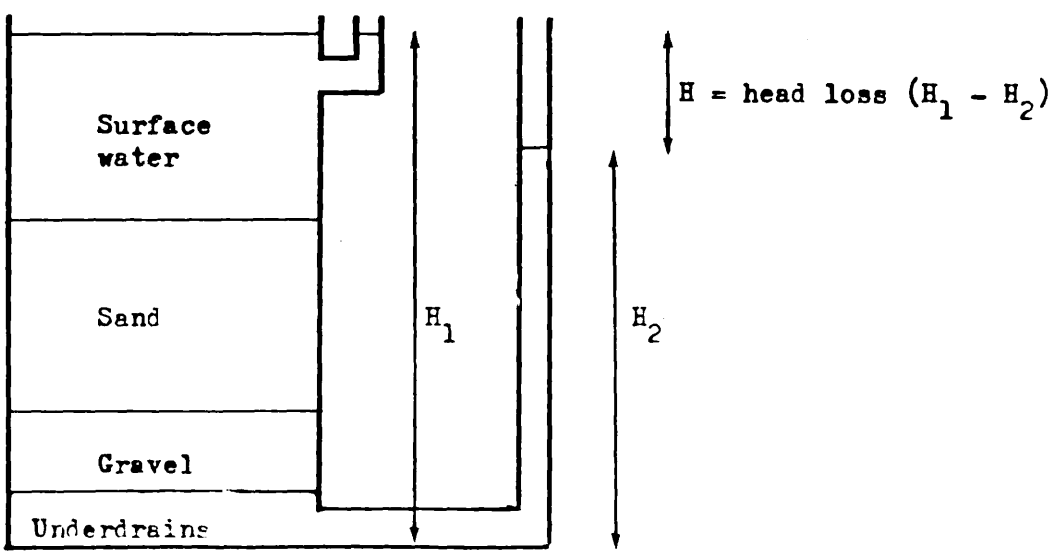


Figure 2.3 DIAGRAMMATIC EXPLANATION OF "HEAD LOSS"



than 4000 μm, the most abundant size range (by weight) being 250 - 500 μm with the distribution of grain sizes being positively skewed (see chapter 3.7).

The porosity of the sand is 37% (chapter 7.), a factor which causes water travelling on to the filter at a rate of x cm.hr^{-1} to have an increased velocity of 2·7x cm.hr^{-1} (i.e. $x.\frac{100}{37}$) whilst passing through the interstices.

2.5 Basic functioning of the filter

A newly sanded filter will have a sand depth of approximately 0·75m. The first process in making the filter operational is to slowly backcharge the filter from below. Here secondary filtered water is carried under low pressure into the underdrains from where it slowly percolates upwards through the gravel and into the sand layer. The effect of this procedure is to remove air from the interstices so that when the filter is working properly there will be no pockets of air present that would reduce the efficiency of the filter.

When this water has reached above the surface of the sand, the backcharging procedure stops and primary filtered water is allowed to pass onto the surface of the sand. When the surface water has reached the maximum level in the filter (approximately 1m in a newly sanded bed), the water is allowed to percolate through the sand and out through the underdrains.

Small particles in suspension in the water pass through the interstices until they come into contact with the surface of a sand grain. Particles too large to pass through the interstices settle on the surface of the sand, which also supports growth of benthic organisms. With time a skin develops at the sand/water interface which is composed of a matrix of algae, bacteria and detritus, together with benthic animals. This skin is often referred to as the "schmutzdecke" (filth layer) or "zoogleal film", but for reasons of simplicity the words 'surface skin' will be used to describe this layer in subsequent discussions.

This build-up of surface skin contributes to the screening efficiency of the bed. However, this also results in an increase in head loss of the bed, thus increasing the resistance to the down-

ward passage of water and consequently decreasing the efficiency of the bed. After a period of time it is necessary to drain the bed and remove this surface skin, in order that the bed may again be run efficiently. This is done by using mechanical skimmers which remove the top 1·5cm of sand. The bed is then backcharged with water from below to remove air and then is supplied with top water again. The depth of sand in the bed is thus reduced gradually until it becomes too shallow to be effective in removing bacteria and suspended particles - this is at between 30 - 45cm depth. At this stage more sand is added to bring the sand up to its original level; this resanding procedure can take place every 1 - 2 years depending upon the length of individual runsand the depth of sand removed at each cleaning operation. The duration of any one filter run before surface skimming is required varies between about 30 - 60 days, depending upon the season.

2.6 Mechanism of filtration

As it passes through a sand filter, a suspended particle is brought into contact with the surface of a sand grain and will be held there by various processes. Those particles consisting of inert material remain there until they are removed during cleaning processes. Those capable of chemical or biological degradation are converted into simpler forms which are either removed in solution or remain until removed during cleaning.

The particles which pass into the interstices of the filter are characteristically smaller than the sizes of the pores in the media. Consequently removal mechanisms within the body of a filter are not concerned with straining. So the ability of a deep filter to retain these particles depends upon two important steps. The first concerns the particle being carried across the liquid flow lines and being brought adjacent to a pore wall. The second concerns the particle being brought to adhere to the pore wall. The former types of step are referred to as "transport mechanisms" and the latter referred to as "attachment mechanisms". Within a slow sand filter there is a very important third step concerning the ability of biota within the filter to react with and alter the degradable particles in suspension. This third step is referred to as "purification" and is the major distinguishing feature between rapid and slow sand filtration. The importance of sedimentation,

hydrodynamic action and interception in mechanisms concerned with transport was demonstrated by Ison and Ives (1969) by the use of kaoline particles in model filters. Attachment mechanisms of importance have been shown by Ives and Gregory (1966, 1967) to include electrical bonding, Van der Waals forces and adsorption.

2.6.1 Transport mechanisms

2.6.1.1 Sedimentation

If the particles have a density significantly greater than that of the liquid, they have a significant constant velocity relative to that of the liquid and which is orientated in the same direction as gravity. The extent to which this deflects particles from streamlines will depend upon the relative orientation of the vectors describing liquid velocity and gravitational velocity. The result is that the larger, denser particles are more readily transported away from the flow lines and towards the pore walls.

2.6.1.2 Hydrodynamic

The liquid flow in the pores is laminar, but with a velocity gradient across the pores. In situations where this gradient is uniform, a spherical particle will be made to rotate, which would cause the particle to migrate across the velocity gradient. However, in a filter this velocity gradient is not uniform and the particle will be deflected, but not in a uniform and predictable way. If the particle is not rigid, but is capable of being deformed, its motion will be even more irregular. The result of all these effects is for the particle to show random drifting movement across streamlines and so it is more likely to be brought into contact with a pore wall.

2.6.1.3 Interception

If there are no forces causing a particle to move across a streamline, the centre of the particle will tend to follow the direction of the streamline. So if the centre of a particle passes within a distance from the pore wall which is less than the length of its radius, it will be intercepted by the pore wall. Large particles are more likely to be intercepted than small ones and so will be intercepted earlier on in the filtration process.

2.6.1.4 Brownian movement

This describes the random movement which very small particles in liquids exhibit, due to the thermal energy of the liquid molecules. Particles $> 1\,\mu m$ in diameter have this movement restricted, due to the viscous drag of the liquid and so the effect here is to cause the particle to move at the most 1 or 2 particle diameters, and so is not of importance in particles of this size. With particles $< 1\,\mu m$ in diameter this movement becomes more significant with decreasing size. The overall effect of this movement superimposed on the direction of the laminar flow, is to increase the probability of a particle being brought into contact with the pore wall.

2.6.1.5 Van der Waals forces

These forces are of more importance in holding a particle to a pore wall once it has come into contact.

To summarise, Brownian movement controls movements of particles of $< 1\,\mu m$ diameter, interception and hydrodynamic mechanisms control particles of 1 - 50 μm diameter and with a density similar to that of water. Particles with densities much greater than water are controlled by gravity.

2.6.2 Attachment mechanisms

2.6.2.1 Van der Waals forces

These forces of mass attraction have only a very minor effect in pulling particles from the liquid flow lines and are more important in holding a particle in contact with a pore wall once the contact has been made. These forces only apply when the separation distance is less than the particle diameter.

2.6.2.2 Electrostatic attraction

This is the attraction of opposite electrical charges. Clean quartz sand has a negative charge and so will attract positively charged colloidal particles as well as cations of various metals. Colloidal particles of organic origin, including bacteria, usually have a negative charge and so in a clean filter these will be repelled by the sand. This is one of the reasons why a clean bed is not effective in retaining impurities of this kind when first put into

service. However, with time, positively charged particles may accumulate on the sand grain, to the extent that oversaturation occurs and the result is that the grain and its attached particles are rendered positively charged. This results in the negatively charged bacteria and colloidal matter of organic origin being attracted and held on the surface of the grain. However, with the build-up of negatively charged ions on the grain surface, oversaturation may again occur and result in reversal of the charge on the complex. These reversals of charge can continue throughout the life of the filter.

2.6.2.3 Adhesions

Particles of organic matter deposited on sand grains can support growth of bacteria and other microorganisms. These micro-organisms plus their wastes, dead cells and organic substrate, can provide a matrix into which particles in suspension can become attached. Bacteria, protozoa and algae can also produce hold-fasts which increase their chances of remaining attached to the inert surfaces.

2.7 Biological purification mechanisms

Some biological purification mechanisms have already been discussed in chapter 1, where a review of slow sand filter studies has revealed that several biological mechanisms are working in slow sand filters to purify the water. Removal of dissolved organic material by bacteria has been demonstrated by experiments with phenols (M.W.B. 1974) and other chemicals (Schmidt 1977). The effect of predatory bacteria, including bdellovibrios (M.W.B. 1974) and of predatory protozoa (Lloyd 1974), has also been shown. However, attempts at quantification of these results were difficult to carry out, due to problems with isolation of these organisms, plus the effect of passive accumulation of bacteria on sand particles by mechanical means which tended to mask the effect of the protozoa.

The importance of ciliates was demonstrated by Barsdate et al (1974), who concluded that in a bacterial population grazed upon by ciliates, mineral cycling was found to be over one hundred times faster than the actual release of nutrients from the material which was being decomposed. Similarly Johannes (1967) found that bacterial breakdown of detritus was more rapid when bacteria feeding

ciliates were present than with bacteria alone.

Rittersbusch (1976) concluded that the effect of rotifers and nematodes on nitrate removed by slow sand filters was negligible, although the direct effects of these organisms were not really measured. Results of studies into the fauna of sewage percolating filters do , however, emphasise the importance of these organisms in purification. Solbé et al (1974) compared the performance of experimental percolating filters with and without populations of macro-invertebrates. The performance of the filter which had been inoculated with fly larvae and oligochaete worms was considerably better than that without these organisms. Similar results were reported by Williams and Taylor (1968) on a percolating sewage filter. The importance of an oligochaete Lumbricillus lineatus was emphasised by Reynoldson (1939a) in maintaining the efficiency of a sewage percolating filter. These effects will be partly due to the grazing activity of the macroinvertebrates preventing the accumulation of too much film and possibly also by improving film characteristics, for example, by enabling oxygen diffusion to cope more efficiently with oxygen demand in the same way that Chironomus larvae may increase the rate of oxidation of organic deposits in polluted rivers (Edwards 1958).

Thus it would appear that the meio and macrofauna of such systems can affect the bacterial populations either by direct removal through cropping and digestion, or indirectly by keeping the bacterial population low and so stimulating the metabolic activities of a population which could, in turn, affect the uptake rates of dissolved organic material by the bacteria. So the effect of detritus and bacteria feeding meiofauna on the efficiency of purification of a slow sand filter would seem to be a possibility, although no work has been attempted to measure its precise effects.

2.8 Operation and maintenance of the filter

2.8.1 Operating velocities

During the cleaning processes which are going on in the filter bed, conditions within the bed have been drastically altered. The flow of water has been stopped and the bed drained. As the surface skin is removed, the top few cms of the bed will begin to dry out. The bed can remain in this condition for 1 - 3 days.

Prolonged exposure of the bed to the atmosphere has been shown (M.W.B. 1971) to cause bacteria, especially the yellow aerobic sporing type, to detach from the sand grains and appear in the filtrate in large numbers when the filter is put back to work. This is also a problem with newly sanded filters. The significance of these organisms lies in the fact that they are normally resistant to normal works' chlorination.

A second problem caused by prolonged exposure to the air is that it has been shown to favour the growth of streptomycetes which cause strong earthy odours in the filter for a period of twenty-four hours after the filter has been put back to work (M.W.B. 1971).

Another problem is that caused by the algal mat which must be quickly removed from the bed, especially in warm weather conditions. In situations where large heaps of Cladophora had been left on the surface of a drained filter, it has been shown (M.W.B. 1971) that temperatures of $32^{o}C$ had developed within the heaps. These conditions were seen to favour the growth of large numbers of E. coli which would seep into the sand, carried in the decomposing liquids from the mounds of Cladophora and hence into the filtrate.

It follows, therefore, that the quality of the filtrate of a slow sand filter, immediately following cleaning, is very poor. The filter is, therefore, not put immediately back to work at the normal filtration rates. Instead, it is normally run at very low rates ($2{\cdot}5 cm.hr^{-1}$) for the first three or four days and then gradually increased to normal operating velocity as the quality of the filtrate improves.

2.8.2 Build-up of head loss

In slow sand filtration the downward movement of water in the interstices is laminar (Ives, 1970). Hence the resistance 'H' offered by a clean filter bed is in accordance with Darcy's law, which states that the approach velocity of flow is proportional to the head loss :

$$V_f = \alpha (H_1 - H_2) \qquad (1)$$

where V_f = approach filtration rate ($m.hr^{-1}$)

H_1 = raw water head (m)

H_2 = filtered water head (m)

α is a coefficient dependent upon the hydraulic characteristics of the complete filter

The head loss $H = (H_1 - H_2)$ can be determined manometrically (figure 2.3) and is a measure of the increased resistance to flow caused by the accumulation of particles on and in the sand.

When the relationship (1) is expanded, it is expressed as

$$H = \frac{V_f}{k} .z \qquad (2)$$

where H = the head loss (m), i.e. the resistance offered by the filter

V_f = filtration rate ($m.h^{-1}$), i.e. total volume passing per hour, divided by the surface area of the bed

z = depth of the bed (m)

The coefficient k has the dimensions of velocity ($m.h^{-1}$) and can be determined theoretically by using one of several formulae available :

$$k = 150(0{\cdot}72 + 0{\cdot}028\ T)\ \frac{p^3}{(1-p)^2}\ \phi^2.d_s^{\,2} \qquad (3)$$

where T = temperature (^oC)

p = porosity (ratio of volume of pores to total volume of filter media), here determined to be 0·37

ϕ = the shape factor (sphericity)

d_s = the specific diameter of the sand grains (mm)

ϕ is the ratio of the surface area of a sphere to that of an average grain of the same material having the same volume. So its maximum value is 1·0 and decreases as the grains become less spherical.

d_s is used to describe the grain size of the sand, with regard to the variation in size of the individual grains. It is defined as the size of an imaginary grain from a uniform sand of which a certain weight has the same gross surface area as an equal weight of the filtering material under consideration. The use of calculations involving specific diameters are normally restricted to rapid sand

filters where the grading of sand has to be more accurately controlled than for slow sand filtration. A simple way of calculating d_s is to use the formula :

$$d_s = d_{10}(1 + 2\log U)$$

where U = coefficient of uniformity and is equal to $\frac{d_{60}}{d_{10}}$

d_{60} = diameter of the seive opening through which 60% of the material will pass

d_{10} = diameter of the sieve opening through which 10% of the material will pass

It is possible, therefore, by substitution of all the unknown quantities with equation (2) to calculate the expected head loss of a bed.

From (2) it can be seen that, for any one run, 'z' will remain constant. The flow rate, V_f, tends to decrease daily as the pores become blocked with particles settling out of suspension, but it can be kept more or less constant by daily opening up the exit valve. So, as the porosity of the bed decreases, with increasing deposition and growth of material in the bed, the value 'k' will decrease and so the resistance to flow in the bed (head loss) will increase.

It is clear that the head loss is also dependent on temperature and hence the density of the water, with low temperatures causing greater head losses. Similarly higher head losses are caused by increased flow rates alone.

The starting head loss of a newly sanded filter, e.g. bed 45 in January, 1975, at a flow rate of 20cm.hr^{-1} was approximately 23cm; towards the end of the first run at a flow rate of 10cm.hr^{-1} it was found to be 84cm. After cleaning, the starting head loss of the second run in March, 1975, had increased, due to deposition of material within the filter, to 46cm at a flow rate of 28cm.hr^{-1} and at the end of this run the head loss had increased to 99cm at a flow rate of 26cm.hr^{-1}. The maximum resistance to flow (head loss) should not exceed 1 - 1·5m and in general the Thames Water Authority make the decision to stop the filter run and clean the bed when the head loss begins to exceed 1 metre.

2.8.3 Cleaning methods

When the head loss has reached a maximum acceptable level, it is necessary to drain the bed in order that the surface skin can be

removed. This is done by closing the raw water outlet and allowing water to drain from the bed, a procedure which takes a few hours and is often done overnight in order to allow cleaning to commence first thing in the morning. (The importance of removing the algal mat quickly and of keeping the cleaning period to the minimum has been emphasised in 2.8.1).

2.8.3.1 Manual cleaning

The large algal mats and bodies of floating Cladophora cannot be removed by mechanical skimmers and so it is necessary to remove them by hand. This is done by workers using large rakes with which they pull the mat masses into rows or heaps in the bed. These are then lifted into dumper trucks and removed from the bed.

2.8.3.2 Mechanical cleaning

This commences as soon as the bed is drained, or when the algal mats have been removed. Ramps are positioned in the bed and machinery is driven onto the bed. In the Thames Water Authority, skimmers mounted on tractors are used. The skimmer consists of a blade which can be mounted between 1 and 3cm below the sand surface and a horizontally positioned screw which removes the dirty sand from the blade and deposits it into a belt loader. The belt loader carries the sand up and away from the skimming area and pours it into a dumper truck travelling alongside the skimmer. When this truck is full it is replaced by another truck and drives its load out of the sand bed and to the washing bays.

Once the bed has been skimmed of its surface skin the surface is smoothed over with the use of a tractor trailing a heavy broad plank behind itself.

This part of the filtration procedure is the most expensive and it has been calculated that for a bed of 0·3 - 0·4 hectares, 85 man hours are required to remove 45 - 75m^3 of dirty sand surface (Ridley 1967). In 1961 (Burman and Lewin, 1961) an 'in situ' method of cleaning a sand filter was developed, but it proved difficult to operate (Ridley 1967) whenever algal growths were present and has not been used since.

When the cleaning process has been completed, secondary filtered water is introduced into the bottom of the filter through the underdrains and the bed is slowly backcharged to remove air from

the interstices. As the water reaches the sand surface, this supply is shut off and water is allowed onto the surface from the rapid sand filters. As has been described above (2.8.1), normal filtration rates are not reached until a few days after the start of the run.

2.8.4 Resanding

After several months use the depth of the sand will have dropped to its minimum acceptable level for efficient removal of suspended material. During operation of the filter some of the suspended material will have penetrated into the bottom 0·3m. In order to prevent this depth of sand from accumulating more and more material without it ever being removed, it is necessary to move the position of this sand with respect to the depth of the filter.

The first procedure in resanding is to dig a trench (using a trenching machine) which is the length of the bed and about 0·5m wide and 0·3m deep. The sand which has been removed in this process is placed to the side of the trench (figure 2.4). The evacuated trench is then filled with washed sand taken from the storage bays. The adjacent strip is then trenched and the sand which is removed is placed on top of the new sand (figure 2.5). This process continues until all of the bed has been trenched. When this has been completed new sand is placed on the bed until the maximum depth of sand is reached (0·75m) and the surface is smoothed down.

When the bed is complete it is backcharged from below and filtration procedures continue as usual.

2.8.5 Sand washing

Sand removed from the filter bed in the cleaning process is transported to the washing bays where it is immediately cleaned. In the Thames Water Authority the washing procedure consists of passing the sand through a series of hoppers (figure 2.6). The dirty sand is fed into the top of one of these hoppers, filtered water is fed, under pressure, into the bottom of the hopper. The effect is to release the organic matter from the sand. This organic matter has a lower density than that of the sand and so it floats to the surface of the hopper and spills over into waste pipes. This sand is then carried from the bottom of the hopper up to the top of the next hopper

Figures 2.4 and 2.5 DIAGRAMMATIC REPRESENTATION OF "TRENCHING"
For explanation see text.

Figure 2.4

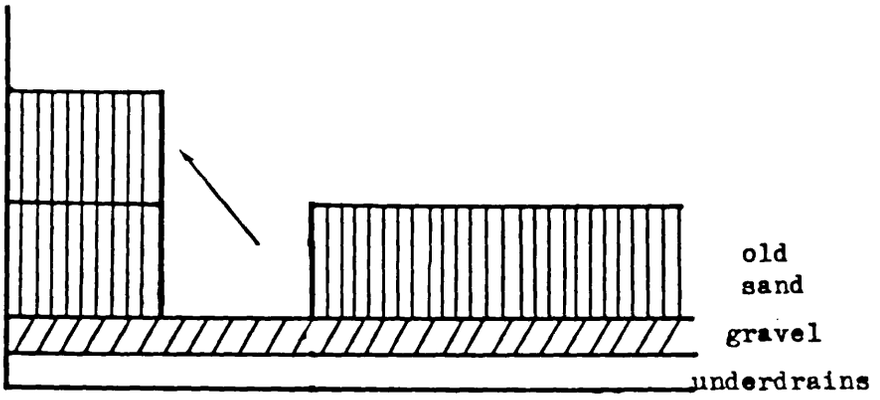


Figure 2.5

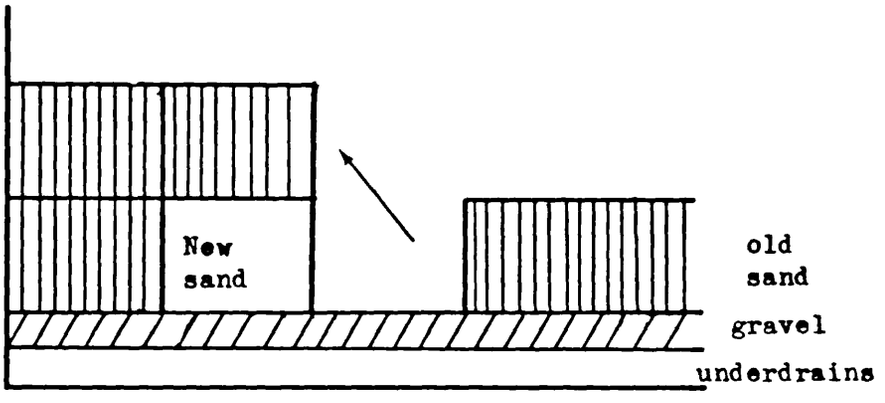
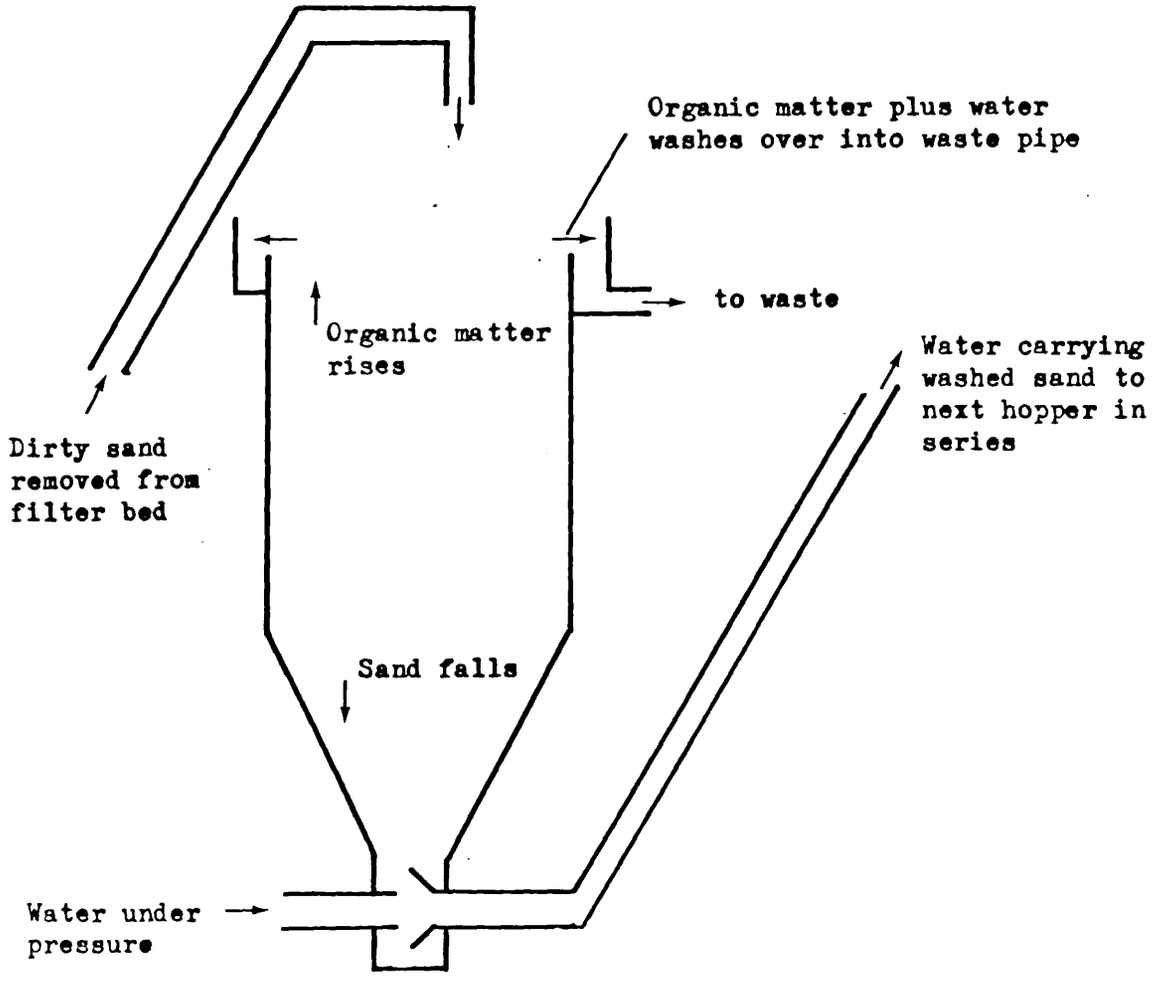


Figure 2.6 DIAGRAM OF SAND WASHING HOPPER, ONE IN A SERIES



in the series and the process repeated until most of the organic matter has been removed. There may be 4 or 5 hoppers in a series.

The washed sand is then stored in adjacent storage bays until it is required for use in resanding a filter.

The washing procedure does tend to reject the finer grains and so a check must be kept on the grading of the sand. In the past strict size gradings, such as those recommended by Hazen (1892) and Holden (1970) have been adhered to. More recently it has been found that the use of builders' sand (Metropolitan Water Board 1974) to replace the lost sand caused no decrease in efficiency of the filtration and was much cheaper than well graded sand.

2.9 Slow sand filtration and the artificial recharge of aquifers

Water pumped from underground aquifers often requires no treatment, apart from a cautionary chlorination to prevent reinfection from breaks in the water distribution system. The reason for this is due to the natural purification which water undergoes as it passes through soil and subsoil strata into porous rock. An obvious comparison can be drawn here between the treatment that natural water receives as it passes into aquifers and that which river derived water receives as it passes through slow sand filters before being put into supply.

With increasing urbanisation, industrial development and rising populations, the demand for water has increased enormously since the start of the century. The result was that the number of groundwater sources began to be inadequate to meet the demands of the consumer. The first experiment with artificial recharge of aquifers took place in Götenborg in Sweden in 1897. Here the amount of water naturally entering the aquifer was artificially increased, but there was found to be no subsequent decrease in the quality of the water later abstracted (Huisman 1967).

It is clear that great importance must be attached to the way in which water is introduced to these underground aquifers. It would, for example, be disastrous if polluted water were directly injected into an aquifer without it having received any form of treatment, as once the aquifer itself has become polluted, it would be very difficult to purify it. Most experience of the use of artificial recharge is found on the continent.

There are 2 basic methods by which artificial recharge is effected.

2.9.1 Induced recharge

This concerns aquifers which are positioned close to surface water bodies which have pervious beds. Ground water is abstracted from these aquifers, thus lowering the ground water level in the surrounding regions and causing an increased downward flow of water from the surface water bodies. This method does, however, suffer from a reduced capacity as the river beds become blocked with deposited organic matter and also cannot be controlled as well as artificial recharge, and so it is less widely practised.

2.9.2 Artificial recharge

This method can consist of flooding a pervious surface with water, or can be caused by pumping water directly into an aquifer via a shaft or well. As mentioned above, the latter method can result in pollution of the aquifer and so must be viewed with reservation. The former method often employs a slow sand filtration system in which water is allowed to percolate through sand beds before it is directed into the pervious substrata of the aquifer.

2.9.3 Use of slow sand filters in artificial recharge

Basins used for artificial recharge can be covered in a layer of sand 0·5 - 1m deep and with a sand grain size distribution equal to that used in slow sand filters used for the direct supply of water. The filter should function very similarly to that of an ordinary slow sand filter with removal of suspended material occurring within the body of the filter. Filtration rates are however slower, at 1 - 10cm.hr^{-1}, than those normally experienced in treatment works (Huismann and Wood 1974) as the water continues percolating through substrata once it has left the slow sand filter itself. When the filter develops a head loss which impairs its efficiency, it is possible to drain the bed and remove the surface skin as described for slow sand filters in treatment works.

Much of the work concerned with slow sand filtration on the continent has been concerned with slow sand filtration as part of the artificial recharge scheme (e.g. Husmann 1968).

CHAPTER 3 FIELD WORK - SAMPLING METHODS

3.1 Sampling area

The slow sand filter bed examined was bed No. 45 at Hampton Treatment Works of the Thames Water Authority. This bed is in a group of four beds (plate 1) which have been used by the Metropolitan Water Board and more recently the Thames Water Authority, for research purposes. All these beds are operational and bed 45 has been used for some research programmes by the Metropolitan Water Board (M.W.B. 1974) and the Thames Water Authority prior to the start of this study.

Whilst this work was in progress various monitoring work was carried out by the Thames Water Authority on bed 45, mainly with respect to recording carbon levels in the sand at the end of a run and also oxygen levels in the surface and outflow water.

3.2 Problems associated with sampling this medium

There were several problems to overcome in attempting to sample the sand filter for the organic material contained within it.

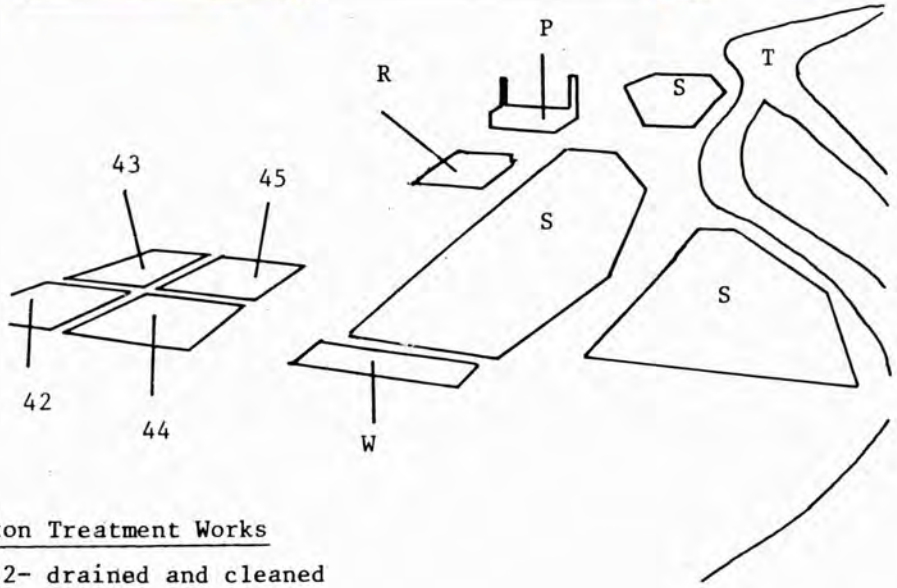
1. The compactness of the sand mediumitself prevented any coring device from being pushed in to a depth greater than 10cm without seriously disturbing the sand within the core.

2. As the sand surface is covered by a layer of water approximately 1m deep, all sampling procedures had to be carried out from a boat. Also, due to the high levels of suspended organic matter and growths of Cladophora in the water during the summer months, it was often difficult to see the surface of the sand bed. As a consequence all sampling gear needed to be of a relatively simple construction and all sampling procedures had to be quick and easy to work from the edge of a boat.

3. As it was necessary to examine the vertical distribution of the meiofauna in the filter, a method was necessary to remove sections from a sand sample that were representative of the depth from which they were removed.

4. In sampling an operational and not an experimental bed, it was necessary that the sampling method in no way affected the quality of the water coming out of the filter. Mainly this meant that the sampling procedure (a) should leave no holes in the filter through which potentially harmful bacteria and viruses could pass and (b) should not allow the relatively clean sand in the body of the filter to get mixed with

Plate 1. Aerial view of Hampton Treatment Works, photograph courtesy of the Thames Water Authority.



Hampton Treatment Works

Bed 42- drained and cleaned

Bed 43- drained only

Bed 44- full of water

Bed 45- full of water

S- slow sand filter beds

T- River Thames

R- rapid sand filter units

P- power house

W- washing hoppers and storage bays

the surface skin.

5. Any sampling method devised would have to cope with the fact that the depth of the sand decreases by about 3cm after each run, due to the method of cleaning the filter, so a bed which started off with clean sand at a depth of 76cm could be reduced to a minimum depth of 30cm when it would need resanding. (Approximately 76cm is the depth of sand used by the Thames Water Authority in the filter beds and this is gradually reduced by cleaning processes to a depth of 30cm, i.e. the minimum depth considered by the Thames Water Authority to be effective for removing water-borne particles).

3.3 Apparatus - discussion of possible designs

The main types of apparatus used for sampling the meiobenthos have been reviewed by Wells (1971). There are basically three types of apparatus used to sample meiofauna : corers, grabs and dredges. The latter two types are really not suitable for use in a slow sand filter as their method of removing sand will inevitably allow surface skin and cleaner deeper sand to become mixed in the bed. They are not very quantitative and do not allow a detailed examination of the vertical distribution of the meiofauna.

The corer type of apparatus is really the most suitable for this particular medium provided that the device does not disturb the sand as it is pushed in. The difficulty of obtaining undisturbed cores has been discussed by several authors and in this context by McIntyre (1973), Wells (1971), Hesslein (1976) and Gilpin and Brunsven (1976). My own experience is that it is only possible to sample below 10cm of slow sand filter sand by quickly rotating the corer around its long axis, a procedure which immediately causes the sand core itself to become disturbed. Several authors have penetrated to depths greater than this: Jansonn (1967), Maitland (1969), Cox (1976), Baker et al (1977), McLachlan et al (1977), but in all cases no conclusive evidence was presented to show that there was no disturbance of the contents during the procedure of obtaining the core. All these methods would cause mixing of adjacent areas of sand outside the corer whilst the corer was inserted and removed.

Once the core has been removed from the medium it is necessary to section it vertically for examination. Methods which use a piston to push the core from the corer will again disturb the surface of the sand and cause compaction of the sample, a fact noted by Rittersbusch

(1974) when sampling a sand filter. Methods which avoid this fall into two groups. The first type uses a released pressure device Maitland (1969), Baker et al (1977), which allows the core to slide slowly from the corer so that it can be sliced as it emerges. The disadvantage with this type is that some mixing of contents may occur as the core slides from the corer. The second type involves the use of an internal tube, fitting tightly inside the outer jacket, which is cut up into rings of varying diameters. These rings can be pushed out of the outer jacket by a piston and the core sliced as they emerge, Jansson (1967), Cox (1976). Alternatively the outer jacket may split longitudinally into two halves,(O'Connor, 1957), thus exposing the rings contained within, which can then be separated.

Hynes (1974) and Gilpin and Brunsven (1976) have solved the problem of compaction and disturbance of the core during insertion of the cores into the sand, by using a different method altogether. They dug a hole in the sand, inserted an empty tube which had holes punched out of its sides and filled it up with sand. This was then left for organisms to colonise it, then it was removed and examined. However this method does allow adjacent sand to fall into the hole left when the core is removed. A similar type of method developed by Coleman and Hynes (1970) used a core which was filled with sand and placed into a hole in the bed of the stream. In this situation, however, the bed of the stream was prevented from collapsing into the hole when the core was removed by the presence of an outer sleeve which surrounded the inner core in the stream bed. Both the outer sleeve and inner core were made of perforated material, but had only a limited success in allowing horizontal migration of animals to take place. Again, the contents of the core had to be mechanically pushed out for depth sectioning to occur. A method devised by Conrad (1977) used several unperforated cores containing sand which were attached to a perspex frame and held in both horizontal and vertical positions. The frame was placed in a hole dug in beach sand and left for organisms to colonise. The method was interesting in that it allowed separate monitoring of horizontal and vertical migrations into the cores, but would be of no use in this present situation, as positioning and removal of the cores would cause considerable disturbance of the sand filter.

Lloyd (1973) and (1974) devised a method specifically to examine the interstitial meiofauna of a slow sand filter. He made a

column from two strips of glass 30cm x 25cm, kept 1mm apart. This was filled with filter sand and inserted into the slow sand filter prior to the start of a run. At intervals these cores were removed and placed under a microscope and the microfauna counted. This had the advantage that there was no compaction of the sand during manipulation of the core and, as the apparatus was so thin, there was no mixing of surface and deeper sand when the apparatus was removed from the bed. One disadvantage of this method is that dried sand was used to be placed inside the apparatus as wet sand would not flow in easily, the result being that the flora and fauna normally present in the sand bed would not be present in this situation. A more serious problem with this method concerns the internal surface area to volume ratio of the apparatus which is very high and could appreciably alter the filter performance within the pores of the sand in the apparatus - see discussion below.

A method used by Doohan (R.H.C. pers. comm.) developed the concepts used by Hynes (1974), Gilpin and Brunsven (1976) and O'Connor (1957). She used a perspex core (I.D. = 55mm, depth 170mm) which was split longitudinally into two halves, one of which was cemented to a circular perforated perspex base plate. When the two halves were fastened together and filled with sand, this inner core could be placed into a hole which had previously been dug in the sand, the sides of which were prevented from collapsing into the hole by the presence of an outer core (I.D. = 70mm, depth = 200mm) positioned inside the hole. Thus it was possible for organisms to colonise the sand core which could be removed at intervals and examined. The fact that the core could be split open meant that the sand column could be cut up into sections without any compaction of the core. This apparatus was developed to examine the rotifers of a slow sand filter and so sampled only the top few cm's of the bed where they were most abundant. For reasons discussed below, the diameter of the inner core was really too small and meant that the very high surface area to volume ratio probably caused erroneous filtration effects within the core. The gap between the inner core and the outer core meant that at this point the depth of the filter was reduced and so there was a possibility of potentially dangerous microorganisms passing out of the filter.

3.4 Apparatus developed

The apparatus consisted of two parts, an inner core and an outer core.

Materials used in construction (appendix 3.1)

Inner core

This was made from 31cm lengths of grey uPVC drainpipes (I.D. = 100mm, wall 3·2mm), sawn longitudinally into two equal halves, towards the end of one of these halves a base plate was attached inside of and at right angles to the long axis of the core (figure 3·1 and plate 2.1 and 2.2).

Base plate

This was circular and made from clear perspex (diameter = 100mm, 6mm thick), holes of 3mm were drilled at distances of 5mm apart. At one point a screwhole was tapped horizontally into the thickness of the plate. This screwhole coincided with a hole at the base of one of the inner core halves and a screw was used to keep them together, so it was possible to detach the two parts if necessary for cleaning. Two small support blocks (35mm x 10mm x 3·2mm), which had been cut from scraps of the inner core, were glued to the inner core just beneath the base plate to help support the weight of the sand.

Outer core

31cm lengths of grey uPVC drainpipes (I.D. = 150mm, wall 4·1mm).

Collar

A collar was made by cutting a plastic funnel so that the small diameter of the collar (diameter = 10·8cm) would just fit round the outside of the inner core and the large diameter (15cm) would just fit inside the outer core. The depth of the collar was 3·8cm.

Assembly of the inner core

The base plate was screwed to one half of the inner core. The two halves of the inner core were held together by two adjustable jubilee clips (9 - 11cm) and the split between the two halves was sealed on the outside of the core with plastic adhesive tape. A single piece of nylon string was attached to the lower jubilee clip and passed inside the upper clip extending for about 10cm above the top of the inner core.

Inner core marker and float

This consisted of a spherical polystyrene float (diameter = 8cm) painted white labelled with red paint for identification, attached to a

Figure 3.1 DIAGRAM OF SAMPLING APPARATUS - INNER CORE, BASE PLATE AND COLLAR (Not to scale)

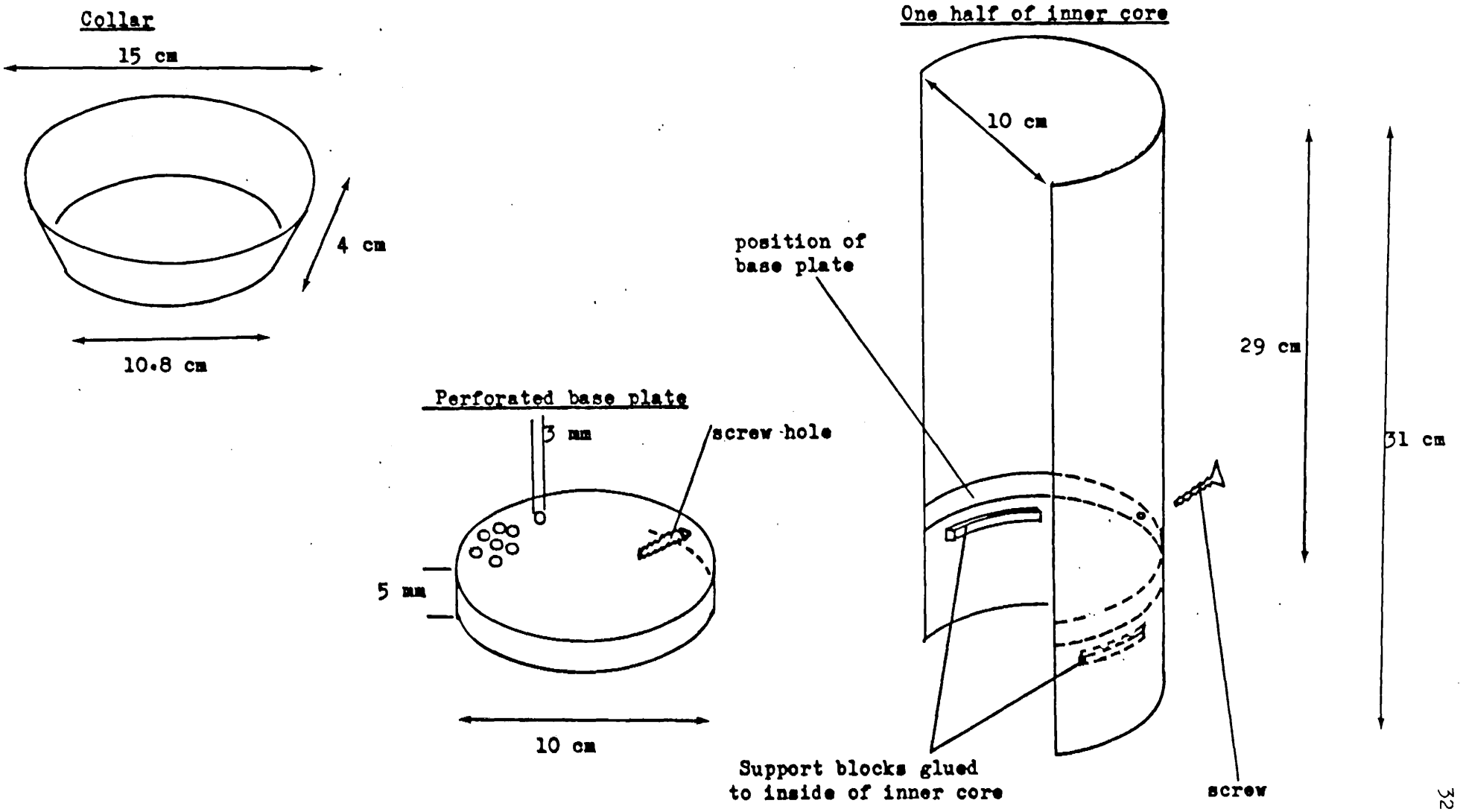


Plate 2.1 Sampling apparatus, inner and outer cores with floats.

Plate 2.2 Details of inner core, assembled and unassembled components.

1m length of cane by nylon string. A second piece of nylon string with a free swinging loop was attached to the opposite end of the cane. As no weight bearing was necessary it was possible to attach this loop to the nylon string of the inner core by some kind of simple fastener, in this case a paper clip was used (plate 2.1).

Outer core marker and float

A small hole was bored at the top of the outer core through which passed a piece of nylon string permanently attached to a 1m length of cane, the opposite end of which was attached to nylon string and a white painted polystyrene float labelled with yellow paint for identification.

3.5 Sampling procedure

3.5.1 Filling the inner cores

Freshly washed sand was collected from the sand storage bays and transported back to the laboratory on the day prior to the start of a new filter run. On the day of a run each inner core was assembled and filled with the damp sand. These inner cores were then placed in empty tanks and slowly backcharged with water from below. The rate was sufficient to wet the sand only, without causing any sorting of the sand particles. When the water level reached the top of the sand, the sand level was seen to have settled a few cm's, at this point more washed sand was added and backcharging continued until no more settling occurred. The wetted inner cores were then transported to Hampton filter works.

When the first cores were removed from the bed for examination, the contents could be seen to have settled a depth of not more than 1cm. This was to be expected following the backcharging processes which occurred prior to the start of the filter run. The contents of cores which were removed on subsequent sampling trips were similarly observed to have settled not more than 1cm. When the time came for the contents of the core to be sectioned for depth examination, the depths chosen were measured from the sand surface and not the top of the sampling apparatus.

3.5.2 Locating random core positions in the bed

It was decided that in order to ensure that the cores were positioned randomly in relation to one another, some system of using random coordinates would have to be developed.

Bed 45 is quite large ($6400m^2$) and if random positions were to be

located, it was necessary for the area to be sampled to be reduced, in order to allow random positions to be located from fixed datum points using a system of random coordinates. The size of each random unit was chosen to be equal to the diameter of the outer core. Thus a sampling square was chosen, the sides of which were equal to 100 times the diameter of an outer core, i.e. 15·84m. The area of the sampled region was, therefore, 250m^2, i.e. 4% of the total bed area.

This sampling square was positioned approximately in the centre of the bed and relocated each time the random grid was set out (i.e. at the start of each new run) from 2 chalk marks on the side of the bed, V and W (figure 3.2). The grid was placed centrally in the bed to avoid any edge effects.

After locating points V and W on the side of the bed a tape measure was stretched between the two and points X and Y were located in the centre of the bed. At these points two wooden 1m length poles were pushed in the sand and the end of a tape measure attached to each.

Previously eighteen sets of random coordinates a and c had been obtained from random figure tables and a and c varied from 0 to 99, with each unit in reality measuring 15·84cm. From these figures the shortest distances of the point from X and Y were calculated, using Pythagoras's Theorem :

x being the shortest distance from X

$$x = (0{\cdot}16.\sqrt{a^2 + c^2}.) \text{ metres}$$

y being the shortest distance from Y

$$y = (0{\cdot}16.\sqrt{b^2 + c^2}.) \text{ metres}$$

The x and y coordinates were recorded on separate pieces of paper and each given to one person whose tape measure was attached to pole X and the other person's tape measure attached to pole Y. Then by joining together the appropriate x and y coordinates, random positions were located in the grid. As each point was located a third person placed an outer core in position (figure 3.3).

When this procedure was complete, all 18 outer cores were in position. Holes were then dug large enough for each outer core (figure 3.4), the outer core placed inside with the top edge level with the top of the filter bed and the sand was firmed down.

Figure 3.2 DIMENSIONS OF FILTER BED 45, WITH POSITION OF SAMPLING SQUARE SHOWN

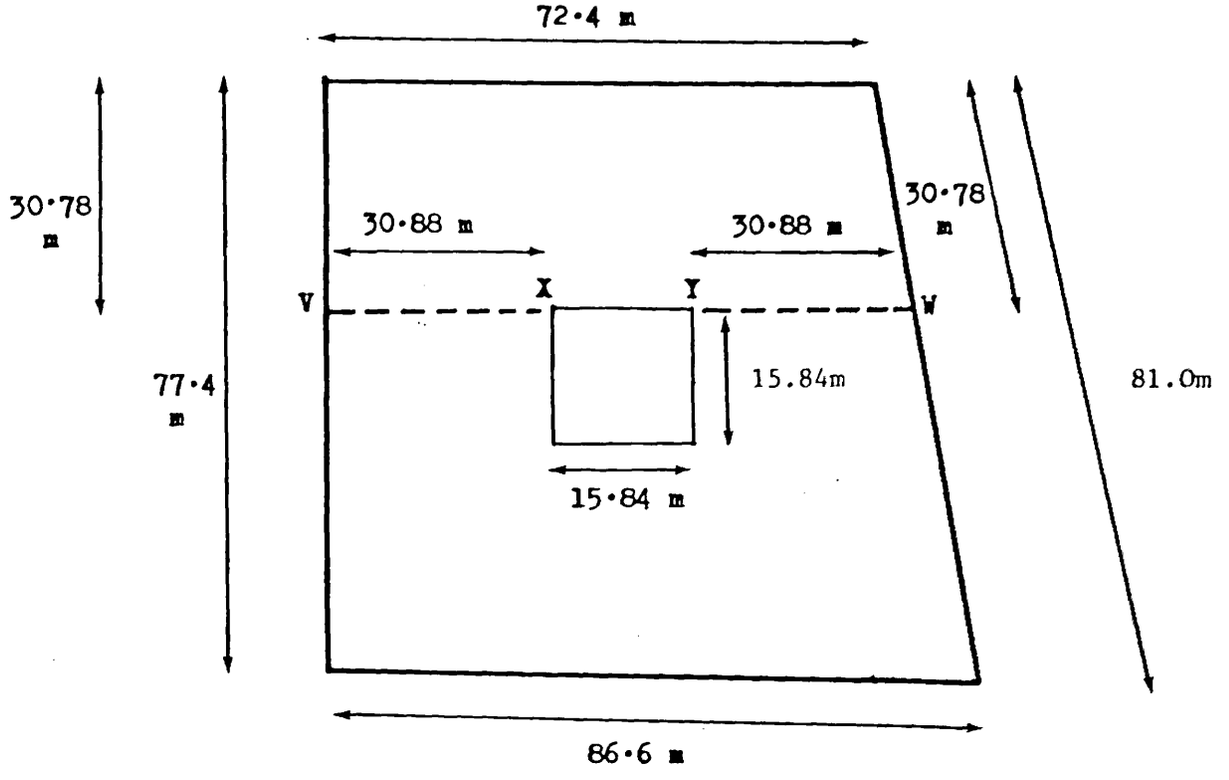


Figure 3.3 LOCATION OF RANDOM SAMPLING UNIT IN SQUARE
For explanation see text.

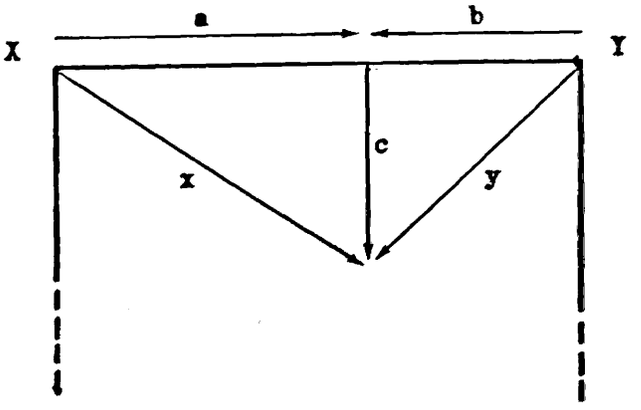
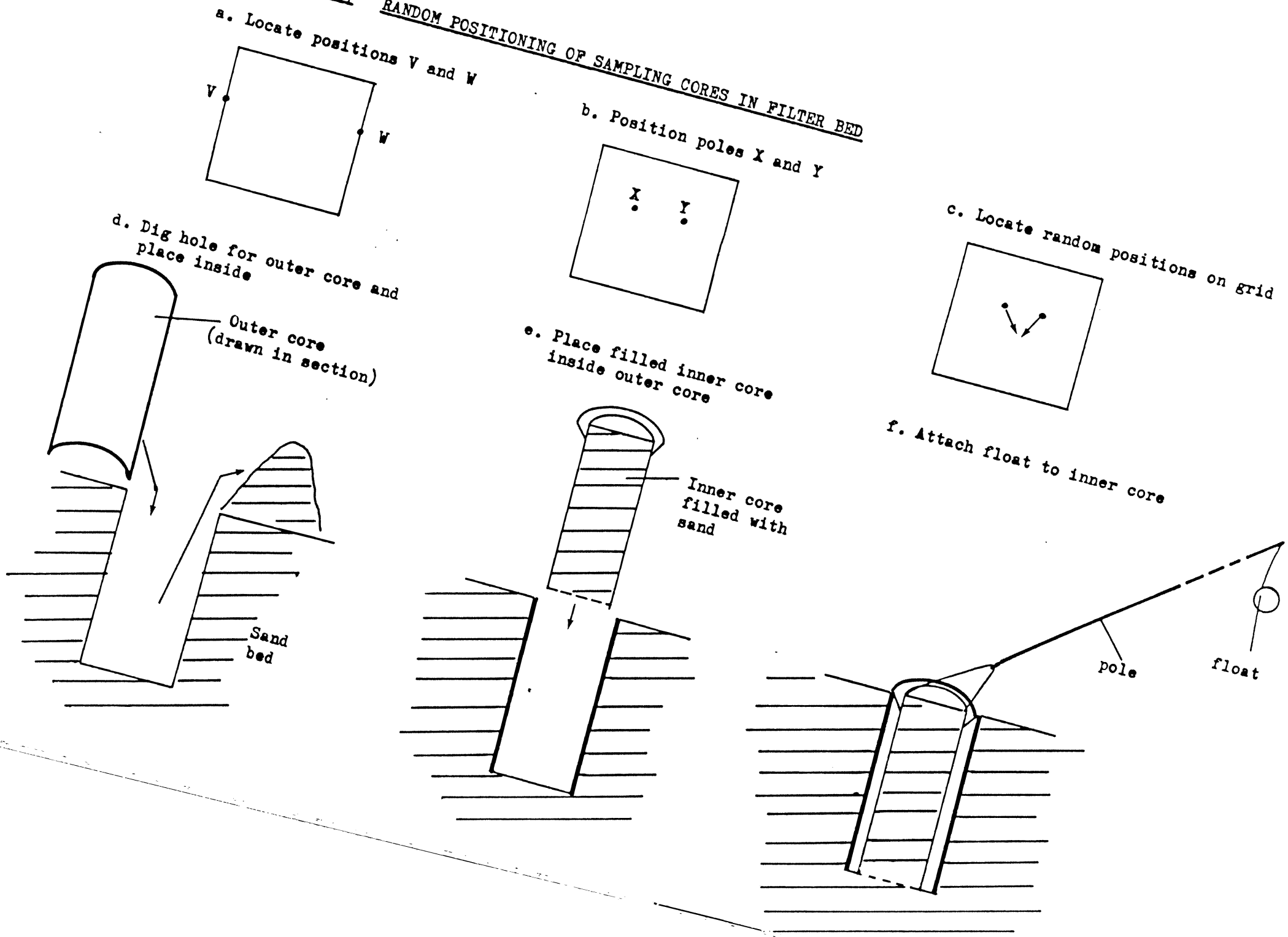


Figure 3.4 RANDOM POSITIONING OF SAMPLING CORES IN FILTER BED



The inner cores were then carried out to their respective outer cores and placed inside them, with their tops level with the surrounding sand.

The float belonging to the inner core was attached to the nylon string of the inner core by means of a paper clip.

Sand removed to produce the hole was scattered around and smoothed down. The bed was then vacated and the filter foreman informed that the procedure was complete and backcharging was started as at the start of a normal filter run.

3.5.3 Sampling procedure proper

The day following the location of random positions for the cores it was necessary to remove three cores for examination. This means that the depth of sand where an inner core has been removed from, would be 30cm shallower than the surrounding sand, a condition which cannot be tolerated in an operational bed. So it was necessary to replace all cores removed for analysis with identical cores containing clean sand.

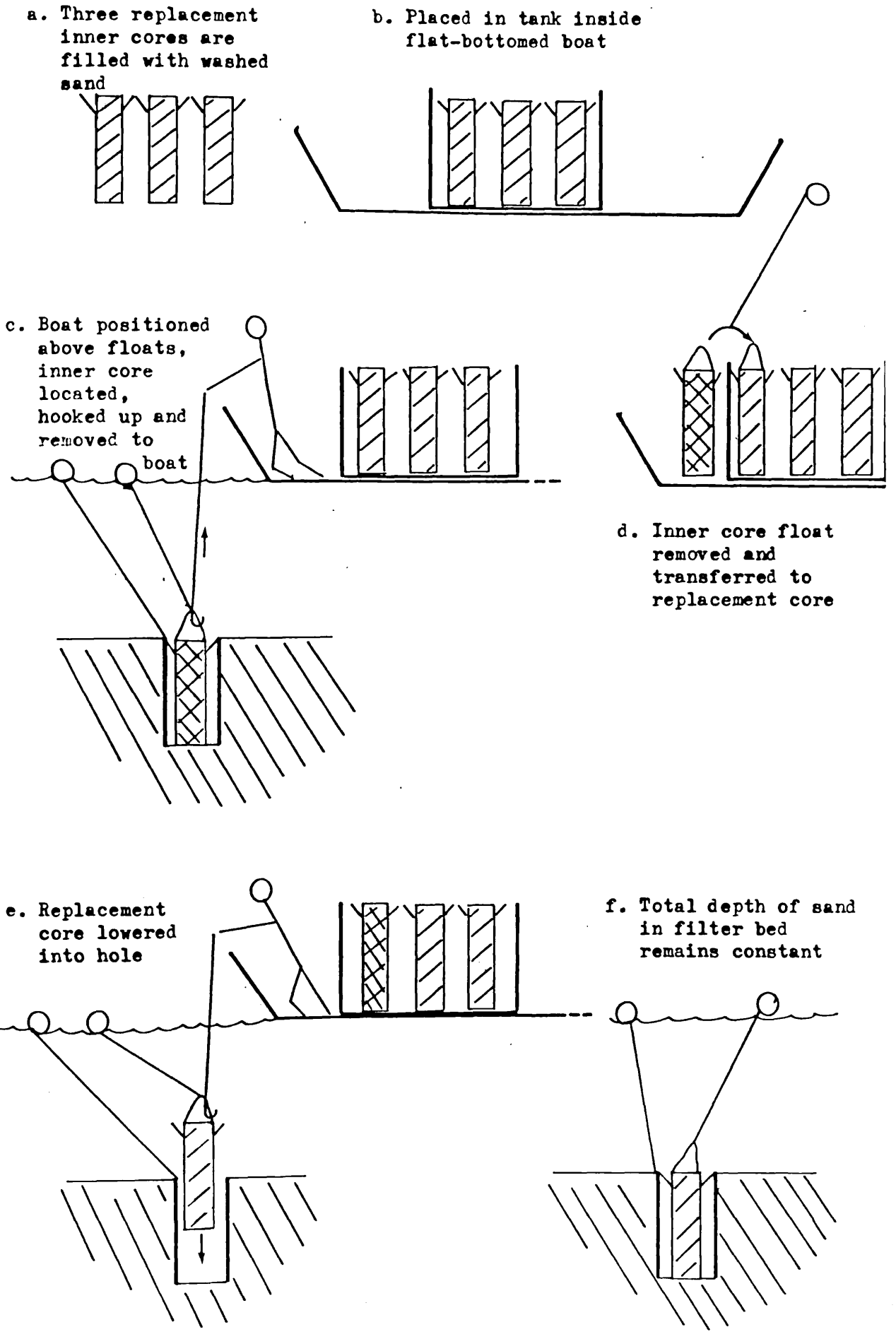
The morning of the sampling trip three inner cores were assembled and filled with washed sand as described above. These were then transported to the filter works. A flat-bottomed boat was used in this process, requiring one person to steer the boat and keep it steady in position over the embedded cores - no anchorage was possible. A second person was required to remove the cores (figure 3.5).

The boat was loaded with a fibreglass tank containing the three replacement inner cores stored so that they would not fall over. The person who was going to be removing the inner cores from the bed required a diving mask - which greatly facilitated viewing the surface of the bed and a pole with a hook on one end. In winter it is necessary for this person to wear a wet suit jacket and helmet.

Three cores had previously been selected for removal. The boat was positioned above the first one. The operator hung over the front of the boat and located the inner core by means of the float. This inner core was then pulled up by hooking around the attached nylon string of the inner core. This was then secured in the boat.

The float from this inner core was then removed and attached to the replacement inner core in the tank. This replacement inner core was then lowered with the hook into the water, using the outer core

Figure 3.5 DIAGRAMMATIC REPRESENTATION OF SAMPLING PROCEDURE
(Not to scale)



float to locate the hole into which the inner core was then placed.

This process was then repeated removing two more inner cores from the bed and replacing with inner cores containing washed sand. These cores were then returned to the bank for examination.

At chosen intervals of time - usually 1 day after the bed run started, at fortnightly intervals, and on the last day of a run - three inner cores were removed for examination, so that the ages of the inner cores were identical with the age of the run at that time.

When a slow sand filter run finally ended, the bed was allowed to drain for cleaning, but before this could commence it was necessary to remove all outer cores and inner cores from the bed. No sand collapsed into the holes at this stage as it was still quite wet. The holes were then filled with washed sand collected from the storage bays and the filter foreman informed that cleaning could commence. The sampling apparatus was then transported back to the laboratory where it was dismantled, cleaned and if necessary repaired, ready for replacing in the bed prior to the start of the next run in two to three days' time.

3.5.4 Vertical sectioning of the core

The inner cores were returned to the bank and immediately cut up. The time which elapsed between removing a core and laying it horizontal prior to cutting up, varied, depending on the ease with which the inner cores could be removed from the bed. This was normally related to visibility of the water and the presence or absence of suspended growth of Cladophora. But an average time from removal of first core to the cutting up procedure would be about ten minutes, during which time migration of animals from the column could occur. However, an examination of vertical distribution of most groups indicates that this does not seem to occur to a very large extent, as can be seen from the high numbers of some groups in the top 1 - 3cm. Also, due to capillarity the core did not dry out during this period and was still very wet when cut open. The procedure was as follows (figure 3.6) :

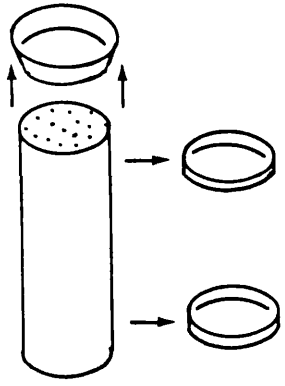
a) The collar and jubilee clips were removed.

b) Two perspex hemicircles (diameter = 10cm) were placed on the surface of the sand - to prevent sand spilling.

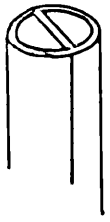
c) The core was then placed horizontally on two wooden support blocks. The plastic adhesive tape was removed and by pulling a piece

Figure 3.6 SECTIONING OF THE SAMPLING APPARATUS

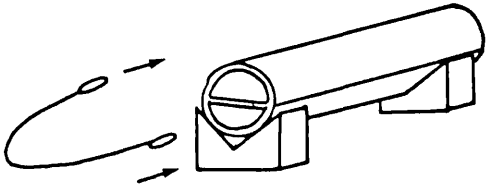
a. Collar and jubilee clips removed from inner core



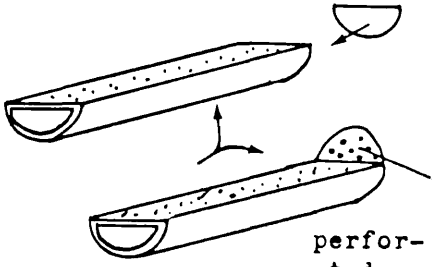
b. Two perspex hemi-circular pieces placed on upper sand surface



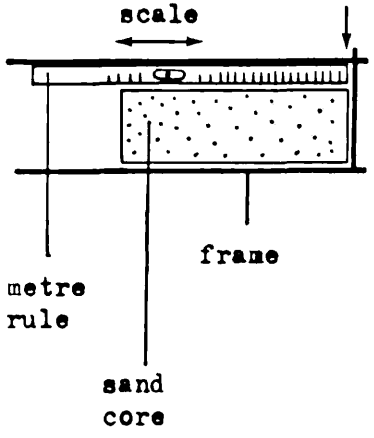
c. Core placed on two wooden supports and sliced longitudinally using wire cutter



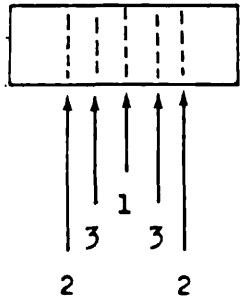
d. Core is separated into two. One hemicircular perspex piece placed on end with no support



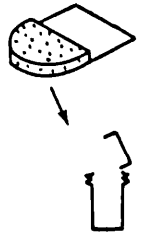
e. Core placed in frame with adjustable side. Top surface of core aligned with zero on scale



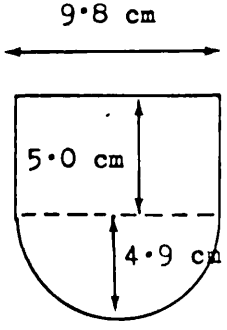
f. Sequence of placing slicers in core



g. Sand slice is placed into screw top jar



Dimensions of aluminium slicer



thickness = 0·9 mm

of wire through the gap between the two halves of the inner core, it was possible to separate the sand core into two equal halves.

d) A third piece of perspex hemicircle was positioned at the bottom of the half core which had no base plate attached. To reduce the possible effect of animals migrating from the core due to water draining from it, procedures c and d were carried out on all cores so that they were lying in a horizontal position before any core was sectioned.

e) One half was then placed in a frame containing a movable metre rule which was zeroed in line with the top of the sand core.

f) Using a second rule held at right angles to this movable rule and with the use of aluminium slicers, it was possible to slice up the core into 1cm thick sections. As the sand would not compact easily, addition of these slicers tended to make the core move sideways, but the effects of this could be reduced by placing the slicers into the core in a certain sequence - see figure 3.6.f and chapter 3.5.5.

g) Each slice was then removed and placed in a labelled bottle with a screw top lid.

h) The samples were then returned to the laboratory for examination (plates 3.1 and 3.2).

3.5.5 Vertical sectioning of core - errors

There is a possibility that by adding all the aluminium slicers to the core that sand may be shifted as each one is added, so that by the time the final slicer is pushed in some of the sand may be positioned in a very different place from its original position in the filter.

This was tested by measuring the volume of the sand grains within each slice. This particular method was also compared with a method subsequently used by Marian Goddard (R.H.C. pers. comm.) in order to improve this particular part of the procedure. This apparatus consisted of a piece of perspex (30 x 12 x 0·6cm) with slits drilled in at 1cm intervals, the size of the slits being just wide enough for one slicer to be pushed through. This apparatus was placed flat on top of the cut side of a core, the top edge aligned with the top of the core and slicers pushed through the slits into the sand in the same way as in figure 3.6.f.

When metal cutters are placed in a core containing sand, the

Plate 3.1 Top- one half of an inner core immediately after being cut in half longitudinally.
Bottom- inner core contained within slicing frame, with slicers in position.

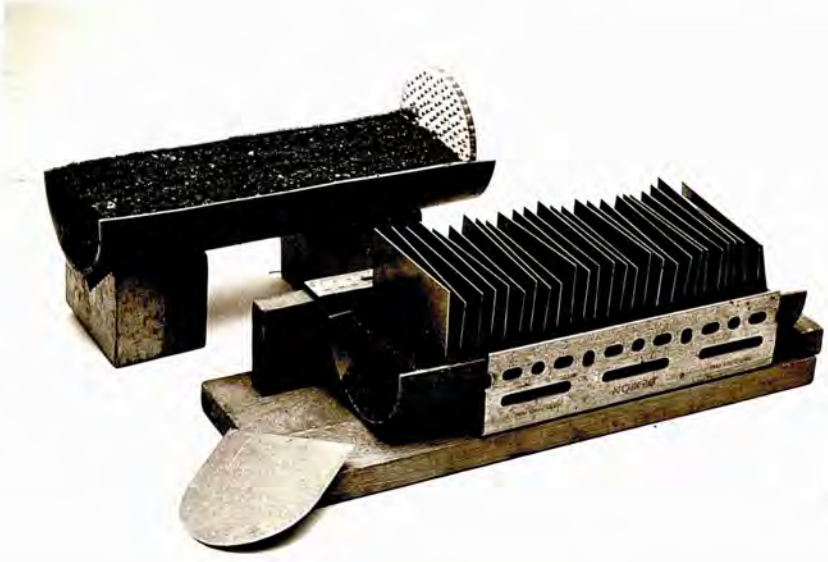
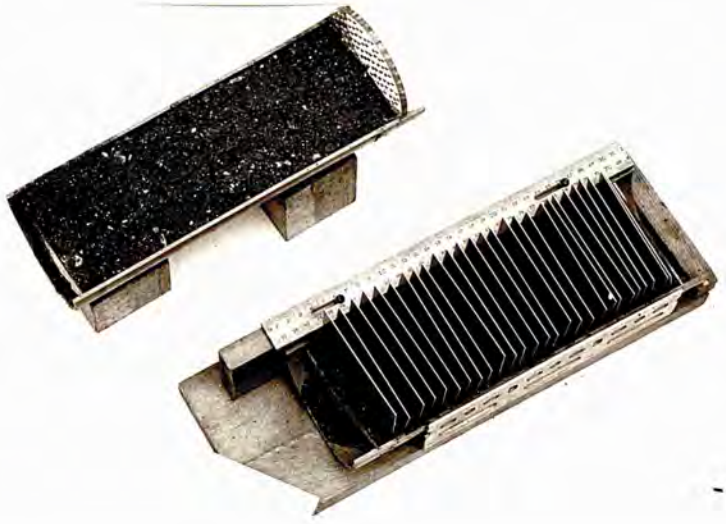


Plate 3.2 View of contents of plate 3.1 from above.



'sand' is compressed, i.e. the volume of the pores is reduced, but the volume of the sand grains is not. So to examine whether or not this sectioning of the sand tends to displace sand to either side of the cutter it is better to measure some parameter related to the characteristics of the sand grains themselves, rather than the total volume contained by the slicers. In this case the volume of the sand grains was measured.

Method

One inner core was removed from the bed, returned to the bank and sliced longitudinally into two halves as described above. One of these halves was then sliced up using the adjustable scale method, to position the slicers, the second half was sliced using the frame method to position the slicers. In each case the sand grains from each slice were removed from the core and dried at 60°C for 48 hours. A specific gravity bottle was filled with distilled water at 20°C (= ag.). Half of this volume was poured out, reweighed, the sand grains added and reweighed. The weight of sand was calculated (b.g). The mixture was then boiled gently to release trapped air, cooled to room temperature, filled with distilled water and reweighed (c.g).

The increase in weight (c - a) was due to the difference between bg of sand and the weight of an equal volume of water. The volume of sand is therefore equal to b - (c - a)ml. For results see table 3.1, using a comparison of means t-test. It would appear that as far as the volume of the sand contained within the slicers is concerned, there is no significant difference ($p > .05$) between the two methods used to slice the core up. However, the method using the perspex frame did produce less variability about the mean (SE = 3·3% of $\bar{X}$) compared with the adjustable scale method (SE = 5·5% of $\bar{X}$). But in both cases this is very low and indicates that there is very little displacement of the sand during the sectioning procedure. However, the perspex frame method is slightly quicker to perform - it took two minutes to complete, compared with the three minutes with the adjustable scale method, but this difference is small compared with the ten minutes which elapsed after the core was removed from the bed and before it was sectioned.

3.6 Comments on sampling apparatus

3.6.1 Dimensions of apparatus - diameter

From a consideration of fundamental mechanisms of filtration, it can be seen that it is the behaviour of the particles inside the pores

Table 3.1 Results of comparison of sand cutting procedures.

	Sand volume	
	Adjustable scale method	Perspex frame method
n	27	26*
$\bar{x}$ ml.	23.159	24.463
S	3.159	1.994
d.f. (n-1)	26	25
t,(p=0·05)	2.056	2.060
S.E.	1.265	0.806
S.E. as % of $\bar{x}$	5.5%	3.3%

* one bottle cracked during experiment.

Comparison of means, t-test
d.f.=51, t=1.789, p=0.1

which governs filter performance. So long as the pore scale and particle size are not altered, the size of the vessel containing the filter media is not important. However, at the boundary of the vessel and filter media atypical pores will be formed, (Ives, 1966) which could seriously affect filter performance if they occupy too great a percentage of the total pore space in the filter.

It has been empirically established that if the diameter of the model is at least 50 times the largest grain size, such boundary effects are negligible, (Ives 1966). For sand removed from Hampton the largest appreciable grain size is the 2mm fraction and so a 10cm diameter tube is applicable here (Ives 1966 and 1970).

This is a condition which the Lloyd (1973) and Doohan samples do not fulfil and suggests that the amount of atypical pores in these samplers may affect filter performance in the sand contained within the cores.

3.6.2 Dimensions of apparatus - depth

Ideally the whole depth of the filter should be sampled, i.e. the depth of the cores should be 75cm. However, due to the cleaning process the depth of the bed is reduced at a rate of about 3cm per run, so that the shallowest depth used to filter water is 30cm. It is obvious that using 75cm cores it would not be long before the cores were seriously protruding from the bed, thus affecting conditions within the sand core. This problem could be solved by using several sets of apparatus of decreasing depths, but this would have meant making 15 sets of apparatus and would have been too expensive and so was not feasible.

From a preliminary depth examination of the sand core it would appear that when sampled the majority of organisms occur in the top 15cms and many do not appear below the top 5cm. So it was decided to use cores of 30cm depth in order that the top 30cm of any slow sand filter could be sampled and only one set of equipment was necessary.

Of course this inevitably means that for some runs the whole depth of sand was not sampled and so some events may have been missed.

3.6.3 Sand used to fill cores

Ideally the inner cores should be filled with sand of the same age as that in the bed. However, the fact that the holes left at the end of a run, after removal of all apparatus, were filled with washed sand

meant that certain areas in the bed contained sand of differing ages. As it was not possible to mark these areas one could not be sure where these regions were and so alternative arrangements had to be made.

It was decided to fill the inner cores with washed sand at the beginning of a run as one was then assured that this sand at least had a reasonable degree of uniformity.

However, the use of this procedure means that in monitoring this sand one is monitoring changes occurring within the sand placed in the inner cores and not necessarily the changes occurring within the slow sand filter itself. On the other hand migration into this sand is possible at all times and can be seen to happen.

3.6.4 Monitoring over the whole 'life' of a bed

As all the cores have to be removed for the cleaning processes to be carried out, and all cores have to be filled with new sand at the start of a run, it was impossible to monitor the 'ageing' processes of the bed as a whole - over a period of two years before it needs resanding. The only way this would be possible would be to have several complete sets of apparatus embedded at various depths within the slow sand filter which emerged as the cleaning process revealed successive layers of sand. This would be very expensive and would very likely result in many of the cores being broken by heavy cleaning machinery travelling over them.

3.6.5 Horizontal migration of animals in the core

Due to the nature of the inner core it is clear that horizontal migration of animals into or out of the core will be prevented. This procedure was allowed to occur in samplers of the Hynes (1974) and the Gilpin et al (1976) types due to the presence of holes in the sides of the cores. However this type of apparatus needed to be in intimate contact with adjacent sand outside the core. The result of this was that when the core was removed adjacent sand would collapse into the hole left behind and so was not applicable in a slow sand filter situation.

Despite the constraint of limited horizontal migration there is nothing preventing vertical migration occurring between the core and the adjacent sand bed through the holes in the base plate.

3.6.6 Main advantages of the sampling apparatus

a) The sand filter can be sampled without disturbing the sand in the process.

b) Sand samples can be removed during the course of a slow sand filter run and not just at the start and end - which previously was the only time available.

c) The use of the outer core prevents adjacent sand from collapsing into the hole vacated by removal of an inner core and thus prevents skin and deeper clean sand from being mixed.

d) The overall depth of a slow sand filter itself remains constant due to the refilling of the hole after sampling by an inner core containing washed sand.

e) The collar present around the inner core was primarily used to prevent suspended organic matter from sedimenting out through the gap between the inner core and outer core and thus prevents material being introduced to a lower level than is normal. The collar also holds the inner core upright inside the outer core and ensures that filtration occurs vertically downwards through the core.

f) The sampling procedure is very quick and easy to carry out under difficult weather and bad conditions.

g) As it is possible to split the sand core into two halves on removal from the bed, no compaction is involved in removal of sand from the core; it has been shown that the sample removed from the sand core was reasonably representative of the depth from which it was in position in the bed.

h) In fulfilling the requirements of the Thames Water Authority (regarding sand collapse and the maintenance of a constant depth over the whole area of the filter) it was possible to use this apparatus on a full scale operational filter. The alternative if these requirements were not met would be the use of a small scale non-operational filter, the parameters of which were not necessarily identical with those of an operational filter.

3.7 Sand grain analysis

A sand grain analysis was carried out using a) sand taken from storage bays and used to fill inner cores, b) sand samples taken from bed 45 and compared with sand grain data, supplied by the Thames Water Authority, taken from bed 45 in 1972.

It should be emphasised here that sand collected from the storage bays and used to fill inner cores may well be sorted into size fractions because of the nature of the washing procedure (chapter 2). At all times sand was collected from areas which appeared to be uniform, although it would not have been feasible to mix the sand in any way to produce a 'normal composition', as mechanical handling of this nature would disturb and probably harm the organisms which had managed to survive the washing and storing processes. When this sand was used to resand a bed it was not mixed in any way, but placed straight back into the bed using dumper trucks, so there may well have been pockets in the bed where the sand was not uniformly distributed.

A second factor which may affect sand sorting is the backcharging process which occurs regularly in an operational bed prior to the start of a run and also occurs in the preparation of inner cores before being placed in a bed.

It was necessary to find out -

a) If the process of filling the inner cores caused the sand composition of the inner cores to be different from that in the sand bed.

b) Whether backcharging processes occurring in the sand bed and in the inner cores caused sand to become sorted - a factor which could cause selective distribution of meiofauna in the bed.

3.7.1 Method

Sand cores

Five cores were filled with sand in the manner described above and placed inside outer cores in positions randomly chosen in the bed. They were left for the duration of a slow sand filter run. At the end of a run they were removed, sectioned and 1cm deep ($40cm^3$) sections taken at 1, 5, 10, 15, 20 and 25cm depths in the core. They were dried at $60^{\circ}C$ for 96 hours to constant weight then placed in an Endecott test sieve shaker - sieve sizes (British Standard Sieves BS410) 4000, 2000, 850, 500, 250, 63 μm diameter - and shaken for ten minutes. Each fraction was removed and weighed.

Sand bed

At the end of the run samples were taken of the sand in the bed outside the area sampled by the grid, so as to avoid sampling sand of different ages. This was done by digging a 50cm deep hole, half of an inner core was then pushed horizontally into the side of the bed and the

sand contained within it removed. This was done four more times to produce five half cores which were then sectioned and treated as above.

Thames Water Authority data

Bed 45 was resanded in 1974 just prior to the start of this study and so data supplied by the Thames Water Authority of a sand grain analysis of bed 45 in 1972 may be considered to be a separate analysis of sand from Hampton Treatment Works. The analysis procedure carried out by the Thames Water Authority was similar to that described above, but no depth examination was carried out. Sieve aperture diameters were slightly different from those used in this study and were 4760, 3353, 2411, 1676, 1204, 853, 599, 422, 295 and 211 μm.

Results

Numerous methods are available in the literature for describing grain size distributions of sediments, but no one system has been adopted which allows direct comparison between findings of different authors.

A simple histogram (figure 3.7) describing size frequency distributions of the various size fractions shows a positively skewed distribution with the modal size fractions being of smaller diameter than the median fraction. This is the case in the majority of sand sediments and can be seen here in data from bed 45. Size fractions vary from $<63\mu m$ to $>4000\mu m$ with the largest size fraction being the $250 - 500\mu m$ diameter, the distribution is very skewed.

By logging the size fraction data it is possible to produce a more symmetrical curve for data of this type. In many cases these curves approach that of a normal distribution curve. Some authors use the ϕ scale ($- \log_2$) of Krumbein (1936) for this purpose.

When cumulative percentage values are plotted on semilog paper a curve is produced (figures 3.8, 3.9, 3.10) from which it is possible to extract information about the characteristics of the curve. It is possible to produce a straight line from a normal frequency distribution by plotting the cumulative percentage values on a probability scale. However, in this case the frequency distribution was overskewed and so even the use of probability - logarithmic scales would not produce a straight line. So for this analysis cumulative percentage data were plotted on a log scale and information for use in analysis was extracted from these curves.

The use of these curves alone can be of some use in comparing

Figure 3.7 PERCENTAGE COMPOSITION BY WEIGHT OF SAND IN ALL CORES

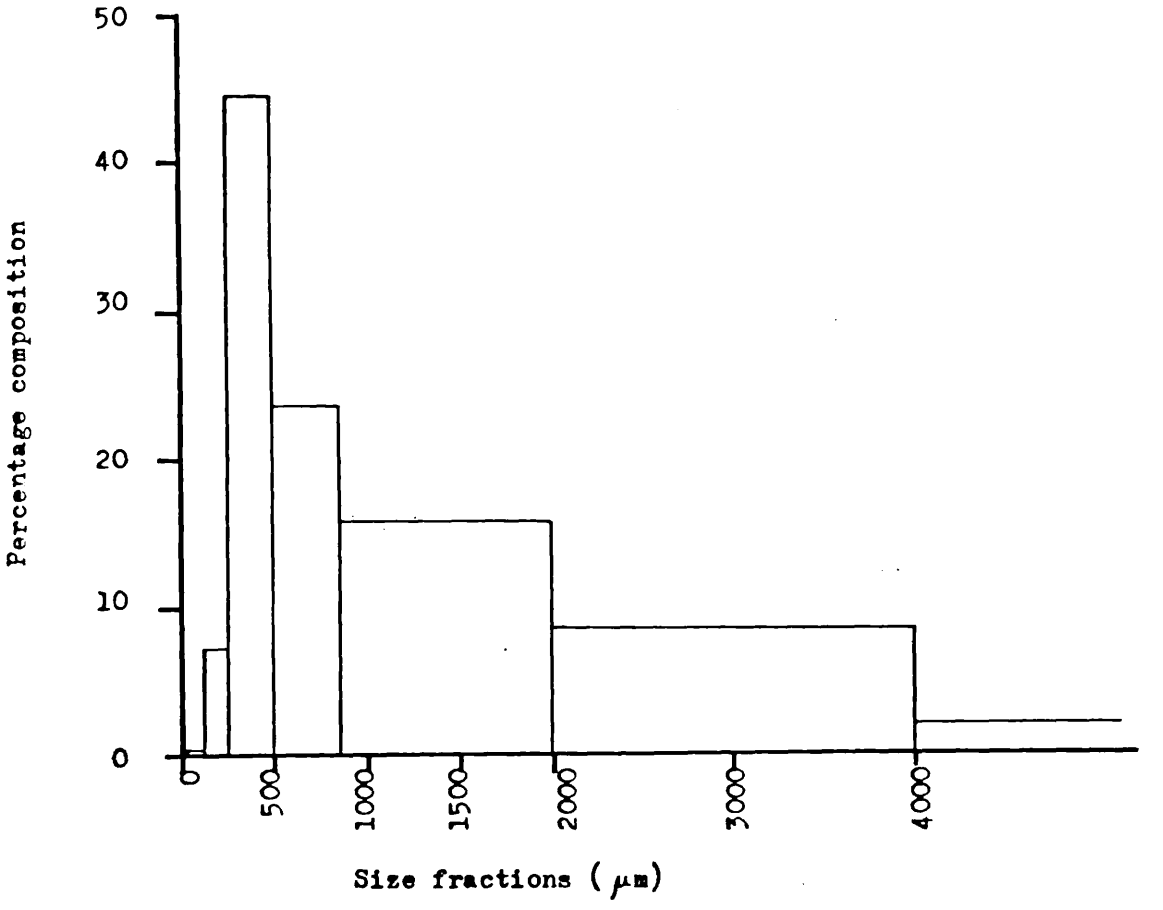


Figure 3.8 SIEVE ANALYSIS: SAND FROM SAMPLING CORES. FIVE SEPARATE ANALYSES

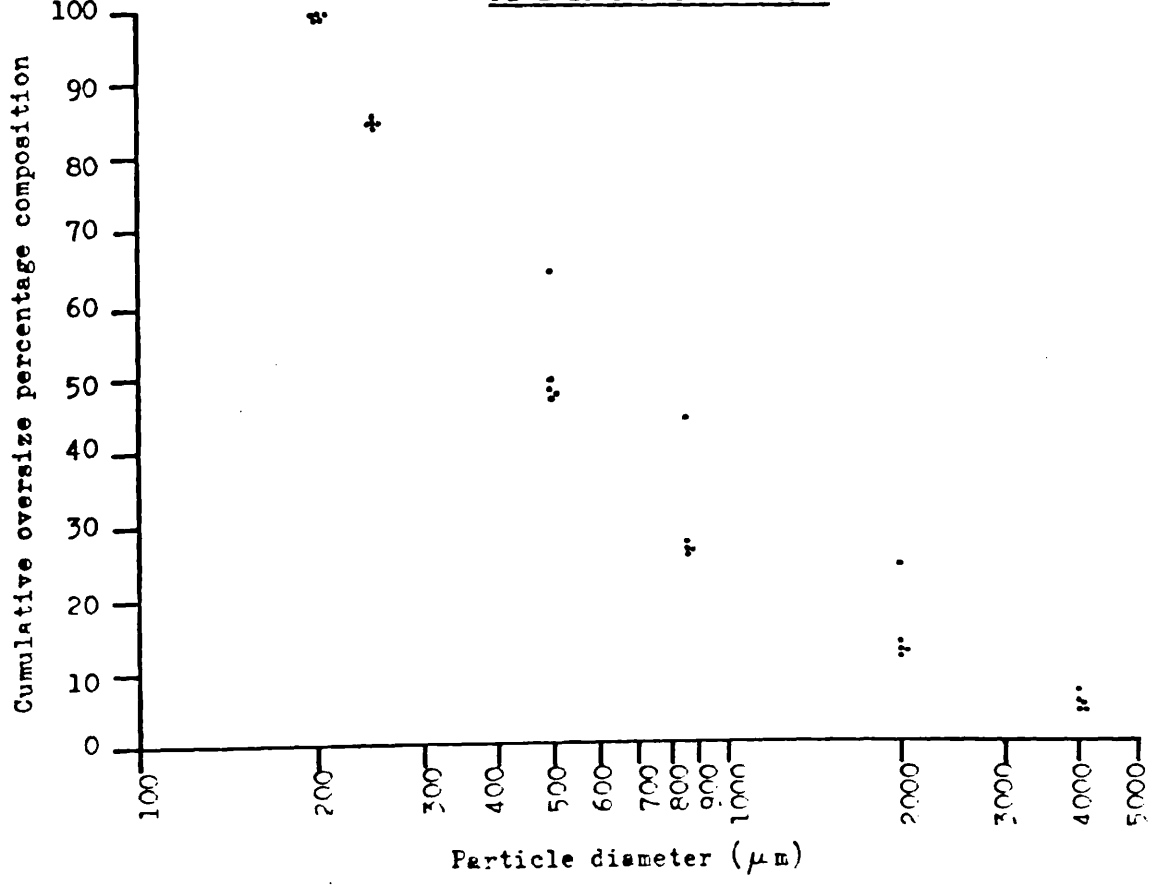


Figure 3.9 SIEVE ANALYSIS: SAND FROM BED 45. FIVE SEPARATE ANALYSES

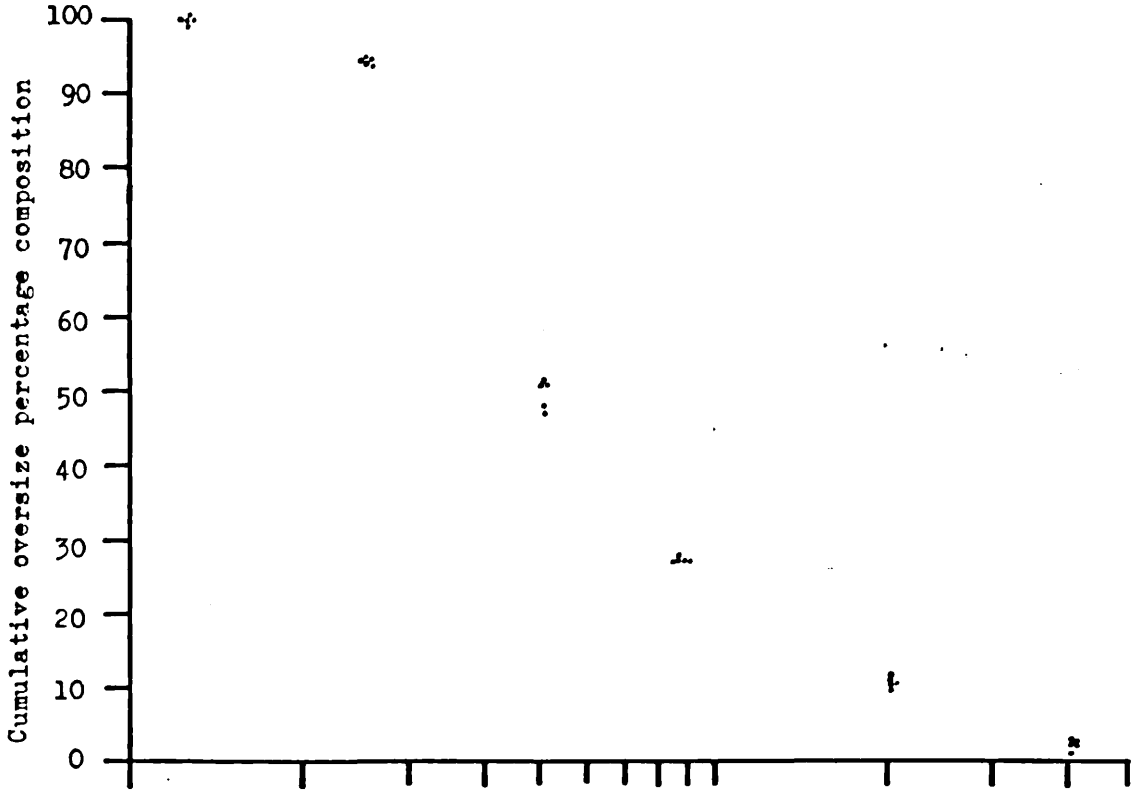
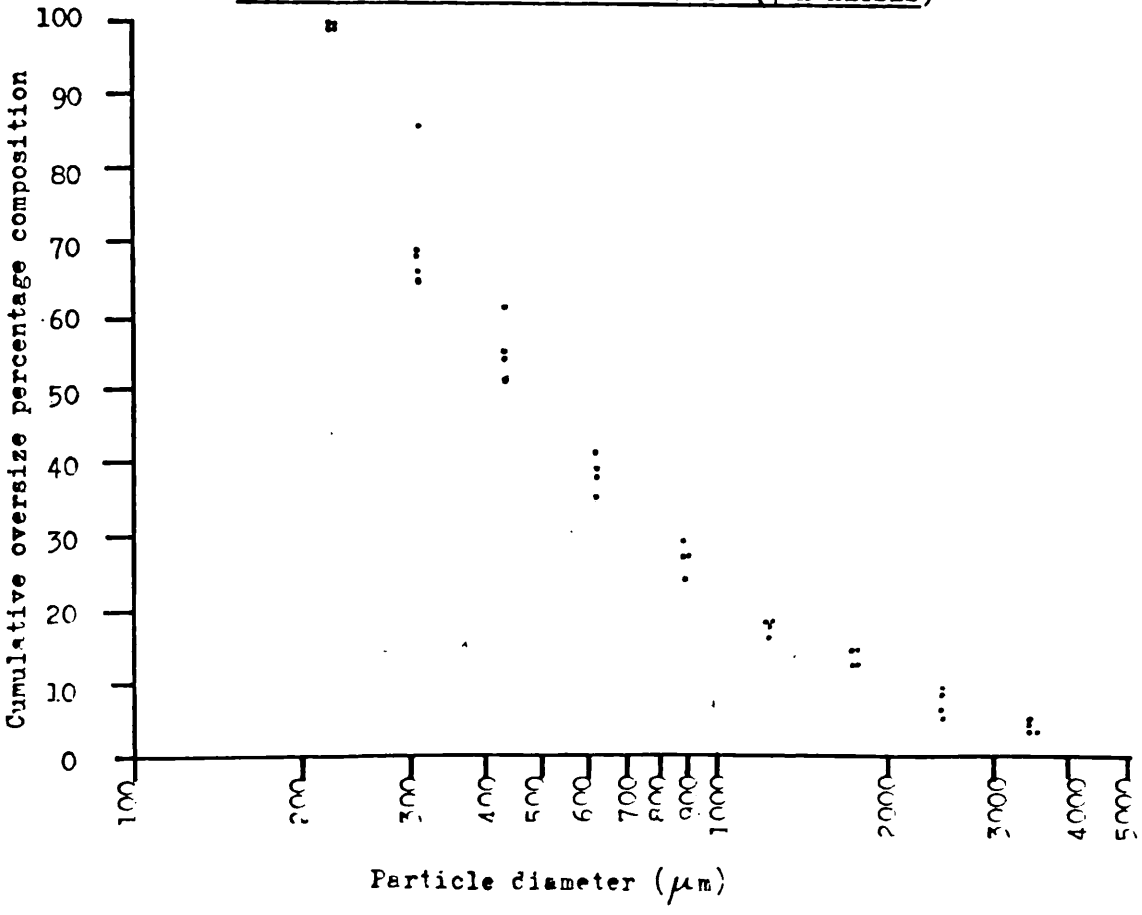


Figure 3.10 SIEVE ANALYSIS: SAND FROM BED 45 (1972) FROM THAMES WATER AUTHORITY DATA (4 ANALYSES)



sediments which have very different size characteristics. However, when the size characteristics are very similar (figures 3.8, 3.9, 3.10) the use of the curves alone does not allow critical comparisons to be made. To solve the problem several authors as reviewed by Morgans (1956), Inman (1952) and Folk (1968) have tried to reduce the characteristics of a particular sediment to a single figure. A comparison of the homogeneity of the various sand samples can be carried out by using an index of sorting. Several commonly used indices of sorting, e.g. coefficient of uniformity, U (Hazen 1892), Trask's sorting coefficient, S_0 (Trask 1932), Phi quartile deviation, QDϕ (Morgans 1956), can be criticised as they only take into account 50% of the sediment distribution and so significant differences shown in the other 50% are not manifest in these figures.

The Graphic Standard Deviation G is a good measure of sorting :

$$G = \frac{\phi 84 - \phi 16}{2}$$

where ϕX indicates the value of ϕ at the X cumulative percentage point - taken from a graph. However, this only takes into account $\frac{2}{3}$rds of the curve. A better measure is the cumulative standard deviation, σI

$$\sigma I = \frac{\phi 84 - \phi 16}{4} + \frac{\phi 95 - \phi 5}{6{\cdot}6}$$

This formula includes 90% of the distribution and is the best overall measure of sorting (Folk 1968). It is the average of (a) the standard deviation computed from $\phi 16$ and $\phi 84$ (this interval includes 2σ and so the standard deviation = $\frac{\phi 84 - \phi 16}{2}$) and (b) the standard deviation computed from $\phi 5$ and $\phi 95$ (this interval includes $3{\cdot}3\sigma$ and so the standard deviation = $\frac{\phi 95 - \phi 5}{3{\cdot}3}$. The two are then simply averaged together.

A two-way analysis of variance was carried out on σI values (table 3.2) obtained for each of the depths in the five cores in bed 45 and was also carried out on similar σI data taken from the sampling cores. This was not possible with the Thames Water Authority data as depth data were not available.

Two-way analysis of variance (table 3.3) on σI values in the sand bed samples indicated that there was no significant difference ($p > {\cdot}1$) between values of σI measured at the various depths in the sand, and also indicated there was no significant difference ($p > {\cdot}05$) between values of σI obtained for the five different cores.

Two-way analysis of variance on values of σI in the inner cores indicated that there was no significant difference ($p > {\cdot}25$) between

Table 3.2 Summary of 'Cumulative standard deviation' (σI), determined on sand samples removed from 1. Bed 45, and 2. Sampling cores placed in Bed 45.

Site	Bed 45 sand					Sampling cores placed in bed 45				
Replicates	A	B	C	D	E	F	G	H	I	J
Depth 1 cm	1.161	1.153	1.142	1.067	1.096	1.261	1.255	1.244	1.107	1.399
5	1.178	1.215	1.112	1.105	1.144	1.205	1.175	1.260	1.281	1.394
10	1.154	1.161	1.099	1.126	1.148	1.272	1.439	1.208	1.231	1.491
15	1.147	1.190	1.081	1.266	1.096	1.199	1.387	1.180	1.199	1.349
20	1.136	1.131	1.116	1.138	1.144	1.248	1.210	1.209	1.292	1.353
25	1.178	1.277	1.172	1.172	1.167	1.258	1.222	1.049	1.249	1.419

Table 3.3 Two-way analysis of variance of data in table 3.2

Source of variation		d.f.	F	p
Bed 45 sand	Between depths	5,29	1.824	0.25
	Between replicates	4,29	2.452	0.1
Sampling cores in bed 45	Between depths	5,29	0.926	0.5
	Between replicates	4,29	7.651	0.001

values of σI measured at the various depths in the core, but that there was a significant difference ($p < \cdot001$) between values of σI obtained for the five different cores.

This result demonstrates that the backcharging processes employed in the operational bed and in the inner cores does not cause any sorting of the sand. However there would appear to be some heterogeneity in the composition of the sand placed in the inner cores. This may well be due to the washing procedures described above and needs to be further examined by including data from another sand bed, i.e. bed 45 in 1972. As the sand from both the sand bed and the inner cores shows no vertical sorting the different depth fractions can be grouped in order to produce one σI value for each core, which can then be directly compared with the Thames Water Authority data (table 3.4).

The results of the one-way analysis of variance (table 3.5) comparing σI in the sand bed, inner cores and Thames Water Authority data indicate that there is no significant difference ($p > \cdot1$) between the values of σI in the three localities.

The results of all these analyses have demonstrated :

a) The backcharging procedure to which the sand is subjected does not cause the sand to become significantly sorted - a factor which will be important in a consideration of meiofauna distributions.

b) Similarly, the procedures used to fill the inner cores with sand do not cause this sand to become significantly sorted in relation to depth.

c) Indicates that the composition of sand used to fill the sampling cores did show some heterogeneity from core to core, probably due to the nature of the washing procedure, although as a whole it was not significantly different in composition from that already present in the sand bed or in other parts of the works (bed 45, 1972, Thames Water Authority data).

Table 3.4 Summary of 'Cumulative standard deviation' (σI), determined on sand samples removed from 1, Bed 45 sand, 2. Sampling cores placed in Bed 45, 3. T.W.A. data available.

Site	Bed 45 sand	Sampling cores	T.W.A. data
Replicates	1.131	1.239	0.992
	1.150	1.267	1.297
	1.171	1.233	1.292
	1.141	1.209	1.104
	1.122	1.404	

Table 3.5 One-way analysis of variance of data in table 3.4

Source of variation	d.f.	F	p
Between sites	2,13	2.627	0.25

CHAPTER 4 - FIELD WORK - METHODS OF ANALYSIS

4.1 Extraction of organisms from sand

4.1.1 Extraction procedure

The implications of quantitative aspects of studies concerning distribution, abundance and other dynamic processes occurring in ecosystems are very dependent on the efficiency of the extraction and counting procedures practised.

The use of different extraction techniques has been reviewed by Uhlig et al (1973) and Hulings and Gray (1971). It is clear that the choice of the type of extraction procedure which is most efficient is dependent on the size characteristics of the sand and the nature of the organisms involved. The amount of time involved in extraction procedures is important, especially when a lot of samples need to be processed, as in this study where 45 sets of sand samples were processed following each sampling trip.

The most efficient method found by Uhlig et al (1973) as far as removal of organisms is concerned, was a simple method of agitating the sand and water mixture and decanting the suspended material; a similar method is used by Wieser et al (1974). Various elutrication methods were tested by Uhlig, but found to be less efficient and required the use of equipment, all of which needed to be cleaned between each extraction procedure in order to ensure no animals remained behind to alter subsequent counts. Floatation methods have a limited success for, although they do succeed in removing organisms from the sediment, heavier organisms tend to sink. Also, they are time consuming - taking 2 - 3 minutes for each floatation procedure which must be repeated several times before an acceptable percentage of organisms is removed. An important point, as discussed by Howmiller (1972), is that the use of floatation liquids may cause weight loss in some organisms due to osmotic processes occurring. A floatation method used by Koosman and Newburgh (1977) did not involve the use of chemicals, but was selective to some extent, selection being related to the weight of the organisms concerned.

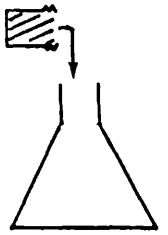
Extraction method (figure 4.1)

a) The sand sample - corresponding to a 1cm depth section ($40 cm^3$) was emptied from the screw top jar and placed in a 250ml conical flask.

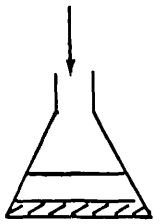
b) Approximately 50ml of distilled water was added to the flask.

Figure 4.1 EXTRACTION OF MEIOFAUNA FROM SAND

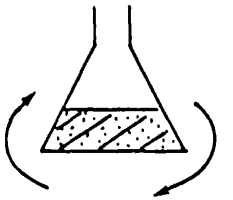
a. Sand sample placed into 250ml conical flask



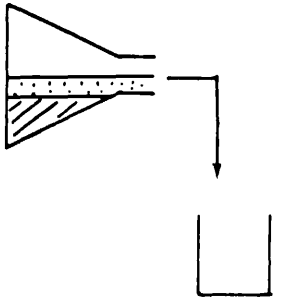
b. Approximately 50ml water added



c. Flask is rotated for approximately 15 seconds to bring organic matter into suspension



d. Meiofauna and detritus in suspension is poured off into beaker



Procedures b - d are repeated for a total of five times

c) The flask was then gently rotated for about 15 seconds.

d) The supernatant containing suspended meiofauna and detritus was quickly decanted into a beaker.

Procedure b) to d) are repeated a total of five times until the majority of organisms are removed.

The method is quick - taking no more than two minutes to remove the majority of the organisms. It required the use of no complicated equipment or chemicals which would require cleaning and did not damage the organisms concerned, however the more delicate protozoa would be damaged by this procedure.

4.1.2 Efficiency of extraction procedure

Method

Organisms were extracted from sand using the technique outlined above. Each washing from sand was then examined using the enumeration technique discussed below. The extraction procedure was then repeated on the same sand until no more organisms were observed. The sand fraction was then examined to see if any organisms remained.

This was then repeated on ten different sets of sand. Results can be seen in appendix 4.1 and figure 4.2.

It can be seen that after five washings the majority of organisms are removed ( ) 99%), so all sand samples were washed five times before the supernatant was counted.

4.2 Enumeration of organisms

4.2.1 Enumeration procedure

The problem here is one of attempting to separate meiofauna from the detritus, both of which have been removed from the sand sample. Floatation techniques are of no use here as the density of many organisms present is often the same as that of the detritus. Ruttner-Kolisko in Edmonson and Winberg (1971) suggests washing the sand several times, the water filtered through a plankton net and then the filtrate centrifuged to collect Protozoa and other small organisms passing through the net.

Experience has shown that filtering the water through sieves serves no useful purpose as several organisms pass through even the smallest sieve size and so the filtrate of all these sieves needs to be examined. Detritus particles vary very much in size and tended to get retained

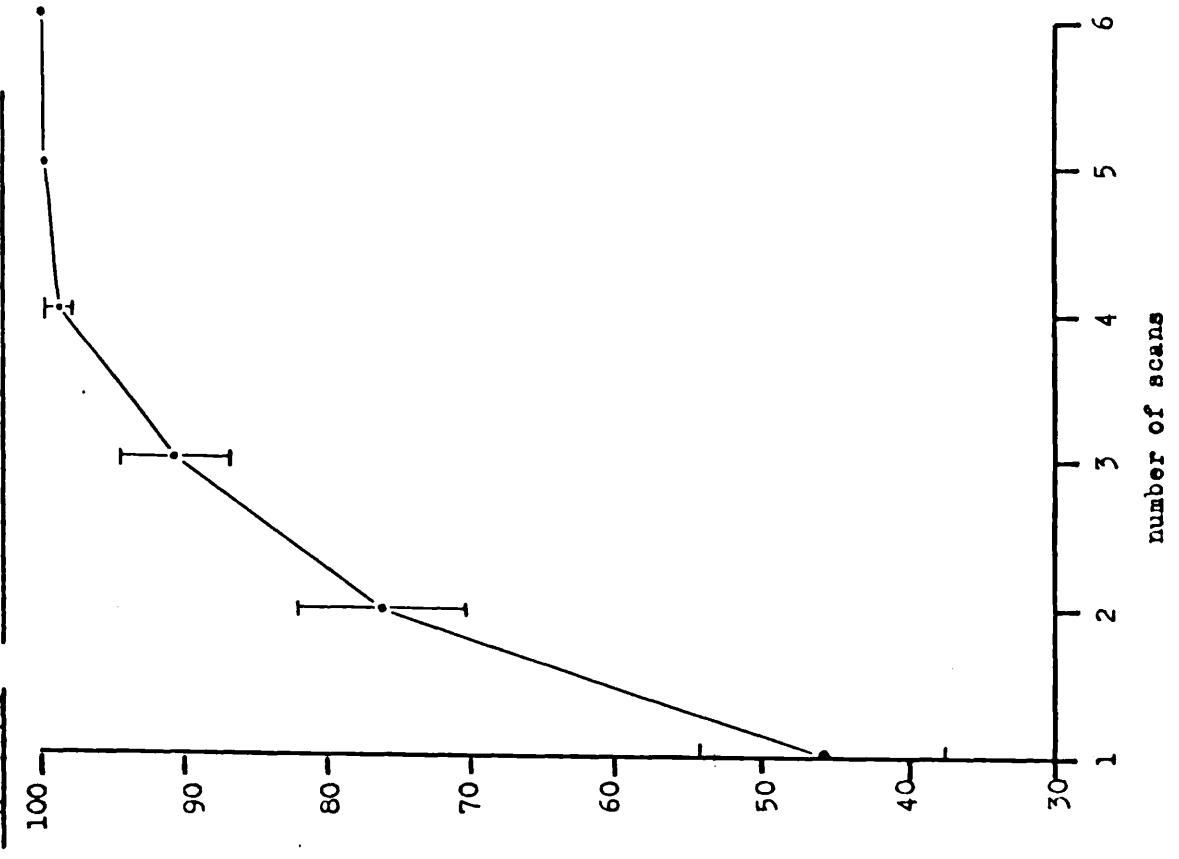


Figure 4.3 EFFICIENCY OF ENUMERATION PROCEDURE

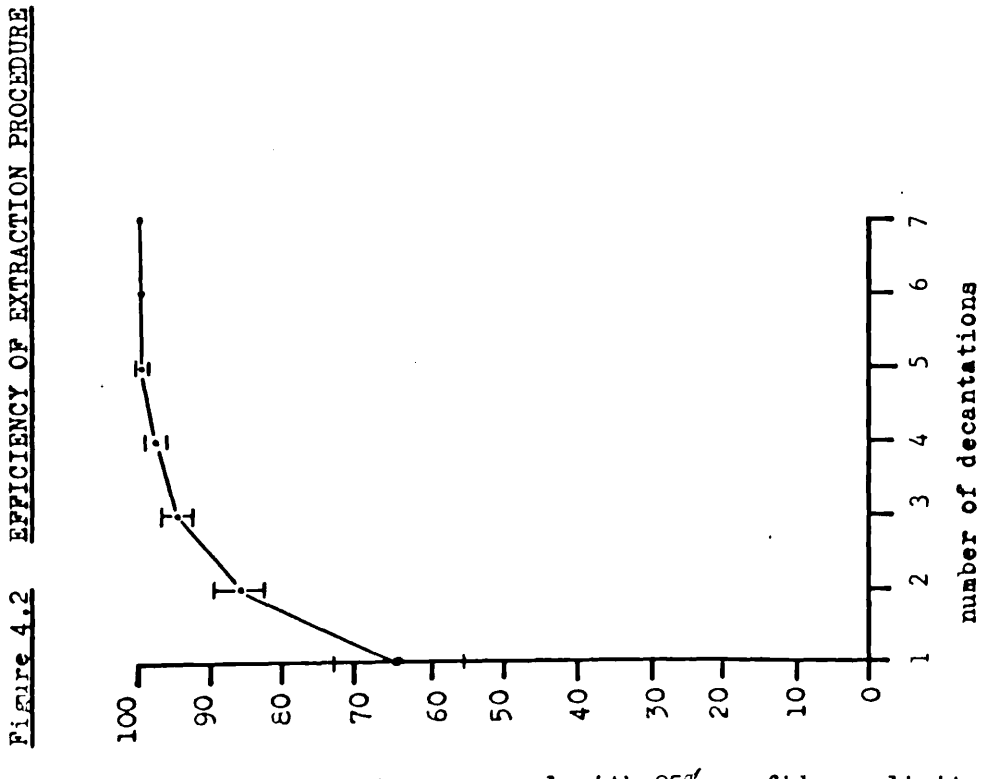


Figure 4.2 EFFICIENCY OF EXTRACTION PROCEDURE

with the fauna by the sieves anyway. Also, it was often found to be difficult and time-consuming to remove organisms which got trapped in the mesh of the sieve and often resulted in damage to the organisms involved.

An ultrasonic method (Thiel et al 1975) has been used to break up sediment aggregations. The aim of the treatment was to make the material more easy to sieve so that organisms could be more quickly counted in their sieve fraction sizes. The problem here is again one of sieving with organisms getting trapped in the sieve itself.

Enumeration method

The technique which was used was simply to pour a fraction of the suspended meiofauna and detritus from the beaker, in which it had been stored after extraction, into a 9cm petri dish. This was then examined under a binocular low-power microscope by moving the dish from side to side on the moveable microscope stage and gradually covering every part of the dish. As each organism was observed it was counted and removed to a watch glass. The procedure was then repeated on the same petri dish until no more organisms could be observed.

The suspension needed to be diluted with distilled water, according to the amount of detritus present, so that samples collected from the surface of a core needed considerable dilution, whereas 25cm depth samples often needed no dilution.

If the petri dish containing the organisms and detritus was allowed to stand for too long the detritus tended to clump together and so could cover an organism and prevent it from being viewed. To prevent this the sample was stirred after each viewing under the microscope.

The number of times that the same petri dish was examined under the microscope depended on the amount of organisms and detritus present. For ease of counting it was often necessary to dilute the original sample so that organisms could be more easily picked out with a pipette. The efficiency of the technique was tested.

4.2.2 Efficiency of enumeration procedure

Organisms were removed from the 9cm petri dish as they were counted using a Watson x 2·5 objective and x 7 eyepiece.

The petri dish was scanned and the total number of organisms removed were recorded. This was then repeated on the same petri dish

until all organisms were removed.

This whole procedure was then repeated using ten new samples of meiofauna and detritus. The results (appendix 4.2) are presented in the form of a graph (figure 4.3) where the cumulative percentage of organisms removed after each scan of the petrie dish are presented. The 95% confidence limits are included to represent the variability in percentage removal caused by the examination of samples with varying amounts of detritus and macrofauna.

Although 99% of organisms were removed after five scannings of a single petri dish, the number of scannings required obviously depends on the relative concentration of detritus and meiofauna present in the suspension and it was not always possible to keep this relatively constant. So in practice, although the minimum number of scannings was five, sometimes it was necessary to increase this if more detritus was present. The procedure became quite quick to perform and the length of time spent on each scan obviously decreased as fewer organisms remained to be counted.

4.3 Organic carbon determination

4.3.1 The use of organic carbon as a parameter in ecological studies

The material present in a slow sand filter which may be available to feed detritus and filter feeding organisms includes bacteria, algae and dead and decaying organic matter. To separate and measure the individual components of the detritus would be a very time-consuming business and in a situation where the food requirements of the meiofauna are not known, information of this kind might be too detailed.

A more useful parameter to measure would be the energetic value of the detritus. Consideration of this flow of energy through ecosystems is accepted by many authors as being important. Here it must be borne in mind that, due to the refractory nature of some of the organic material, not all of the measured energetic components will be utilisable by the meiofauna. Energy, however, is an inconvenient parameter to measure in the field, especially when many samples need to be analysed.

An alternative is the use of organic matter expressed in terms of organic carbon which is an easily measured constituent of all ecosystems. Again, it must be emphasised that the measurement of organic carbon present in a sample produces a 'blanket value' indicating the

total amount of material available to the organism present - just how much of it is capable of being utilised is another matter.

4.3.2 Determination of organic carbon

There are several methods available for the determination of carbon in organic matter. The one described below was found to be accurate, relatively quick to perform and did not require the use of expensive equipment.

There are two parts to the analysis, the first part involves the digestion of the carbon by a wet oxidation procedure using concentrated sulphuric acid and excess potassium dichromate. The second part involves the use of a potentiometric end point detection procedure in a titration to determine the amount of dichromate remaining at the end of the digestion step.

Reactions

The organic matter is heated with excess potassium dichromate in the presence of concentrated sulphuric acid. The dichromate remaining unreduced at the end of the reaction is determined by titrating with $(NH_4)_2SO_4FeSO_4$, ferrous ammonium sulphate (FAS) to an end point located by a simple potentiometric circuit.

Principle of wet oxidation procedure

The organic matter is oxidised to yield its original inorganic constituents. Thus the amount of carbon present can be calculated from the amount of oxygen used in the reaction :

$$C_xH_{2y}O_z + (x + \frac{y-z}{2})\ O_2 \longrightarrow xCO_2 + yH_2O \quad \text{(Golterman 1971)}$$

So it is necessary to know the composition of the organic matter and hence the values of x, y and z. In most cases this is not known, but many authors assume the equation :

$$C_6H_{12}O_6 + 6O_2 \longrightarrow 6CO_2 + 6H_2O$$

where 1mg O_2 $\equiv$ 0·3754mg C

In this case the oxidising agent used is potassium dichromate, $K_2Cr_2O_7$

where 1·0ml N/8 $K_2Cr_2O_7$ $\equiv$ 1mg O_2

and so 1·0ml N/8 $K_2Cr_2O_7$ $\equiv$ 0·3754mg C

Apparatus and reagents - appendix 4.3

Procedure - see also figure 4.4

1. Place $1cm^3$ sand sample in a glass vial.
2. Dry at $80^{o}C$ for 24 hours if storage is necessary.
3. Add 5ml (Vw) distilled water to sand sample.
4. Shake two hours.
5. Remove 1·0ml (Va) supernatant and pipette into reaction vessel.
6. Add 1·0ml 0·2N dichromate and 4·0ml concentrated sulphuric acid.
7. Digest in oven at $125^{o}C$ (i.e. reaction temperature of 6) for three hours.
8. Remove from oven and cool for three hours.
9. Add 20ml distilled water to reaction vessel to increase volume of liquid in order to cover Pt probe during titration.
10. Place reaction vessel on magnetic stirrer and add stirrer bar. A constant stirring speed is essential.
11. Lower Pt-calomel reference electrode into solution and titrate with 0·1N FAS until the end point is reached using a potentiometric circuit (see below). The approach of the end point is indicated by "flickering" of the needle. Add more FAS until a constant steady mV reading is obtained.
12. Run in more FAS until two more steady readings are recorded. Draw on graph.
13. Extrapolate back to zero line to obtain end point.
14. To obtain a blank reading repeat from 6 onwards, but using 1ml of distilled water in reaction vessel instead of sample.

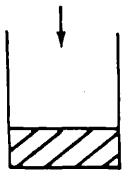
4.3.3 Discussion of the wet oxidation procedure

4.3.3.1 Preservation of the sample prior to analysis

Samples which cannot be analysed immediately can and should be preserved by drying. In this case all samples were dried as it was not possible to analyse the material immediately. Lovegrove (1966) showed that varying the drying temperature between 49 and $100^{o}C$ did not cause significant changes in lipid value, and lipid values after drying for 24 hours in the oven were comparable with those in a desiccator.

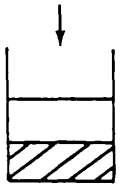
Figure 4.4 PROCEDURE OF WET OXIDATION OF ORGANIC CARBON

1. Place $1cm^3$ sand in vial

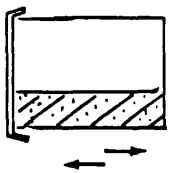


2. Dry at $80^{o}C$ for 24 hours

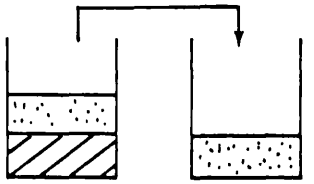
3. Add 5ml (Vw) distilled water



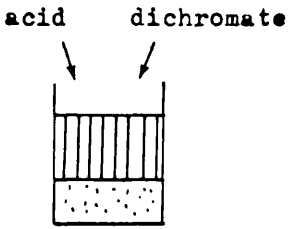
4. Shake for 2 hours



5. Remove 1 ml supernatant to reaction vessel (Va)



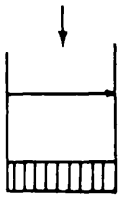
6. Add reagents to reaction vessel



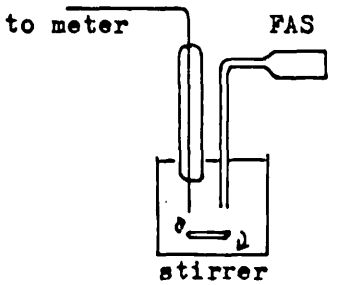
7. Digest in oven $125^{o}C$ for 3 hours

8. Cool 3 hours

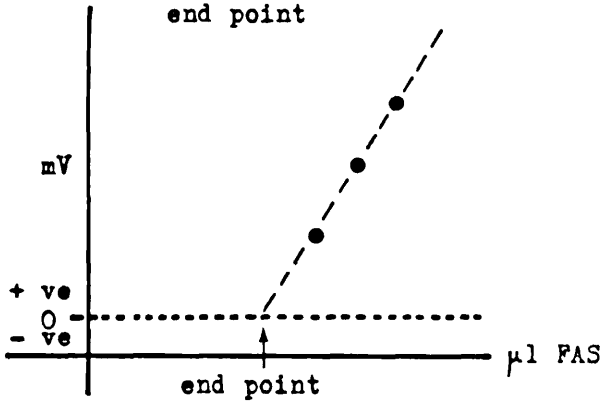
9. Add 20ml distilled water to increase volume



10. Titrate against ferrous ammonium sulphate



11, 12, 13. Extrapolate back to zero line to obtain end point



However, he did show that prolonged heating in an oven at 100°C affects the fat content to the extent of approximately 4% of the dry matter. Curl (1962) showed that drying plankton samples at temperatures higher than 80°C will volatalise some lipids and amines as well as denaturing many proteins. Temperatures of 80 - 90°C have been shown to be optimum for drying material (Maciolek 1962), above this temperature there is a decrease in carbon levels found present in the sample. All sand samples were dried at 80°C for 24 hours and then stored in the dark until analysis could proceed.

4.3.3.2 Resuspension of organic matter

Prior to digestion it was necessary to resuspend the organic matter in distilled water so that a sample could be removed for analysis. This was done by adding 5ml of water to the 1ml sand sample and then the sample was shaken for two hours. This time was found to be sufficient to resuspend the majority of the organic matter.

This shaking period had previously been investigated by adding 5ml distilled water to a $1cm^3$ sand sample in a vial. This was shaken for up to a period of four hours. At intervals of time 3 x 0·5ml aliquots of suspended material were removed, placed into three separate reaction vessels and digested as described above. After this removal of 3 x 0·5ml aliquots for digestion, 1·5ml distilled water was added to the vial to ensure that the total volume of water in the vial remained constant. The amount of carbon in suspension was determined for each length of shaking period. Samples were removed for digestion after 5,30 mins and 1, 2, 3, 4 hours. The results (appendix 4.4) are plotted; it can be seen (figure 4.5) that after two hours there is no appreciable increase in amount of carbon resuspended. This period of time was chosen for all subsequent treatments.

4.3.5.3 Choice of oxidising agent

The two main wet oxidising agents are potassium permanganate and potassium dichromate. The main disadvantage of permanganate as an oxidising agent is that it is subject to autooxidation and because of this instability it cannot be employed in the rigorous reaction conditions necessary for complete oxidation of organic matter. Potassium dichromate has been found to be a very effective oxidising agent for organic matter. Its main attributes are that it is extremely stable in neutral and acid solution and does not decompose appreciably during

Figure 4.5 AMOUNT OF ORGANIC CARBON REMOVED FROM SAND IN RELATION TO LENGTH OF SHAKING PERIOD

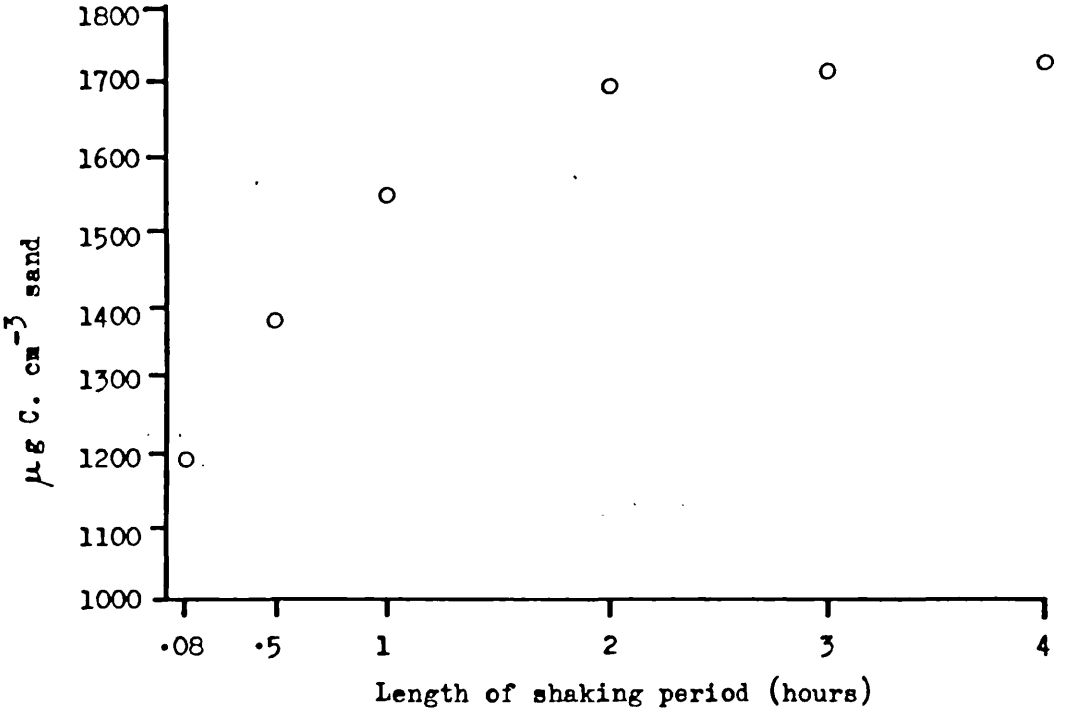
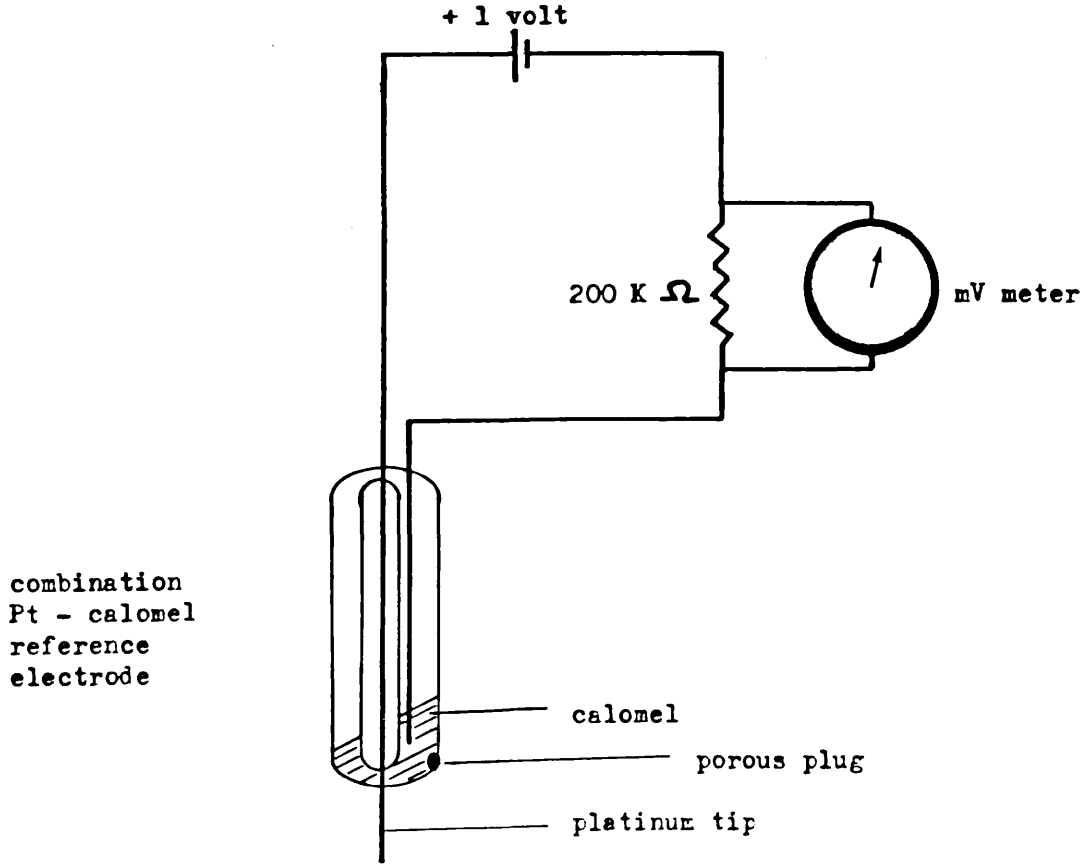


Figure 4.6 CIRCUIT DIAGRAM OF POTENTIOMETRIC END POINT ANALYSIS PROCEDURE USED IN ORGANIC CARBON ANALYSIS



a reaction even at high temperatures and acid concentrations.

4.3.3.4 Concentration of reactants

The concentration of dichromate used varies very much in the literature. The initial dichromate concentration gives a specified oxidation potential which decreases as the reaction proceeds according to the extent to which the dichromate becomes reduced. So, ideally, a constant dichromate concentration should be maintained throughout the reaction in order to expose all unoxidised organic matter to the same oxidation potential. A maximum normality of 2·0N - i.e. the solubility limit for dichromate in the reaction mixture and a minimum normality of 0·05N - i.e. minimum for useful oxidation, have been established (Maciolek 1962). So within these limits the choice of dichromate normality is often a compromise depending upon factors such as carbon content of the sample, nature of sample (wet or dry) and nature of titration following digestion. In general, not more than 50% of the dichromate should be reduced at low normalities. In this case a normality of 0·2N dichromate was found to be convenient.

A dose of 1ml 0·2N dichromate would be reduced by 60% by 1ml aliquot removed from a sample originally containing approximately 1800 $\mu gC.cm^{-3}$ sand and 20% by a sample containing approximately 600 $\mu gC.cm^{-3}$ sand, and so allows a fairly wide range of carbon values to be digested before the addition of extra reagent doses. At carbon concentrations greater than 1800 $\mu gC.cm^{-3}$ it was necessary to double the reagent doses in order to lower the percentage of carbon reduced in the reaction.

4.3.3.5 Acid to dichromate ratio, length and temperature of digestion period

The ratio of volume of acid to volume of dichromate solution used varies considerably in the literature. In general a higher acid volume is used as this produces a more efficient oxidation of material. The 2 : 1 volume ratio of concentrated sulphuric acid to aqueous dichromate was approved by Walkley (1947) and is advised by most modern procedure methods (Maciolek 1962, H.M.S.O. 1972, Golterman 1969).

The mixing of the two components in the 2 : 1 ratio produces a temperature of 115 - 125°C (Walkley 1935) but this does not produce a complete oxidation of the organic matter unless it is maintained by external heating. Temperatures cited in the literature vary from no

heat at all (Gaudette et al 1974, Walkley and Black 1934, Peech et al 1947) through a vague description of 'boiling' used by Konrad et al (1970), Allen et al (1975), Mebius (1960), H.M.S.O. (1972), Golterman (1969), to a temperature of 140°C used by Canelli et al (1976). It should be borne in mind, however, that spontaneous decomposition of dichromate increases with temperature causing a definite loss in precision above about 150°C (Maciolek 1962).

The length of digestion period is another factor to vary in the literature, varying from 30 minutes (Konrad et al 1970, Mebius 1960, Peech et al 1947) up to 3 - 6 hours (Golterman 1969) with an intermediate period of two hours used by Talling (pers. comm.), Stones (1974), Moore (1976) and H.M.S.O. 1972). An optimum period of three hours was determined by Maciolek (1962) using three different carbohydrate groups.

In this study the maximum reaction temperature of 125°C was maintained for a period of three hours.

4.3.3.6 Catalysis

Evidence for the effectiveness of a silver catalyst would not yet appear to be conclusive. The effectiveness of the silver catalyst is due to the fact that it renders refractory molecules, such as acetic acid, completely degradable. Gaudette et al (1974) compared results of a wet oxidation procedure containing no silver catalyst with results from a combustion carbon analyser using organic matter samples from the same source. He found almost perfect correlation between the two methods and interpreted this as indicating that the combustion carbon analyser could not account for the refractory material present. Increases in oxidation of 15% (Stones 1974) and 10% (Moore 1976) have been shown to be due to the addition of a silver catalyst in the digestion procedure. The use of a silver catalyst is also suggested by Canelli et al (1976) and Golterman (1969 and 1975). A discussion of the use of silver as a catalyst by Maciolek (1962) concluded that the catalytic effect of silver becomes less evident as the oxidising conditions were made more efficient.

In this study no silver catalyst was used, but with hindsight it may have been useful to include in the method. However, attempts have been made to optimise the use of all other components in the procedure. It has also been shown that different organic compounds are oxidised to varying degrees in the digestion procedure (Fleet et al 1972, Maciolek 1962, Canelli et al 1976, Stones 1974) and so the results of this oxidation procedure will depend ultimately on the type of organic compounds

present. The use of a silver catalyst may improve the percentage oxidation of the more refractory compounds, but it would appear that in no case is 100% oxidation achieved. On the other hand, not all organic compounds are immediately available, in one form or another, for biological metabolism and so the results of these determinations must be viewed accordingly.

Carbon values, of material analysed by the Thames Water Authority and quoted in this study, were arrived at with the use of a silver catalyst (A. Steel, pers. comm.). It is possible, therefore, that there may be slight discrepancies in the results of carbon analyses of material processed by the two methods.

4.3.3.7 Interferences

The main interference is from chloride, if concentrations greater than 100mg.L^{-1} chloride ion are present then precautions must be taken, In the River Thames at Walton, the chloride ion concentration is less than 40mg.L^{-1} (Metropolitan Water Board 1974) and so no precautions were necessary.

4.3.3.8 Titrating solution - ferrous ammonium sulphate (F.A.S.)

Ferrous solutions are rather unstable under some conditions. They should be acidified (approximately 2% by volume) to increase stability and storage life. Sunlight and high temperatures also affect stability which can be improved if the ferrous solution is stored in a stoppered brown glass bottle in a cool place. All titrations were carried out in a room with a constant temperature of 15°C. FAS should be standardised regularly before use.

4.3.3.9 Precision, limits of detection and accuracy of method

The minimum detectable carbon value was determined by digesting glucose solutions with known carbon concentrations. Glucose solutions containing approximately 1, 5, 10, 20, 50, 100, 200 μgC.ml^{-1} were prepared. 1ml of each concentration was placed in a reaction vessel and 1ml 0·2N potassium dichlorate and 4ml concentrated sulphuric acid were added and the analysis procedure completed as described above. Five replicates of each concentration were prepared (table 4.1).

The precision of the method was good at concentrations greater than 20 μgC.ml^{-1}, where the precision (standard deviation as a percentage of the mean) was shown to be ± 3%, which compares with Allen et al

Table 4.1 Precision of organic carbon analysis method.

Theoretical carbon conc. $\mu g.ml^{-1}$	1.06	5.32	10.64	21.28	53.21	106.40	212.8
Carbon conc. determined by analysis. $\mu g.ml^{-1}$	1.90	6.36	8.66	20.87	52.97	105.26	205.63
	0.15	4.22	9.64	20.44	52.94	104.28	202.47
	0.89	6.61	9.85	19.37	53.43	108.24	211.75
	0.34	6.15	8.97	20.59	52.45	109.34	211.17
	1.47	5.05	8.63	20.68	51.96	106.21	210.25
n	5	5	5	5	5	5	5
$\bar{x}$	0.95	5.68	9.15	20.49	52.75	106.67	208.25
S as % of $\bar{x}$ i.e. precision	$\pm$77.9	$\pm$17.8	$\pm$6.2	$\pm$3.1	$\pm$1.1	$\pm$2.0	$\pm$1.94
% recovery of theoretical carbon conc.	90%	105%	86%	96%	99%	100%	98%

(± 2%, 1975) and Canelli (± 4%, 1976). Below concentration of $20\,\mu g.C.ml^{-1}$ precision decreases, with values of ± 78% at $1\,\mu gC.ml^{-1}$ which is not acceptable. At $5\,\mu gC.ml^{-1}$ precision is ± 18% and indicates that $5\,\mu gC.ml^{-1}$ is the minimum determinable value.

The maximum theoretical carbon value which can be determined is $600\,\mu gC.ml^{-1}$ but it is recommended that samples containing more than $400\,\mu gC.ml^{-1}$ should be treated with at least twice the reagent doses so that the amount of dichromate reduced does not much exceed 60%.

Accuracy (defined as the percentage recovery of known standards) in situations where precision was good, showed recoveries of greater than 96% of the amount of carbon supplied, compared with 97% and 104% (Fleet et al, 1972 - two methods), 98% (Stones 1974) and 100% (Canelli et al 1976).

4.3.3.10 Time involved

Using this method of analysis it was possible to process 90 samples in a total of 18 hours.

4.3.4 Potentiometric end point determinations

4.3.4.1 Method used

All potential differences between reference electrodes and indicator electrodes can be used to follow a titration to its end point. In this case use is made of a combined calomel reference and platinum indicator electrode with a high resistance (e.g. 200K Ω) across it.

Quite a few of the redox couples familiar in titrimetric analysis e.g. Fe^{3+}/Fe^{2+}, are slow in establishing a steady potential at a Pt indicator electrode when the measurement is made in the ordinary manner with a balanced (zero voltage) potentiometric circuit. To eliminate long waiting periods the recommended procedure is to make use of a polarised indicator electrode at which electrolysis is forced to occur at a small rate (Lingane 1958). In this case the Pt electrode is polarised anodically keeping the Pt electrode at a potential of +1 volt (see figure 4.6).

A second advantage of polarising the Pt electrode is that there is no increase in potential caused by electrode reactions with Cr^{6+}, Cr^{3+} and Fe^{3+} ions, which are present in the solution (Kolthoff and May 1946).

So when an acid solution of potassium dichromate is titrated with ferrous ammonium sulphate there is no change in potential of the Pt electrode until all the dichromate has been reduced and the solution contains

an excess of Fe^{2+} ions. At the end point surplus Fe^{2+} ions caused the measured potential to increase from a previously low constant level. Here it is possible to zero the small steady potential recorded on the meter before the end point is reached, by the use of the variable back-off potential control, i.e. "buffer control" of a pH meter.

The rate of change of the potential after the end point is reached is directly proportional to the volume of ferrous solution added. So by measuring potential and obtaining two or more values in the presence of excess ferrous ions the end point is reached by extrapolation to the zero potential. The amount of ferrous solution added at that point is equal to that at the end point (figure 4.4). In practice the amount of FAS added at the end point was determined by using a regression analysis calculator programme which determined the best straight line relationship between the points using the method of least squares. However, this linear relationship holds true only up to recorded mV potentials of + 1·5mV (figure 4.7). So it was necessary to obtain three mV readings before this value was reached and in practice readings above 1·3mV were not accepted.

4.3.4.2 Electrode problems

a) Loss in sensitivity was corrected by electrolysing the electrode. The platinum electrode is connected to the negative pole of a D.C. source. An inert auxiliary electrode (iron nail) is connected to the positive pole and electrolysis allowed to proceed for three minutes in 0·1N sulphuric acid with a current of 10mA. It was found necessary to carry out this procedure monthly.

b) Drift was caused by impurities on the platinum electrode and was corrected by gently cleaning the electrode with scouring powder.

4.3.4.3 Calculation

$$C\ \mu g.cm^{-3} = (Vb - Vs).\frac{Nf.Vw.}{Va}\ 3{\cdot}0029$$

where Vb = vol of FAS used to reach end point (BLANK) µl.

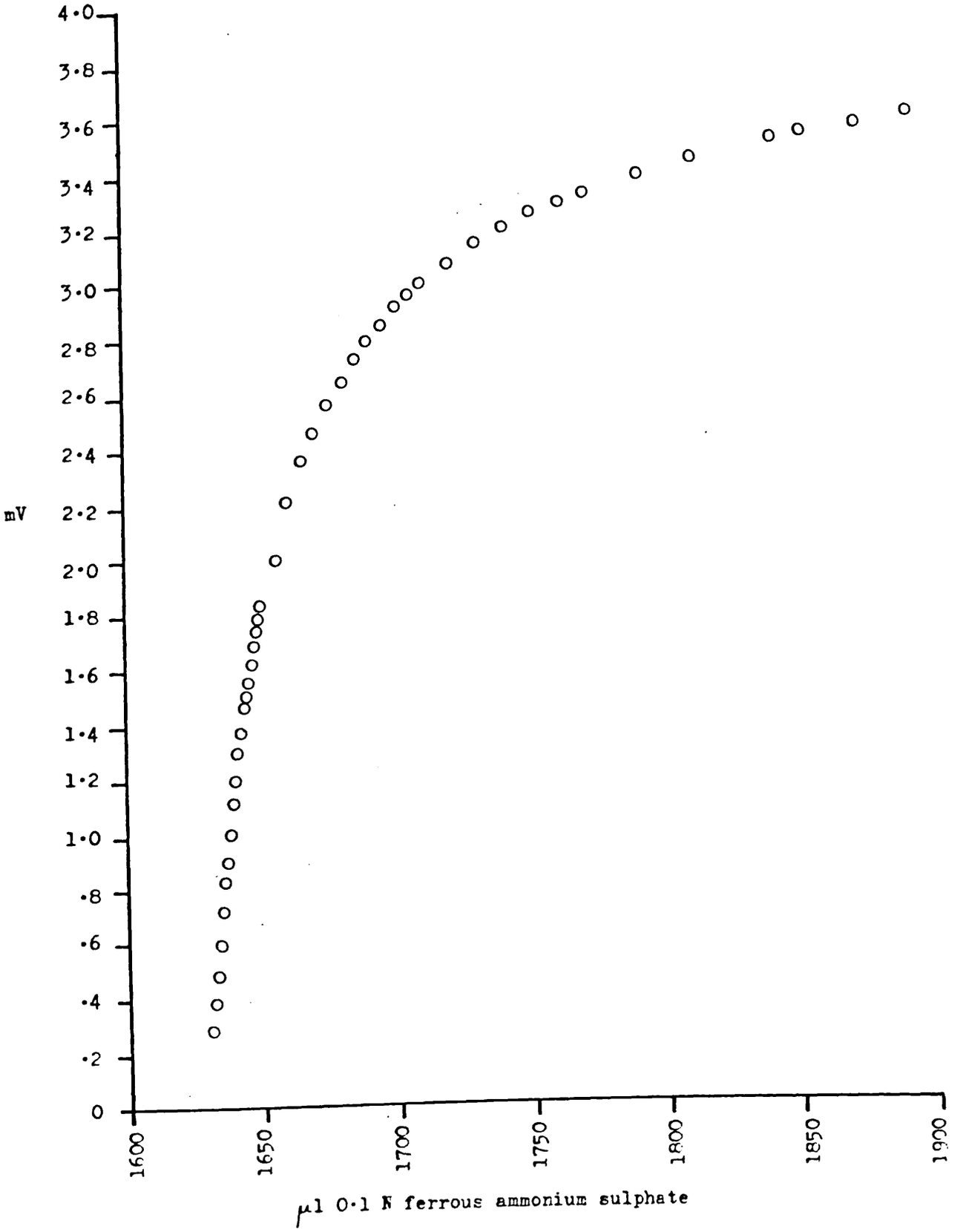
Vs = vol of FAS used to reach end point (SAMPLE) µl.

Nf = normality of FAS.

Vw = vol of water in shaking bottle ml.

Va = vol of aliquot of suspended organic matter removed for reaction ml.

Figure 4.7 RELATIONSHIP OF POTENTIAL (mV) RECORDED, PAST THE END POINT, AND VOLUME OF FERROUS AMMONIUM SULPHATE ADDED IN TITRATION OF FAS AGAINST POTASSIUM DICHROMATE SOLUTION



The fact that organic matter consists of many organic compounds, all of which are oxidised to varying degrees in the wet-oxidation procedure, has been discussed above. In measuring the carbon content of such organic matter it is only possible to give accurate estimations of carbon content when the various components have been identified. These investigations are not really feasible in a study of this kind and so the calculation used above was based on the reduction of glucose where $1mgO_2 = 0{\cdot}3754mgC$, this principle is used by Talling (pers. comm.).

4.4 Statistical analyses

4.4.1 Means

Means quoted in the text are arithmetic unless otherwise stated. The 95% confidence limits for arithmetic means quoted were calculated as follows :

95% confidence limits of mean = mean ± t.SE

where SE = standard error of the mean

t = value of student's 't' when p = 0·05 at the appropriate degrees of freedom

When the data did not follow a normal distribution it was necessary to transform these data in order that a mean might be obtained. When this was necessary the transformation used was quoted in the text. The result of these transformations is that the derived means are slightly smaller than the arithmetic means.

4.4.2 Linear regressions

Linear regression by the method of least squares was used to define relationships between two variables. The general form of the regression is :

$$y = bx + a$$

where y is the dependent variable

x is the independent variable

b is the regression coefficient

a is the intercept on the y axis

and details of the calculations are given in Sokal and Rohlf (1969)

4.4.3 Covariance analysis

Comparisons of several regression lines were possible with the use of covariance analysis. Firstly it was necessary to make a comparison of the residual variances. This was done with the use of an F test, or in the case of more than two samples, by Bartlett's test, a type of analysis of variance. If these residual variances were found to be homogenous then it was possible to compare the slopes or regression coefficients and also the elevations of the lines by the use of F tests. Details of all these tests are given in Snedecor and Cochran (1967). If there was no significant difference between slopes and elevations of the regression lines then it was considered reasonable to combine the data, if this was required.

4.4.4 Numerical integration

In order to obtain values of numerical abundance of organisms from cores where alternate depths were examined, numerical integration was used. In this case a Hewlett Packard Program Tape was used which accepted y (numerical abundance) and x (depth) data and calculated the formula of the smoothest curve through the series of given points and then calculated the area beneath the curve by integration of the formula. Details are given in Ralston and Wilf (1967).

4.4.5 Analysis of variance

Analysis of variance tests were used to compare the variation in a set of data which had been partitioned into components associated with the possible sources of variation. Both one-way and two-way analyses of variance wereused. The former was employed when comparisons were made between a number of independent random samples, one sample from each population. The counts were classified in one direction and the number of counts in each section could be different. The latter method was employed when the counts of related samples were matched in groups, so that comparisons were made between groups and also between samples. Details of the calculations are given in Sokal and Rohlf (1969).

CHAPTER 5 SPECIES COMPOSITION OF SLOW SAND FILTERS AND SIZE CHARACTERISTICS OF ENCHYTRAEUS BUCHHOLZI

5.1 Meiofaunal organisms present in slow sand filters

The meiofauna found in bed 45 are listed in table 5.1., together with a comparison of those found by Lloyd (1974) in slow sand filters at Ashford Common treatment works (Thames Water Authority), Husmann (1958) - slow sand filters in Bremen, and Rittersbusch (1974) - slow sand filters used for ground water recharge.

The types of organisms found in slow sand filters are characteristic of those found in other interstitial habitats. In general they are small, either in length or cross section and fairly mobile. In bed 45 the largest organism encountered was Enchytraeus buchholzi which reached maximum lengths of 4 - 5mm. Some stages of the chironomid larvae would reach similar lengths, but in general these did not penetrate below the top few cms of the filter. The nematodes and naidids were in general less than 3mm in length and individuals of Aelosoma hemprichi were less than 1mm long. The microturbellaria and harpacticoids were similarly small sized at less than 1mm. The occasional cyclopoid copepod and ostracod would also be found in the surface samples, though these were not true inhabitants of the interstitial system, but were possibly feeding on material deposited on the filter surface.

Table 5.1 shows similar species compositions found by Lloyd (1974) although there are slight discrepancies which could well be due to the fact that the sand in the two treatment works is of a slightly different quality, Hampton sand being of slightly less exact specifications than sand at Ashford. Husmann (1958) and Rittersbusch (1974) list similar taxonomic groups, although there are slight differences in the exact species.

As far as the oligochaetes are concerned, the most numerically abundant was E. buchholzi. This was present in relatively large numbers throughout the year and was the only worm present which was actively reproducing sexually. Three naidid worms were present - Pristina idrensis, Pristina foreli and Nais elinguis, of which P. idrensis was the most abundant, the other two species being present very occasionally. Sexually reproducing forms of the naidids were never encountered, although varying proportions were actively repro-

Table 5.1 Meiofaunal organisms found in Bed 45, Hampton Treatment Works (T.W.A.) 1975-1976, and from other sources.

Major groups			Species	This study	Lloyd 1974	Husmann 1958	Rittersbusch 1974
Platyhelminthes	Turbellaria	Microturbellaria	Catenula lemnae	*			
			Microstonum lineare	* R	* S		
			Stenostonum sp.	* R	*		
			Castrella truncata	* R			
		Tricladia			*		
Rotifera				*	*	*	*
Gastrotrichia					*		
Nematoda				*	*	*	*
Annelida	Oligochaeta	Aelosomatidae	Aelosoma hemprichi	*	*		*
			A. travancorense				*
		Naididae	Pristina idrensis	*			
			P. foreli	*			
			P. bilobata				*
			Nais elinguis	*			
			N. communis		*	*	
			Stylaria fossularis		*		
			Chaetogaster sp.		* R		
		Tubificidae	Phallodrilus aquaedulcis			*	
		Enchytraeidae	Enchytraeus buchholzi	*		* S	
			Marionina riparia				*
Tardigrada			Macrobiotus sp.		*		
Arthropoda	Crustacea	Copepoda Harpacticoida	Canthocamptus staphylinus	*	* S		*
			Nitocra hibernica			*	*
			N. reducta			*	

Table 5.1 continued.

Major groups			Species	This study	Lloyd 1974	Husmann 1958	Rittersbusch 1974
Arthropoda	Crustacea	Copepoda Harpacticoida	Phyllognathopus viguieri			*	
			Epactophanes richardi			*	
		Cyclopoida		* F		*	
		Ostracoda		* F	* R		
	Diptera	Chironomidae		*			
	Arachnida	Acarina		* RF	*		

* -indicates that the species has been recorded by the various authors

R -indicates rarely occuring groups or species

S -indicates that an unidentified species of the genus has been recorded

F -indicates that this species is found only in the surface film, is really an inhabitant of the surface water but may browse off the filter skin

ducing by asexual budding. A. hemprichi was the only member of the Aelosomatidae present and this also reproduced asexually by budding.

E. buchholzi is a very cosmopolitan species and has been recorded from terrestrial soil environments (Springett 1970, Abrahamsen 1971, Dash and Cragg 1972), interstitial aquatic habitats (Botea 1963), percolating sewage filters (Learner 1972, Solbé 1975), as well as in this slow sand filter.

N. elinguis, P. idrensis and P. foreli have been recorded in sewage percolating filters (Solbé 1975, Learner et al 1978) and gravel beds (Botea 1963, Ladle 1971). In addition, N. elinguis has been reported from brackish beaches (Beschindt and Noack 1976). It would appear that N. elinguis is more characteristic of detritus rich (Botea 1963, Beschindt and Noack 1976, Learner et al 1978) or "organically polluted" situations (Wachs 1967, Ladle 1971, Eyres et al 1978). The fact that it is found only occasionally in the slow sand filter suggests that the filters are not sufficiently rich in detritus to allow the development of large populations of this species. For although the top 1 and 2cm of the filter do accumulate high levels of organic carbon, the remainder of the filter experiences little overall change in the level of detritus present.

The fifth oligochaete recorded, A. hemprichi, has also been recorded in interstitial situations (Botea 1963, Ladle 1971) and detritus rich environments (Beschindt and Noack 1976).

5.2 Dry weight length, segment number relationships of Enchytraeus buchholzi

5.2.1 Dry weight determinations

Biomass measurements of organisms can be expressed in several ways, including energy content, carbon content, dry weight and wet weight. It is possible to interchange these measurements provided precautions are taken to ensure the reproducability of methods used in the determinations. Apart from energetic studies few authors directly determine the energetic value of their material. Wet weight is often criticised because of the difficulty in ensuring removal of all adhering water from organisms. On the other hand, it is impossible for wet weight determinations of some very small organisms to be made, due to the fact that they desiccate within seconds of being removed from water. Despite these problems many authors still record

wet weight.

Some of these problems have been overcome by measuring the body volume and density of organisms (Edwards 1967, Abrahamsen 1973). The former parameter requires the taking of many measurements, a process which can be time consuming and the latter requires the use of solutions of differing densities which must be carefully chosen, to ensure that the water content of the organisms is unaffected by osmotic processes. Because of these problems dry weight determinations are probably more useful than wet weight determinations, provided that care is taken to ensure good reproduc bility of method used (Lovegrove 1966).

Due to their small size, regular dry weight determinations of E. buchholzi were difficult to carry out. A more convenient method would be to regularly measure some anatomical feature of the worm which could be used to relate to dry weight. The lack of rigid features, apart from the chaetae, means that most parts of oligochaete worms are difficult to measure. This difficulty is more pronounced when measurement of live animals is required, as is the case in culture experiments. The most well used method of obtaining worm length is by measuring preserved length under a microscope, but this can be difficult when curling of the worm is caused by the various chemicals used. Kosiorek (1974) surmounted this problem by using ethyl ether to kill the worms, which kept the worms straight. In this study the number of segments per worm were counted and related to dry weight. This method allowed worms to be examined alive, without handling and was quick and easy to carry out with use of a x 70 magnification microscope.

Methods of obtaining dry weight measurements vary very much in the literature. In most cases no information is given as to how full the gut is when weighed. Brinkhurst (1970) has shown that in a situation where the sediment from which oligochaetes are removed contains a high proportion of finely divided mineral material, the ratio of dry weight of the worm plus gut contents, to dry weight of worms with empty guts, could be as high as 2·5 : 1. In this case guts were allowed to empty prior to weighing.

The use of preservatives prior to dry weight determinations has been criticised, as it has been shown that use of these materials causes changes in subsequent dry weight determinations over periods of time. The most common effect is reduction in dry weight

(Howmiller 1972, Jonasson and Thorhange 1972, Stanford 1973, Donald and Paterson 1977), but increases in weight have also been recorded (Lovegrove 1966). In this study 5% formalin was only used to kill the organisms - exposure period was 5 seconds and then the worm was immediately removed and washed in 3 changes of water to remove any traces of formalin, before being dried.

With small organisms the use of an oven for drying the material is not necessary (Lovegrove 1966) and it was observed here that the worms began to desiccate within seconds of being removed from water and placed onto a platinum pan. The dried material is very hygroscopic and increases in weight due to absorption of atmospheric water were examined.

5.2.2 Reproducibility of measurements taken on a Cahn balance

A clean platinum pan was placed in a desiccator for 96 hours. At the end of this period it was transferred using fine forceps to the pan of a Cahn Gram Electrobalance and weighed. The pan was then reweighed a total of 50 times over a period of 40 minutes. Precision of method was very high with the standard deviation being 0·082% of the mean, the mean pan weight was 694·7μg (appendix 5.1). There was no change in weight of the pan caused by reaction of atmospheric water with the platinum.

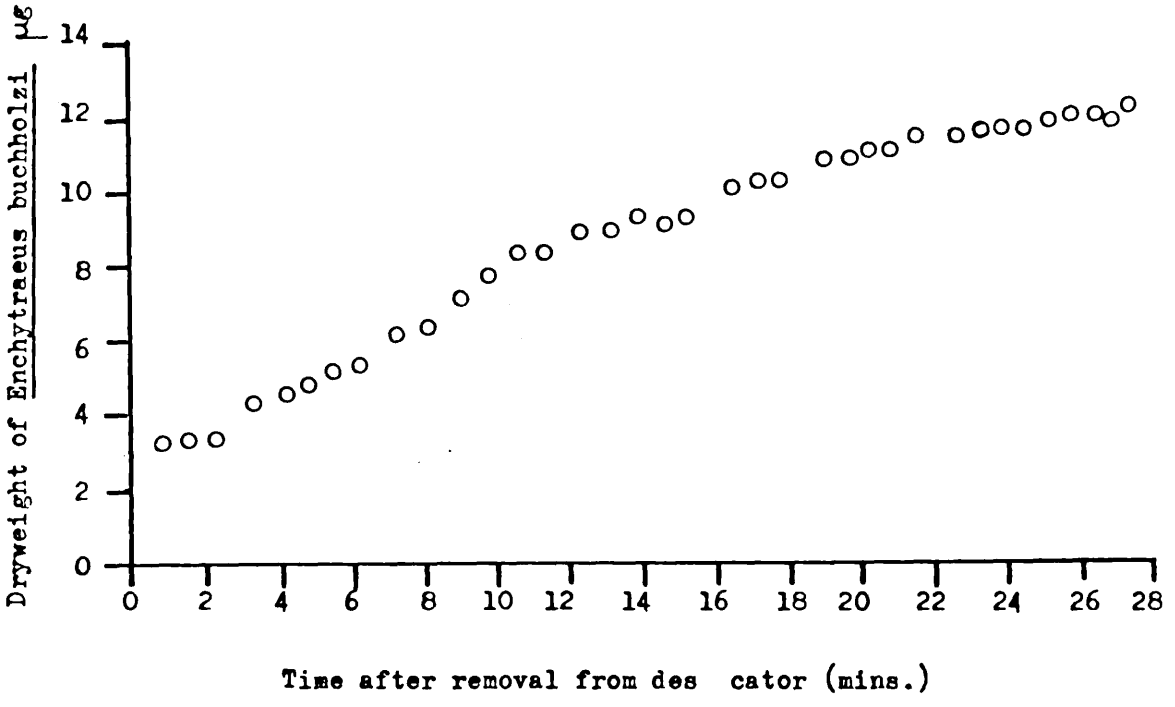
5.2.3 Uptake of atmospheric water by a dried worm

The worm was treated as described in 5.2.4 and dried on a platinum pan for 96 hours. The pan and worm was then transferred to a Cahn Gram Electrobalance containing a pan of calcium chloride crystals, using fine forceps and weighed at intervals over a period of 28 minutes. After the 28 minutes period the weight of the worm had increased by 260% (figure 5.1). This underlines the speed necessary for an accurate dry weight determination of the worm. In all subsequent weighings the weight of the worm was determined within 60 seconds of being removed from the desiccator, any measurements which took longer than this to record were rejected.

5.2.4 Method of obtaining dry weights

Using fine tungsten needles individual worms were placed into separate haemaglutination trays containing water, for the guts to empty. The worms were examined under a low power binocular microscope

Figure 5.1 INCREASE IN DRY WEIGHT OF E. BUCHHOLZI DUE TO UPTAKE OF ATMOSPHERIC WATER



and those with empty guts were removed, to be used in the weight determinations. Others with gut contents were discarded. Each worm was then dropped into 5% formalin for 5 seconds to kill it, removed and given 3 washes in distilled water to remove traces of formalin solution. The worm was then placed into a haemaglutination tray containing water, segment number was counted, reproductive state noted and the outline of the worm was drawn, using a camera lucida attachment on a Leitz microscope at magnification x 50. The length of the worm was determined using a map measurer.

The worm was then removed from the tray, using a tungsten needle and transferred under a microscope to a small clean platinum pan, approximately 1mm square. This was placed in a dry haemaglutination tray in a desiccator containing freshly prepared calcium chloride crystals for 96 hours. After this period the pan and worm were removed and weighed on a Cahn Gram Electrobalance (sensitivity 0·2 μg). The weighing cabinet of the balance contained a tray filled with freshly prepared calcium chloride crystals in an attempt to reduce the atmospheric water content of the air in the apparatus.

5.2.5 Length - dry weight relationship

The relationship between body length and dry weight in E. buchholzi is plotted on a double log scale (figure 5.2). The relationship is described by the equation $W = 1{\cdot}37.10^{-5}.L^{1{\cdot}70}$, where W = dry weight in μg, L = body length in μm. The relationship was investigated by regression analysis and was found to be significant ($p < {\cdot}001$).

It has been well documented (e.g. review in Winberg 1971a) that when there is no alteration in body shape during growth, then the slope of the relationship b = 3, i.e. weight increases are proportional to the cube of the length. However, if the ratio of linear measurement to weight increases during growth then a slope of less than 3 will be recorded. This is the situation with oligochaetes which exhibit a high increase in length with respect to weight. The value of b = 1·70 in this study compares well with data available for similar organisms in table 5.2.

With E. buchholzi it was possible to separate individuals into those which were mature (eggs could be seen in the body) and immature (no eggs present). Although with other species it is possible to use the presence of a clitellum to indicate maturity,

Figure 5.2 ENCHYTRAEUS BUCHHOLZI: LENGTH-DRY WEIGHT RELATIONSHIP

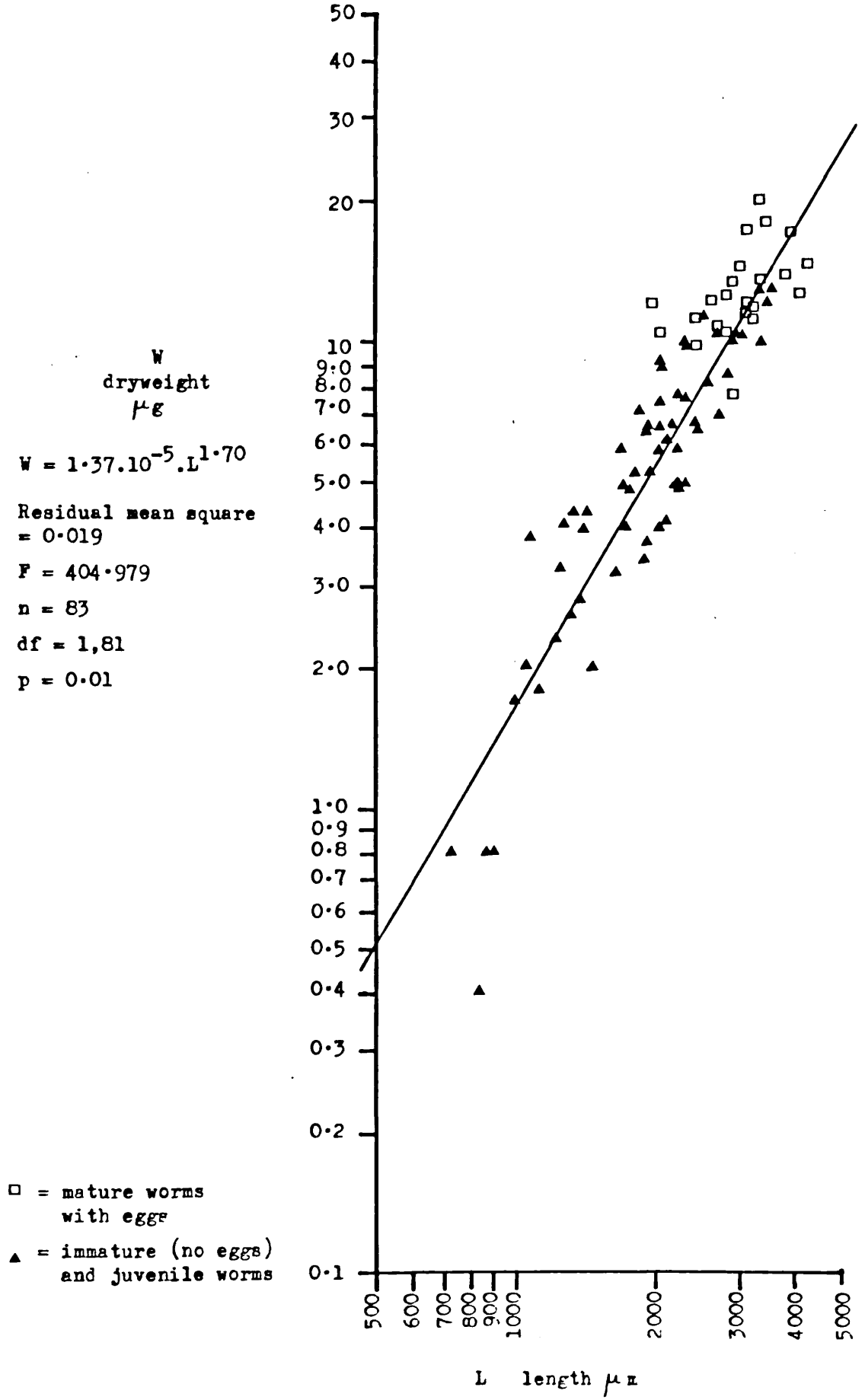


Table 5.2 Regression coefficient (b) of length-weight relationships (double logarithmic scale) of various oligochaetes.

Organism	b	Author
Tubifex tubifex	1.76	Kosiorek 1974
Enchytraeus buchholzi	1.70	Present study
Stylaria lacustris	2.10	Kamlyuk and Kovalchuk 1972
Isochaetides newaensis	1.80	Poddubnaya 1972

in this case the clitellum only extends over segments 12 and 13 and is very difficult to see. However, due to the transparent nature of the worm it was possible to observe eggs present in the body and this was used to indicate maturity. A further subdivision of the immature class was made to indicate individuals which had recently hatched from cocoons. From laboratory data (chapter 5.3), it was observed that hatching worms never exceeded 16 segments in number and so all worms counted with 16 or less segments were recorded as "juvenile". Thus it was possible to separate worms into mature (eggs present), immature (no eggs present) and juvenile (16 or less segments). The mean weights and lengths of these three groups are given in table 5.3.

5.2.6 Segment number - dry weight relationship

The relationship between segment number and dry weight in E. buchholzi is plotted on a double log scale (figure 5.3) and is described by the equation $W = 2{\cdot}24.10^{-4}.S^{3{\cdot}19}$, where W is dry weight in μg and S is segment number. The relationship was investigated by regression analysis and judged significant ($p < {\cdot}001$). This demonstrates the good relationship between segment number and dry weight in this species and indicates the usefulness of segment number as a parameter which can be measured quickly and with ease in living as well as dead animals. It is useful here to compare the residual mean squares of the length/dry weight and segment number/dry weight relationships. The residual mean square of the former relationship was found to be 0·019 and of the latter 0·011, indicating that slightly fewer errors are met in counting segment number than in measuring length.

5.3 Enchytraeus buchholzi - cocoons, field data and laboratory hatched worms

The number of eggs present per cocoon were recorded from all field samples between May, 1975, and March, 1976 (filtration rate 20 - 35cm.hr^{-1})(appendix 5.2). The relationship between egg number per cocoon (field data) and cocoon length of a sample (figure 5.6) was investigated by regression analysis, the relationship was significant ($F = 14{\cdot}602$, df = 1, 48, $p < {\cdot}001$).

In order to have some idea of the segment number of juvenile worms on hatching from the cocoon a sand sample was removed from

Figure 5.3 E.BUCHHOLZI: SEGMENT NUMBER - DRY WEIGHT RELATIONSHIP

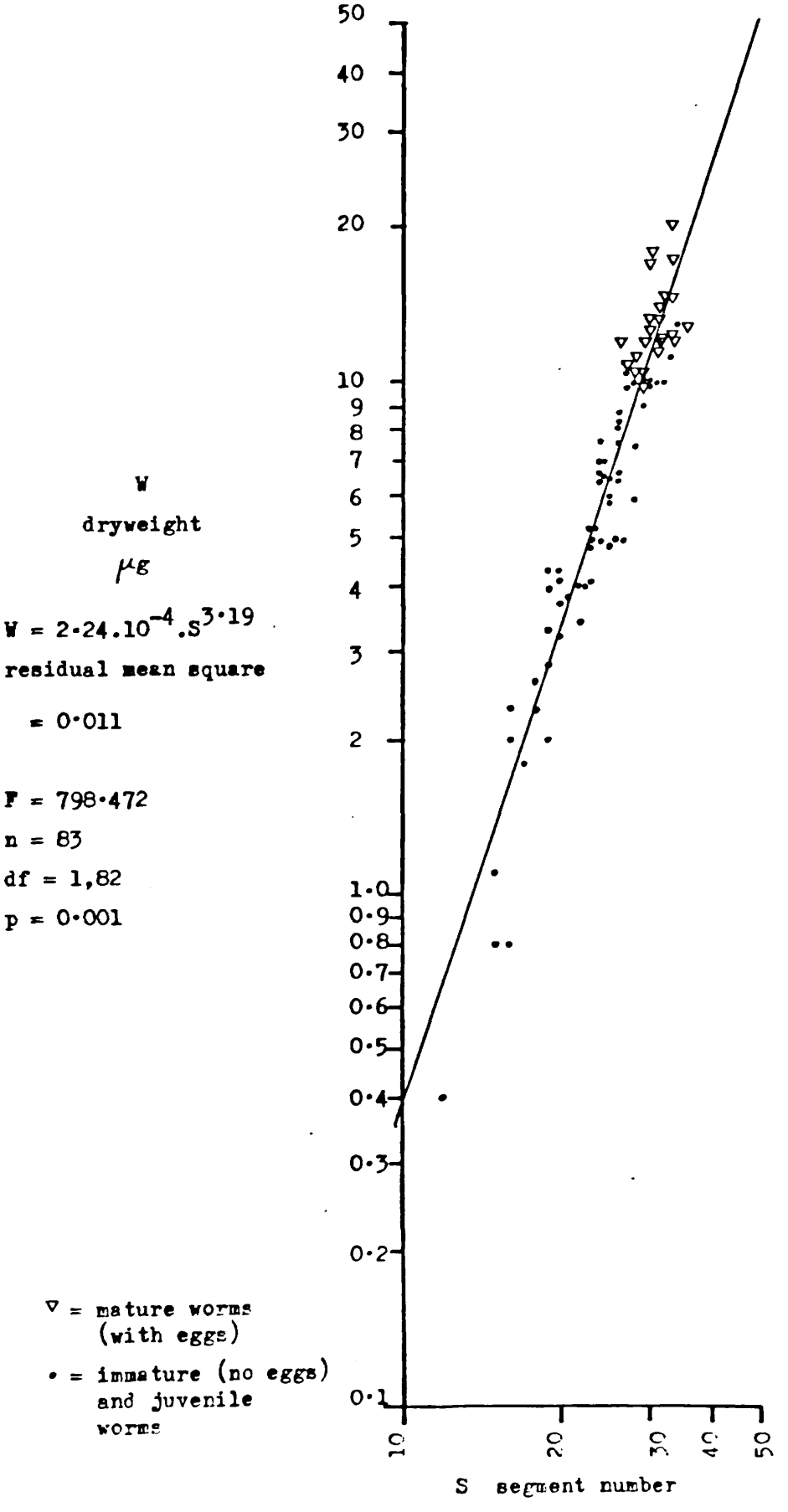


Table 5.3 Mean dry weights and lengths of Enchytraeus buchholzi.

	n	Weight (µg) $\bar{x}$	Weight (µg) 95% confidence limits	Length (µm) $\bar{x}$	Length (µm) 95% confidence limits
Mature worms	23	13.1	11.8-14.4	3231	2942-3520
Immature worms	53	6.4	5.6-7.2	2134	1959-2309
Juvenile worms	7	1.2	0.5-1.8	912	757-1067

Figure 5.4
E. BUCHHOLZI
FIELD DATA
MEAN EGG NO.
PER COCOON,
WATER
TEMPERATURE
RELATIONSHIP

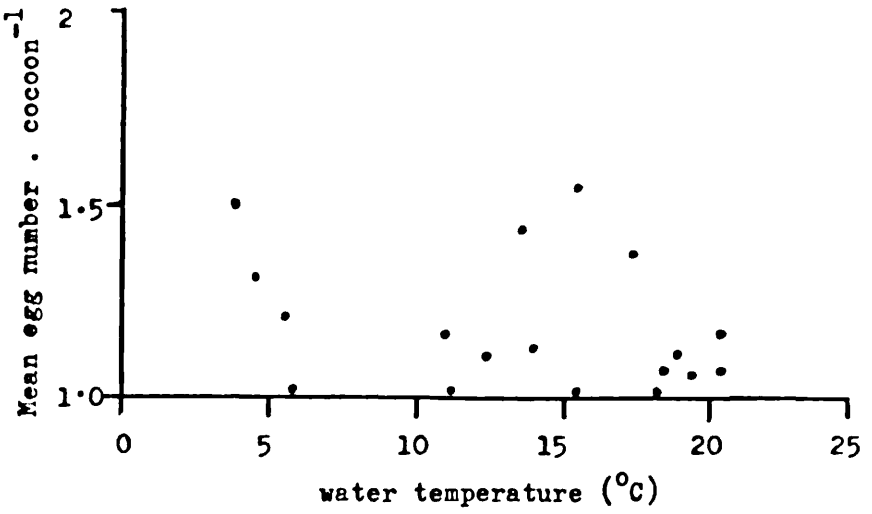


Figure 5.5
E. BUCHHOLZI
LABORATORY
HATCHED WORMS
15°C
NO. OF JUVENILES
PER COCOON,
COCOON LENGTH
RELATIONSHIP

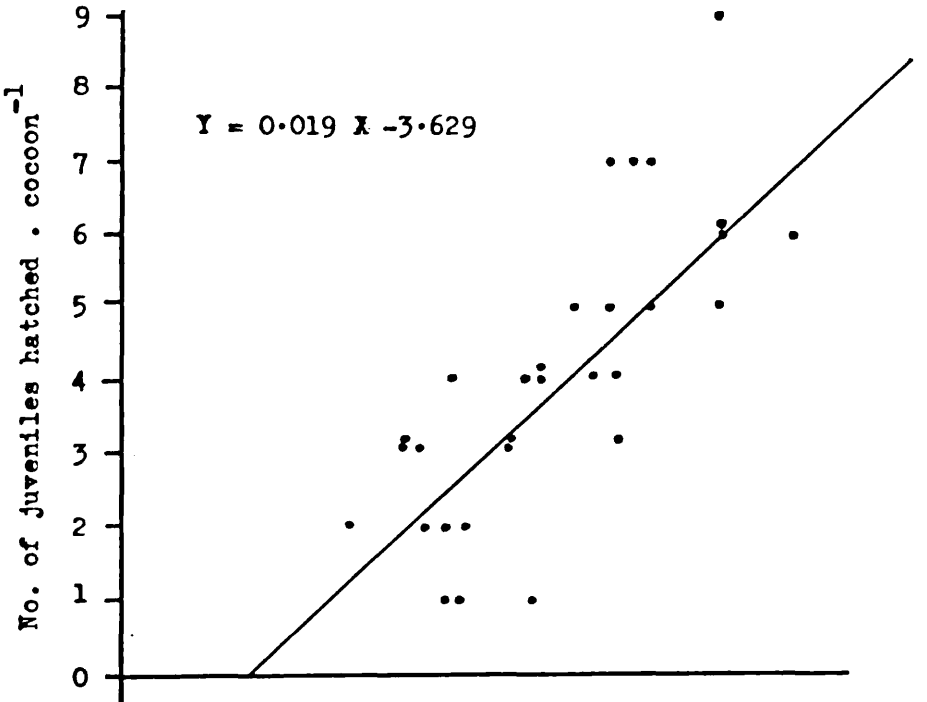
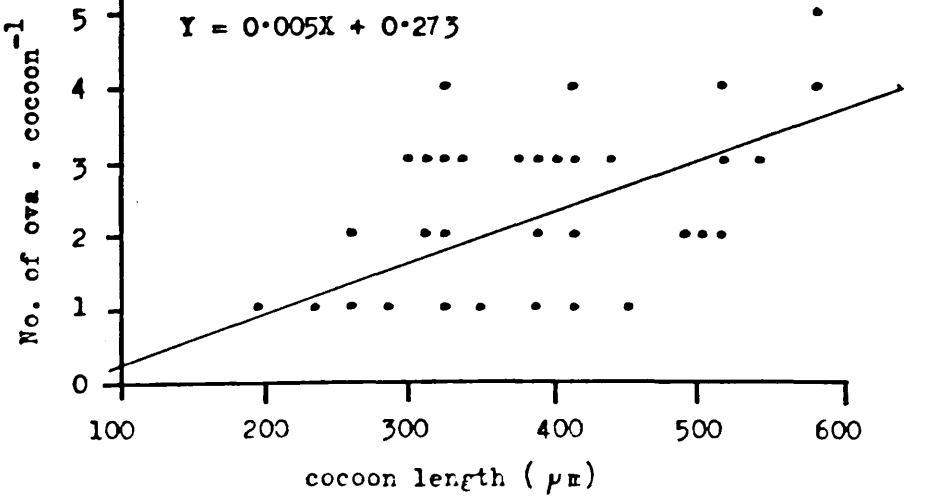
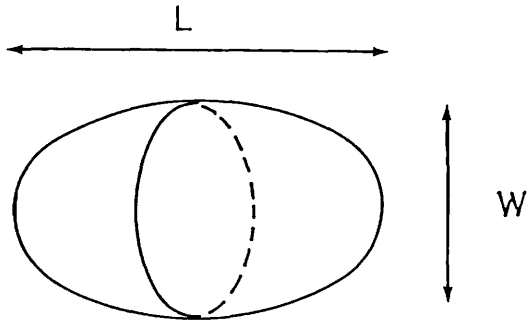


Figure 5.6
E. BUCHHOLZI
FIELD DATA
NO. OF OVA PER
COCOON, COCOON
LENGTH RELATION-
SHIP



bed 45 in May, 1977 (water temperature 15°C). This sample was removed from a bed which had been run at over 40cm.hr^{-1} since the beginning of March, 1976. Length and width of each cocoon was recorded (appendix 5.3). Each cocoon was placed in a separate container in water and kept in the dark at ambient water temperature (15°C). Every 24 hours the containers were removed and the number of any worms which had hatched and their segment number, were recorded. Some growth may have occurred in these juveniles between hatching and being observed. The volume of each cocoon was determined by the use of the following formula, based on the assumption that the cocoons are egg shaped (prolate spheroid) formed by rotating an ellipse about its major axis.



$$\text{Volume} = \frac{4}{3}\pi\left(\frac{L}{2}\right)\left(\frac{W}{2}\right)^2$$

where L = length and W = width

The relationshipsbetween numbers of juveniles hatched and length, width and volume of cocoon were investigated by regression analysis. In all 3 cases there is a significant ($p < .001$) relationship between the 2 parameters concerned (table 5.4). The relationship between numbers of juveniles hatched and cocoon length is illustrated in figure 5.5. Similar relationships between numbers of eggs per cocoon and cocoon length have been demonstrated by Williams et al (1969) for Enchytraeus coronatus and Lumbricillus rivalis.

The mean egg number per cocoon (field data) does not seem to be related to temperature (figure 5.4), although there is some suggestion of reduced egg number at 20°C. Learners data (1972) on E. buchholzi (table 5.5) would also suggest a slightly reduced egg number per cocoon at 20°C. All of Learners (1972) data was derived from laboratory cultured animals originally removed from sewage

Table 5.4 Regression analyses of number of ova per cocoon and number of juveniles hatched per cocoon, of E. buchholzi, on various cocoon dimensions.

	Abscissa	Ordinate	d.f.	F	p	b
Laboratory hatched data	Cocoon length µm	No. juveniles hatched	1,28	38.030	.001	.019
	Cocoon width µm	" " "	1,28	43.078	.001	.028
	Cocoon volume µm	" " "	1,28	40.320	.001	.141
Field data	Cocoon length µm	No. ova per cocoon	1,48	14.602	.001	.005

Table 5.5 E. buchholzi, summary of available data.

	Christensen 1956	Springett 1970	Learner 1972	This study
Egg no. per cocoon		2 *	1-9 * $\bar{x}$=3.5(8°C) 3.3(15°C) 2.9(20°C)	1-9 $\bar{x}$=1.188 $\bar{x}$=3.7 **
Cocoon length				382-439µm $\bar{x}$=411µm
Segment no. on hatching		9 ± 2		12-16 ** $\bar{x}$=14.5
Length on hatching		1900µm		757-1067µm $\bar{x}$=912µm
Segment no. at maturity		30 ± 4		26-35
Length at maturity	5-10mm	9mm *	3-4mm *	2942-3520µm $\bar{x}$=3231µm
Habitat	Terrestial	Limestone grassland	Sewage percolating filter	Slow sand filter

* =laboratory data, ** =laboratory hatched data.

percolating filters.

The percentage frequency of egg number per cocoon (field data) is compared with laboratory hatched worms and Learners (1972) data in figures 5.7 and 5.8. There is considerable variation between field data obtained in this study and both Learners (1972) percolating filter data and laboratory hatched data obtained from a slow sand filter operating at a slightly higher filtration rate. This can also be seen in figures 5.5 and 5.6, although the cocoon length range is similar in both sets of slow sand filter data it is apparent that the number of ova per cocoon is much lower in the field situation than in the laboratory hatched (faster rate) situation. This would seem to suggest that in the field situation some of the ova may have died and decomposed during the incubation period. Exactly what are the reasons for these discrepancies is not known, although there is the possibility that the slightly higher filtration rates in the slow sand filter may have produced more favourable conditions, possibly due to a better degree of aeration in the bed, for both adults and cocoons and this was reflected in higher egg numbers per cocoon being laid.

To summarise, the field data in this study indicated a range of egg number per cocoon from 1 to 5 with a mean of 1·188, whereas the laboratory hatched (15°C) data demonstrated a range of 1 to 9 with a mean of 3·7. Learners (1972) laboratory data, taken from sewage percolating filters, indicated a similar range (1 - 9) with a mean of 3·5 (8°C), 3·3 (15°C) and 3·9 (20°C). In all cases the means were obtained using the transformation log x to account for the positively skewed distribution of data.

Segment number per newly hatched worm ranged from 12 to 16 (figure 5.9) with a mean of 14·5. Occasionally the worms had different segment numbers even though they hatched from the same cocoons, but in no case was the difference greater than 1 segment. From a terrestrial population of *E. buchholzi*, Springett(1970) found the mean segment number per worm on hatching was 9 (standard deviation ± 2).

From table 5.5 it can be seen that in general Learners (1972) data agrees well with the results of this study as far as the size of the organisms are concerned, although there are some discrepancies regarding egg number per cocoon. Springett (1970) and Christensen (1956) found larger sized individuals than were found in this study or in Learners data, differences which may well be due to the different environments (table 5.5) from which the worms were removed.

Figure 5.7 E. BUCHHOLZI
PERCENTAGE FREQUENCY OF
EGG NO. PER COCOON

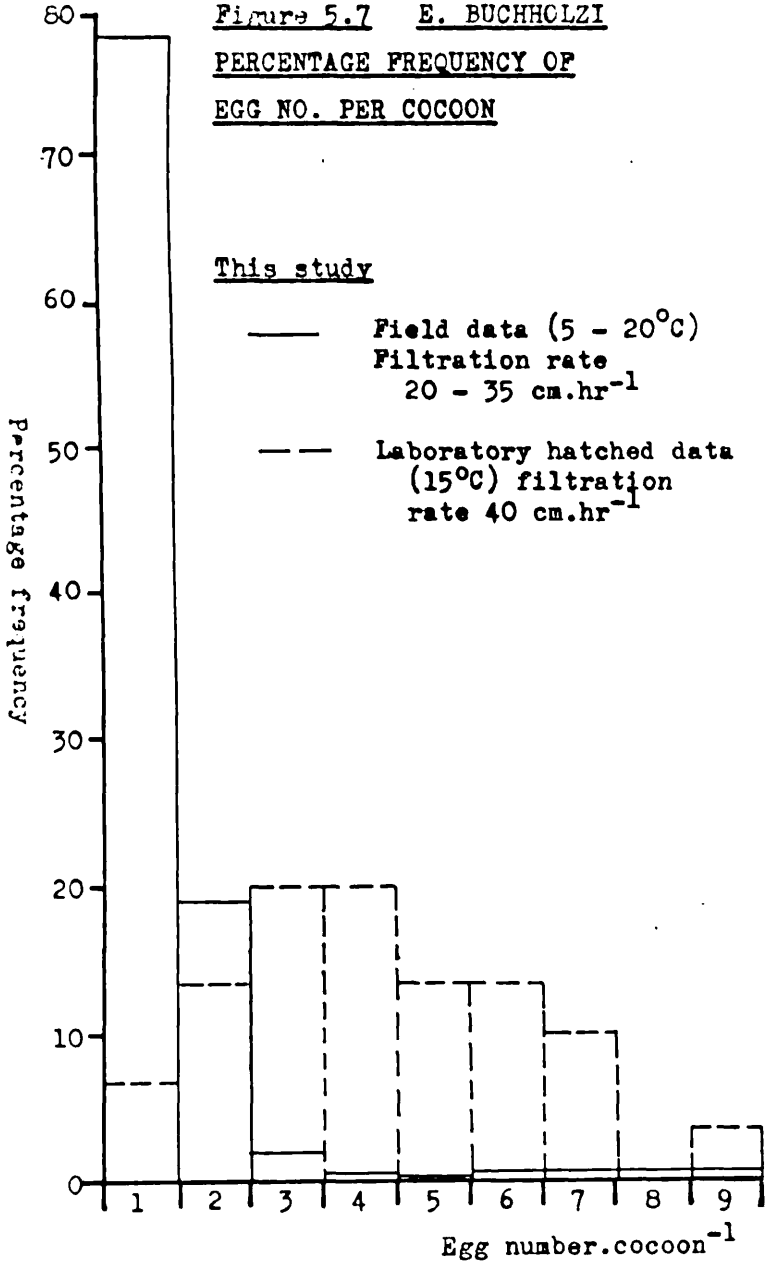


Figure 5.8 E. BUCHHOLZI
PERCENTAGE FREQUENCY OF
EGG NO. PER COCOON
LEARNER'S (1972) LABORATORY
HATCHED WORMS

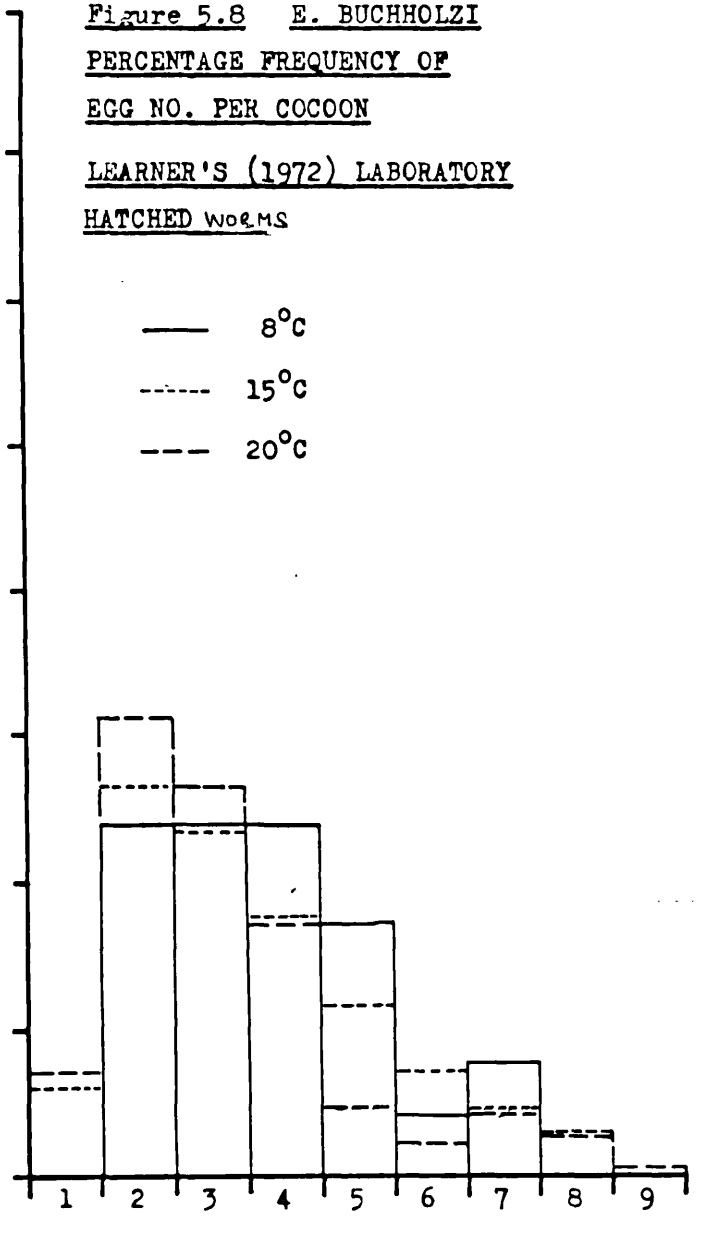
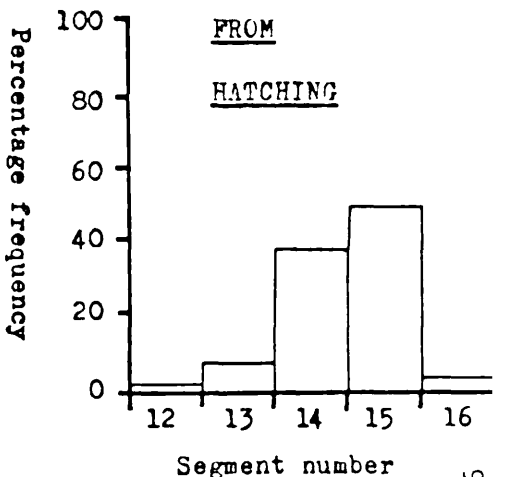


Figure 5.9
E. BUCHHOLZI
SEGMENT NUMBER
WITHIN 24 HOURS
FROM
HATCHING



CHAPTER 6 FIELD WORK RESULTS

6.1 Sampling dates

Sampling dates are given in appendix 6.1. A few points should be noted here :

1. The numbering of these runs is related to the sequential order in which bed 45 was sampled, for example run 1 was the first run that was sampled. It must be pointed out that bed 45 was resanded in December, 1974, and the first run which this bed was subjected to was in January/February, 1975. So the second run which this bed was subjected to corresponds with run 1 of the sampling program.

2. No data are presented for run 1, as this was a trial run used to test sampling procedure, methods used to fill inner cores with sand, extractions of meiofauna and the enumeration procedure.

3. No carbon data are available for runs 2 and 3 as the carbon analysis method was developed, tested and tried out whilst these runs were in progress.

4. All data collection on run 6 were suspended in order to carry out analysis of previously collected data, the results of which were of use in the design and building of the experimental filter column.

6.2 Carbon data

6.2.1 Investigation of the distribution of carbon values within cores

An experiment was conducted to investigate the heterogeneity of carbon values within the same core and also between separate cores removed from the bed on the same sampling trip.

Three cores of the same age were removed at random from a filter bed. From one half of an inner core, three sand samples (each $1cm^3$) were taken at random from each depth examined. This was repeated on the two remaining cores. Results can be seen in appendix 6.2.

In order to treat these data using parametric tests three conditions are necessary (Elliott 1977) :

1. That the data should follow a normal distribution.
2. That the variance should be independent of the mean.
3. That the components of the variance should be additive.

In certain situations it is possible to transform the data so that they conform to a normal distribution. Here it is useful to consider the use of Taylor's Power Law (Taylor 1961). The relationship between S^2 and $\bar{x}$ has been shown to obey a power law, which is expressed by :

$$S^2 = a\,\bar{x}^{\,b} \qquad \text{or} \qquad \log S^2 = \log a + b \log \bar{x}$$

where a and b are constants, with b being an index of aggregation of the parameter involved. If the data can be shown to obey this power law, with variance demonstrated to be dependent on the mean, it is possible to remove this dependence by transforming the data. The correct transformation to apply is x^p where $p = 1 - \frac{b}{2}$ (Elliott 1977, Southwood 1975). Generally, when b = 1, the distribution is random and a square root transformation is appropriate; when b = 2, the distribution is lognormal and the transformation required to normalise the data is logarithmic; and when $b > 2$ the distribution is very overdispersed and a reciprocal transformation is necessary.

Three regression analyses of $\log S^2$ on $\log \bar{x}$ were carried out (table 6.1), (i.e. one regression per core) and the results subjected to covariance analysis which indicated that there was no significant difference ($p > \cdot 5$) between the regression lines (table 6.2). This result indicates that all the data may be grouped to produce one regression line (table 6.3), where the relationship between S^2 and $\bar{x}$ was shown to be significant ($p < \cdot 001$) with the slope b = 1·37. This indicates that the data are tending to follow a poisson distribution and in order to remove the dependence of S^2 on $\bar{x}$ it would be necessary to transform the data using $\sqrt{x}$. The adequacy of this transformation was tested by calculating $\log S^2$ and $\log \bar{x}$ values of the transformed data and subjecting it to regression analysis (table 6.3). The results showed that the relationship between the two variables was now not significant ($p > \cdot 5$), indicating the removal by the transformation of the original dependence of S^2 values on $\bar{x}$ values.

The use of this $\sqrt{x}$ transformation allowed the data to be tested using an analysis of variance technique - a parametric method which demands that the data should follow a normal distribution. A one-way analysis of variance was thus carried out on the transformed data to investigate the heterogeneity of carbon values between the separate cores, at any one depth (table 6.4). It can be seen that at the higher (1, 2, 3cm) and lower (23, 25cm) depths, there is a significant difference ($p < \cdot 05$) between the carbon values determined in the

Table 6.1 Regression analysis of data, in separate cores, in appendix 6.2

Data	Abscissa	Ordinate	d.f.	F	p	b
Core 1	$\log \bar{x}$	$\log S^2$	1,13	10.096	0.001	2.161
Core 2	"	"	1,13	8.026	0.025	1.703
Core 3	"	"	1,13	0.683	0.5	0.417

Table 6.2 Covariance analysis of data in table 6.1

Analysis	d.f.		p
Homogeneity of mean squares	2	$X^2=0.749$	0.9
Comparision of slopes	2,39	F=1.554	0.25
Comparision of elevations	2,41	F=0.301	0.75

Table 6.3 Regression analysis of total data in appendix 6.2, untransformed and transformed.

Data	Abscissa	Ordinate	d.f.	F	p	b
All cores, all depths	$\log \bar{x}$	$\log S^2$	1,43	15.590	0.001	1.370
As above. transformed ($\sqrt{x}$)	"	"	1,43	0.258	0.75	

Table 6.4 One way analysis of variance of organic carbon data in appendix 6.2

Source of variance	Depth cm	d.f.	F	p
Between cores	1	2,6	8.523	0.025
"	2	"	5.726	0.05
"	3	"	5.679	0.05
"	4	"	2.293	0.25
"	5	"	3.713	0.1
"	7	"	0.447	0.75
"	9	"	1.302	0.5
"	11	"	1.926	0.25
"	13	"	0.509	0.75
"	15	"	0.635	0.75
"	17	"	0.137	0.75
"	19	"	0.193	0.75
"	21	"	1.445	0.5
"	23	"	11.675	0.01
"	25	"	10.344	0.025

separate cores, but that in the remaining depths there is no significant difference ($p > .05$) observed.

Two important points have emerged here. The first is that, from the regression analysis, it would appear that the degree of dispersion of the carbon data within the three separate cores remains the same and is random. Secondly, from the results of the one way analysis of variance test, it has been shown that there are significant differences in the amount of carbon found in the three separate cores, at certain depths - mainly at the top of the core. This second point is further investigated in 6.2.2, for all the carbon data available from field results.

These conclusions emphasise the necessity of obtaining carbon values from each core sampled in order to relate to any possible heterogeneity of meiofaunal densities. However it was considered less necessary to take more than one sample from each depth in each core as, on the basis of these results the distribution appeared to be random within each core. On each sampling trip 1 x $1cm^3$ sand sample was removed for carbon analysis from each depth examined in each core removed from the bed.

6.2.2 Investigation of the distribution of carbon between cores and between depths

Carbon data determined in runs 4, 5 and 7 are presented in appendix 6.3. It should be noted here that data from run 4, day 41, core I, was not used in this investigation - several large leaves had deposited on the surface of this core preventing penetration of material into the core and thus causing low carbon values to be accumulated. The carbon penetration within this core was thus not representative of sand from a run which had reached 41 days.

First it was necessary to investigate whether there was any greater heterogeneity in carbon values at the higher depths compared with those below, as was suggested by the results of 6.2.1 and possibly related to the depositing organic matter in the filter water. $\log S^2$ and $\log \bar{x}$ values from depths 1 and 2cm on dates other than the first sampling date were subjected to regression analysis (table 6.5). On the first sampling date there will have been little opportunity for accumulation of carbon to have occurred and the relationship was found to be significant ($p < .001$) and the slope $b = 2{\cdot}228$. Secondly, the $\log S^2$ and $\log \bar{x}$ values of all other data were similarly subjected to regression analysis (table6.5) where the relationship was also sig-

Table 6.5 Regression analysis of carbon data in appendix 6.3

Data	Abscissa	Ordinate	d.f.	F	p	b
Carbon data from depths 1 and 2 cm, excluding first sampling trips	$\log \bar{x}$	$\log S^2$	1,14	27.986	0.001	2.228
All carbon data, excluding that at depths land 2 cm, apart from first sampling trips	"	"	1,99	21.515	0.001	1.67

Table 6.6 Covariance analysis of regressions in table 6.5

Analysis	d.f.	F	p
Homogeneity of mean squares	99,14	1.296	0.5
Comparision of slopes	1,113	0.898	0.5
Comparision of elevations	1,114	0.516	0.5

Table 6.7 Regression analysis of carbon data in appendix 6.3, transformed and untransformed data.

Data	Abscissa	Ordinate	d.f.	F	p	b
All data	$\log \bar{x}$	$\log S^2$	1,115	99.70	0.001	2.011
All data, transformed (log x)	"	"	1,115	3.020	0.1	

nificant ($p < \cdot001$) and the slope was slightly less at $b = 1\cdot67$. This discrepancy in the value of b might indicate that in the higher depth there was a greater degree of overdispersion of carbon. An analysis of covariance test was used to compare these two sets of data. The results (table 6.6) would seem to indicate that there is no signficant difference ($p > \cdot25$) between the two sets of data and so no significant difference in the degree of dispersion of carbon values at the different depths. These two sets of data may therefore be grouped together.

The relationship between S^2 and $\bar{x}$ for all depths on all sampling dates was investigated by subjecting log S^2 and log $\bar{x}$ to regression analysis (table 6.7). The relationship between the two was significant ($p < \cdot001$) and the slope $b = 2\cdot111$. So the dependence of S^2 on $\bar{x}$ is demonstrated and a transformation of the data is indicated if parametric tests are to be employed in the next analysis. The slope of the relationship $b = 2\cdot111$ indicates that a logarithmic transformation should be used. The adequacy of this transformation was tested by calculating log $\bar{x}$ and log S^2 of the transformed data and subjecting it to regression analysis (table 6.7) where the dependency of S^2 on $\bar{x}$ was shown to be removed as the relationship between the two variables was shown to be not significant ($p > \cdot05$).

Summarised, this result indicates that there is no significant difference with depth in the degree of dispersion of carbon in the bed, and so all depths may be considered together for statistical purposes. Secondly, the results indicate that the carbon data are overdispersed and may be following a log-normal distribution.

6.2.3 Comparisons of carbon data with that determined by the Thames Water Authority

Thames Water Authority carbon analyses of sand removed from bed 45 are given in appendix 6.4. The Thames Water Authority samples were taken at the end of each filter run after the bed had been drained for cleaning purposes. A hole was dug in the bed and samples of sand removed at known depths. Comparisons are only possible between the Thames Water Authority carbon data and the carbon values determined in this study at the ends of runs5 and 7, no Thames Water Authority carbon data are available for the end of run 4. A problem here is that the depths examined by the Thames Water Authority do not exactly correspond with those depths examined in this study. It was possible to construct

depth profiles with the Thames Water Authority data by using the uppermost of the two depth sections quoted for one carbon value as the depth against which to relate the carbon value (e.g. in run 7 the carbon value for depth section 5 - 7·5cm is plotted at depth 5cm).

Carbon values were plotted on a log scale against depth (figure 6.1) and subjected to regression analysis (table 6.8). For run 5, in both cases the straight line relationship between carbon value (log scale) and depth is significant: Thames Water Authority data, $p < \cdot 025$; this study, $p < \cdot 001$. In run 7 there is a significant straight line relationship between carbon (log scale) and depth ($p < \cdot 001$) in this study, but the Thames Water Authority data do not show a significant relationship ($p > \cdot 05$).

With the use of an analysis of covariance test (table 6.9) it was possible to compare the two regression lines in each run. In run 5 there was no significant difference ($p > \cdot 75$) between the two regression lines, but in run 7 there was a significant difference ($p < \cdot 01$)(table 6.9).

It is difficult to conclude anything of any meaning on the basis of only two comparisons of this kind, especially considering the wide difference in the number of data points provided from the two locations (this study $n = 45$, Thames Water Authority data $n = 6$). At the end of run 7 it would appear that there is a significant difference between levels of carbon in the sand cores and in the bed (Thames Water Authority data). This is to be expected considering the fact that washed sand had originally been placed in the cores prior to the start of a run, whereas in the bed (Thames Water Authority data) carbon has been accumulating throughout the depth of the sand since the bed was resanded. However, at the end of run 5 there is no significant difference demonstrated between carbon values found in the two locations. There may well be a time factor operating here with greater penetration of detritus with depth having occurred in the later run. This would be causing higher carbon values to be recorded at greater depths thus causing the difference between the two regression lines to be significant.

6.2.4 Vertical distribution of carbon in the sampling cores

The vertical distribution of carbon was investigated by plotting carbon values, on a log scale, against depth in the core (figure 6.2). An examination of these plots indicates that penetration of incoming organic carbon does not go further into the sand than the top 1 - 2cm. It can also be seen from these plots that towards the end of a filter

Figure 6.1 DEPTH PROFILES OF ORGANIC CARBON DATA DETERMINED IN THIS STUDY AND FROM T.W.A. DATA

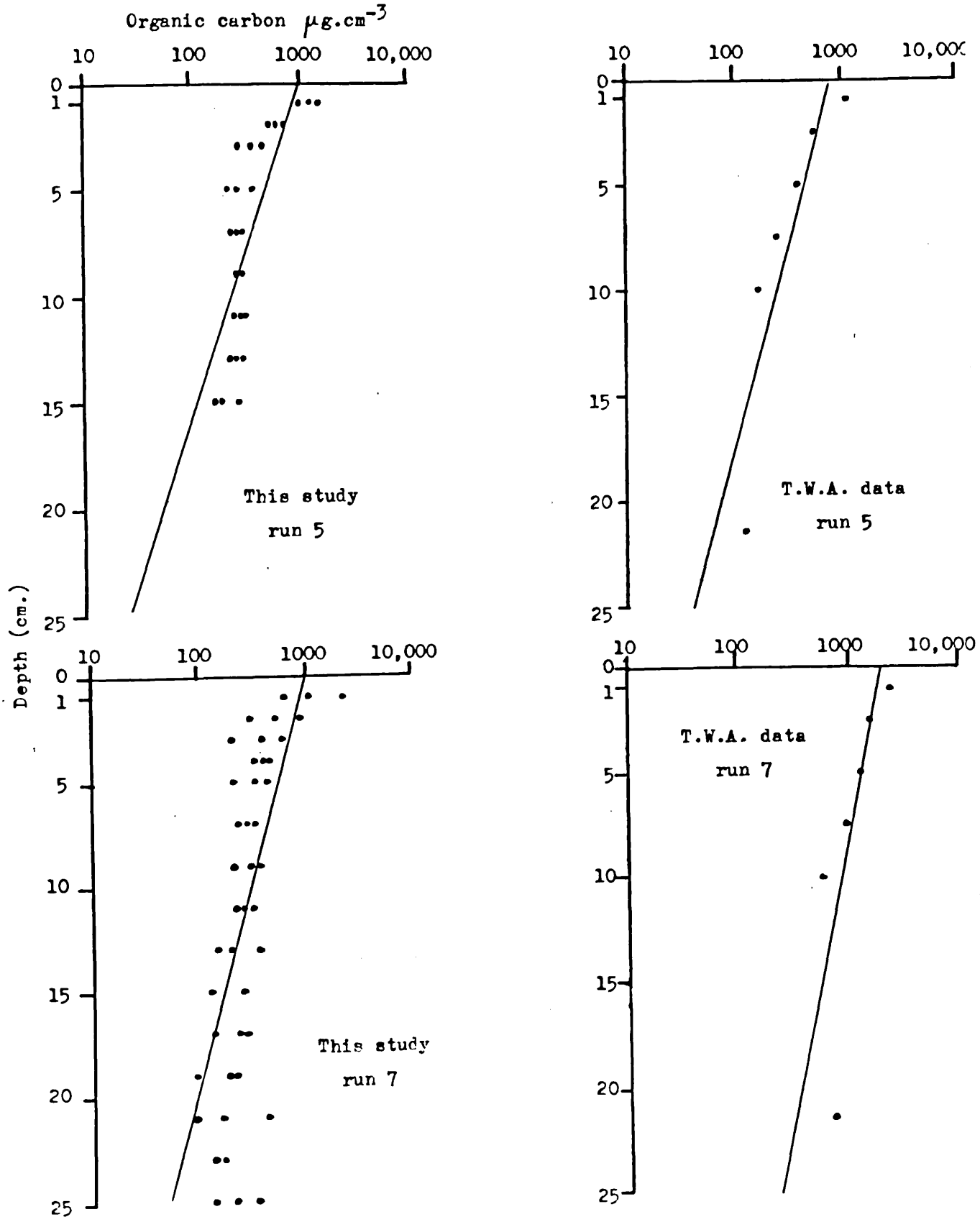


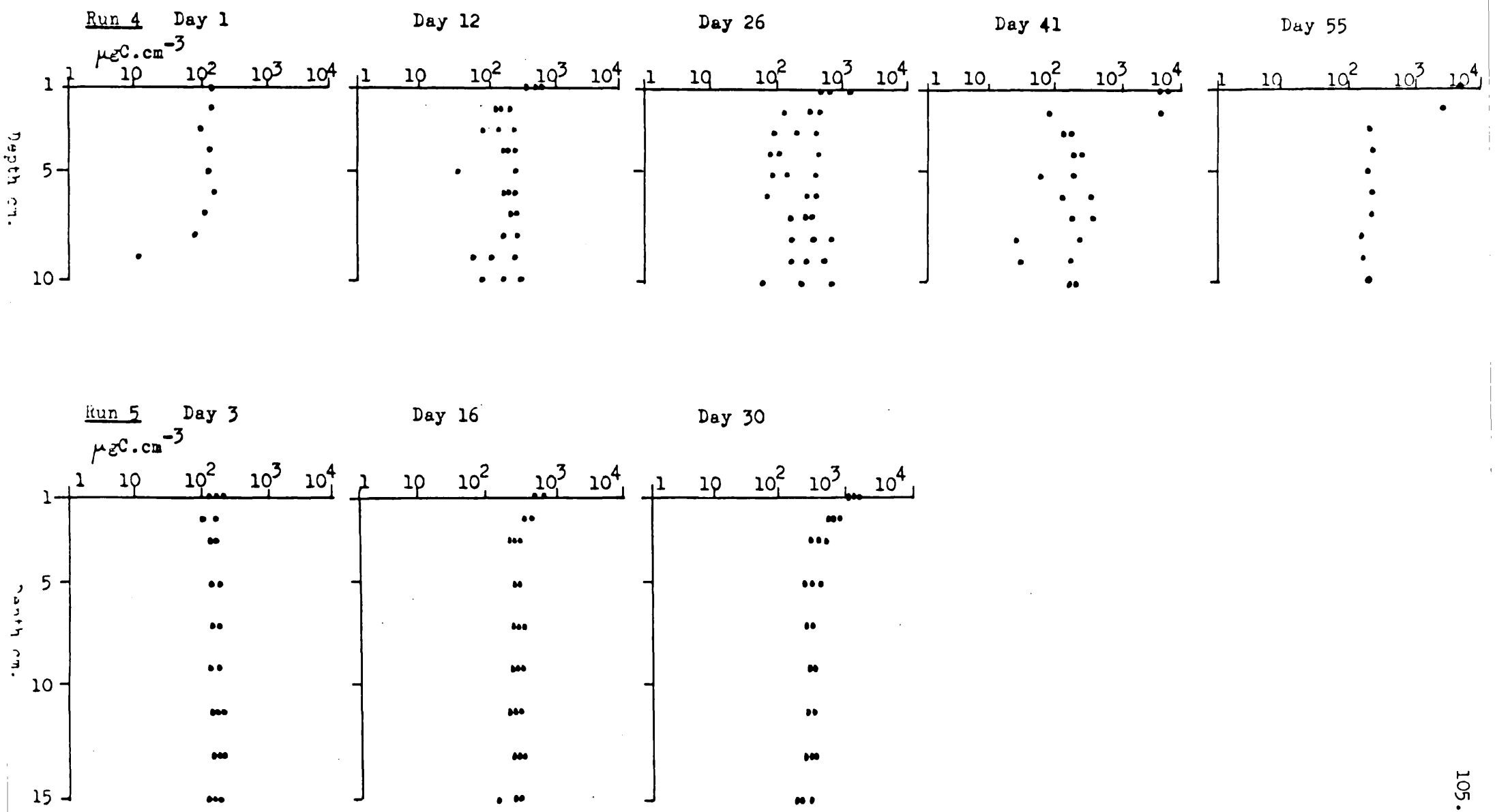
Table 6.8 Regression analysis of carbon data presented in figure 6.1

Data	Abscissa	Ordinate	d.f.	F	p
Present study, Run 5	log C	depth	1,25	32.503	0.001
T.W.A. data, Run 5	"	"	1,4	13.962	0.025
Present study, Run 7	"	"	1,43	39.257	0.001
T.W.A. data, Run 7	"	"	1,4	6.102	0.1

Table 6.9 Covariance analysis of regressions in table 6.8

Data	Analysis	d.f.	F	p
Run 5	Homogeneity of mean squares	4,25	1.488	0.5
	Comparision of slopes	1,29	0.453	0.75
	Comparision of elevations	1,30	0.059	0.75
Run 7	Homogeneity of mean squares	43,4	1.212	0.5
	Comparision of slopes	1,47	0.267	0.75
	Comparision of elevations	1,48	7.778	0.01

Figure 6.2 DEPTH PROFILES OF ORGANIC CARBON DATA



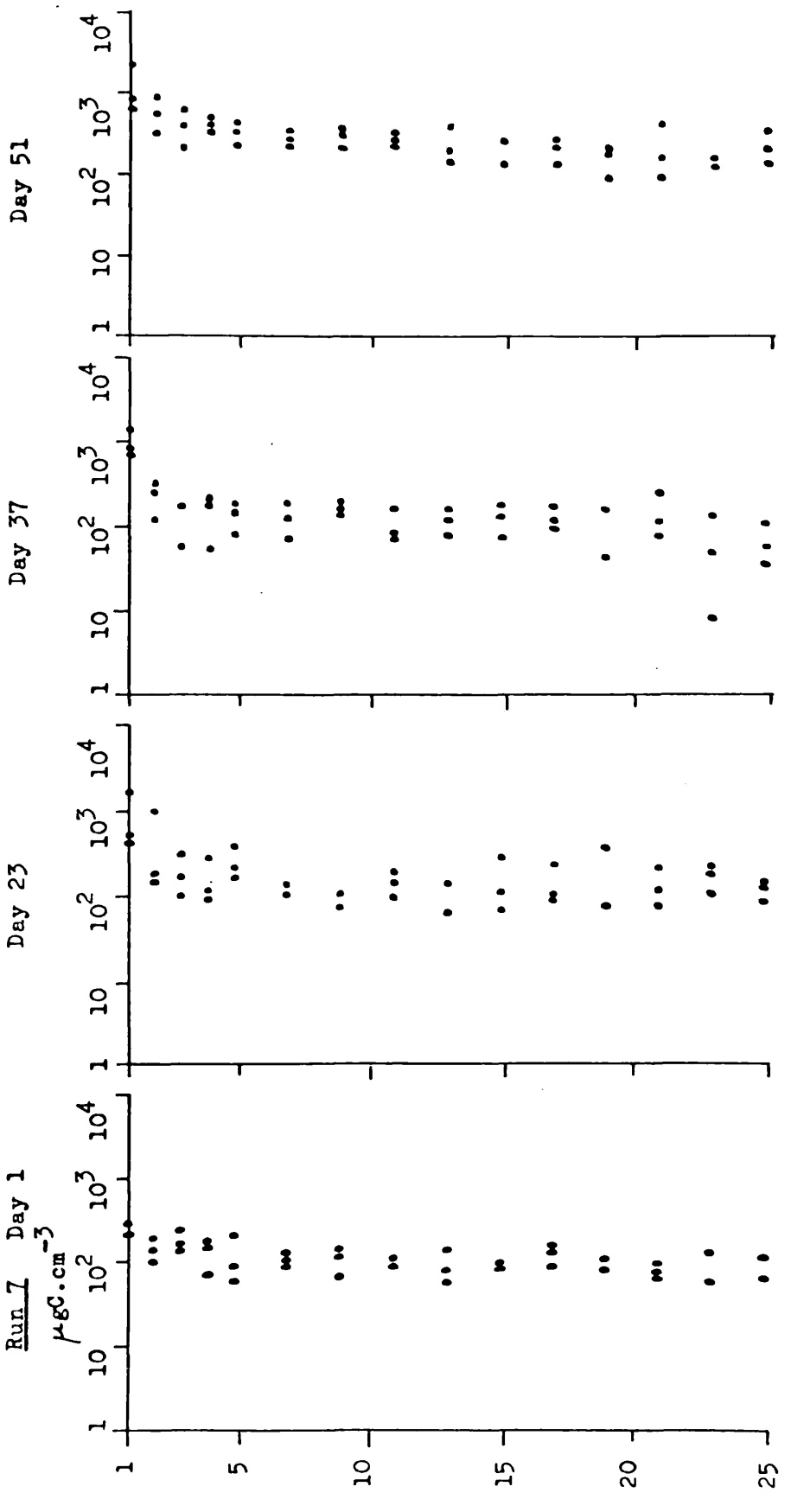


Figure 6.2 (Continued)

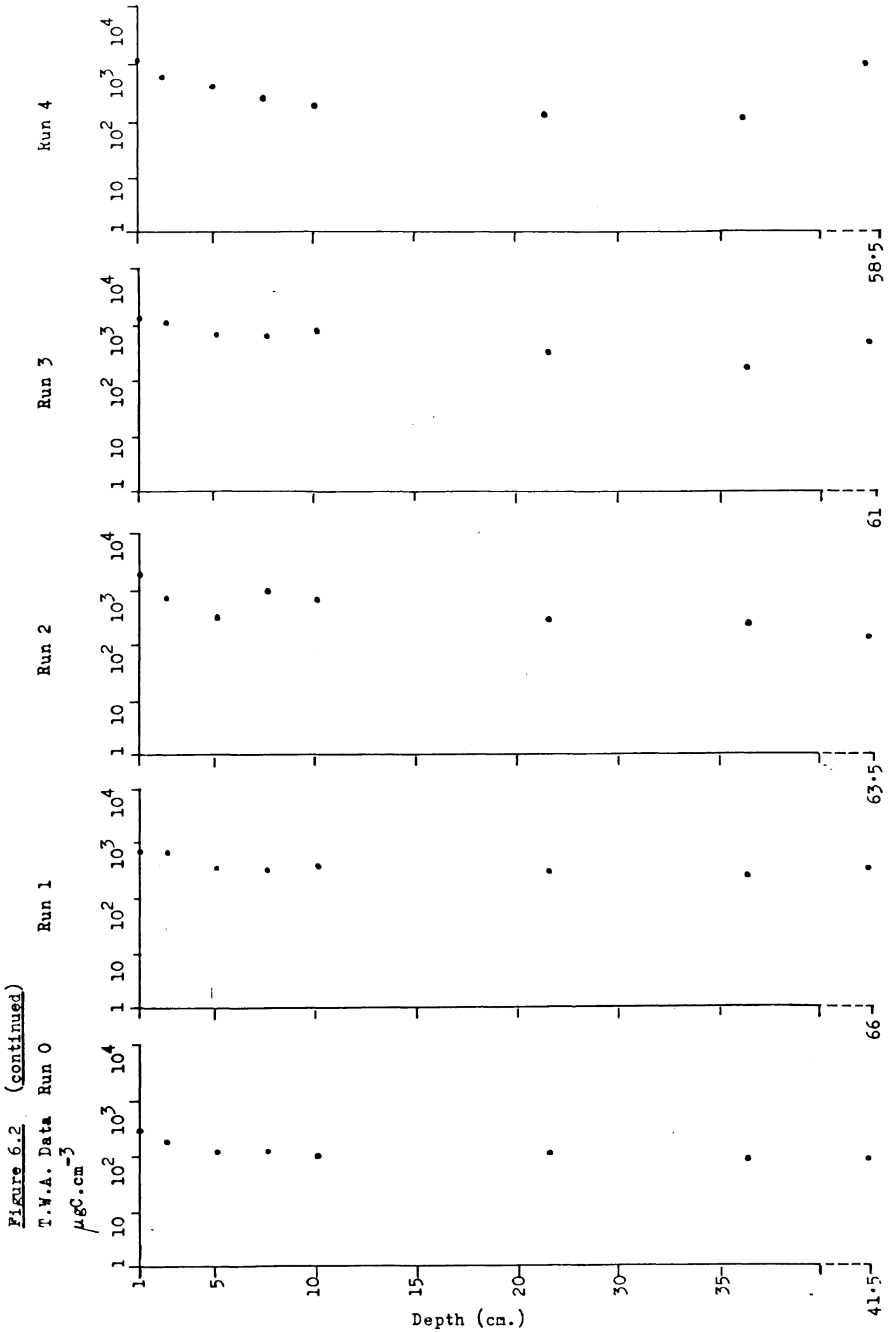


Figure 6.2 (continued)

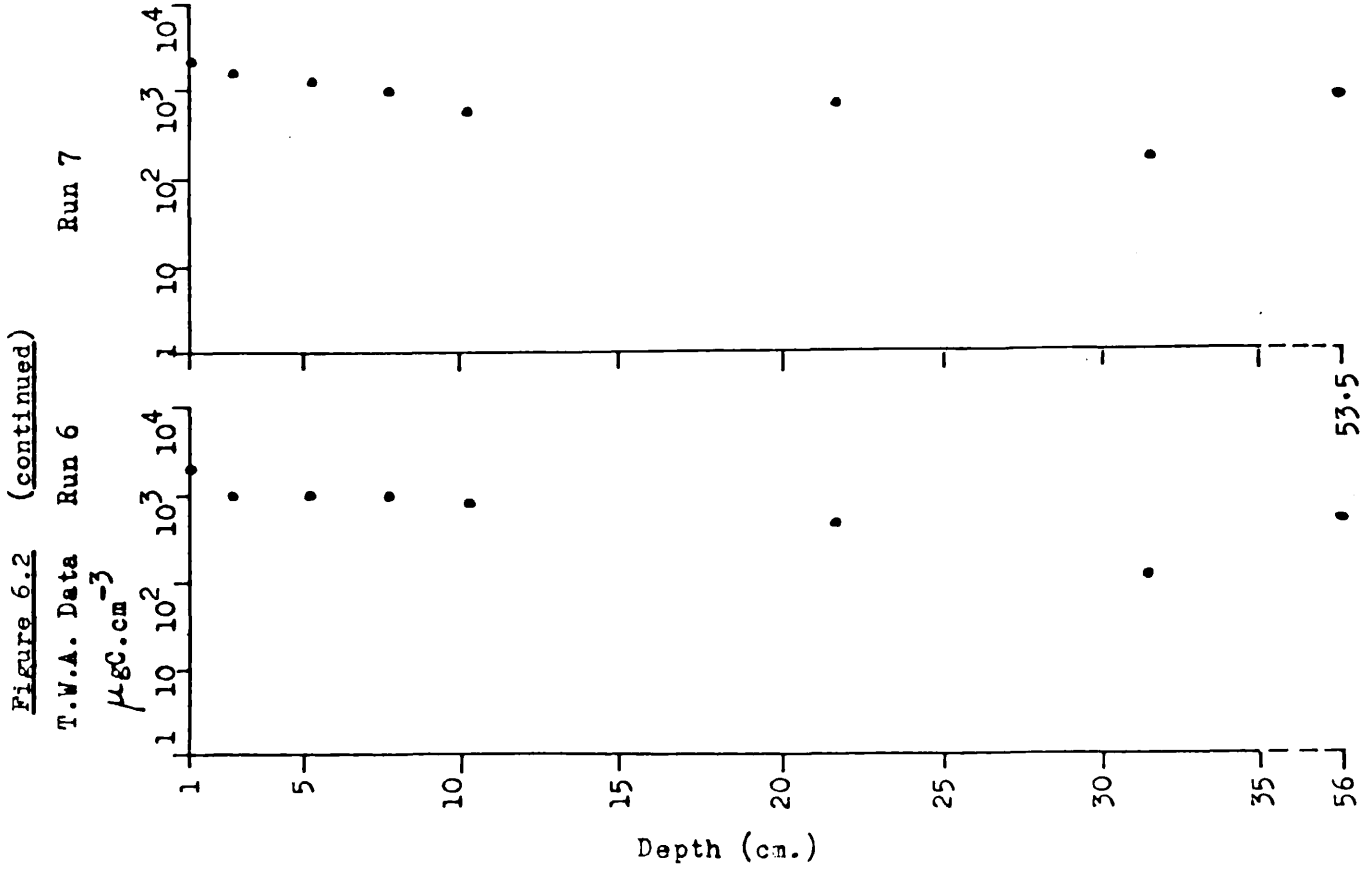


Figure 6.2 (continued)
T.W.A. Data
Run 6
Run 7
$\mu gC.cm^{-3}$
1
10
10^2
10^3
10^4
1
5
10
15
20
25
30
35
53.5
56
Depth (cm.)

bed run carbon levels in the top 1 and 2cm of sand are much higher than those in the body of the sand below. It was decided to investigate whether there was any difference in the values of carbon at 1cm, 2cm and below 2cm.

6.2.4.1 Values of carbon below 2cm

A one way analysis of variance on transformed (log x) data revealed (table 6.10.1) that there were no significant differences ($p > .05$) between the values of carbon found at different depths on the same date. The carbon data collected on the same date were then grouped and a one way analysis of variance test on transformed (log x) data revealed significant differences ($p < .05$) between values of carbon collected on different dates in the same run (table 6.10.2).

6.2.4.2 Values of carbon at 1 and 2cm

It was necessary to investigate whether the carbon data collected from 2cm depth were different from that below 2cm. A one way analysis of variance test on transformed (log x) data indicated (table 6.10.3) that on occasions (mostly after the first sampling trip) there were significant differences in carbon value ($p < .005$).

Secondly a one way analysis of variance test on transformed (log x) data revealed significant differences ($p < .001$) between carbon values determined at 1cm and below 2cm, again this was mostly after the first sampling occasion in each run (table 6.10.4).

A comparison of carbon values at 1cm and at 2cm (one way analysis of variance on transformed, log x, data) indicated that in 40% of each there were significant differences ($p < .05$) observed (table 6.10.5). It was therefore decided to treat these two depths separately.

With time there were significant differences ($p < .05$) observed in carbon values determined at 1cm depth in each run (fig. 6.10.6). However, at 2cm depth significant differences were only found in run 5 (fig. 6.10.7). The former result here agrees with the results of carbon values below 2cm depth, but the latter result contradicts this. This disagreement is probably not of much significance as all other depths indicate changes in carbon value with time.

6.2.4.3 Thames Water Authority data

Thames Water Authority carbon data were subjected to a two way

Table 6.10 One way analyses of carbon data (transformed-log x) in appendix 6.3

Sub table	Source of variation	Run no.	Age of run(days)	d.f.	F	p
6.10.1	Between depths (below 2cm)	4	12	7,16	1.296	0.5
			26	7,16	0.342	0.75
			41	7,8	0.683	0.75
		5	3	6,12	0.362	0.75
			16	6,14	0.867	0.1
			30	6,14	2.419	0.1
		7	1	12,25	1.026	0.5
			23	12,26	0.622	0.75
			37	12,26	1.035	0.5
			51	12,26	1.863	0.1
6.10.2	Between separate dates on each run, (below 2cm)	4		4,75	2.612	0.05
		5		2,58	43.835	0.001
		7		3,155	21.558	0.001
6.10.3	Between the 2cm depth and below 2cm	4	12	1,25	0.006	0.75
			26	1,25	0.172	0.75
			41	1,16	3.385	0.1
		5	3	1,20	1.157	0.5
			16	1,22	20.462	0.001
			30	1,22	39.848	0.001
		7	1	1,39	2.150	0.25
			23	1,40	6.362	0.025
			37	1,40	3.401	0.1
			51	1,40	8.454	0.01
6.10.4	Between the 1cm depth and below 2cm	4	12	1,25	10.770	0.005
			26	1,25	3.638	0.1
			41	1,16	38.916	0.001
		5	3	1,20	0.199	0.75
			16	1,22	51.265	0.001
			30	1,22	128.91	0.001
		7	1	1,39	20.743	0.001
			23	1,40	32.388	0.001
			37	1,40	37.682	0.001
			51	1,40	32.709	0.001

Table 6.10 continued.

Sub table	Source of variation	Run no.	Age of run(days)	d.f.	F	p
6.10.5	Between 1cm	4	12	1,4	31.056	0.01
	and 2cm		26	1,4	4.265	0.25
			41	1,2	1.132	0.5
		5	3	1,4	0.613	0.5
			16	1,4	4.778	0.1
			30	1,4	22.475	0.01
		7	1	1,4	9.354	0.05
			23	1.4	1.432	0.5
			37	1,4	25.106	0.01
			51	1,4	2.820	0.25
6.10.6	1cm depth,	4		2,5	18.647	0.005
	between different	5		2,6	84.038	0.001
	dates on each run	7		3,8	5.774	0.025
6.10.7	2 cm depth,	4		2,5	0.487	0.75
	between different	5		2,6	38.861	0.001
	dates on each run	7		3,8	2.153	0.25

analysis of variance test, using transformed (log x) data (table 6.11). There was found to be a significant difference ($p < .001$) between values of carbon found at difference depths and a significant difference ($p < .001$) between values of carbon found on different sampling dates. The latter result agrees with the present study. The former result, however, is contradictory, but it must be emphasised that the Thames Water Authority examined deeper depths but with no replication. This significant difference with depth in Thames Water Authority data is probably due to the fact that the sand is continually ageing and accumulating detritus, whereas sand used in this study has been freshly washed prior to being placed in the cores.

It can also be seen that with the Thames Water Authority data the value of carbon found at the bottom of the filter is very much higher than at 10 to 20cm above it. This may be due to the fact that at this point there is a change in the nature of the filter sand where it meets the underlying gravel. At this point the size in the interstices in the gravel would be much larger than those in the overlying sand and may cause a change in the velocity of water here, thus affecting the nature of settlement of inert and living matter.

6.2.5 Discussion

The degree of dispersion of carbon within sampling cores has been shown to be random, but from core to core it can be seen that the carbon is overdispersed and is tending to follow a log-normal distribution. This may be due to several factors :

1. Initial heterogeneity in the carbon distribution contained in newly washed sand placed in the bed after trenching.
2. Variation in filter performance over the area of the bed.
3. Heterogenous spatial distribution of organisms inhabiting the bed.

The first of these factors has not been investigated and so any discussion must be speculative, although some heterogeneity of sand grains between cores has been demonstrated (chapter 3.7) in sand removed from the washing bays. The second factor is a possibility and could itself be related to the heterogenous distribution of particles in the bed which would cause local differences in water velocity. The heterogenous distribution of algae, both in number and in species, on the bed surface has been demonstrated by Bellinger (1968). He related this to the position of the water inlet in the bed causing local scour-

ing and also to the direction of the prevailing wind causing algae to be carried in suspension further away from the inlet before sedimenting out.

The third factor is feasible and was investigated. The degree of overdispersion mentioned above remains the same irrespective of depth, but significant differences in the values of carbon found at different depths have been demonstrated. The vertical distribution of carbon reveals significant differences in value of carbon at 1cm, 2cm and below 2cm. However, below 2cm the carbon values appear to be fairly homogenous. This may indicate that most of the incoming carbon is deposited in the top 1cm and to a lesser extent at 2cm. Alternatively this might also indicate accumulation of organisms at this depth.

Changes in the value of carbon determined at all three depths can be seen to occur with time, emphasising the dynamic process occurring within the whole body of the filter.

Comparisons with Thames Water Authority data are difficult. This may be due to the different sampling techniques involved and the statistical validity of comparing samples with widely differing values of n, but it does tend to emphasise the ageing effect on the sand in the bed over the course of several runs compared with that of the newly washed sand used to fill the sampling cores at the start of each run.

6.3 Analysis of spatial distributions of meiofauna

6.3.1 Quadrat size and number of sampling units

Much has been written (Gray 1971, Elliott 1977, Hulings and Gray 1971, Southwood 1975) about the need to obtain an optimum quadrat size for use in sampling procedures in order that an accurate estimate of the population of a particular habitat may be obtained. In the present study the minimum diameter of the sampling apparatus was determined by a consideration of liquid flow characteristic (3.6) and the maximum size was very much related to the cost of the material from which it was constructed. Similarly the number of sampling units which could be removed at any one time for analysis was dependent on the cost of the equipment, but to a greater degree on the amount of time required to process each sample. This processing time varied very much from one sample to another, depending upon the amount of detritus and number of organisms present, both of which increased with the age of the filter run, as discussed in 4.2.1. Towards the end of

a filter run there were very high levels of carbon present in the top 1 and 2cm and high numbers of organisms present. Consequently it could take as long as 5 days to examine the organisms present in one core. The result of this was a realisation that if one set of samples must be processed (including enumeration procedures and carbon analyses) before the next set was collected, then there must be a limit on the number of samples removed at any one time.

A consideration of the 95% confidence limit became important here. An examination of the statistic 't' (Student's t-test) shows that at a probability level of p=0·05 there is not much of an improvement (i.e. decrease) in the value of t caused by increasing the number of degrees of freedom above 2. Thus if the number of sampling units is 3, the use of this statistic t (with d.f. = 2 and p = 0·05) in the calculation of the 95% confidence limits, does not cause these limits to be increased much more than if the degrees of freedom were greater than 2.

It was found that in the majority of cases, 3 sampling cores could be processed within the time intervals between sampling trips and so 3 cores were removed from the sampling bed for analysis on each sampling trip.

6.3.2 Diurnal migrations

Much evidence is offered for increased diurnal drift at night in streams (reviewed by Hynes 1970) and was also noted by Kovalak (1978) on both artificial and natural substrates. Clifford (1972) however, noted that there was no difference between day and night in the number of animals on stream beds. Although the above is not evidence of vertical migrations in stream beds, it is perhaps evidence of increased nocturnal activity. Little work has been done on the diurnal vertical migration of organisms in substrates, but what evidence there is suggests that little activity takes place. Brinkhurst (1970) found no evidence of diurnal migrations of oligochaetes in the benthos of a freshwater lake and Arlt (1973) found similar results in stream meiofauna over a 24 hour period.

No examination of diurnal vertical migrations was investigated in this study, mainly because of the intense sampling that would be necessary over a relatively short period of time. However, all cores in this study were removed between the hours of 10.00a.m. and 12.00 noon, in order to reduce any migration effects on the number of organ-

isms removed from the cores.

6.3.3 Discussion of the concept of the washed sand, contained within the sampling cores, as an artificial substrate

A consideration of the above concept is appropriate at this stage as it precedes a consideration of the migration of organisms into the cores.

It is reasonable to consider the sand used to fill the cores as an artificial substrate, as it has been subjected to various washing procedures before being used. This sand can be compared with various artificial substrates used in ecological studies. As far as colonisation by organisms of these artificial substrates is concerned they are not as successful as the natural substrate in yielding representative numbers of organisms in short periods of time. The reasons for this are complex, but are mainly related to the fact that the artificial substrate is often very different in nature from the surrounding substrate.

The fact that several authors (e.g. Boaden 1962, Gray 1966, Jansson 1967, Fenchel et al 1978) have shown substrate size preferences by organisms emphasises the need to reproduce the physical characters of the natural substrates when using artificial substrate samplers. A second factor of importance in substrate selection is the maintenance of a natural bacterial flora and associated layers of detritus (Wieser 1956, Meadows and Williams 1963, Meadows 1964, Gray 1966). So in order that the colonisation of invertebrates on an artificial substrate should be similar to that of a natural substrate, it follows that the two should be as close in nature as possible.

Time has been shown to be an important factor in reaching maximum colonisation rates of artificial substrates and it would appear that the more natural is the substrate, then the shorter is the time taken to reach stable population levels similar to those of the natural substrate. Mason et al (1973) using limestone and porcelain spheres found it took 8 weeks to reach stability, whereas the porcelain ball samplers of Roby et al (1978) took two to four weeks to reach maximum diversity and species number. The use of a natural substrate (Coleman and Hynes 1970) which had been washed and kept dry for some time before being placed into the sampling pots, demonstrated that numbers were still increasing after 28 days. However, in a situation where the only treatment was removal of organisms from a substrate by agita-

tion (Waters 1964) recolonisation to previous levels was seen to occur in 1 - 2 days.

Although the sand which was used to fill the cores has the same size characteristics (section 3.7) as that in the bed, it has been subjected to washing procedures which will have removed a considerable amount of the living organisms and detritus contained within its interstices. However, it has not been completely denuded of its biological characters by drying or cleaning in acid, so it is reasonable to suppose that it still retains some of its "attractive features" to organisms for recolonisation.

6.4 Horizontal distributions of meiofauna

6.4.1.1 Methods available for the analysis of horizontal distributions

Various methods are available for the analysis of horizontal distributions (reviews in Elliott 1973, Southwood 1975). Between one method and another there can be a great deal of variation in the degree of complexity involved in treating the data. It is obviously desirable to condense sample data so that any given population may be described by a few simple parameters which can be easily compared with those of another population. However, the computing of these parameters should not be an end in itself and it must be borne in mind that the study of distribution patterns can only be of any ecological significance if it produces insight into the biology of the organism concerned. Often it is useful to compare the frequency distribution of the data with that of a known mathematical frequency distribution, it is also possible to produce a single figure index of dispersion for the data.

At this point it is useful to discuss the various terms used to describe heterogeneity in an environment. Much use is made of the term "contagious", "aggregated" and "patchy". All of these terms have more or less the same meaning and indicate the presence of an overdispersed distribution. It is, however, unfortunate that the word "contagious" is so often used in the literature, as it does tend to suggest that the presence of one organism in a sample influences the probability of others occurring there, where biologically this may not be the case.

6.4.1.2 Indices of dispersion

Elliott (1977, modified from Green 1966) has listed the attributes which an ideal index of dispersion should possess :

1. It should provide real and continuous values over the range from maximum regularity, through randomness, to maximum contagion.
2. It should not be influenced by variation in the size of the sampling unit, the number of sampling units, the sample mean, and the total numbers in the sample.
3. It should be easy to calculate from large amounts of data.
4. It should enable differences between samples to be tested for significance.

He concludes that there is no perfect index of dispersion which fulfills all of these conditions.

Indices based on the variance to mean ratio have been shown to be good statistical tests for agreement with a poisson distribution, but are not good comparative measures of the degree of overdispersion as they can only be used when Σx, $\bar{x}$ and n have the same value in each sample. They are, however, often quoted in the literature. One such index is $S^2/\bar{x}$ (table 6.12) which varies from < 1 for a binomial distribution; through 1, indicating a poisson distribution; to > 1 for an overdispersed distribution, with increasing values of the index indicating greater overdispersion of the individuals concerned in the habitat. The probability of the distribution agreeing with a poisson distribution can be determined by calculating a χ^2 value which can be compared with tables of χ^2 at the appropriate degrees of freedom.

An index which is independent of the sample mean ($\bar{x}$) and the total numbers in the sample (Σx) is Morisita's Index I_σ (table 6.12). It is, however, a strong function of the number of sampling units (n) at both ends of its range. In this study, in the majority of cases, the number of sampling units removed on any one occasion was constant. So for this situation it would be possible to use I_σ for comparative purposes. Like the $S^2/\bar{x}$ ratio, it is possible to test whether the distribution concerned agreed with a poisson distribution, by calculating a χ^2 value.

The parameter 'b' of Taylor's Power Law (see also section 6.2) is an index of dispersion and varies continuously from 0 for a regular distribution, through 1 for a random distribution, and 2 for a log-

Table 6.11 Two way analysis of variance of carbon data in appendix 6.4

Source of variance	d.f.	F	p
Between depths, all runs	5,30	16.119	0.001
Between dates, all runs	6,30	19.356	0.001

Table 6.12 Summary of various indices of dispersion.

Index of dispersion	Values of : Max. regularity	Random	Max. contagion	Test of agreement with Poisson X^2	d.f.
$\frac{S^2}{\bar{x}}$	0	1	$\leq x$	$\frac{S^2}{\bar{x}}(n-1)$	n-1 *
Morisita's Index $I_\delta = n.\frac{\leq(x^2)-\leq x}{(\leq x)^2-\leq x}$	$1-\frac{(n-1)}{\leq x-1}$	1	n	$I_\delta(\leq x-1)+n-\leq x = \frac{S^2}{\bar{x}}(n-1)$	n-1 *
Taylors Power Law b	0	1	∞		

* departures from randomness are judged significant ($p<.05$), when X^2 is outside the appropriate 5% confidence levels for n-1 degrees of freedom.

So if calculated $X^2_{(0.975)} <$ tabulated $X^2_{(0.975)}$ the distribution is regular

if " $X^2_{(0.025)} >$ " $X^2_{(0.025)}$ " dist. is overdispersed

if tabulated $X^2_{(0.025)} >$ calculated $X^2 >$ tabulated $X^2_{(0.975)}$ the distribution is random.

normal distribution to infinity for highly overdispersed distributions. This index is independent of n, Σx and $\bar{x}$, however it does require several estimates of S^2 and $\bar{x}$ and so 'b' cannot be calculated for one sample. In this situation it is possible to calculate several estimates of S^2 and $\bar{x}$ by using values of x at the same depth from the three separate cores removed on the same sampling trip. So it would be possible to calculate a figure for 'b' and hence an index of dispersion for the organisms removed on each sampling trip.

Using these methods it is therefore possible to indicate whether a distribution is regular, random or overdispersed. If one wants more information about the degree of overdispersion, then it is necessary to compare the distributions with known mathematical frequency distributions.

6.4.1.3 Mathematical frequency distributions

Several mathematical frequency distributions which are applicable to biological distributions are listed in table 6.13, in order of increasing overdispersion. By plotting these overdispersed distributions on arithmetic graph paper in the form of a frequency histogram they can be seen to be positively skewed and the degree of skewness can be seen to increase in value with increasing overdispersion of the components involved.

The biological explanation for distributions of this type have been discussed as follows :

Thomas - Based on the assumption of randomly distributed colonies whose individual populations are slightly more overdispersed than random.

Neyman Type A - This is fairly similar to the Thomas distribution and describes the dispersion of larvae recently hatched from randomly distributed clumps of eggs. There is an arbitrary limit to clump size depending on distance crawled by the larvae.

Polya-Aeppli - Here the habitat is initially colonised at random by parent organisms all arriving simultaneously and later producing clumps of offspring. The number of individuals per cluster have a geometric distribution.

Negative Binomial (N.B.D.) - There may be several explanations offered for this distribution (Elliott 1977, Southwood 1975).

1. True contagion. The presence of one individual or event

Table 6.13 Mathematical frequency distributions.

Frequency distribution	Degree of dispersion	Shape of frequency histogram
Positive binomial	regular	normal
Poisson	random	
Thomas Neyman Type A Polya-Aeppli Negative binomial Poisson lognormal Lognormal	overdispersed with increasing overdispersion	positively skewed, increasing skewness

increases the chance that another will occur in the same unit (e.g. laying of eggs in masses).

2. Constant birth/death/immigration rates. Growth of a population with constant rates of birth and death per individual and of immigration per unit of time leads to a N.B.D. for population size.

3. Compounding of poisson and logarithmic distributions. If clumps of individuals are distributed at random and the numbers of individuals in the clumps are distributed independently in a logarithmic distribution, then a N.B.D. will result - this has been seen to occur in bacterial counts.

4. Heterogenous poisson sampling. Where the mean of a poisson distribution varies from occasion to occasion then on certain occasions a N.B.D. will result. This was shown by Ito et al (1962) where a series of counts of gall wasp on chestnut trees were distributed as a poisson for each single tree, however when the counts for all trees were combined they described a N.B.D.

Logarithmic - This model describes situations for which the N.B.D. would be underdispersed. The log-normal distribution is one which, when the individual values are transformed logarithmically, produces a normal distribution. In many cases in the literature, the N.B.D. and log-normal distribution are used synonymously.

Poisson log-normal - Described by Cassie (1962). In certain cases the action of a series of environmental factors leads to the population being distributed in a succession of poisson series, themselves being distributed according to the log-normal model.

6.4.1.4 Discussion

As discussed above, the calculation of a χ^2 value can be used to decide whether or not a distribution is regular, random or overdispersed. However, if one wants to be more precise about the degree of overdispersion the data must be compared with selected mathematical frequency distributions.

The Thomas and Neyman distributions are fairly similar and the Thomas distribution does not vary much from a poisson. At the other end of the scale the log-normal distribution is scarcely distinguish-

able from the N.B.D. (Cassie 1962) when a small number of samples are considered. The poisson-lognormal distribution is a special case which can be detected if the data does not agree well with the log-normal distribution.

It would appear, then, that the more useful distributions to consider would be the Neyman Type A, Polya-Aeppli and the N.B.D. or log-normal distributions. To do this it is possible to calculate the expected terms of the mathematical distribution in order that a χ^2 value might be calculated. However, it is well known that the efficiency of the χ^2 test falls down when the value of x in any category is less than 5 (Sokal and Rohlf 1969). In this study, for Enchytraeus buchholzi alone, out of 156 sets of samples counted, 138 had values of x of less than 5. The use of the χ^2 analysis method for goodness of fit testing is therefore probably too optimistic in this situation where n is only equal to 3 and values of x are often less than 5.

Alternative tests, for comparison with Neyman Type A, Polya-Aeppli and N.B.D. are available. The "T" and "U" tests described by Evans (1953) are based on a comparison of observed and expected moments. Values of T and U and their standard errors can be calculated for agreement with these three distributions. Although tables of probability have not been produced, it is possible to compare the statistic with its standard error in order to decide whether or not the data agrees with that of the mathematical distribution concerned.

However the use of such tests, based on analyses of moments, are really designed for situations which have very high values of n. So the use of the T and U statistics in relation to Neyman-Type A, Polya-Aeppli and N.B.D. are really not reliable in situations where the values of n are as low as 3. So tests of agreement with these three distributions were not pursued.

In conclusion, it would appear that the most useful measure of spatial dispersion, as far as this data is concerned, is 'b' of Taylor's Power Law which does indicate the degree of overdispersion concerned. In situations where b was impossible to calculate - due to less than three sets of data being available for the regression - Monisita's index of dispersion and associated χ^2 value were calculated.

6.4.2 Results

Taylor's Power Law was used to examine the relationship between variance and mean of the samples removed on each sampling trip. It was

possible, by calculating a $\bar{x}$ and S^2 value for each depth examined in the three cores removed, to produce several points on a graph which could be investigated by regression analysis. No regressions were obtained where less than three cores were removed, or where there were less than three sets of $\bar{x}$ and S^2 values calculated on each sampling trip. Tables of numbers of organisms counted in each core are given in appendices 6.5 - 6.14 and results of regression analyses are given in tables 6.14 - 6.23.

It can be seen that as far as each group is concerned, in the majority of cases the relationship between variance and mean is significant ($p < .05$), indicating that on any one date, at all depths the degree of dispersion of the organisms remains constant.

Enchytraeus worms (table 6.14)

The value of the slope 'b' in most cases is equal to 2, indicating that the organisms are following a lognormal distribution (the low value of b = 1·4 in run 2, day 29 is correlated with very low numbers on this date). However, on the first sampling dates of each run it can be seen that numbers are very much lower than later on, and on the first sampling trip of runs 3 and 5 it can be seen that the slopes of the relationships are lower (b = 1·6 and 1·3), indicating a more random distribution of individuals. The high b = 1·998 of run 7, day 1, may be confusing, as an examination of the data in appendix 6.5 again shows very low numbers present in the core, and on inspection tend to be exhibiting a random distribution.

It would appear then that at the start of a run very low numbers of organisms are present in the core and these appear to be randomly distributed. With time numbers increase and the species becomes more overdispersed, tending to conform to a lognormal distribution. An examination of the relationship between b and length of run can be seen in figure 6.3. If the two possibly confusing values of b (determined on run 2, day 29 and run 7, day 1) are ignored, it would appear that there is a general increase in the value of b up to day 15, beyond this time the value of b does not vary much. Interpreted, this result indicates an increase from a random to a lognormally distributed population within the first 15 days of a filter run being started. This could possibly be related to the rate of migration of organisms from the sand below the cores into the cores, which possibly slows down or stops after 15 days or so. Numbers, however, continue to oscillate after this time although the degree of dispersion remains more or less constant.

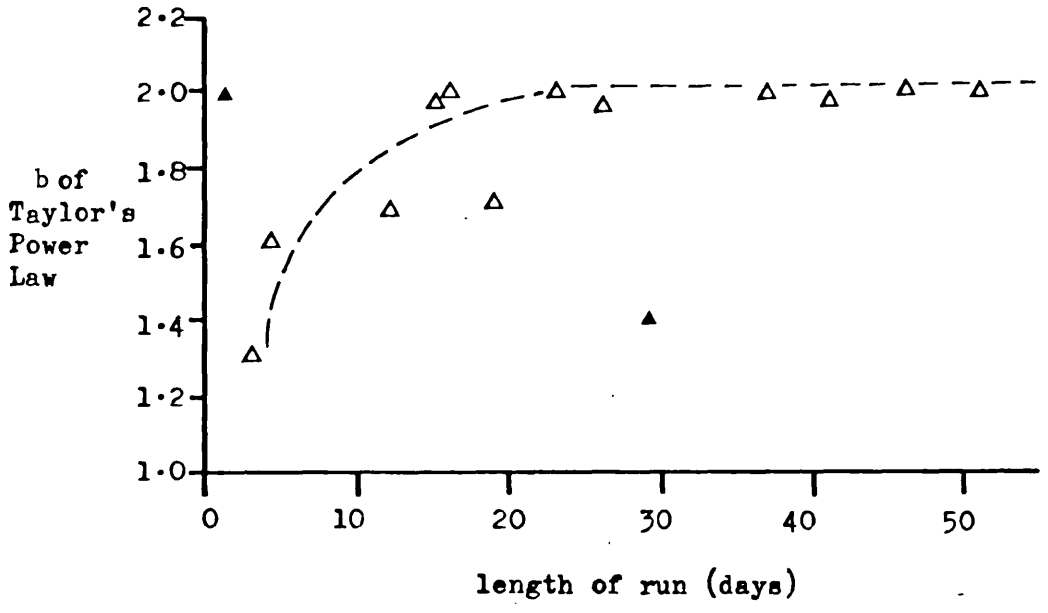
Table 6.14 Determination of "b" of Taylor's Power Law for E. buchholzi worms.

Run	Day	Abscissa	Ordinate	d.f.	F	p	b
2	1	$\log \bar{x}$	$\log S^2$				
	15	"	"	1,3	544	0.001	1.976
	29	"	"	1,3	47.8	0.01	1.435
	46	"	"	1,4	168	0.001	2.015
3	4	"	"	1,4	33.350	0.005	1.638
	19	"	"	1,3	27.000	0.025	1.747
4	12	"	"	1,8	4.888	0.1	1.690
	26	"	"	1,8	51.399	0.001	1.977
	41	"	"	1,8	5.704	0.05	1.980
5	3	"	"	1,13	10.829	0.01	1.307
	16	"	"	1,13	52.122	0.001	2.002
	30	"	"				
7	1	"	"	1,5	9170728	0.001	1.998
	23	"	"	1,13	180	0.001	2.023
	37	"	"	1,13	323	0.001	2.078
	51	"	"	1,13	552	0.001	2.064

Table 6.15 Determination of "b" of Taylor's Power Law for E. buchholzi cocoons with eggs.

Run	Day	Abscissa	Ordinate	d.f.	F	p	b
2	1	$\log \bar{x}$	$\log S^2$	1,4	80.555	0.001	1.893
	15	"	"	1,4	30.102	0.01	2.041
	29	"	"				
	46	"	"	1,4	6.947	0.1	1.840
3	4	"	"	1,3	13.970	0.05	2.295
	19	"	"	1,4	5.356	0.1	1.436
4	12	"	"	1,8	4.537	0.1	1.344
	26	"	"	1,8	81.409	0.001	1.879
	41	"	"	1,5	9.800	0.05	1.503
5	3	"	"				
	16	"	"	1,11	132.55	0.001	1.812
	30	"	"				
7	1	"	"				
	23	"	"	1,7	548	0.001	1.917
	37	"	"	1,13	18.877	0.001	1.243
	51	"	"	1,13	188	0.001	1.831

Figure 6.3 E. BUCHHOLZI RELATIONSHIP OF b OF TAYLOR'S POWER LAW AND LENGTH OF RUN



▲ For explanation of these figures see text

Line drawn by eye

Enchytraeus cocoons (tables 6.15 and 6.16)

An examination of the data in appendix 6.6 reveals that there can be quite high numbers of both full and empty cocoons in the sand cores at the start of a filter run. It is unlikely that the full cocoons have been laid by the very few numbers of worms present at that time. What is more likely is that these cocoons managed to survive the sand washing processes and storage in the outdoor bays before the sand was collected and placed in the cores prior to the start of each run.

The distribution of full cocoons seems to vary from random (b = 1·243) to overdispersed (b = 2·295) although in the majority of cases (8 out of 12) they do seem to be approaching a lognormal distribution. Empty cocoons seem to be similarly distributed with some dates showing a more random (b = 1·229) distribution, but with 8 out of 13 cases showing more overdispersed distributions. These distributions do not seem to be following the same sort of pattern which the worms are exhibiting, i.e. increasing overdispersion from random to lognormal with increasing age of the bed. Several factors are important here, the proportion of sexually mature individuals in the population, length of development time of the eggs and hatching success. These interrelating factors would tend to complicate the picture of spatial dispersion of the cocoons.

Aelosoma (table 6.17)

In all cases the spatial distribution of Aelosoma is overdispersed and is tending towards a lognormal distribution with b ranging between 1·502 and 1·893.

Pristina idrensis (table 6.18)

Occurrence of P. idrensis is limited to run 4 and the start of run 5; it would appear that at the start of run 5 the distribution is approaching randomness (b = 1·375) whilst on run 4, day 26, the distribution is more overdispersed (b = 1·894). The high value of b (1·999) on run 4, day 12, is misleading, very few numbers are present here and on inspection of appendix 6.8 it would appear that the data is not overdispersed.

Nematodes (table 6.19)

Nematodes are more abundant in runs 4 and 5, the distributions

Table 6.16 Determination of "b" of Taylor's Power Law for E. buchholzi empty cocoons

Run	Day	Abscissa	Ordinate	d.f.	F	p	b
2	1	$\log \bar{x}$	$\log S^2$	1,4	8.462	0.05	1.573
	15	"	"	1,4	16.454	0.025	1.771
	29	"	"	1,3	50.652	0.01	1.783
	46	"	"	1,4	17.339	0.025	1.918
3	4	"	"	1,3	1.884	0.5	1.329
	19	"	"	1,3	19.608	0.025	1.472
4	12	"	"				
	26	"	"	1,8	113	0.001	2.188
	41	"	"	1,8	15.506	0.005	2.685
5	3	"	"				
	16	"	"	1,8	11.210	0.025	1.229
	30	"	"				
7	1	"	"				
	23	"	"	1,4	$2(10)^{12}$	0.001	1.997
	37	"	"	1,3	2.400	0.25	1.331
	51	"	"	1,3	60.630	0.005	1.516

Table 6.17 Determinations of "b" of Taylor's Power Law for A. hemprichi.

Run	Day	Abscissa	Ordinate	d.f.	F	p	b
2	1	$\log \bar{x}$	$\log S^2$				
	15	"	"	1,4	518	0.001	1.867
	29	"	"				
	46	"	"	1,2	178.771	0.01	1.708
3	4	"	"	1,4	22.182	0.01	1.502
	19	"	"	1,3	39.667	0.01	1.723
4	12	"	"	1,7	160	0.001	1.804
	26	"	"	1,8	199	0.001	1.893
	41	"	"	1,2	12.070	0.1	1.669
5	3	"	"	1,10	61.579	0.001	1.84
	16	"	"				
	30	"	"				
7	1	"	"				
	23	"	"				
	37	"	"				
	51	"	"				

Table 6.18 Determinations of "b" of Taylor's Power Law for P. idrensis.

Run	Day	Abscissa	Ordinate	d.f.	F	p	b
2	1	$\log \bar{x}$	$\log S^2$				
	15	"	"				
	29	"	"				
	46	"	"				
3	4	"	"				
	19	"	"				
4	12	"	"	1,3	6924068	0.001	1.999
	26	"	"	1,5	60.639	0.001	1.894
	41	"	"				
5	3	"	"	1,12	7.651	0.025	1.375
	16	"	"				
	30	"	"				
7	1	"	"				
	23	"	"				
	37	"	"				
	51	"	"				

Table 6.19 Determinations of "b" of Taylor's Power Law for the nematodes.

Run	Day	Abscissa	Ordinate	d.f.	F	p	b
2	1	$\log \bar{x}$	$\log S^2$				
	15	"	"				
	29	"	"	1,4	1051198	0.001	1.989
	46	"	"	1,2	137	0.01	1.929
3	4	"	"	1,3	21.073	0.025	1.650
	19	"	"	1,2	59.348	0.025	1.551
4	12	"	"	1,6	84.8	0.001	2.156
	26	"	"	1,8	82	0.001	1.884
	41	"	"	1,8	30.695	0.001	2.494
5	3	"	"	1,11	9.376	0.025	1.196
	16	"	"	1,9	76	0.001	1.714
	30	"	"				
7	1	"	"				
	23	"	"				
	37	"	"				
	51	"	"	1,3	3.737	0.25	1.463

appear to be generally overdispersed with b varying from 1·551 to 2·474, but with a random distribution being demonstrated at the start of run 5 (b = 1·196), and a less overdispersed distribution at the end of run 7 (b = 1·463). Again, no trends in the degree of distribution can be seen with increasing age of the bed.

Chironomid larvae (table 6.20)

The chironomid larvae do not often penetrate into the depth of the filter and are usually restricted to the top 1 and 2cm of sand and because of this it was only possible to calculate a value for b on three separate occasions. On each date it can be seen that the value of b lies around 1·6, indicating a more overdispersed distribution than random, however the data does not tend to follow a lognormal distribution. To supplement this information Morisita's index of dispersion and associated χ^2 value were calculated for each depth in the sand on a number of dates (table 6.21). It can be seen that in the majority of cases the distributions are overdispersed, although on run 5, day 16, with greater penetration of individuals into the sand it can be seen that the distribution becomes random.

Harpacticoids (table 6.22)

These were mainly present at the end of run 2 and during run 4. From table 6.22 it can be seen that, apart from run 4, day 26, in which a lognormal distribution is indicated, the distributions appear to be random with b ranging from 1·168 to 1·397.

Flatworms (table 6.23)

Due to the low numbers of individuals present values of b for total flatworms were calculated and it can be seen that they show a lognormal or more overdispersed distribution.

6.4.3 Summary

In conclusion it may be said that all groups, apart from the harpacticoids, show overdispersed distributions with most tending towards the lognormal. When numbers of individuals are low they may show a more random distribution. Enchytraeus buchholzi worms is the only species with sufficient data to show change of the value of b with time, indicating a random distribution at the start of run and a lognormal distribution after 15 or so days. This may suggest that after

Table 6.20 Determinations of "b" of Taylor's Power Law for the chironomids.

Run	Day	Abscissa	Ordinate	d.f.	F	p	b
2	1	log $\bar{x}$	log S^2				
	15	"	"				
	29	"	"				
	46	"	"	1,2	125.601	0.001	1.592
3	4	"	"				
	19	"	"	1,3	576.171	0.001	1.659
4	12	"	"				
	26	"	"				
	41	"	"				
5	3	"	"				
	16	"	"	1,8	163.6	0.001	1.656
	30	"	"				
7	1	"	"				
	23	"	"				
	37	"	"				
	51	"	"				

Table 6.21 Values of Morisita's Index of Dispersion for the chironomid larvae, selected data from appendix 6.12

Run	Day	Depth cm	I_δ	X^2	Dispersion
2	15	1	1.571	10.000	overdispersed
	29	1	2.576	65.024	"
	46	1	1.248	73.621	"
3	4	1	1.143	17.582	"
	19	1	1.157	575.574	"
		2	2.047	244.798	"
4	26	1	2.315	32.250	"
5	16	1	1.700	173.439	"
		2	1.105	5.895	random
		3	1.000	2.000	"
		4	1.800	5.200	"
		5	1.000	2.000	"

In all cases n=3, d.f.=2

From tables of X^2, the distribution is regular if $p > 0.975$ and $X^2 < 0.051$, random if $0.025 < p < 0.975$ and $0.051 < X^2 < 7.378$, overdispersed if $p < 0.025$ and $X^2 > 7.378$

Table 6.22 Determinations of "b" of Taylor's Power Law for the harpacticoids.

Run	Day	Abscissa	Ordinate	d.f.	F	p	b
2	1	$\log \bar{x}$	$\log S^2$				
	15	"	"				
	29	"	"				
	46	"	"	1,3	19.688	0.025	1.168
3	4	"	"				
	19	"	"				
4	12	"	"	1,8	4.884	0.1	1.256
	26	"	"	1,6	9.597	0.025	2.094
	41	"	"	1,7	15.222	0.01	1.397
5	3	"	"				
	16	"	"				
	30	"	"				
7	1	"	"				
	23	"	"				
	37	"	"				
	51	"	"				

Table 6.23 Determinations of "b" of Taylor's Power Law for the flatworms.

Run	Day	Abscissa	Ordinate	d.f.	F	p	b
2	1	$\log \bar{x}$	$\log S^2$				
	15	"	"				
	29	"	"				
	46	"	"				
3	4	"	"				
	19	"	"				
4	12	"	"				
	26	"	"	1,7	25.508	0.005	2.441
	41	"	"				
5	3	"	"				
	16	"	"	1,7	67.005	0.001	2.111
	30	"	"				
7	1	"	"				
	23	"	"				
	37	"	"				
	51	"	"				

15 days there is some alteration in the rate of migration of individuals into the cores from the sand beneath, possibly indicating that at this stage the organisms have recolonised the sand as a response to changes in environmental conditions caused when the filter was put back into operation.

Results from section 6.2 demonstrated the overdispersed distribution of detritus in the cores, a distribution which tended towards the lognormal. As several of the meiofaunal groups also showed a tendency towards a lognormal distribution, the possibility arises that the horizontal distribution of the meiofauna may be dependent on the distribution of the detritus.

This possible relationship was investigated in data available from runs 4, 5 and 7. To remove any effect which the vertical distribution of carbon may have on species abundance, total numbers for each core were determined, by integration. These values were then corrected to give a mean figure for 1cm depth ($40cm^3$) for each core, this was necessary as the depths examined in the three runs differed. Mean carbon values ($\mu gC.cm^{-3}$) were similarly calculated, the resultant data can be seen in appendix 6.15.

The relationships between mean carbon values present in each core and the various meiofaunal groups were investigated by regression analysis (table 6.24). It can be seen that only in two cases (i.e. Enchytraeus worms and flatworms other than Catenula) was the relationship significant ($p < .05$). This relationship may be obscured by the fact that all of the groups examined show some seasonal periodicity, a factor which can cause variations in abundance which may not be correlated with total detritus present.

6.4.4. Discussion

There is a considerable quantity of literature available showing that populations of many meiofaunal species are distributed in an overdispersed fashion. This is shown in many types of environment including slow sand filters used for artificial recharge (Rittersbusch 1974), marine beaches (Gray and Reiger 1971, Rosenberg 1974, Cox 1976, Gerlach 1977, Lee et al 1977), fresh water streams (Egglishaw 1964, 1969, Ulfstrand 1967, Paterson and Fernando 1971, Ladle 1971, Arlt 1973, Fahy 1975, Friberg et al 1977, Rabeni and Minshall 1977 and Minshall and Minshall 1977), the benthos of freshwater lakes (Alley and Anderson 1968, Brinkhurst et al 1969, Darlington 1977). Terrestrial en-

Table 6.24 Regression analysis of data in appendix 6.15

Abscissa	Ordinate	d.f.	F	p	b	R.M.S.
log carbon	log E. buchholzi	1,24	5.645	0.05	1.078	0.299
"	log E. buchholzi cocoons(full)	1,20	0.058	0.75	0.124	0.338
"	log A. hemprichi	1,12	0.214	0.75	0.322	0.547
"	log P. idrensis	1,8	3.595	0.1	1.373	0.279
"	log chironomid larvae	1,8	0.596	0.5	0.875	0.345
"	log harpacticoids	1,9	2.362	0.25	0.678	0.202
"	log Catenula	1,6	0.836	0.5	0.937	0.469
"	log other flatworms	1,13	5.733	0.05	1.816	0.295
"	log nematodes	1,18	2.783	0.25	1.290	0.756

vironments also demonstrate overdispersed distributions, particularly with regard to enchytraeid worms (Nielsen 1954, O'Connor 1957, Peachey 1962, 1963 and Abrahamsen and Strand 1970). There is, however, some evidence to contradict these findings, e.g. McIntyre (1973) who found good correlations between the numbers of organisms removed from replicate cores taken from the sea bed. Milbrink et al (1974a and b) found chironomid larvae to be randomly distributed on the surface of a freshwater lake, although they also found that the distribution of tubificids agreed with that of a negative binomial distribution.

On a wider basis, Alexsson et al (1975) found that by fitting a number of theoretical mathematical frequency distributions to ten empirically found distributions of animals in freshwater, marine and terrestrial environments, in nearly all cases the negative binomial distribution gave the best fit to the empirical data.

Much evidence is available (Boaden 1962, Swedmark 1964, Jansson 1967, Husmann 1968, de March 1976 and McLachlan et al 1977) concerning the influence of substrate particle size in the colonisation of habitats by organisms. In the slow sand filter situation there has been shown (section 3.7) to be no discontinuities in the size grading of the sand particles in both a horizontal and vertical direction in the filter. It seems reasonable, therefore, to presume that, although the overall size characteristics of the sand grains will determine the species which can inhabit a slow sand filter, any spatial heterogeneity in the numbers of organisms found in the filter will not be a function of particle size.

Alternative explanations for these overdispersed distributions have been considered, Gerlach (1977) found experimentally that overdispersed distributions of marine meiofaunal nematodes was due to the "attractiveness" of the decaying organisms causing mobile nematodes to migrate to centres where the decaying organisms were situated. He surmises that the attractive substances probably do not emanate from the decaying organisms themselves, but from the bacteria and flagellates associated with the initial stages of decay. Lee et al (1977), Ulfstrand (1967), Alley and Anderson (1968), Fahy (1975) and Egglishaw (1964) all found that the spatial heterogeneity of organisms in their environments was significantly correlated with the patchy distribution of food material in the environment. Minshall and Minshall (1977) attributed variation in the numbers of organisms on experimental trays in streams to the particle size of the substrate and also to the level of detritus in each tray, with higher numbers found where more detritus

was present. Rabeni and Minshall (1977) concluded that, all things being equal, if stream organisms were offered two habitats which vary in particle size but have equal amounts of detritus present, then there was no preference shown for either substrate, thus emphasising the influence of food distribution in the spatial distribution of the organisms concerned.

The lack of an overall significant relationship, in this study, between species abundance and total carbon present in the environment, demonstrated in these results, may indicate that the distribution of species is related to some unmeasured component of the detritus, for example the bacteria, protozoa or algae. This bacterial component, however, is very variable depending upon the abundance of detritivorous organisms and also the ageing of the inert detrital material itself.

There is much evidence (Newell 1965, Brinkhurst and Chua 1969, Fenchel 1970 and 1972, Hargrave 1970, Wavre and Brinkhurst 1971, Calow 1974) to show that it is the bacterial component of the detrital assembly which provides nutrition for detritivorous organisms. Several organisms have been shown (Kajak and Warda 1965, Coler et al 1967, Hargrave 1970 and Calow 1974) to demonstrate a preference for certain bacterial components of the detritus. It is difficult to see how organisms could select certain bacteria when faced with a mixture of assorted species within a detrital aggregate. It is more likely that selection would be related to the dimensions of the food particles and the feeding mechanisms concerned.

This situation is further complicated by the fact that some organisms have been shown to demonstrate selective digestion of consumed bacteria which is possibly related to the bacterial type and the enzyme system of the organisms concerned (Brinkhurst and Chua 1969, Wavre 1970, Wavre and Brinkhurst 1970, Whitley and Seng 1976).

A further factor to consider is that the abundance of various bacterial groups has been shown to vary within a detrital assembly, depending upon the state of decomposition of the detrital material present (Rodina 1966, Olah 1972, Fenchel and Harrison 1976).

Another view is suggested by Rosenberg (1974) who found that high densities of organisms were often correlated with aggregation in the environment. This was taken further by Taylor and Taylor (1977) who proposed that, as the relationship between spatial variance and population density is a power function averaging more than 1 ($1{\cdot}48 \pm 0{\cdot}39$,

for a range of species), spatial behaviour must change as density changes. The effect being to produce more extremes of concentration and isolation of individuals at high densities than could be forecast from low densities by simple proportions. They regard this as convincing evidence that spatial behaviour is density dependent.

In a review, Southwood (1975) observed that the dispersion of the initial insect invaders of a crop is often random, becoming more heterogenous as the organisms reproduce. So, changes in the densities of an insect often leads to apparent changes in the distribution. When the population is very sparse, the chance of individuals occurring in a sampling unit is so low that the distribution is apparently random. Similarly, Friberg et al (1977), who found lognormal distributions in the majority of species in south Swedish streams, considered that species with less overdispersed distributions are characteristic of those coming from perturbed streams where less time has been available for recolonisation. So he suggested that lognormal distributions are the norm with less dispersed distributions found in situations where organisms are recolonising disturbed areas.

On the other hand, however, two authors Berthet and Gerard (1965), who found that 13 species of soil mites examined followed negative binomial distributions, also found that as the density of the individuals falls there was an increase in heterogeneity of the distribution. This they interpreted as the animals probably being clumped in the most favourable sites. This situation, however, is caused by some environmental change and should not be compared with that found in newly recolonised areas.

On the basis of the available evidence it would seem that in the absence of spatial discontinuities in substrate particle size, then the distribution of detrital and other food particles very often influences the distribution of organisms present. Due to various factors the distribution of food particles is often patchy with the result that the organisms themselves are overdispersed. It would appear that in a perturbed situation, for example where the substrate has been disturbed and the organisms removed, then the distributions of organisms recolonising these areas is often random, a phenomenon related to the low densities of organisms encountered at this stage. A parallel may be drawn here with the recolonisation of the sampling cores by E. buchholzi, where the distribution of the initial colonisers is more or less random, corresponding with low densities at this stage. With time, and as con-

ditions in the filter become more stable, the distribution of the organisms can be seen to approach that of the lognormal. (The importance of recolonisation of an area from within the substratum was shown by Williams and Hynes (1976) who calculated that migration from this direction contributed to 18% of the total movements into a disturbed area of stream bed). This situation is not quite so apparent with other species, the reason for this may be that whereas E. buchholzi occurs at relatively high densities throughout the year, other species are more seasonal in their occurrence in the filter, a situation which leads to fewer statistical analyses being possible.

The horizontal distributions of the organisms have been shown not to correlate significantly with the horizontal distribution of the carbon in the filters. This underlines how the food - species abundance relationship is by no means a simple one, but is most likely dependent upon the assimilatable components of the detrital assembly which were not investigated in this study.

6.5 Vertical distributions

The numbers of organisms found in each $40cm^3$ section examined are plotted on a logarithmic scale against depth (arithmetic scale) in the core (figures 6.4 - 6.12). In the majority of cases three values are plotted at each depth, corresponding to the three cores removed on any one sampling trip. The logarithmic scale was used in order to reduce the horizontal variability in abundance observed in the majority of species (section 6.4) which tended to show a lognormal distribution. A consequence of this use of the logarithmic scale is that it does tend to mask some discontinuities in the depth distribution of the organisms, more significance should therefore be given to any observable trends in vertical distribution.

6.5.1 Results

First sampling trip of each filter run

It can be seen that as far as all groups are concerned there is a considerable variation in the number of organisms present in the cores at this time. As little time has been available for colonisation of the cores from below in the filter, the abundance of these organisms represents what was present in the sand when it was removed from the sand bays. The variation in abundance of organisms could be due to many factors including the efficiency of the washing procedures to which

the sand is subject after removal from the cleaned beds. It will also be related to the ability of the organisms to survive storage procedures, depending upon temperature in the piles of stored sand and length of time for which it is stored.

Enchytraeus worms (figure 6.4)

The high variability in abundance in runs 2, 3 and 4 makes it difficult to see any trends in vertical distribution. In runs 5 and 7 an avoidance of the top 1cm of the core can be clearly seen. Below this depth numbers increase down to 10cm (run 5) and 5cm (run 7) and below these depths there seems to be little change in abundance.

Enchytraeus cocoons (figure 6.5)

The vertical distribution of cocoons does not seem to correspond with that of the worms. It can be seen that in the majority of cases in runs 2 and 3, cocoons are more abundant in the top 1cm, decreasing in abundance below this. This could suggest that cocoons are laid at a time of day or night when the worms are not sampled. In runs 4, 5 and 7 this vertical distribution is not so clear cut.

Aelosoma, P. idrensis, P. foreli (figures 6.6, 6.7, 6.8)

These species do not seem to show any trends in vertical distribution.

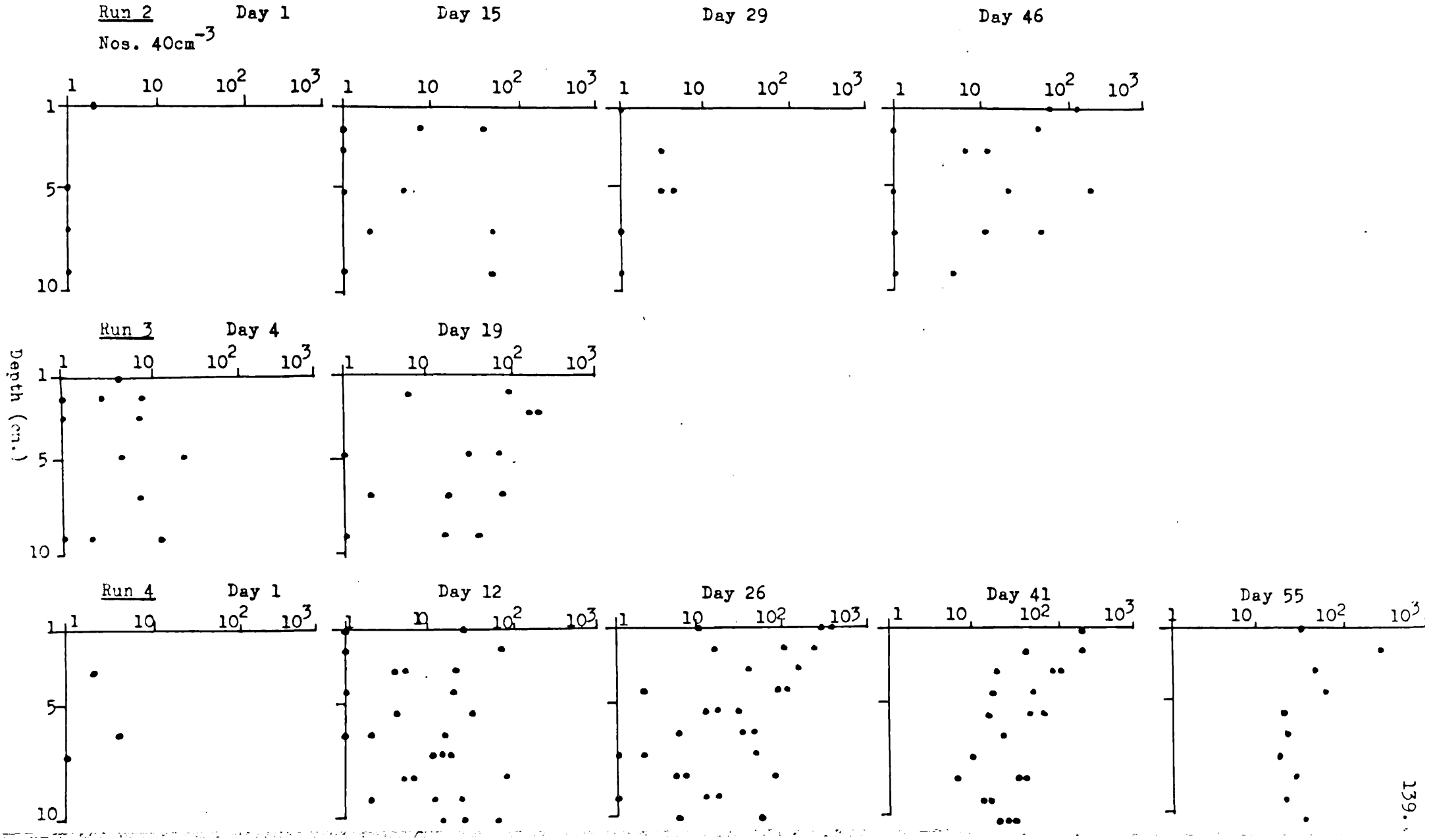
Nematodes (figure 6.9)

During run 4 the nematodes were at their most abundant and demonstrated highest numbers in the top 1cm, decreasing in abundance with depth. On other occasions nematodes were much less abundant and did not show any discernible trends with depth.

Chironomid larvae (figure 6.10)

These were most abundant in the top 1cm of sand and decreased markedly in numbers below this depth. Rarely did they penetrate below 5cm although in run 5 quite a high number (34 inds.40cm^{-3}) were counted as far down as 12cm. This vertical distribution is most likely correlated with the chironomids' life cycle, they are not interstitial organisms and are more truly benthic in habit, here colonising the sand/water interface layer which is rich in detrital deposits.

Figure 6.4 DEPTH PROFILES OF E. BUCHHOLZI



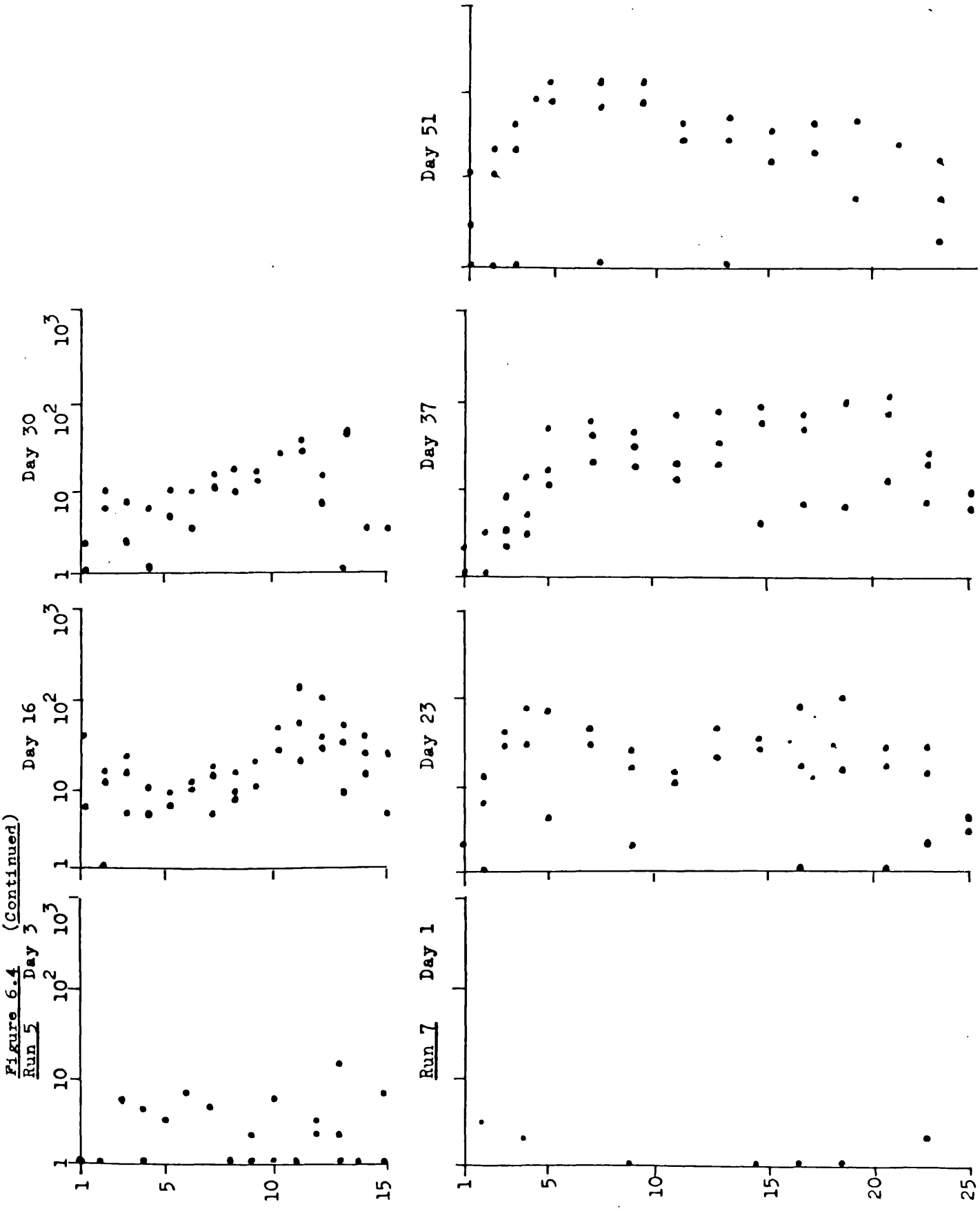
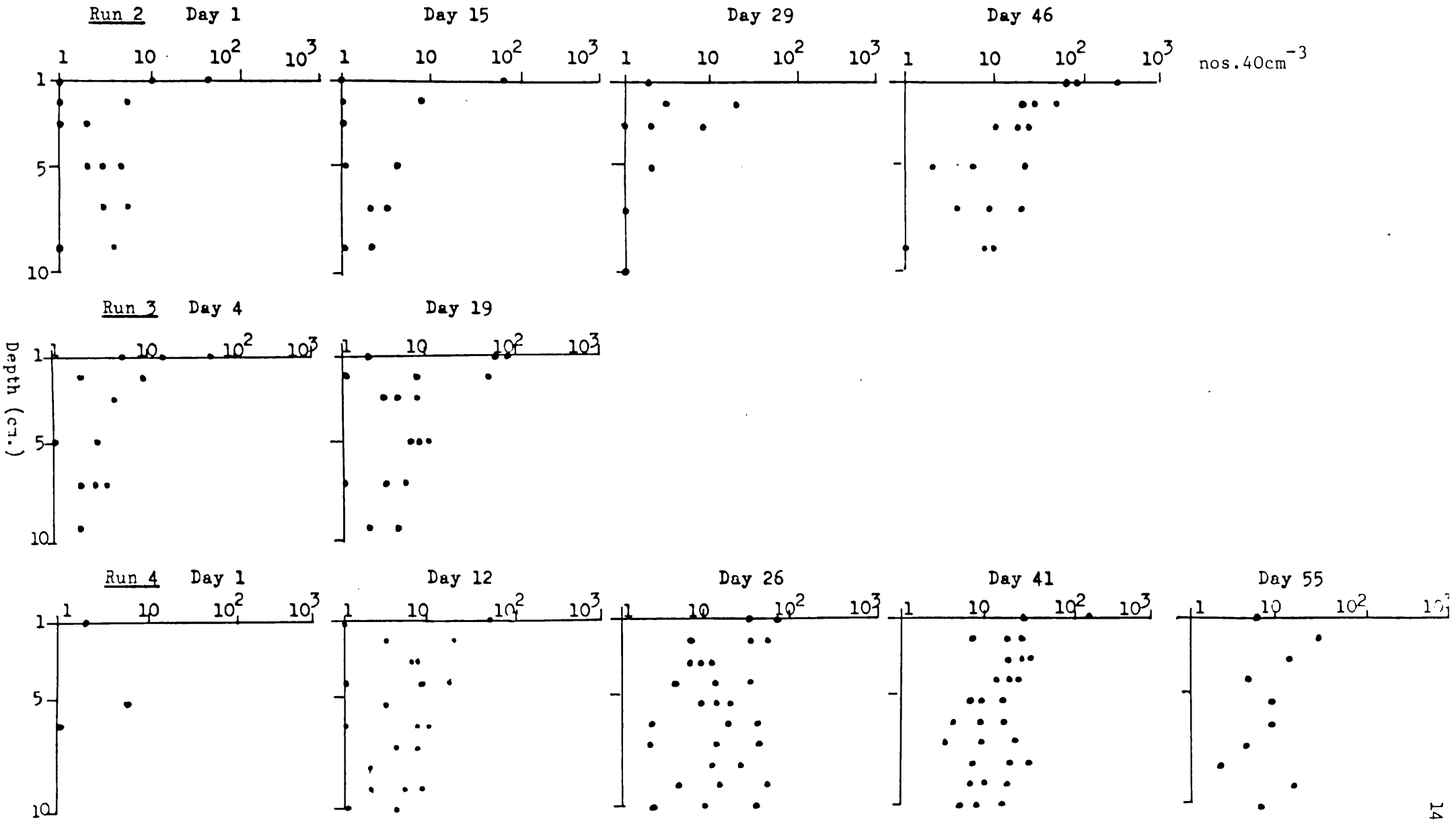


Figure 6.4 (Continued)

Figure 6.5 DEPTH PROFILE OF E. BUCHHOLZI COCOONS (FULL AND EMPTY)



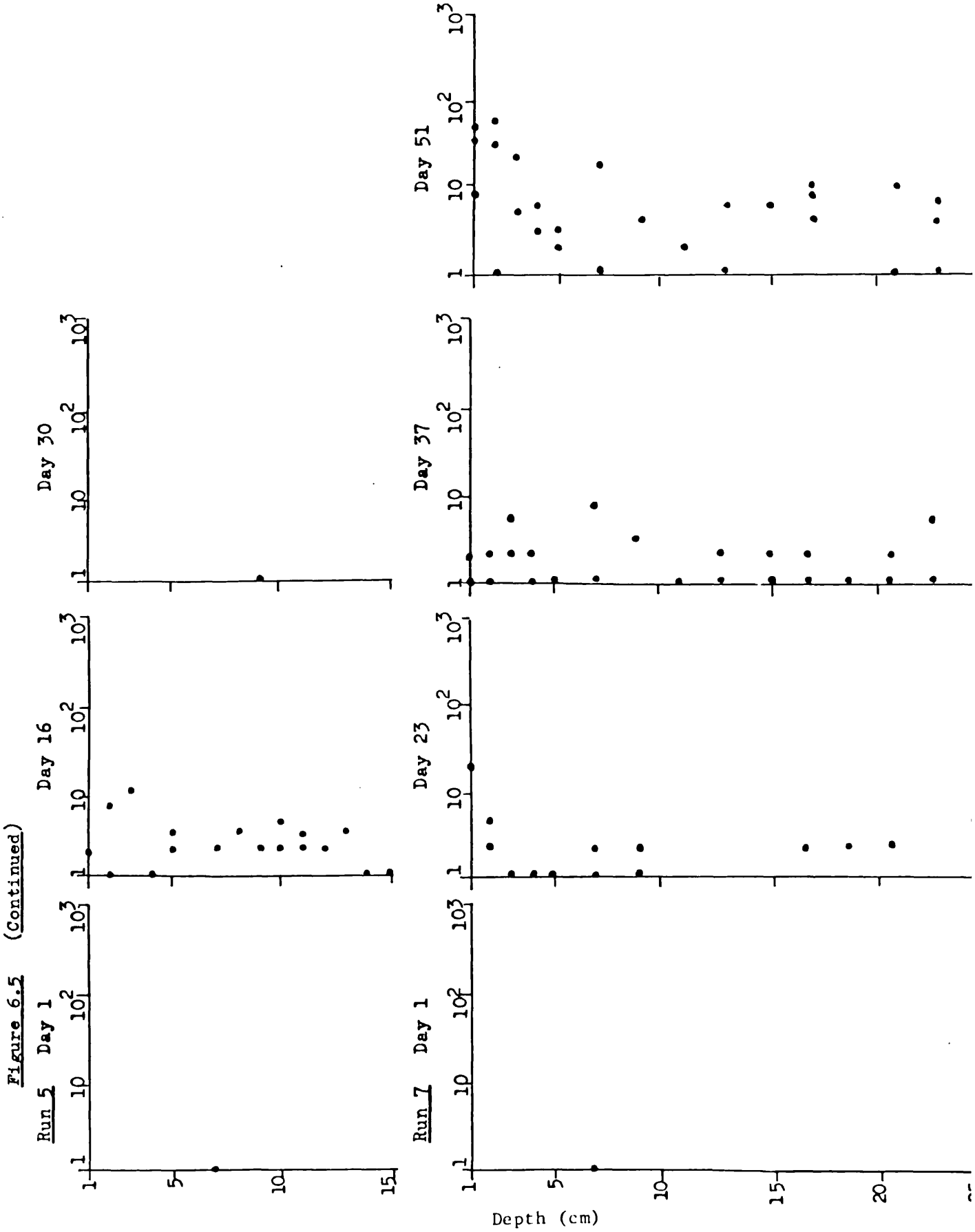
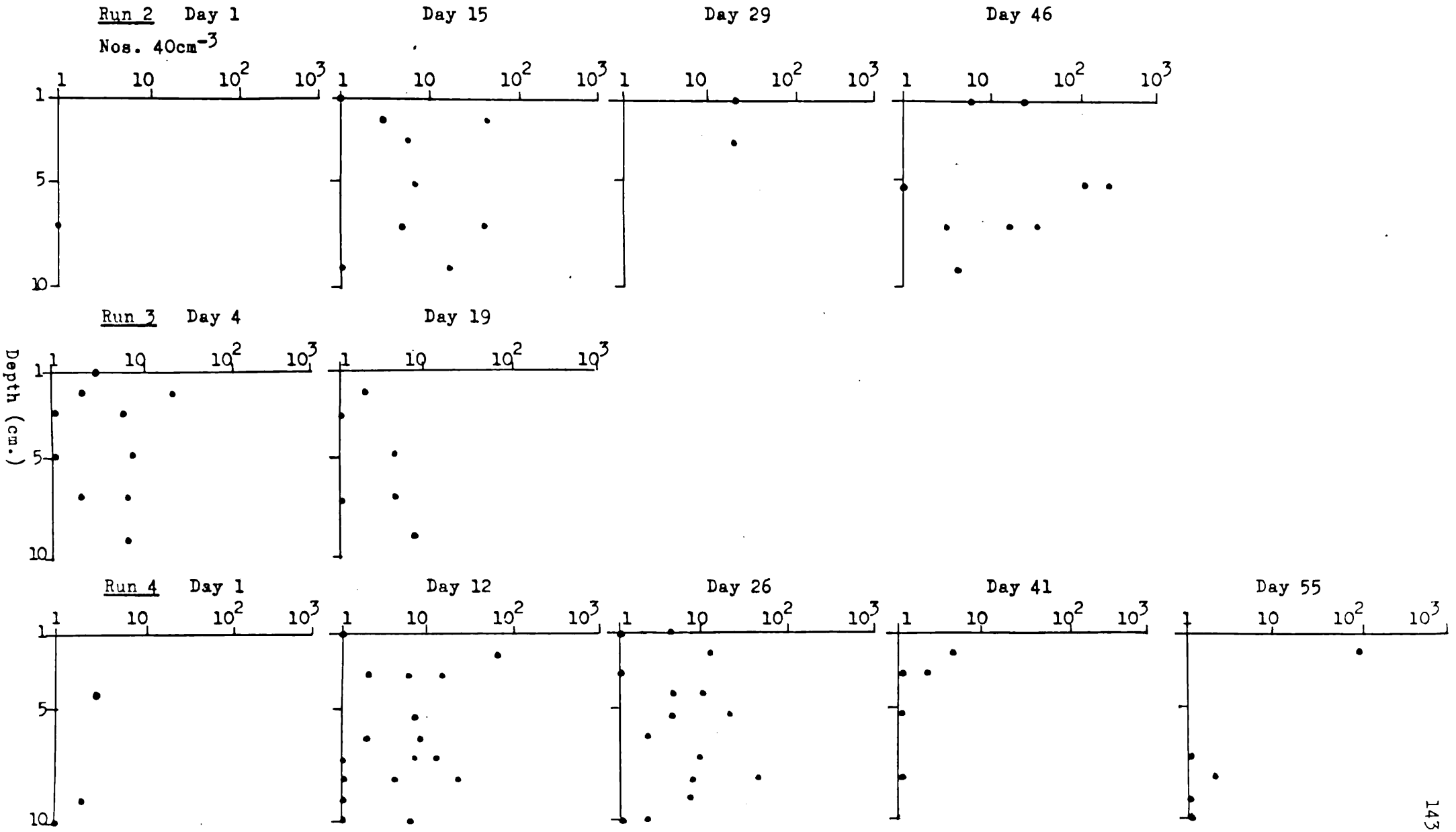


Figure 6.5 (Continued)

Figure 6.6 DEPTH PROFILES OF A. HEMPRICHI



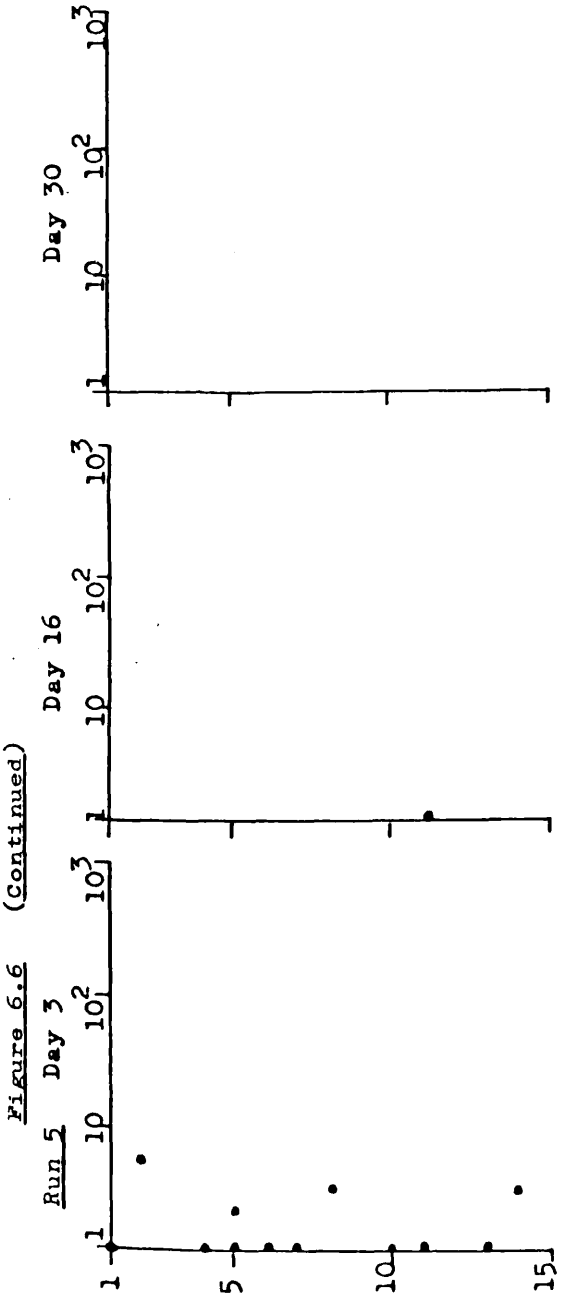


Figure 6.6 (Continued)

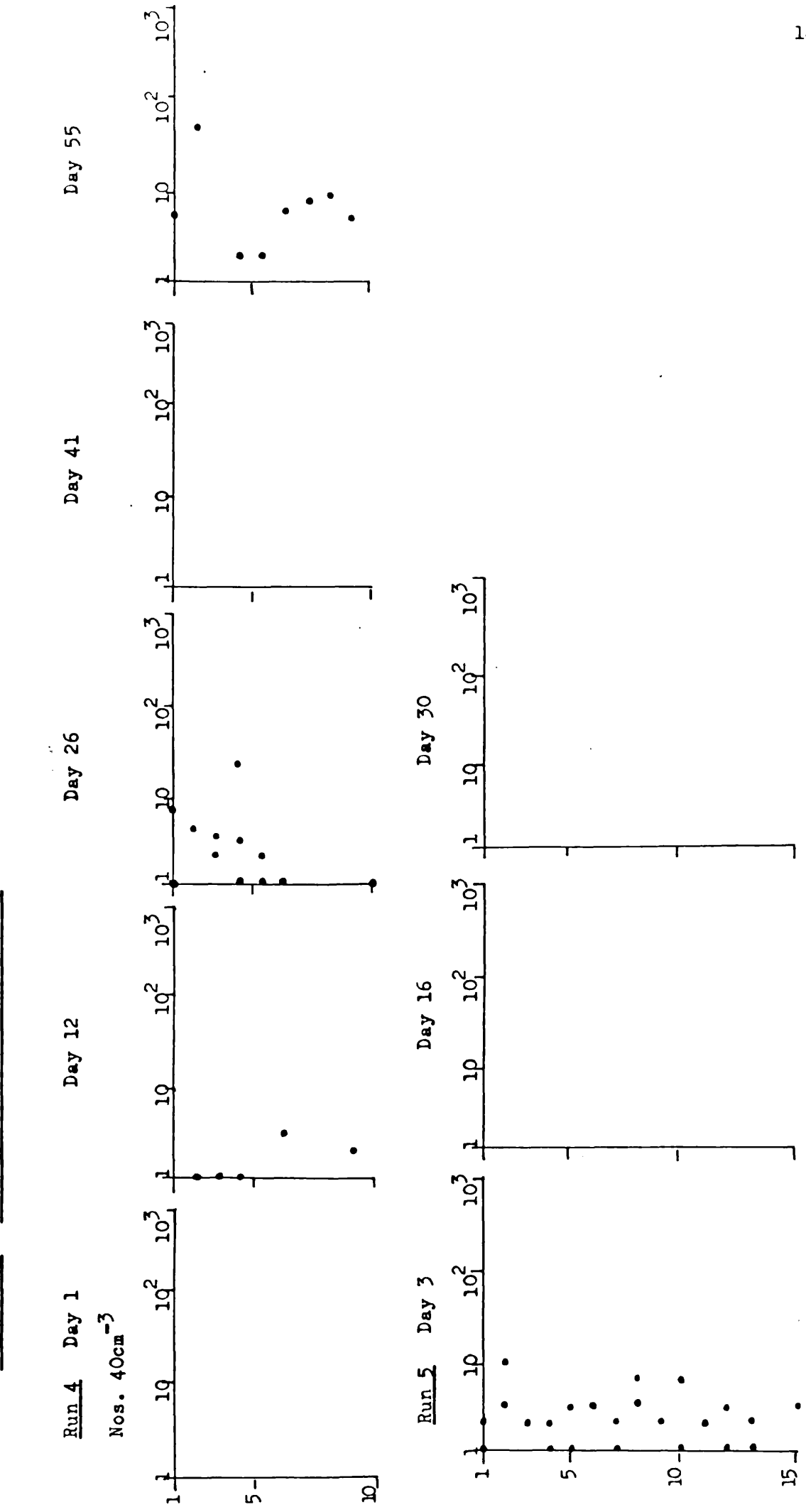


Figure 6.7 DEPTH PROFILES OF P. IDRENSIS

Figure 6.8 DEPTH PROFILES OF P. FORELI

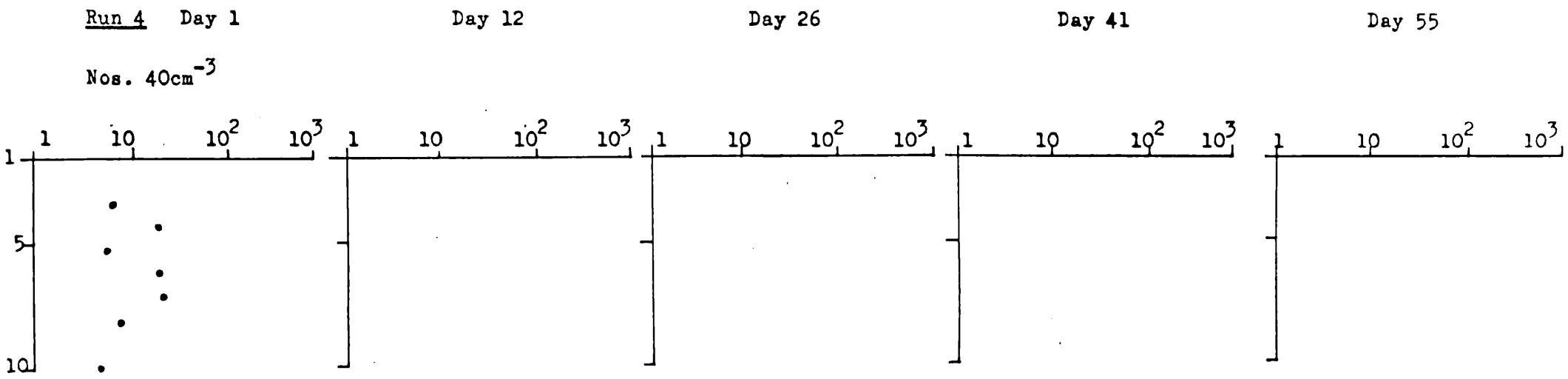
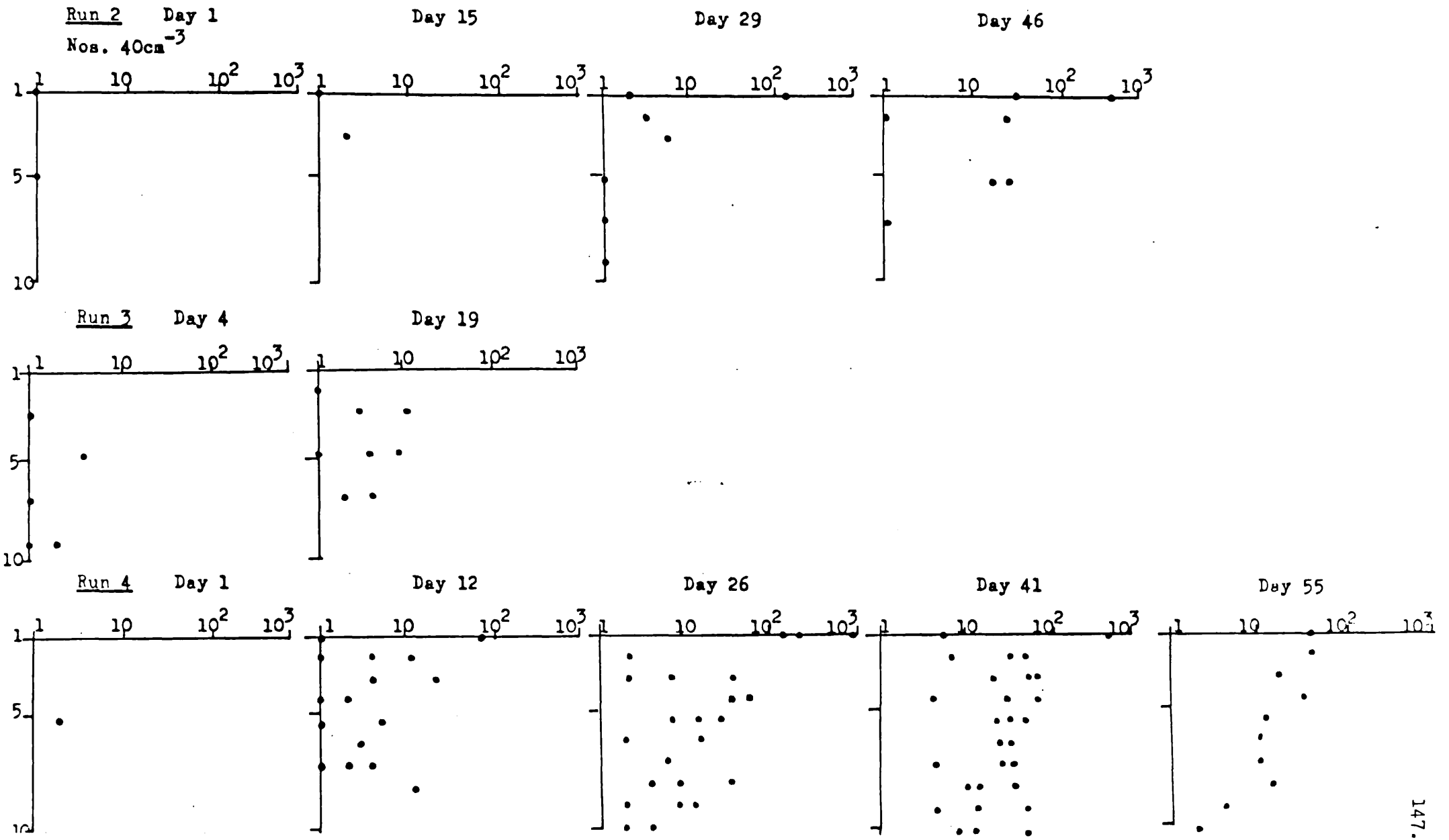


Figure 6.9 DEPTH PROFILE OF NEMATODES



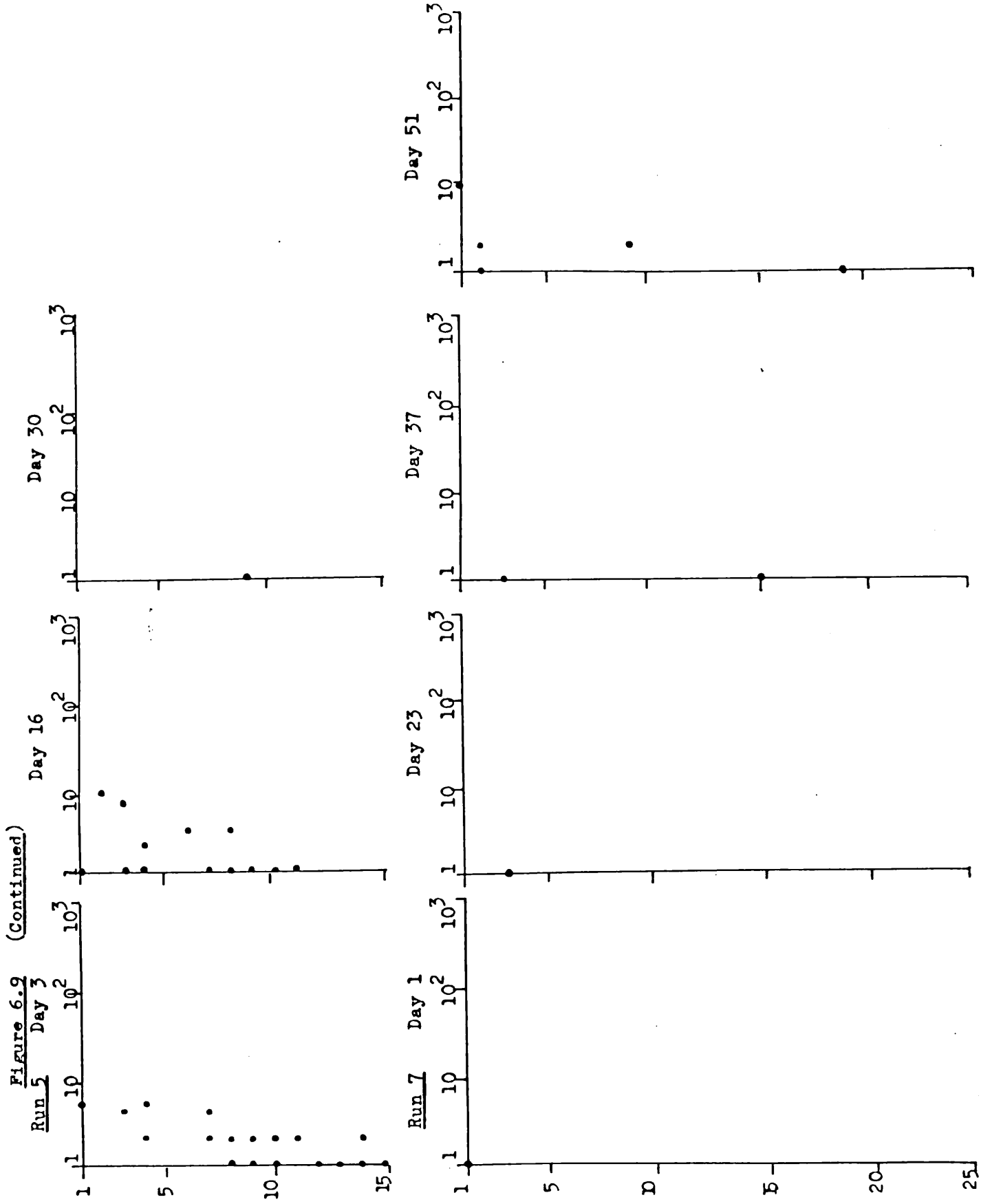
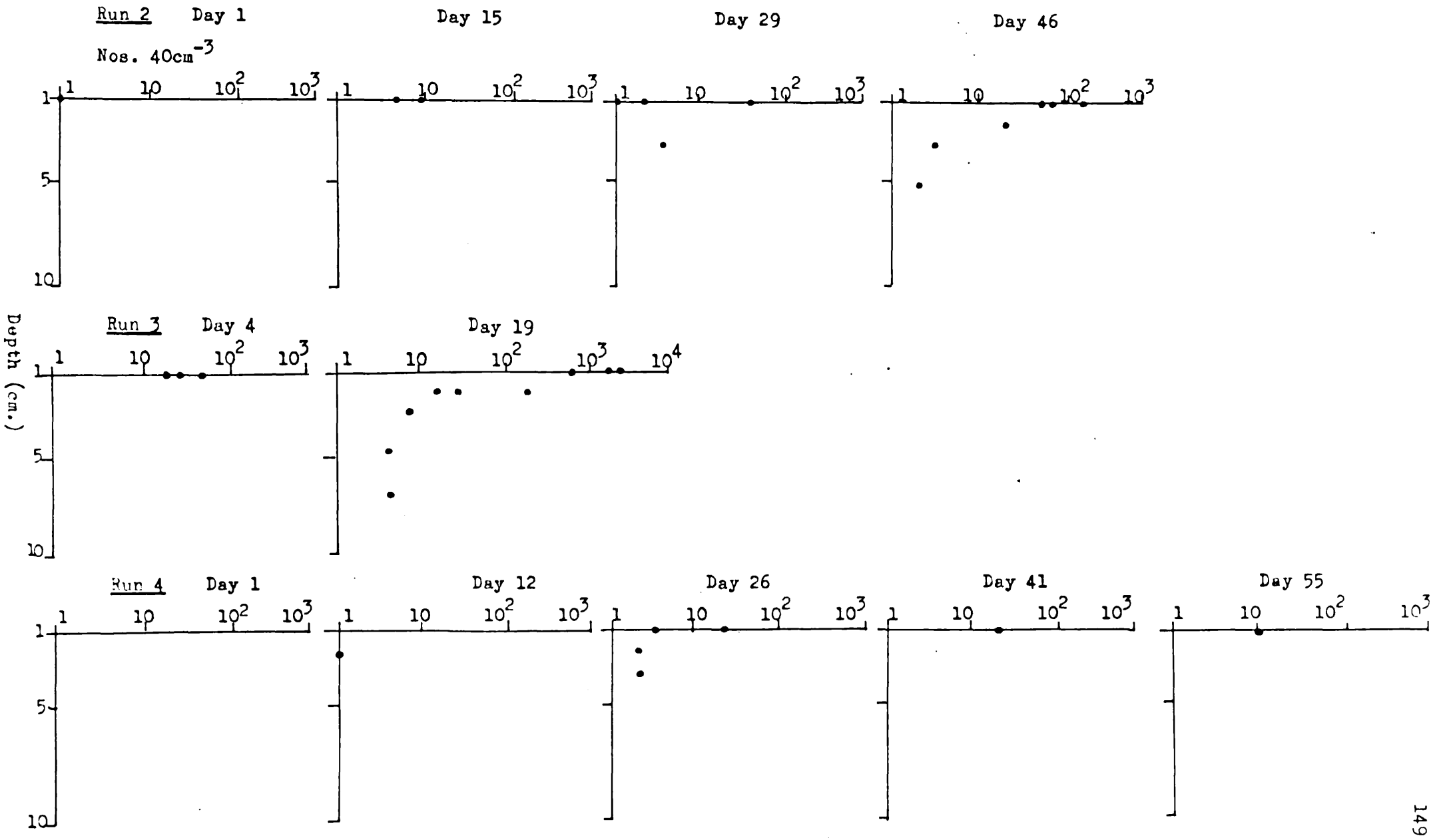


Figure 6.9 (Continued)
Run 5 Day 3
Day 16
Day 30
Day 51
Day 23
Day 37
Run 7 Day 1
1
10
10^2
10^3
5
10
15
20
25

Figure 6.10 DEPTH PROFILES OF CHIRONOMID LARVAE



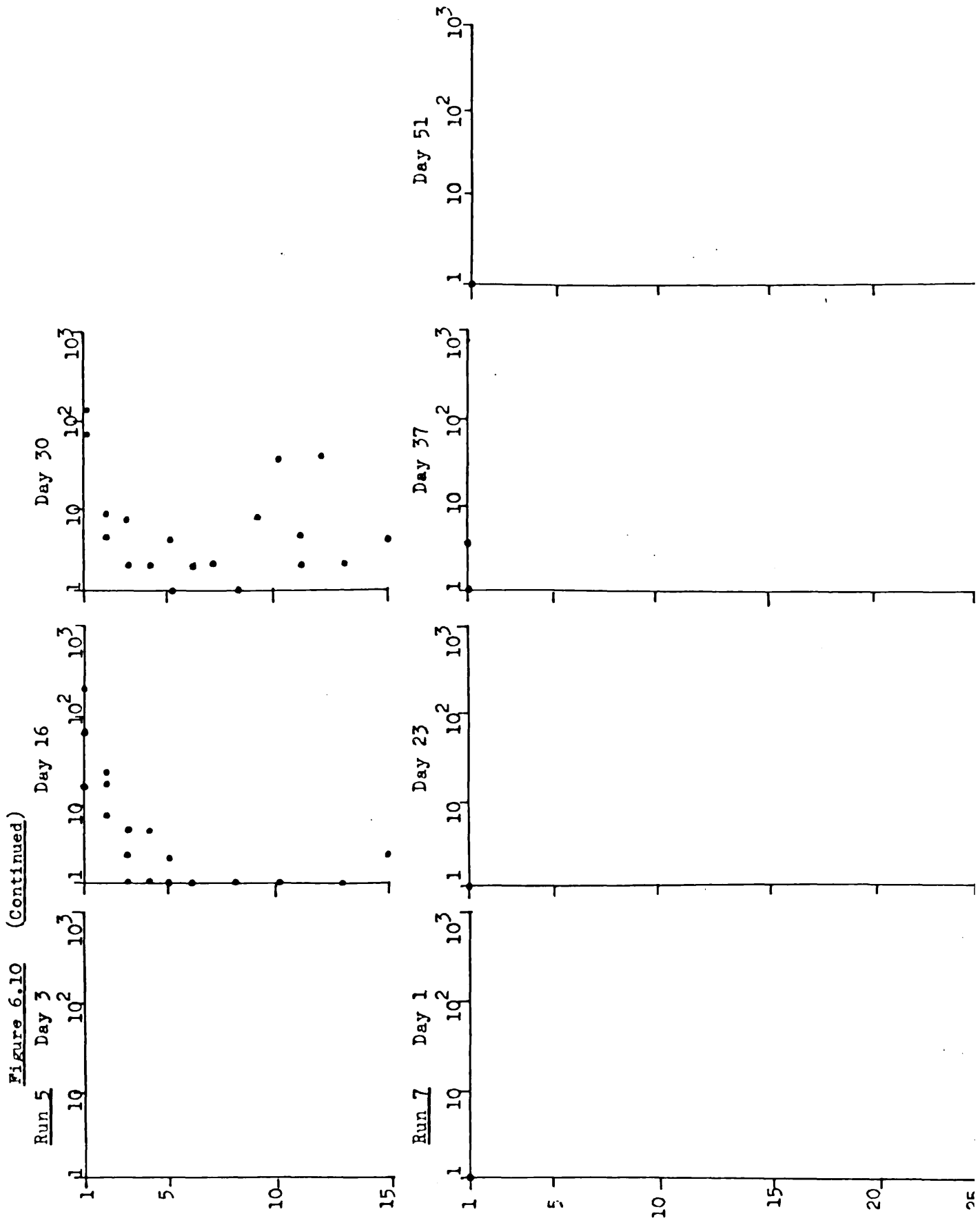


Figure 6.10 (Continued)
Run 5
Day 3
Day 16
Day 30
Run 7
Day 1
Day 23
Day 37
Day 51

Figure 6.11 DEPTH PROFILES OF HARPACTICOIDS

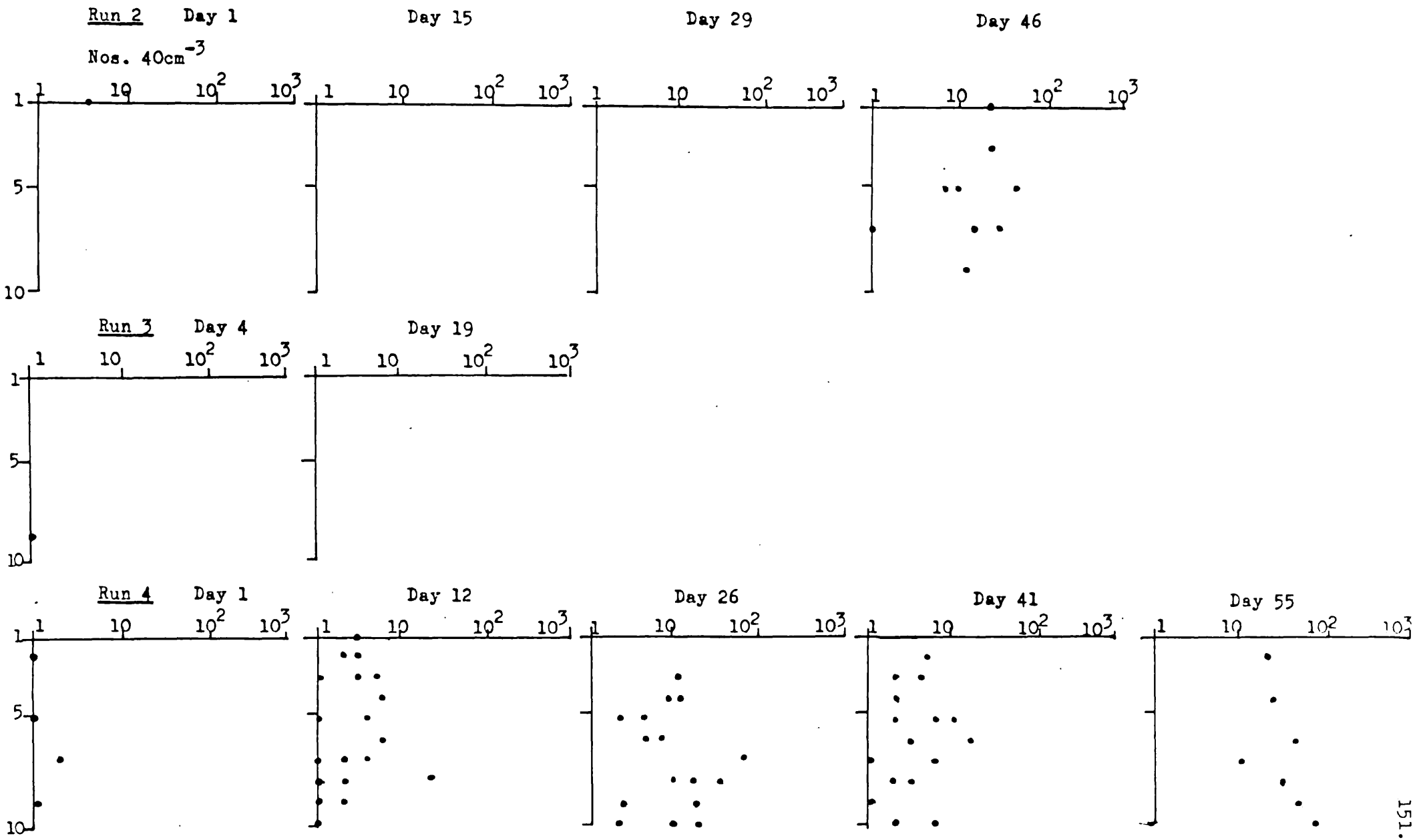
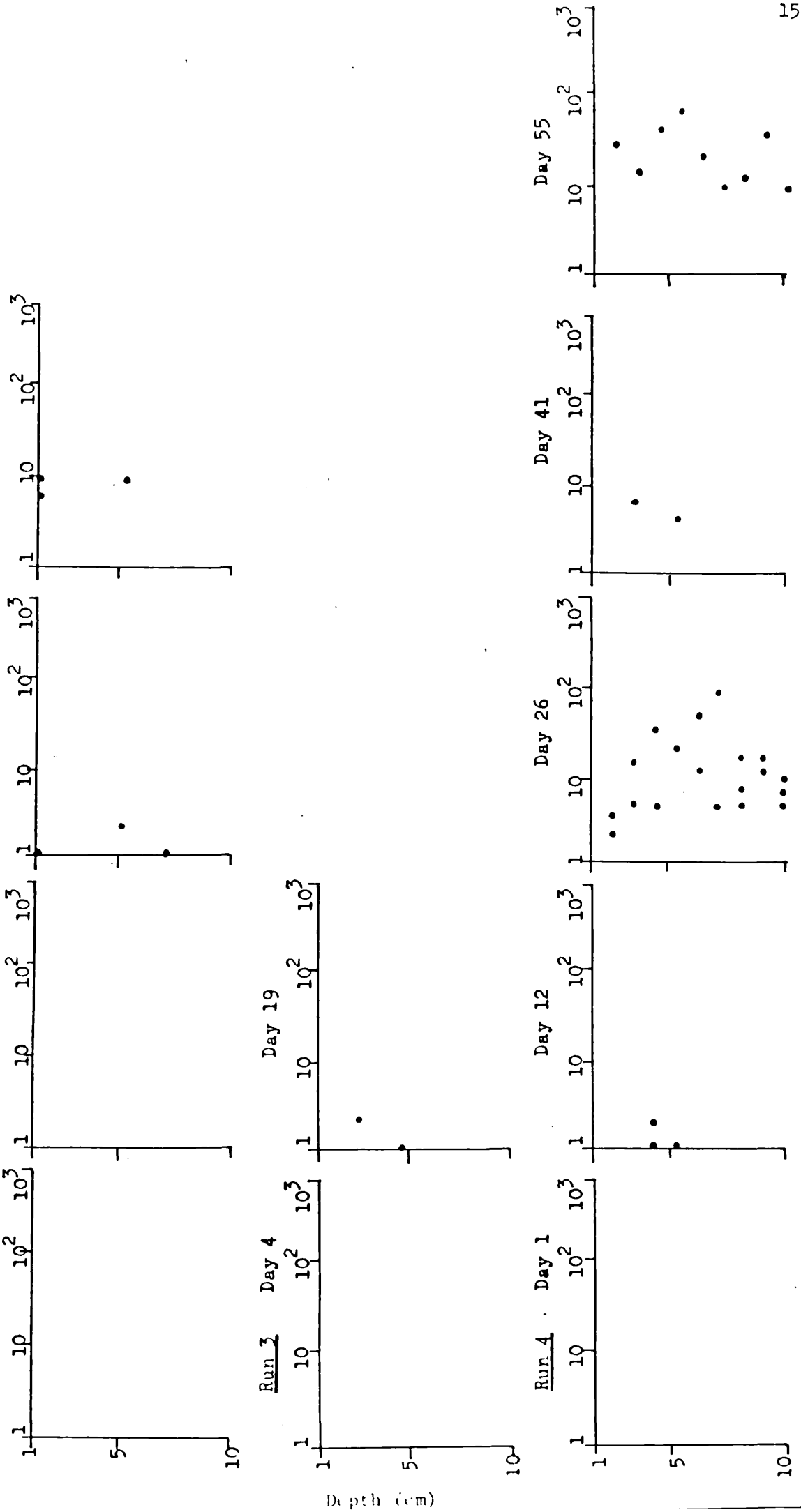


Figure 6.12 DEPTH PROFILE OF FLATWORMS



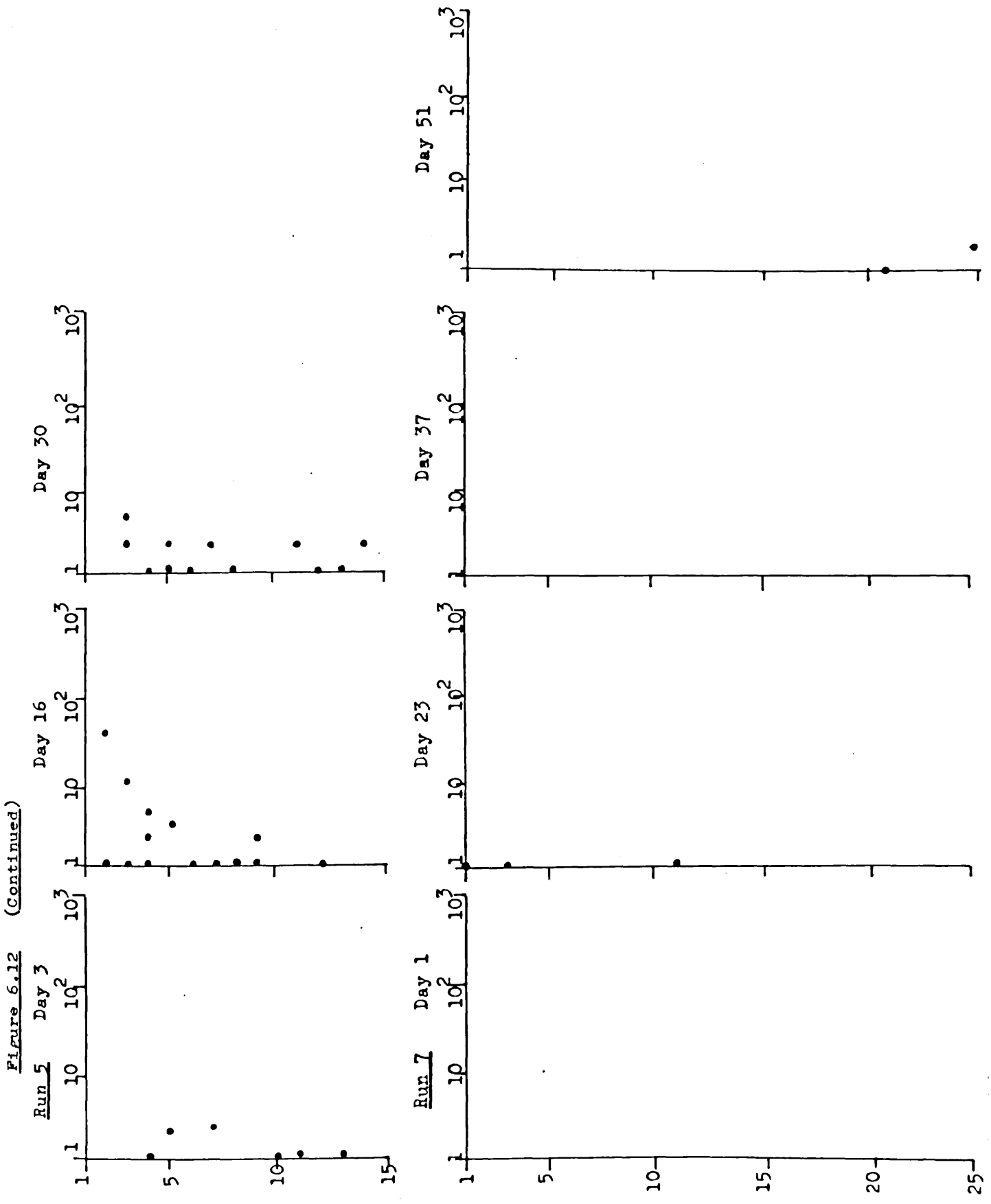


Figure 6.12 (Continued)

Harpacticoids and flatworms (figures 6.11, 6.12)

Both these groups show more or less no vertical zonations.

6.5.2 Discussion

In the slow sand filter situation where there are no spatial discontinuities in the substrate there should be no reasons, on grounds of the physical nature of substrate alone, why organisms should not be found throughout the whole depth of the filter. As far as the depths examined in this study are concerned, organisms can certainly be seen to penetrate at least to 25cm. However, as the filter run proceeds, incoming organic detritus from the rapid sand filters deposits in the top 1 and 2cm and also on the surface of the sand. The major effect of this is to increase the head loss of the bed and reduce its efficiency. A consequence of this is that the interstices, mainly of the top 1 and 2cm become filled with organic matter, a factor which could limit the types of organisms here to those which are capable of living in these conditions. A further effect of this increase in organic matter is that there may be high numbers of associated bacteria which will be respiring at this level in the bed. This will cause the dissolved oxygen levels to be reduced drastically (section 8), thus again limiting the organisms to those which can survive these conditions. However, a more attractive consequence of these high levels of detritus and associated bacteria is that organisms may well be induced into these regions due to the possibly high level of food at these depths. The explanations for the types of vertical distributions shown by different organisms in this habitat may thus be very different.

A discussion of the available literature regarding vertical distributions of meiofauna will be limited to those concerned with interstitial sand systems. The reasons for this are due to the big difference in the types of organisms which inhabit an interstitial and a benthic mud situation. The maintenance of an interstitial community, as pointed out by Cox (1976) depends upon sediment mobility. In marine and freshwater lakes this is due to wave action and in streams due to current. If this mobility is not maintained then the substrate will tend to fill in with finer sediments, resulting in a less penetrable and habitable interstitial system. The effect of this will be a change from the interstitial organisms to those with a burrowing mode of life. In a slow sand filter the incoming water brings in detrital particles which penetrate the top 1 and 2cm and fill in the pores. The mainten-

ance of the interstitial system in a slow sand filter is due to the constant attention from man who removes this accumulating material at regular intervals.

There is much evidence in the literature to show that organisms tend to show distinct patterns of vertical distribution in interstitial systems. In the majority of cases, organisms can penetrate to the limit of the interstitial system. For example Hynes et al (1976) showed penetration to 25cm, Harris (1972a, b, c) down to 50cm, Hynes (1974) 50cm, Ferencz (1974) 60cm, Cox (1976) 27cm. Rittersbusch (1974) showed penetration of meiofauna as deep as 260 cm in the sand layers underlying a slow sand filter used for artificial recharge.

In natural interstitial systems, most organisms tend to congregate in the top layers. As far as total numbers of organisms are concerned Arlt (1973) found 48% in the top 1cm, McLachlan (1977) showed the majority were located between 1 and 5cm, Fenchel (1969) demonstrated most abundance at 1 - 2cm, Ferencz (1974) in the top 1 and 2cm and Wieser (1960) in the top 10cm.

Vertical distributions of meiofauna in slow sand filters used for water purification were examined by Husmann (1976)(figure 6.13) and Lloyd (1974)(figure 6.14). Rittersbusch (1974)(figure 6.15) made similar examinations of both the slow sand filter and underlying sand components of an artificial recharge basin (the sand grain size composition of the upper slow sand filter component was very similar to that used by the Thames Water Authority at Hampton). Both Husmann's and Rittersbusch's filters operated at $5cm.hr^{-1}$ and Lloyd's at $15cm.hr^{-1}$.

Hynes (1974) showed naidids to be most abundant in the top 12·5cm. In a slow sand filter, Lloyd (1974) showed that Stylaria sp was most abundant in the top 2cm, however Nais sp showed most abundance at 22cm. In this study, no zonation with depth can be seen in the naidid species present. Lloyd (1974) also showed that Aelosoma was most abundant in the top 3cm of a filter, whereas in this study no zonation with depth can be seen. The problems which arose from a consideration of the sampling apparatus used by Lloyd have been discussed in section 3.

Total oligochaetes were shown by Rittersbusch (1974) to be most abundant in the top 5 and 10cm although some avoidance of the top 5cm can be seen on one occasion - a situation demonstrated in this study by E. buchholzi and also with regard to total oligochaetes by Cox (1976) and Ferencz (1974). However, Husmann (1976) showed the

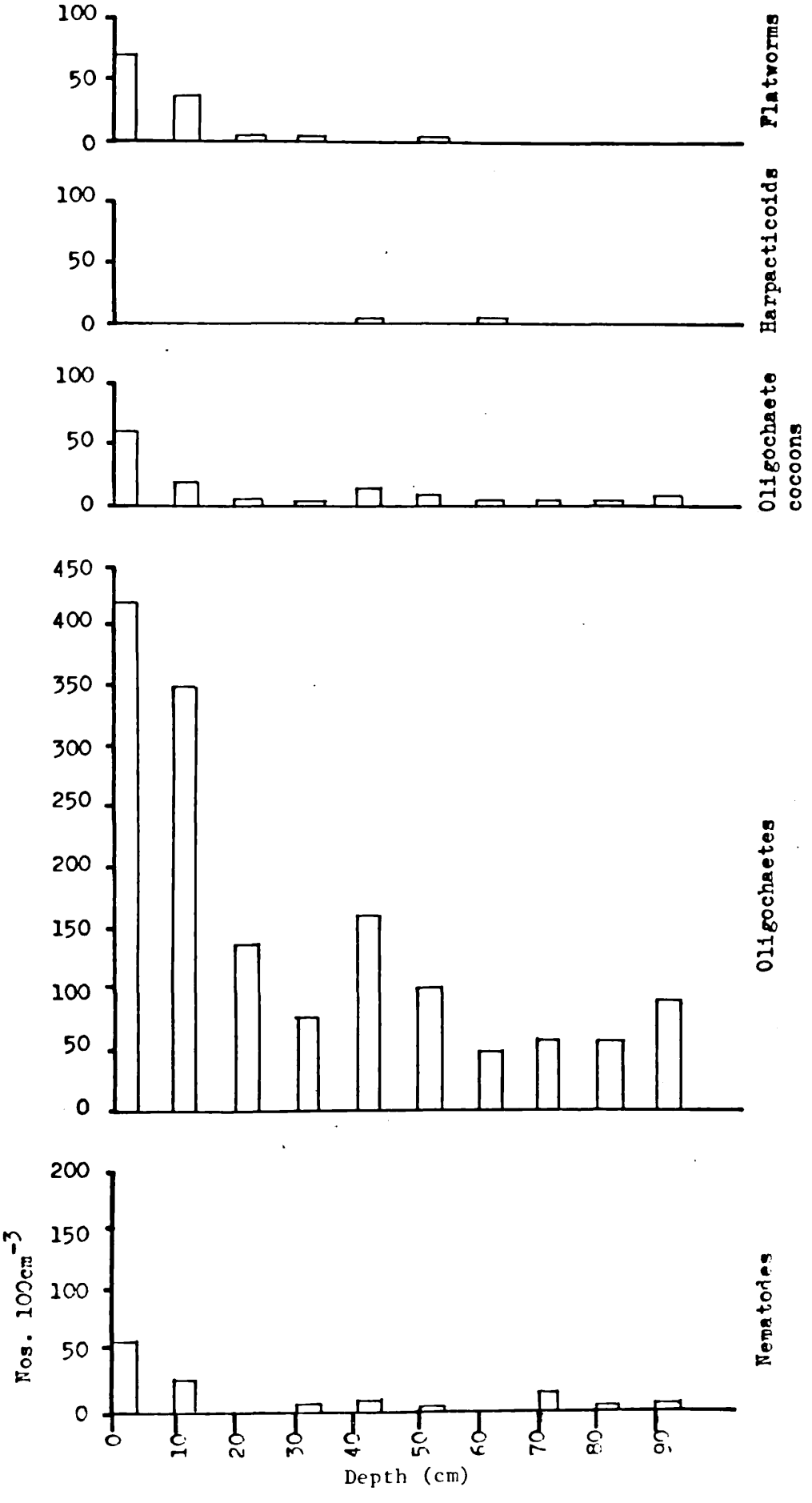


Figure 6.13 DEPTH PROFILES OF MEIOFAUNA IN SLOW SAND FILTERS. FROM HUSMANN (1976)

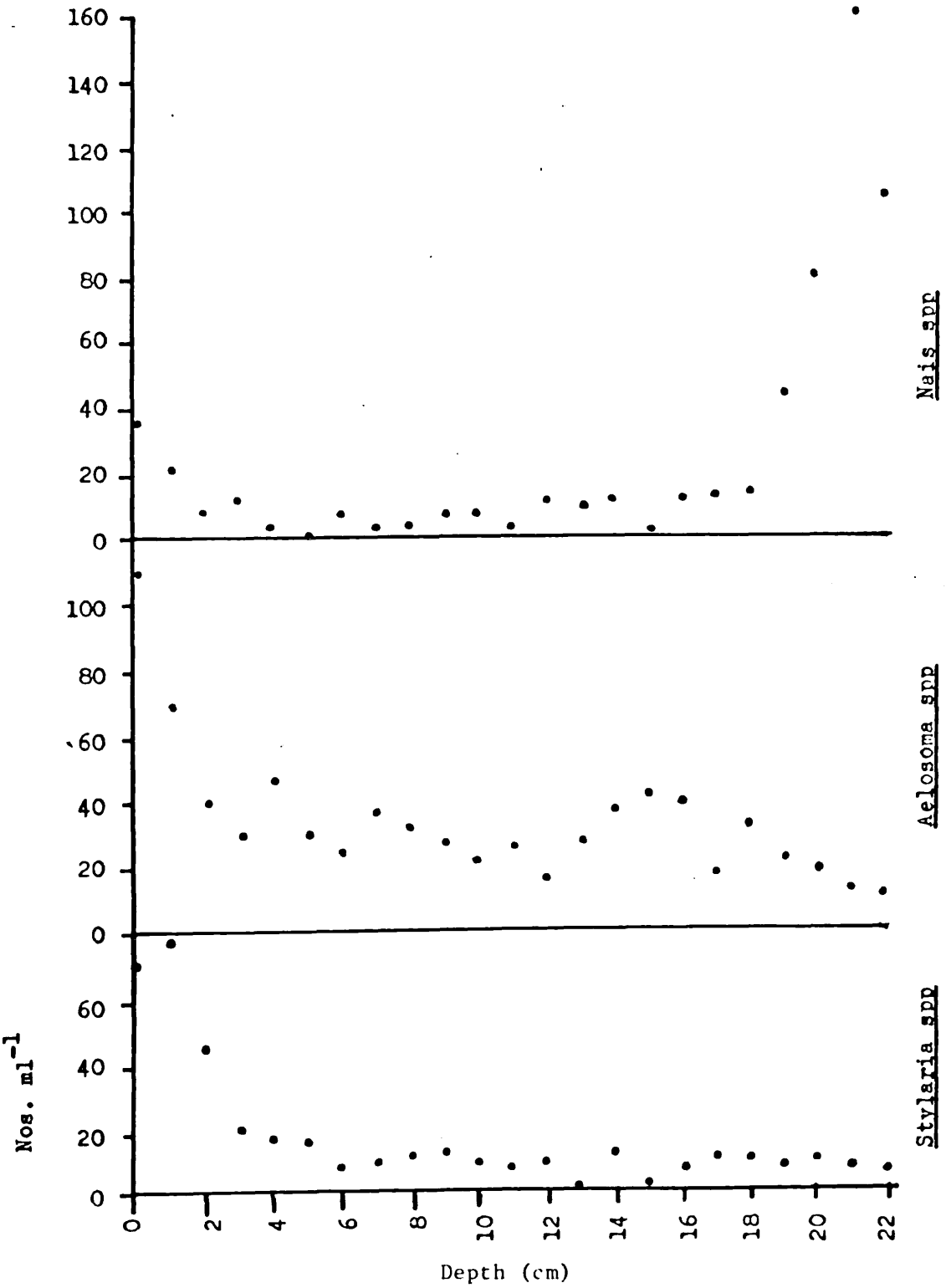


Figure 6.14 DEPTH PROFILES OF OLIGOCHAETES IN A SLOW SAND FILTER, FROM LLOYD (1974)

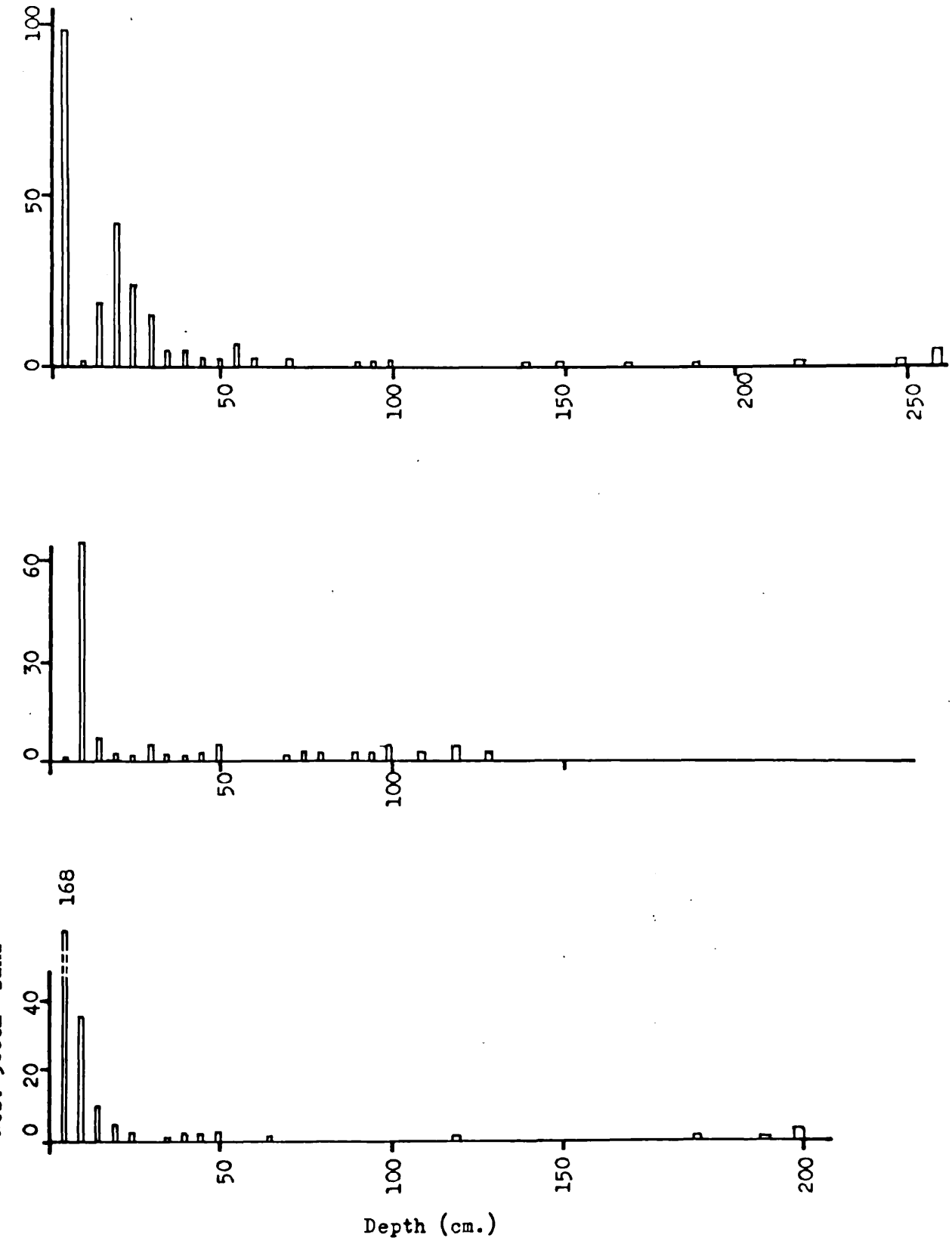


Figure 6.15 DEPTH PROFILES OF MEIOFAUNA IN THREE SLOW SAND FILTERS USED FOR ARTIFICIAL GROUND WATER RECHARGE, FROM RITTERSBUCH (1974). a. OLIGOCHAETES

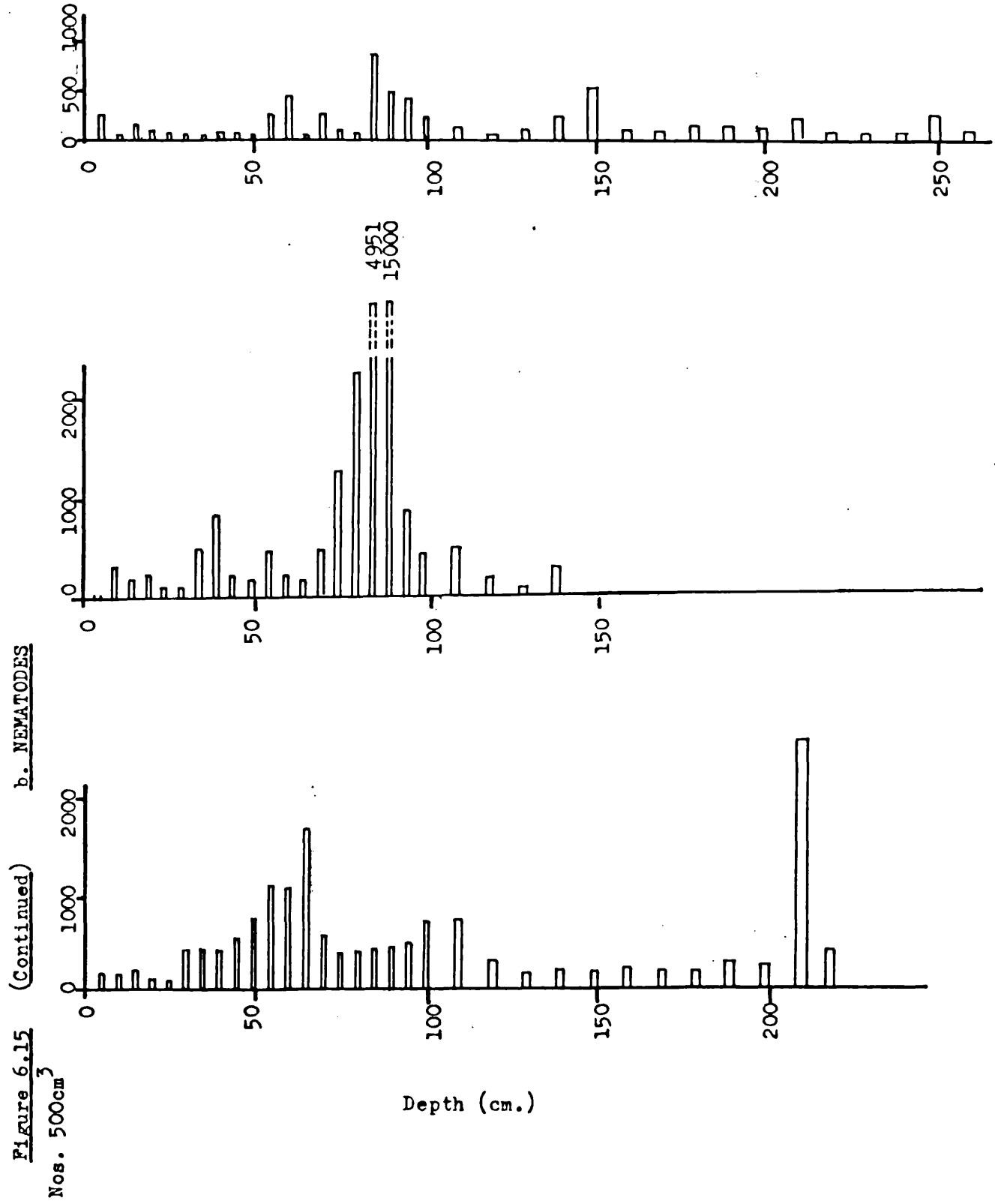


Figure 6.15 (Continued)

Nos. 500cm^3

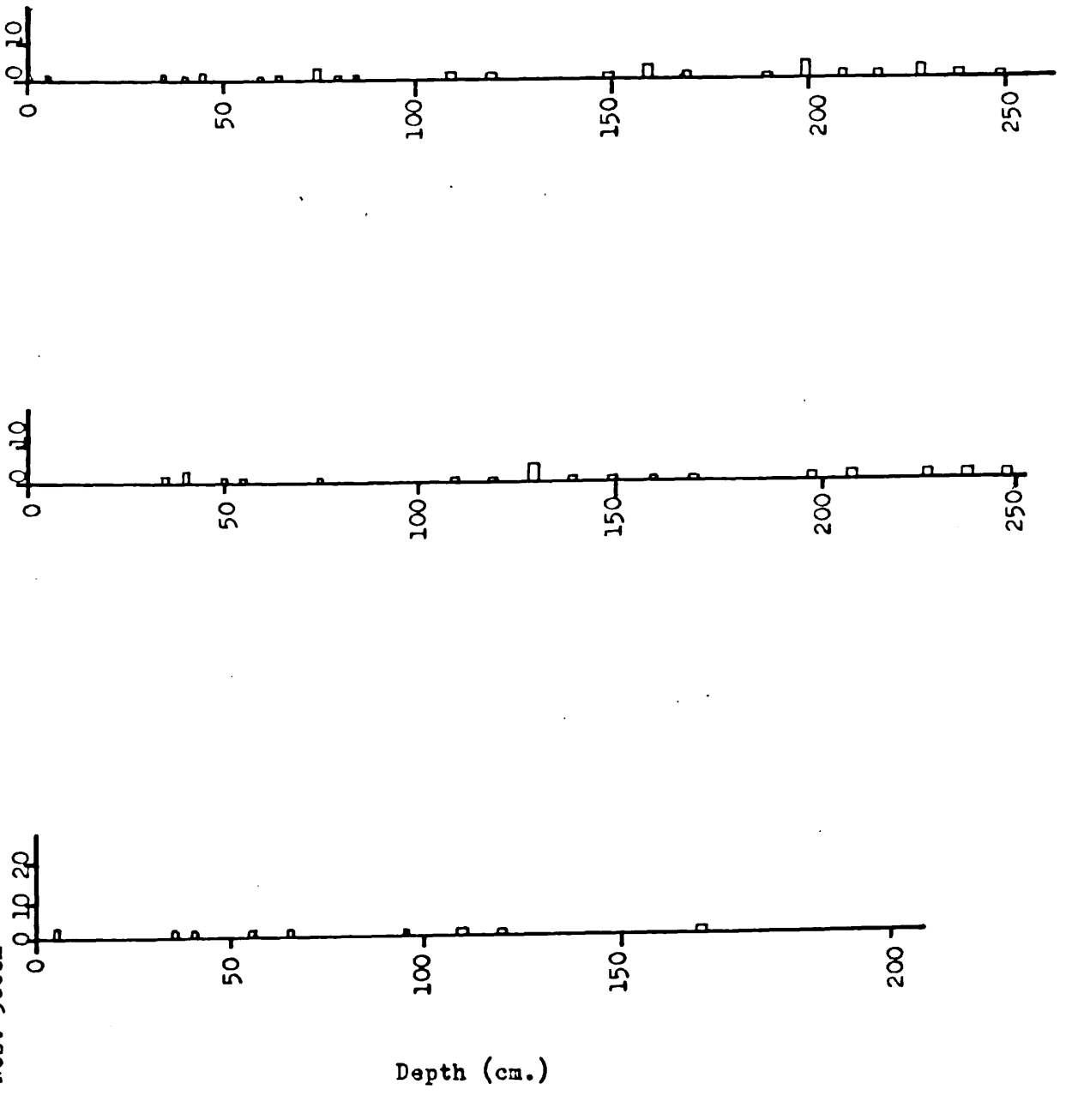


Figure 6.15 (Continued)
Nos. 500cm^3

distribution of total oligochaetes to have maximum numbers in the top 5cm. The distribution of oligochaete cocoons found by Husmann (1976) closely follows that of the oligochaete worms showing maximum abundance in the top layer of sand - agreeing with the results of runs 2 and 3 in this study.

Nematodes were shown by Hynes (1974), Fenchel (1969) and Cox (1976) to congregate in the top layers of the system. In the slow sand filter situation this type of distribution was shown by Husmann (1976) and also in this study. However, the findings of Rittersbusch (1974) disagreed, with numbers being low at the surface and higher numbers found between 30 and 100cm depth.

The distribution of chironomid larvae in this study agrees with that of Husmann (1976) and Hynes (1974).

The lack of vertical zonation shown in this study by both the harpacticoids and the flatworms agrees with the results of Hynes (1974), Husmann (1976) and Rittersbusch (1974)(all harpacticoids) and Cox (1976) (flatworms), although Cox (1976) showed that harpacticoids tended to accumulate between 12 and 27cm.

All of these interstitial systems cannot be directly compared for reasons discussed below and perhaps this may explain some of the disagreements found in published literature regarding vertical distribution. The slow sand filter is a unique system with water flow being vertically downwards ensuring a constant flow rate, with a supply of fresh water from above and with constant removal of any possibly toxic byproducts of metabolism. Changes in the filtration velocity of a slow sand filter can cause changes in the vertical zonation of organisms with faster rates causing deeper depth distributions of some organisms. This was demonstrated for ciliates by Lloyd (1973). As far as a beach is concerned, the stability of the system is dependent upon wave action and the water regime can be very variable. Although stability of the system is not quite so variable in a river or stream, these systems are still subject to varying degrees of erosion depending upon the nature of the water regime.

Reasons for these zonations seem to be dependent on the organic matter content of the system. Animals can perceive detritus as a space restricting medium and a supply of food but indirectly they may also respond to the oxygen demand of the bacteria which forms an integral part of the detrital particles. A correlation between the oxygen content of interstitial water and zonation of meiofauna has been

pointed out by Fenchel et al (1967), Fenchel (1969), Hynes (1974), Elmgren (1975) and McLachlan et al (1977). (The importance of oxygen profiles in this study is discussed in section 8). The significance of food in limiting vertical zonations has been discussed by McLachlan et al (1977), who showed a significant correlation between nematode numbers and the nitrogen content of sand and also by Tietjen (1971) who showed a similar correlation with the carbon content of the sand. The possibility of detritus acting as a space restricting medium is demonstrated by Ferencz (1974) who showed avoidance by oligochaetes of the top 10cm of an interstitial system lying beneath a thick layer of mud.

It is possible to consider the types of vertical distribution in this study as falling into three groups. The first includes those which show maximum abundance at the surface and decreasing abundance with increasing depth, as was demonstrated in this study by the nematodes and chironomid larvae. Reasons for this type of distribution may be due to a high demand for food, corresponding with high levels of carbon in the top few cms; or it may be due to a demand for oxygen (section 8), or may be related to the nature of the life cycle, as is the case with the chironomid larvae.

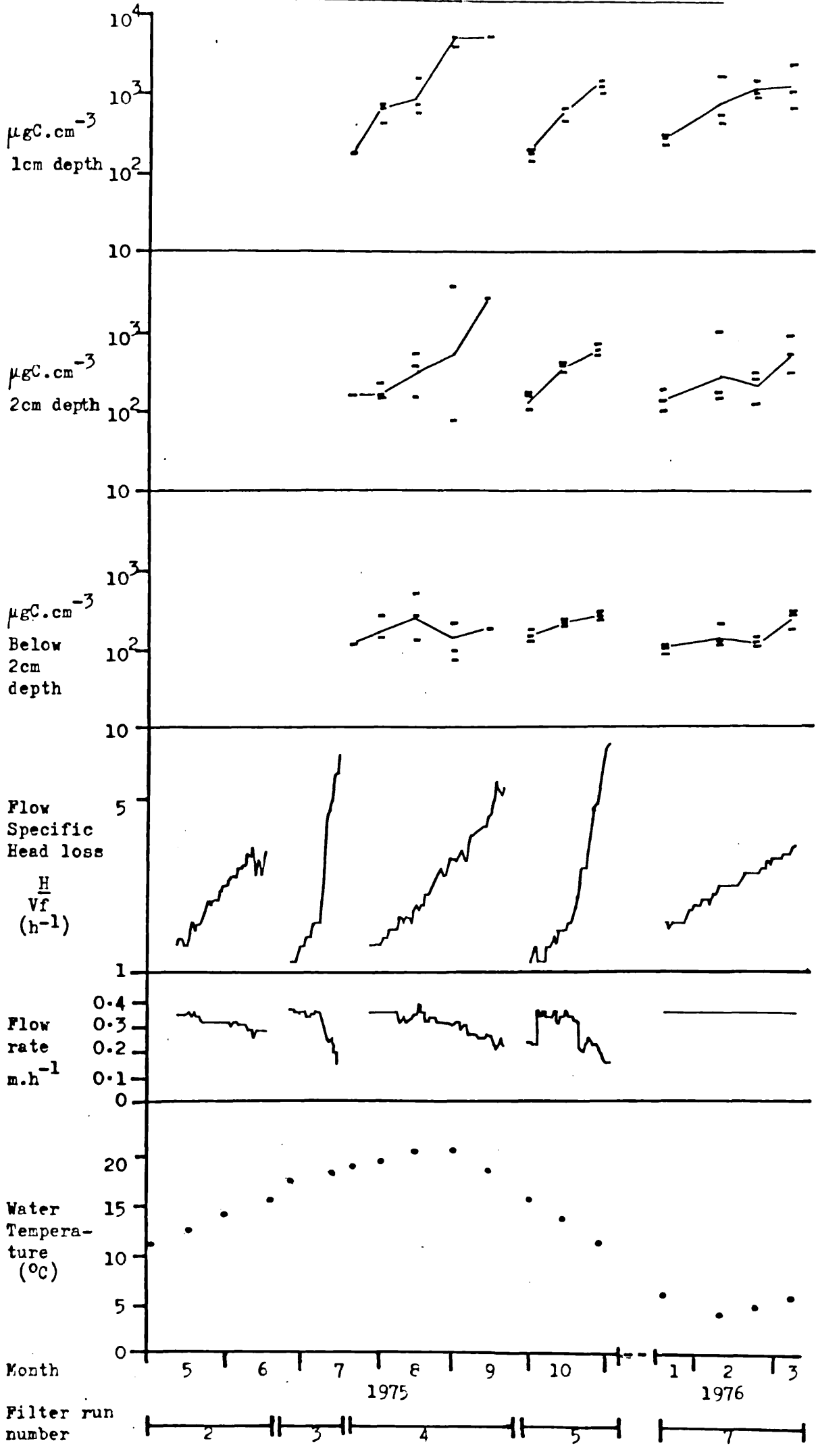
The second type of distribution demonstrated in this study was shown by E. buchholzi which demonstrated avoidance of the top few cms of the filter and with maximum abundance being below this region. This may be due to the physical effect of high levels of detritus in the top few cms of the filter causing the environment to become less interstitial and more benthic. This may also demonstrate that the enchytraeid was able to tolerate lower oxygen levels.

The third type of distribution was shown by the naidids, Aelosoma, the flatworms and harpacticoids which demonstrated more or less no zonation with depth. As far as the latter three groups are concerned, these organisms are relatively small and hence may have a greater mobility in the interstitial system. Because of this they may not be so limited by the availability of food and suggests also that they may be more tolerant than other groups to lower oxygen levels.

6.6 Standing crop data

Head loss and flow rate data for bed 45 (courtesy of the Thames Water Authority) is available in figure 6.16. From Darcy's law it can be seen that head loss (H) is affected by the flow rate (V_f) according

Figure 6.16 SEASONAL VARIATION IN ORGANIC CARBON CONCENTRATIONS



to the relationship $H = \frac{V_f.h}{k}$ (see chapter 2). As it can be seen that flow rate can vary from 0·15 to 0·35m.h^{-1} during the course of one filter run, then it is clear that the head loss recorded will be being continually affected by these fluctuations in flow rate. To overcome this problem the head loss data is adjusted with respect to the flow rate and the resulting value $\frac{H}{V_f}$ is termed "flow specific head loss" and is measured in hours^{-1}. This is plotted on a log scale against time in figure 6.16. This flow rate adjustment to head loss will leave a figure which can be more directly related to changes in porosity of the sand (and temperature/water density changes) due to increases in the amount of detritus deposited in the sand.

In order to present changes in numerical abundance of organisms with respect to time it was necessary to find the mean numbers of organisms present in 1cm depth of sand (40cm^3) removed from one half of the sampling core. When the sand was examined from one of these cores it was more usual to count organisms removed from every other 1cm depth in the core. So in order to obtain an estimate of total numerical abundance for the core a numerical integration program was used. The integrated total was then divided by the total depth of sand in the core to produce a figure representing the mean number of organisms present in 40cm^3 of sand. It would not be correct to adjust this value to find the number present in a more convenient volume, e.g. 10cm^3 or 100cm^3 as one cannot predict that numbers would remain constant when measured over a smaller or larger area. However, figures of abundance were corrected to a m^2 area for comparison to be made with the literature. Thus three values of abundance were available for each meiofaunal group on each sampling date.

In order to obtain a mean it was necessary to transform the data due to the overdispersed distributions of the meiofauna demonstrated in 6.4. So for each of these dates a geometric mean was obtained, using the relationship

$$\text{geometric mean} = \left[\text{antilog} \frac{\Sigma \log(x+1)}{n} \right] - 1$$

where x = number of organisms present, n = number of samples. log (x+1) was used where zero counts were encountered, in which case it was necessary to finally subtract 1 from the antilog.

Carbon data was treated in a similar way but the three levels of carbon suggested in 6.2 were treated separately (i.e. 1cm, 2cm and below 2cm). Figures of mean abundance for the three cores (from

integrated totals - appendix 6.15) and the geometric means (appendix 6.16) are plotted on a log scale with respect to time in figures 6.16, 6.17, 6.19, 6.20, lines are drawn connecting the geometric means for each filter run.

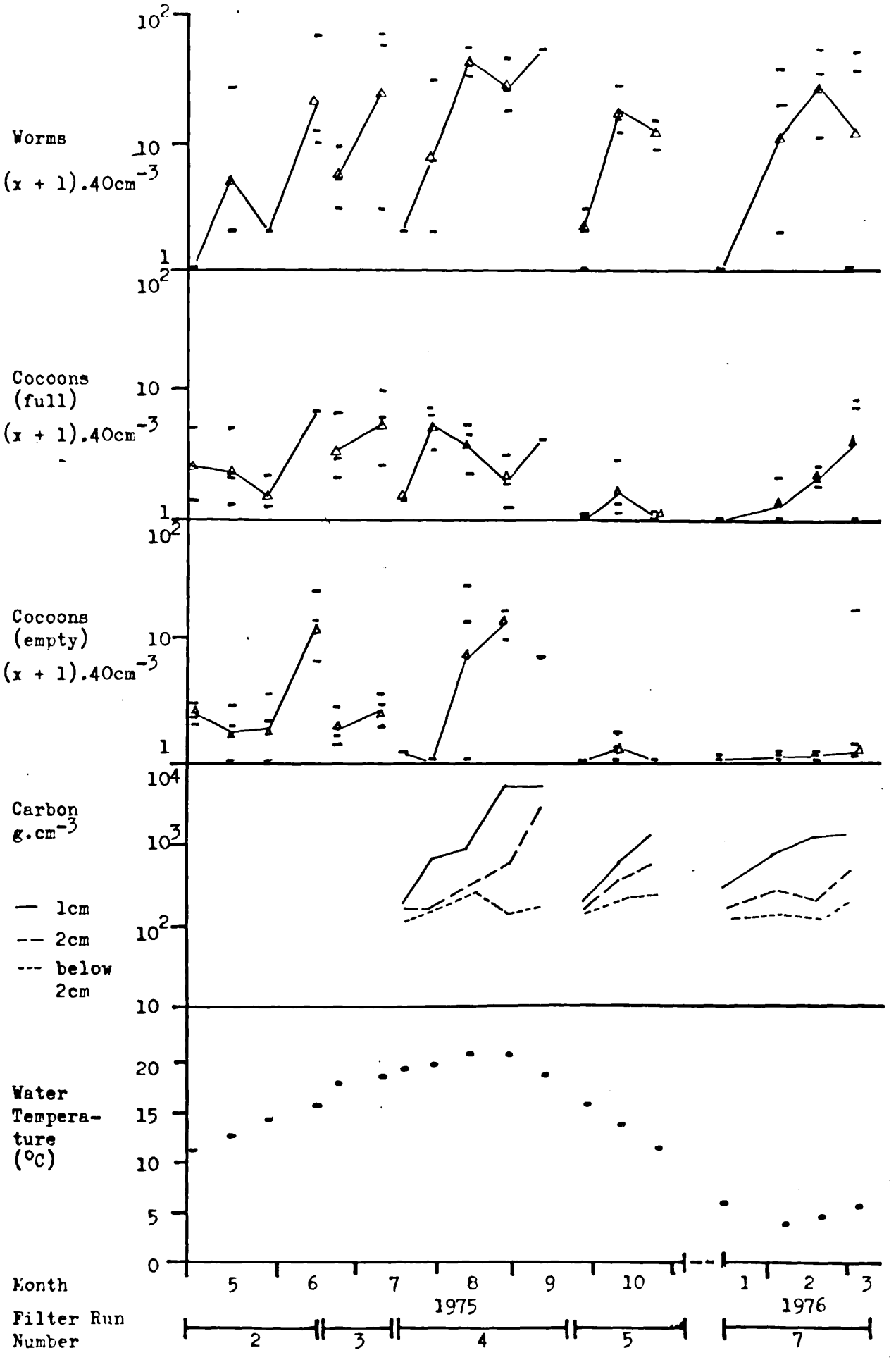
It can be seen that the flow rate of each filter run starts at a maximum of $0{\cdot}35 m.h^{-1}$ and decreases with time to approximately $0{\cdot}2 m.h^{-1}$. Flow rate data for run 7 was not available but the maximum rate did not exceed $0{\cdot}35 m.h^{-1}$ (I.P. Toms. T.W.A. personal communication). Adjusted head loss (flow specific head loss) shows an exponentially related increase with time and on runs 4 and 7 it can be seen that the rate of change of head loss is very similar to the rate of change of organic carbon measured at 1cm depth. With time it can also be seen that there is little increase in organic carbon recorded below 2cm, but that at 2cm depth there is a definite increase in organic carbon although at a lower rate than in the top 1cm of sand. Fluctuations in the amount of carbon present at any one depth in the sand do not appear related to the temperature recorded. This might suggest that contributions to organic carbon levels in the top 1cm of sand by benthic algal growths are obscured by the amount of settling allochthonous detritus in the inflow water.

In order to examine fluctuations in numerical abundance of the various meiofaunal groups it is really necessary to ignore the first value of abundance obtained for each filtration run. This figure is only representative of the organisms which have migrated into the sampling cores during the first 24 hours of the filter run and it has been suggested that it takes longer than this period of time for some of the organisms to reach stability (chapter 6.4).

Figure 6.17 indicates little seasonal fluctuation in abundance of E. buchholzi worms although there may be a suggestion of slightly higher numbers in July/August followed by a decline in September to October. The cocoons with eggs indicate slightly higher numbers in June, July, early August, with a decline in September to February. It is difficult to interpret the fluctuations in numbers of empty cocoons as no information is available as to their rate of decay. However it is apparent that fewer of these empty cocoons were observed during the autumn and winter corresponding with observations of lower numbers of cocoons containing eggs at this time.

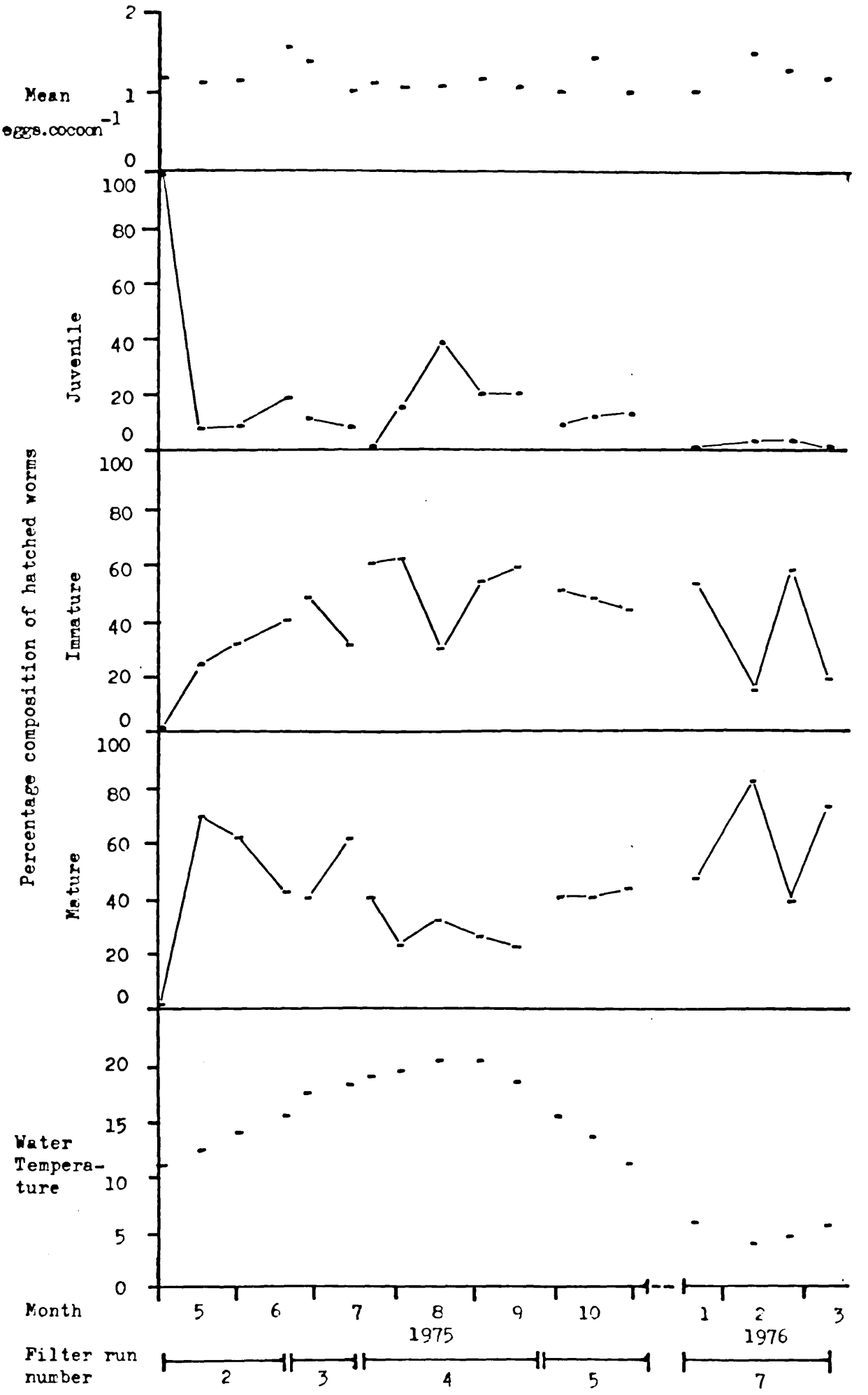
The mean number of ova per cocoon does not fluctuate during the year (figure 6.18)(mean = $1{\cdot}188$ eggs.cocoon^{-1}) although when

Figure 6.17 SEASONAL VARIATION OF E. BUCHHOLZI



Δ Geometric Mean

Figure 6.18 E. BUCHHOLZI, PERCENTAGE COMPOSITION OF HATCHED WORMS



related to temperature there was a slight suggestion of reduced egg number at 20°C (figure 5.4). It has been suggested earlier (chapter 5), from a comparison of field and laboratory hatched worms that there may be considerable mortality of ova in the cocoons before hatching. The suggestion of reduced egg numbers at 20°C may be related to the slightly higher levels of organic carbon at this time (July/August) possibly causing lower oxygen levels in the interstitial water of the filter.

It was possible to investigate changes in the percentage composition of E. buchholzi as far as maturity of individuals was concerned. It has already been discussed (chapter 5.2) that it was possible to separate individuals into one of three groups - juvenile, immature and mature. The percentage composition of these groups was determined on each sampling date (figure 6.18). Percentage composition of recently hatched juveniles tended to vary between 10 and 30%, again there are no very obvious fluctuations in the relative proportions of juveniles in the population although there is a suggestion of a peak in August/September immediately following the increase in cocoon abundance. So it would seem that the laying of cocoon and hatching of juveniles occurs at all times of the year with a possibility of increased cocoon laying during June/July/early August followed by increased hatching in August/September which would agree with incubation periods of 10 - 39 days (Learner 1972) and 35 days (Springett 1970).

It is apparent that between July and October there is a relative decrease in the proportion of mature individuals in the population. The maturation period of E. buchholzi varies between 16 - 50 days (20 - 8°C) (Learner 1972) and 180 days (10°C, Springett 1970), one might therefore expect a higher proportion of mature individuals in the winter months - a fact which is apparent in this study. However the relative decrease in mature individuals during the summer months might also be caused by degeneration of the sex organs of some of the mature worms following the increased breeding rate in June/July/early August . This phenomenon was noted by Timm (1967) and Springett (1970) who found juveniles of Marionina clavata and Cernosvitoviella briganta present in the summer with mature individuals decreasing in the spring. Also O'Connor (1958) found the prevalence of enchytraeid juveniles corresponding with a decrease in the proportion of mature individuals and suggested that mature individuals were undergoing regression of their sexual organs at this time.

O'Connor (1957) has suggested that enchytraeid cocoons are laid in the winter, accumulate in the soil and hatch as a response to rising temperatures in the spring. This is not the case in this situation, however, as it would appear that it is an increased laying of the cocoons which is corresponding with increasing temperatures in June. Solbé (1971) has similarly shown increased cocoon production at 15°C with Dendrobaena rubida subrubicunda and Eiseniella tetraedra and lower rates at 7 and 20°C.

Enchytraeids have been shown to exhibit more obvious seasonal fluctuations in abundance in terrestrial environments. Abrahamsen (1971) has recorded peaks of abundance of E. buchholzi in June. Nielsen (1955), O'Connor (1957), Peachey (1963), in permanently moist soil conditions, found maximum densities in the summer followed by a decline in late winter. They concluded there was a significant relationship between increasing numbers and increasing temperatures. But Nielsen (1955) also considered that seasonal fluctuations in the soil environment were related to soil moisture conditions with low abundance figures corresponding with periods of reduced soil moisture content and also lethally high summer temperatures for both adults and cocoons.

Other factors come into play in the sewage percolating filter where high wash out rates in the summer caused low numbers of enchytraeids at this time (Reynoldson 1939a and 1948). In percolating filters - Williams et al (1969) found cocoons of Lumbricillus rivalis and Enchytraeus coronatus present throughout the year but most abundant in February to May, corresponding with a peak of adult numbers at this time, after which adult numbers decreased considerably. Timm (1967) concluded that enchytraeids will reproduce throughout the year but will have periods of more intense reproductive activity.

With the use of the segment number - dry weight relationship demonstrated for E. buchholzi, it was possible to determine total dry weight of this organism present in each core. This value was converted to μg of organic carbon using the relationships, 1g dryweight of oligochaete is equivalent to 5364 calories (Cummins and Wuycheck 1971) and 1g organic carbon is equivalent to 10K calories (Winberg 1971b); thus 1g dry weight of oligochaete is equivalent to 0·5364g organic carbon. Thus it was possible to obtain biomass (expressed as $\mu gC.40cm^{-3}$) figures for E. buchholzi for each sampling core on each sampling date. The geometric mean of these values (appendix 6.17) is

plotted in figure 6.18.1 to show the seasonal variation. This pattern of seasonal variation of biomass can be seen to closely follow that of numerical abundance.

The percentage contribution of E. buchholzi to the total organic carbon (both dead and living) was determined (appendix 6.17) for filter runs 4, 5 and 7 (figure 6.18.1). The percentage composition of E. buchholzi can be seen to be not greater than 2·3% during these filter runs and at the start of each run the percentage composition is very low - never more than 0·14%. It would seem therefore, that this particular species does not account for much of the build up in head loss experienced over a filter run. Although the standing crop of E. buchholzi does not decrease significantly at the end of the filter run, the percentage composition of this species can be seen to decrease at this time. This is really only emphasising the relative increase of other measurable components of the total organic carbon present at this time.

Aelosoma hemprichi (figure 6.19) is present in the filter from May to October but disappears during the winter months. When present it does not seem to show any pronounced peaks of abundance, although fewer numbers are present in September and October. There is a possibility here the A. hemprichi may be normally present in the depths of the filter lower than those examined in this study and that at certain times of the year it migrates upwards to find more favourable oxygen conditions. This may be further demonstrated at the end of run 3 where slightly higher numbers of A. hemprichi were encountered. This run experienced big increases in head loss with time. It is possible that this was accompanied by high carbon levels and / or high chironamid numbers causing lower dissolved oxygen levels in the interstitial water and thus causing A. hemprichi to migrate into the higher levels of the filter. The proportion of the population found to be actively budding (table 6.25) does not seem to show any seasonality and varied considerably from 0 to 100%, but was more usually between 10 and 44%. The absence of budding individuals on run 3 may be a consequence of the high head loss rates and possible low dissolved oxygen conditions. On no occasion was more than one bud found in each budding chain.

N. elinguis was only present in June/July, P. foreli was found only in August (figure 6.19). P. idrensis was present in larger numbers only in runs 4 and 5 between July and October. It is possible

Figure 6.18.1 SEASONAL FLUCTUATIONS OF BIOMASS OF E. BUCHHOLZI

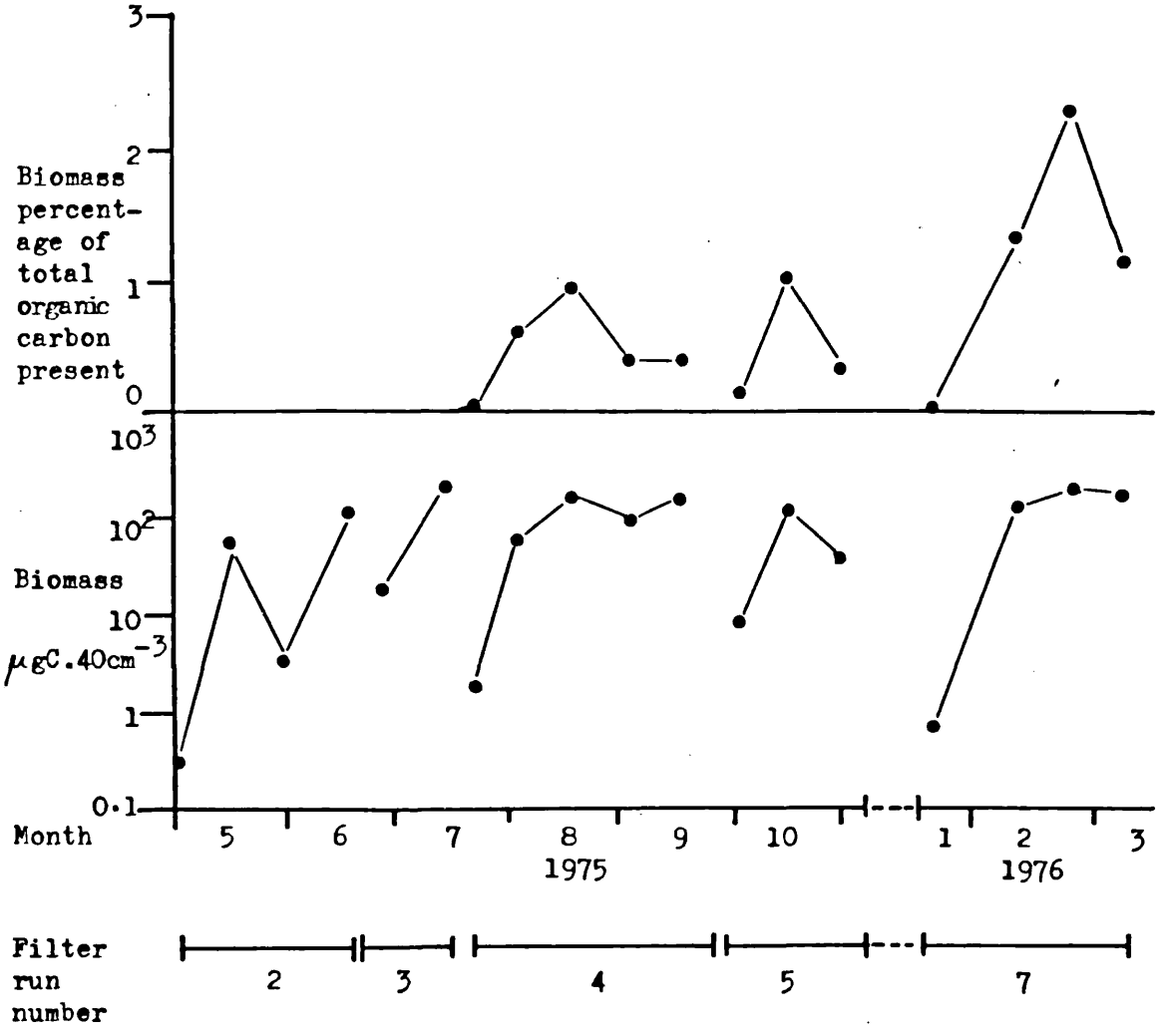
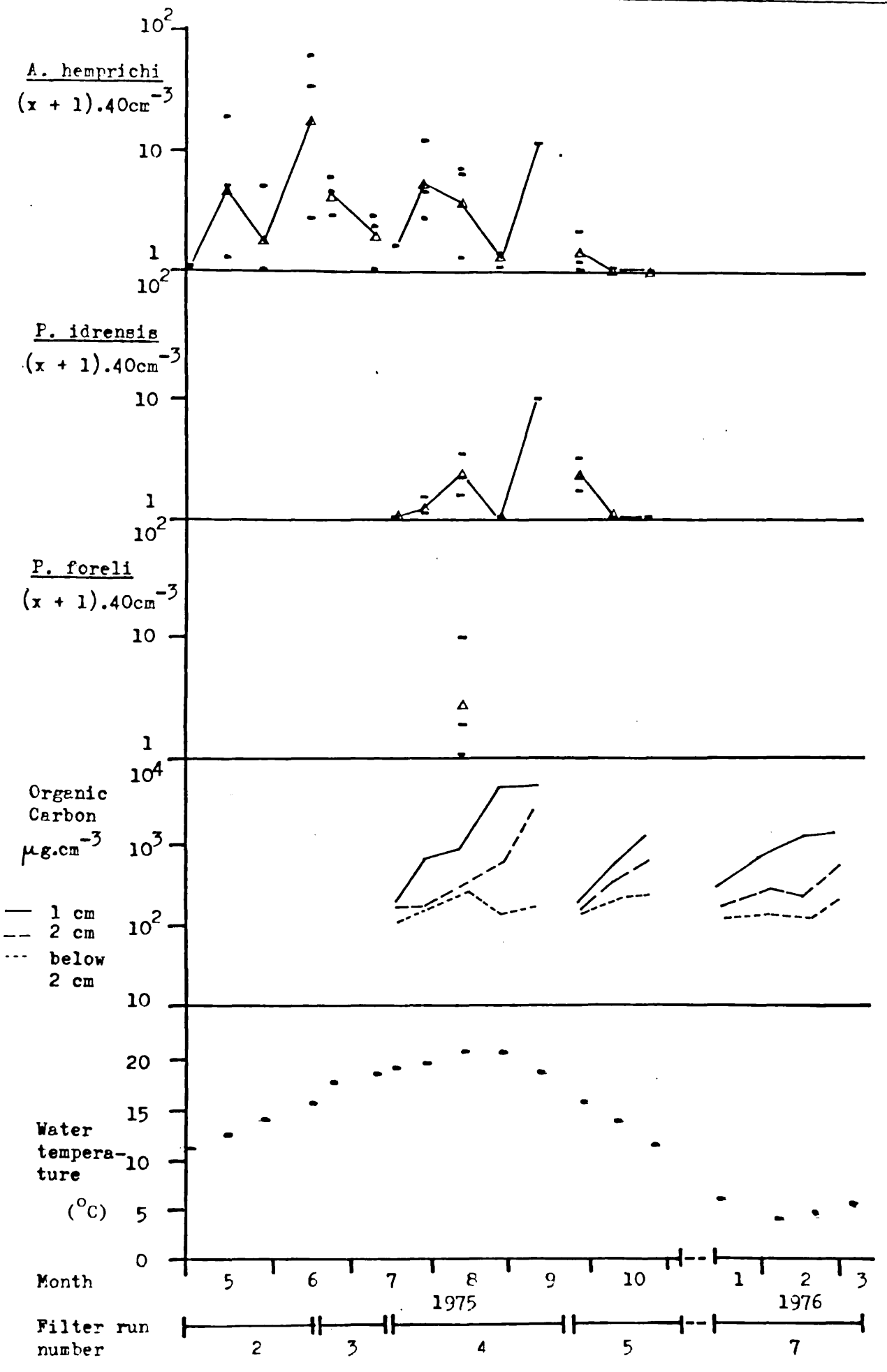


Figure 6.19 SEASONAL VARIATION OF A. HEMPRICHI, P. IDRENSIS AND P. FORELI



Δ Geometric Mean

that these species may also be more normally found deeper in the filter than was examined in this study and that at certain times of the year may migrate upwards to reach more favourable conditions. On the other hand, the apparent absence of these species for the greater part of the year may just be indicating that they are present in very low numbers and that during the summer months reproduce at faster rates stimulated by the higher temperatures.

In general asexual reproduction is the norm amongst naidids (review in Learner et al 1978), although at certain times of the year, usually during unfavourable winter conditions, sexual forms are apparent which produce cocoons (Timm 1967). These cocoons may survive the winter and hatch with the arrival of improving temperature conditions. In this study, however, no sexual forms of these three species were found, suggesting that conditions in the filter were favourable enough to prevent the need to produce these "overwintering" eggs. This may be related to the artificial nature of the slow sand filter ensuring an all-year-round supply of detritus which the naidids may have been feeding on.

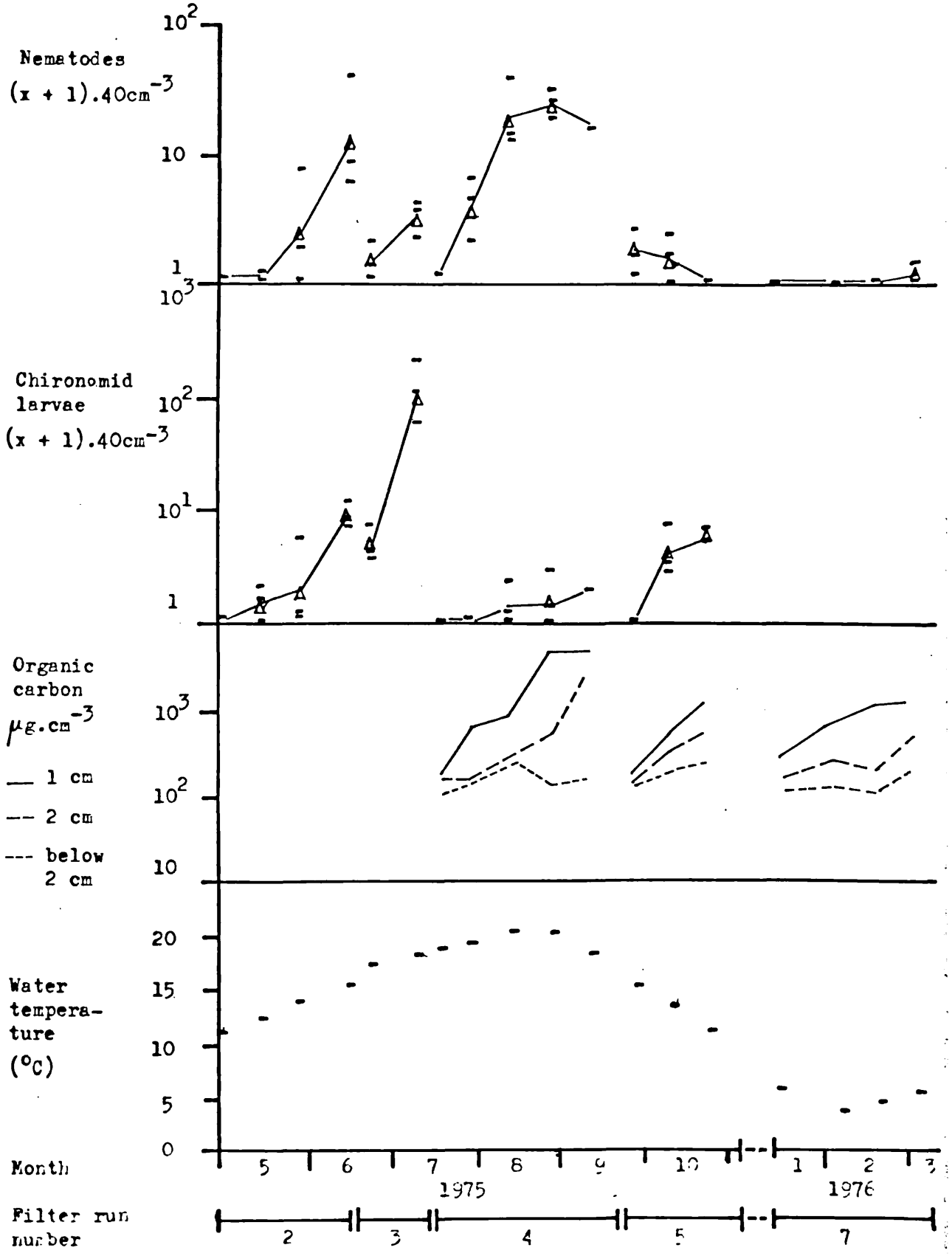
The proportion of the naidid population which was actively budding (table 6.25) did not appear to be related to water temperature although rate of fission has been related to temperature conditions (review in Learner et al 1978). On the few occasions that N. elinguis was present, it was found that every individual was budding. Lower degrees of budding were found in the two Pristina species, between 0 and 16%. McElhone (1978) found that the proportion of N. pseudobtusa which was budding varied considerably throughout the year (40 - 90%) but concluded that the proportions with a bud was significantly correlated with food abundance. However, it is difficult to make any similar conclusions concerning these three naidid species and food availability as the concentration of particulate organic matter in the filter varied very little in the region in which these naidids were found.

The nematodes reach a peak of numerical abundance in August/ September and decline to very low numbers in the winter. This decline may be a direct response to the low temperatures prevalent at this time reducing the reproductive rate. The main reason for this being that the level of particulate organic matter in the region of the filter where the nematodes congregate (i.e. the top 1cm) does not decline significantly during the winter months, which would suggest that food

Table 6.25 Percentage composition of oligochaete populations with respect to their reproductive states.

Run	Day	E.buchholzi % composition of total adults			Budding organisms as a % of the total			
		Juvenile	Immature	Mature	A.hemprichi	P.idrensis	P.foreli	N.elinguis
2	1	100	0	0	0			
	15	7	24	69	10			
	29	8	31	62	16			100
	46	18	40	42				
3	4	11	48	40	0			100
	19	8	31	61	0			100
4	1	0	60	40	17			100
	12	15	62	23	44	16	15	
	26	38	30	32	27	0		
	41	20	54	26	100			
	55	20	59	22	24	6		
5	3	9	51	40	24	4		
	16	12	48	40	0			
	30	13	44	43				
7	1	0	53	47				
	23	3	15	82				
	37	3	58	39				
	51	1	19	73				

Figure 6.20 SEASONAL VARIATION OF NEMATODES AND CHIRONOMID LARVAE



Δ Geometric Mean

is not limiting at this time. It must also be remembered that when the bed is cleaned at the end of a run the top 3cm of sand is removed. This sand will contain the greater proportion of the nematode population thus leaving few individuals in the sand to reproduce, and with low winter temperatures the rate of reproduction may be sufficiently reduced to prevent the build up of a large population. A similar peak of abundance at this time has been noted by Hulings (1974) in the fauna of a sandy beach in the Lebanon.

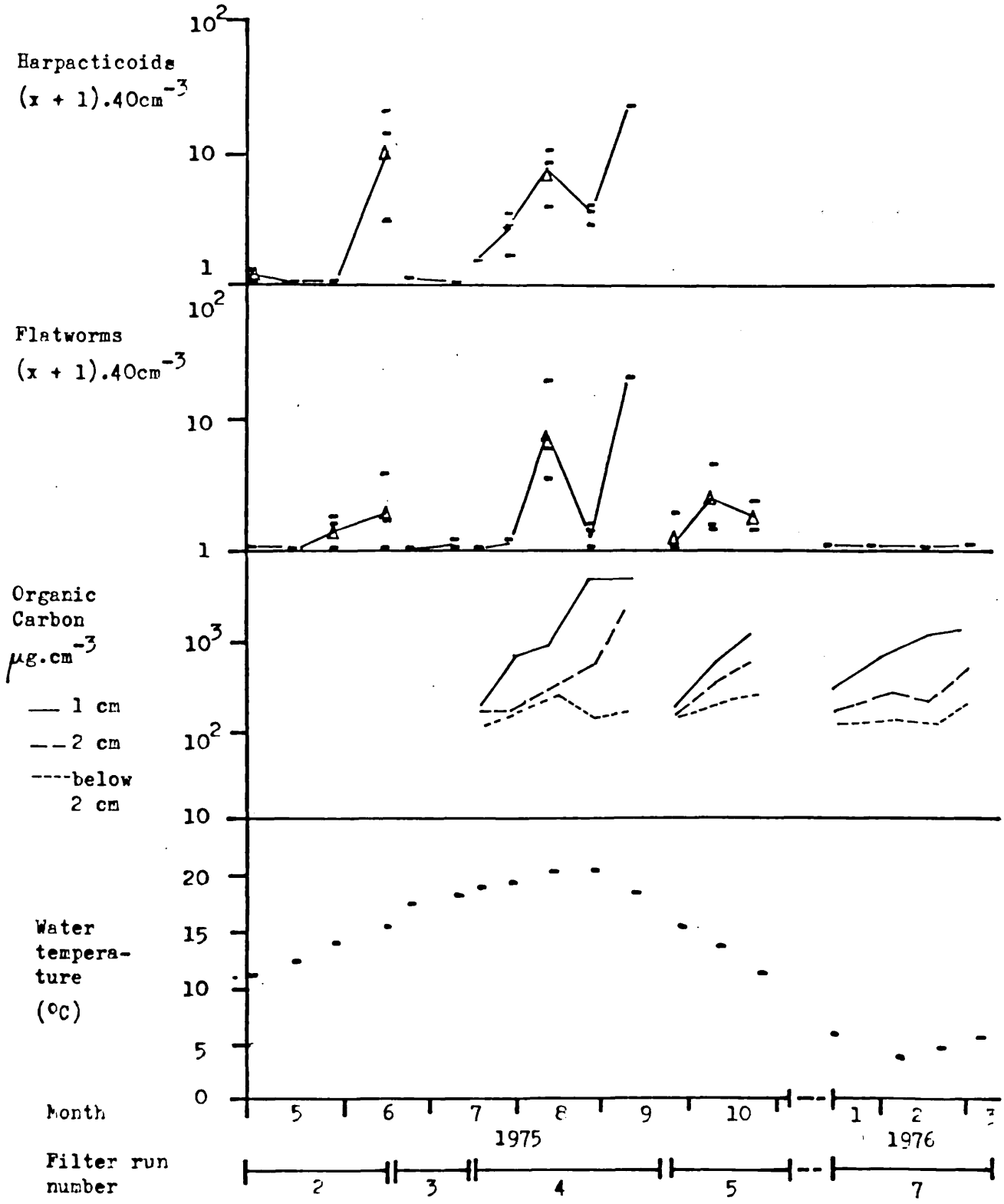
Chironomid larvae (figure 6.20) demonstrate two peaks of abundance, the first in June/July when very high numbers were encountered and the second in October. The first peak in fact corresponded with a very short filter run (run 3) of only 24 days. The "flow specific head loss" on this run (figure 6.16) increased considerably in a short period of time and it is possible that the increase in chironomid numbers during this period may have been mainly responsible for this very short filter run.

Harpacticoids appear absent from the filter during the winter months, but show no obvious peaks of abundance during the remainder of the year (figure 6.21). The very low numbers recorded during run 3 may be due to the very short length of this run preventing the establishment of a larger population. Hulings (1974) similarly found no peaks of harpacticoid abundance in the marine beach fauna.

The flatworms were present throughout the year and demonstrate slightly higher numbers in August. Hulings (1974) found two peaks of Turbellarian abundance in May and August.

Obvious peaks of abundance do not seem to be apparent in this slow sand filter as far as most organisms are concerned. One must here make allowance for the fact that the nature of operation of a slow sand filter, involving cleaning procedures when the top 1·5cm of sand is removed, will tend to obscure any peaks of abundance which might otherwise be apparent. This lack of seasonality has been noted by several authors who have examined interstitial systems. Swedmark (1964) concluded that reproductive periods covering the greater part of the year were observed in many species. Similarly Renauld-Debyser (1963) reported that most meiofaunal interstitial species were reproducing all the year round. Gowing (1972) found no seasonal pattern in total meiofaunal abundance and concluded that this absence of seasonality resulted from continually high organic inputs allowing the maintenance of high meiofauna population densities. Similarly

Figure 6.21 SEASONAL VARIATION OF HARPACTICOIDS AND FLATWORMS



Δ Geometric Mean

McIntyre (1964) reported the absence of seasonal variations of meiofauna from sublittoral muds, regions which were higher in organic matter content and concluded that the presence of continually high levels of food prevented the occurrence of these seasonal peaks.

On the other hand, Hulings (1974) did find that most of the species occurring in the marine sandy beach he examined showed two peaks of seasonal abundance in the spring and autumn. Harris (1972a, b and c) also found two peaks of abundance at these times. Harris concluded that the spring peak was generally associated with an increase in water temperature, whereas the autumn peak occurred at maximum or decreasing temperatures, thus suggesting a differential response to temperature.

Food availability and temperature seem to be the most important factors as far as seasonality in interstitial systems is concerned. It would appear that high year round food availability might tend to mask the effect of temperature fluctuations by allowing reproduction to occur from season to season.

The overall numerical abundance of meiofaunal organisms found in the slow sand filter is compared with other habitats in table 6.26. It was difficult to make direct comparisons with the numbers of organisms present in other habitats mainly because different authors express density in different units. Abundance may be expressed as the numbers found in the volume of sediment examined or beneath a certain area of sediment surface. In this case figures of abundance per $40cm^3$ were adjusted to give those found beneath $1m^2$ of filter surface, some of the information in the literature had to be similarly converted for comparative purposes.

The abundance of N. elinguis in the slow sand filter is low compared with other habitats. This species has been reported from regions which are generally high in organic matter and it may be that the slow sand filter is not sufficiently high in organic matter at all depths to allow the successful development of this species. Numbers of E. buchholzi found in this study are higher than those found in coniferous forest soil, but lower than those in sewage percolating filters, although the value for the percolating filter is a sum of all enchytraeid worms present. A factor to consider here is that the nature of a sewage percolating filter allows the establishment of high organic levels at all depths in the filter whereas in a slow sand filter there are very obvious discontinuities in the level of organic

Table 6.26 Density of meiofauna from various habitats.

Group	Density (nos.10^3.m^{-2})	Habitat	Author
Oligochaeta			
Aelosoma hemprichi	6-241.5	slow sand filter, 60cm deep	This study
Nais elinguis	0.05	" " " " "	" "
	0.1-5.2	stream bed	Learner et al (1978)
	3	brackish beach	Beschindt and Noak (1976)
	0.4-59	organically polluted river bed	Eyres et al (1978)
Pristina idrensis	3-132	slow sand filter, 60cm deep	This study
P. foreli	25.5	" " " " "	" "
Enchytraeus buchholzi	2.5-5.5	coniferous forest soil	Abrahamsen (197
	15-795	slow sand filter, 60cm deep	This study
E. coronatus	1500-12000 ($100-800.L^{-1}$)	sewage percolating filter total depth 1.5m	Williams et al (1969)
Enchytraeidae	1-9	fen	Dash and Cragg (1972)
	2-20	coniferous forest soil	" " "
	30-74	sandy pasture	Nielsen (1955)
	10-290	moorland	Peachy (1963)
	134	coniferous wood soil	O'Connor (1957)
	14100 ($94.10^2.dm^{-3}$)	sewage percolating filters total depth 1.5m	Solbe et al (1974)
Total Oligochaeta	1-5	organic rich lake benthos	Darlington (1977)
	0.2-29.5	brackish beach	Beschindt and Noak (1976)
	56-83	brackish river sediment	Arlt and saad (1977)

Table 6.26 continued.

	159	slow sand filter used for aquifer recharge, 260cm deep	Rittersbuch (1974)
	215-287	sandy marine beach	Cox (1976)
	49.5-1194	slow sand filter, 60cm deep	This study
	900	lake benthos	Hargrave (1969)
	12569	slow sand filter, 95cm deep	Husman (1976)
Nematoda	5.8-133	brackish beach	Beschindt and Noak (1976)
	10-190	oligotrophic lake benthos	Holopainen and Paasivirta (1977)
	92.4	slow sand filter, 94 cm deep	Husmann (1976)
	1.5-382.5	" " " , 60cm deep	This study
	230-700	marine sandy beach	Cox (1976)
	126-814	brackish river sediment	Arlt and Saad (1977)
	800	interstitial sand	McIntyre (1970)
	150-1800	marine sand	Wieser (1960)
	3100	intertidal mud	McIntyre (1970)
	18124	slow sand filter used for aquifer recharge, 260cm deep	Rittersbuch (1974)
Chironomid larvae	0.1-2	organically rich lake benthos	Darlington (1977)
	4.6	lake benthos	Hargrave (1969)
	2.2-23	brackish beach	Beschindt and Noak (1976)
	1-45	oligotrophic lake benthos	Holopainen and Paasivirta (1977)
	88.3	slow sand filter, 95 cm deep	Husmann (1976)
	6-1606.5	" " " , 60cm deep	This study

Table 6.26 continued.

Harpacticoida	0.6-23	brackish beach	Beschindt and Noak (1976)
	2.3	slow sand filter, 95cm deep	Husmann (1976)
	47	" " " used for aquifer recharge	Rittersbuch (1974)
	10-60	oligotrophic lake benthos	Holopainen and Paasivirta (1977)
	1.5-321	slow sand filter, 60cm deep	This study
Turbellaria	1-6	oligotrophic lake benthos	Holopainen and Paasivirta (1977)
	1-7	marine sand	Wieser (1960)
	4-9	brackish river sediment	Arlt and Saad (1977)
	50-100	marine sandy beach	Cox (1976)
	1.5-298.5	slow sand filter, 60cm deep	This study

matter encountered.

Comparisons with numbers of total oligochaetes found in various habitats indicates generally high abundance in slow sand filters. Again this may be due to high levels of detritus in a slow sand filter allowing the development of larger oligochaete populations.

The numbers of nematodes in this study compare with Husmann's (1976) data for a slow sand filter, but are much less than those found by Rittersbusch (1974). The nature of the slow sand filter may be of importance here, as Rittersbusch's filter was very deep (260cm).

Chironomid larvae are more abundant than was found by Husmann (1976), although Husmann's data did not examine changes over a whole year. Generally though, the numbers of chironomid larvae found in a slow sand filter are higher than in several other types of habitat. Similarly higher numbers of harpacticoids and flatworms have been found in the slow sand filter than in other benthic and interstitial environments.

Some factors to consider here are the greater depth in a slow sand filter which is available for colonisation, plus the fact that vertical water flow will allow greater penetration of oxygen into the filter compared with other interstitial habitats. Also this continual water flow will improve the removal rate of any unwanted by-products of metabolism. The artificial nature of the slow sand filter ensured the constant input of allochthonous detritus into the surface layers of the system which may be used as a food source and thus allows establishment of populations at times of the year where in other habitats there may well be a decrease in food abundance.

6.7 Rates of change of meiofaunal populations

The rate of change of a population is proportional to the size of the population at that particular moment, i.e. $\frac{dN}{dt} = r\bar{N}$ where $\bar{N}$ = mean numbers of organisms over the measured time interval, assuming constant recruitment and mortality; and so the instantaneous rate of change of the population (r) was calculated from the logistic equation :

$$N_t = N_o.e^{rt}$$

where N_t = number of organisms present after time t,
N_o = number of organisms present at time zero,
t = time interval in days

For any animal populations, r may be one of several values depending upon environmental factors. Under optimal conditions r will assume a value which is characteristic of the species being examined. However, these optimal conditions are rarely encountered either in the field or even in the laboratory although approximations may be obtained.

Values of daily r were calculated for each meiofaunal group and for each of the three levels of organic carbon in the filter, i.e. at 1cm, 2cm and below 2cm, thus giving a comparable daily rate of change for carbon concentration (table 6.27). It would be unnecessary to plot the seasonal variation in r as any discussion would be similar to that of the standing crop data which is plotted logarithmically. The fluctuations in r (expressed as percentage change per day) are, however, related to temperature in figure 6.22. Of course many other factors affect the survival of a population including food conditions and availability, inter and intraspecific competition predation and oxygen conditions. Most of these factors were not investigated in this study and so their precise effects cannot be evaluated here. The factors which would cause a decline in the population would be more complicated to assess than those causing increases in the population and so only positive rates of change are discussed here. Variations in values of r are difficult to interpret mainly because here one is dealing with a field situation in which the distribution of most of the organisms is heterogenous and so one has to deal with geometric means in order to obtain values of r. This in itself will cause a certain amount of variation in the value of r at any one temperature regardless of all the other environmental factors which are coming into play.

As far as the carbon is concerned, values of r over the first 14 days or so are not necessarily higher than later on in the run and, of course, are very dependent on the input of allochthonous material into the system. The first value of r obtained for the meiofaunal groups for each run over the first 14 or so days will probably be more indicative of rates of migration into the cores and not due to population growth. This would certainly appear to be the case as far as E. buchholzi worms are concerned (figure 6.22), where initial values of r are generally higher than those obtained later on in the run. The initial values of r obtained for E. buchholzi cocoons do not show this pattern which suggests a delay factor regarding the laying of cocoons by these initial enchytraeid worm colonisers. Chironomid larvae also demonstrate initially high values of r which may be due to high hatching rates from eggs deposited on the filter surface after

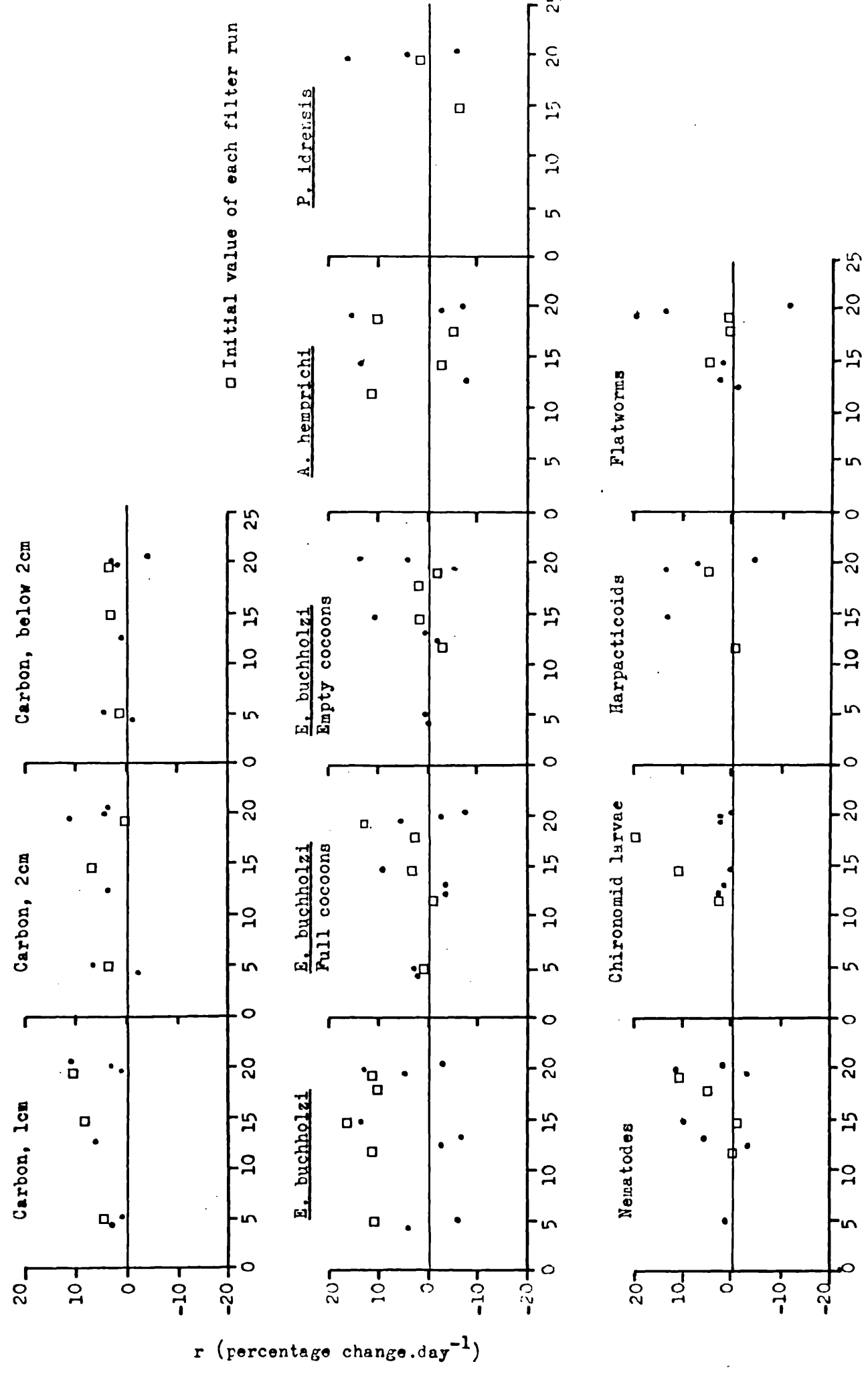
Table 6.27 Instantaneous rates of daily increase (r) of various populations of meiofauna and organic carbon.

$r = \frac{\ln N_t - \ln N_o}{t}$, where N_t is the no. of organisms present after time t (days), N_o is the no. present at time zero.

In this case (N+1) was used for the meiofauna and N was used for the carbon figures.

Run no	Date	Mean T (°C)	t (days)	E.buchholzi worms	E.buchholzi full cocoons	E.buchholzi empty cocoons	A.hemprichi	P.idrensis	P.foreli	nematodes	chironomid larvae	harpacticoids	flatworms	carbon 1cm depth	carbon 2cm depth	carbon below 2cm
2	2/5/75	11.7	14	0.115	-0.006	-0.025	0.114	------	------	0	0.029	-0.007	------	------	------	------
	16/5/75	13.2	14	-0.065	-0.034	0.008	-0.076	------	------	0.056	0.017	------	0.024	------	------	------
	30/5/75	14.8	17	0.135	0.091	0.109	0.136	------	------	0.100	0.003	0.132	0.018	------	------	------
3	24/6/75	17.9	15	0.102	0.031	0.025	-0.051	------	------	0.053	0.208	------	0.006	------	------	------
4	17/7/75	19.3	11	0.114	0.126	-0.017	0.107	0.017	------	0.109	------	0.046	0.009	0.108	0.005	0.035
	28/7/75	20.0	14	0.128	-0.026	0.136	-0.026	0.046	0.071	0.115	0.024	0.071	0.135	0.029	0.043	0.030
	18/8/75	20.5	15	-0.029	-0.072	0.044	-0.068	-0.056	-0.066	0.019	0	-0.046	-0.115	0.110	0.038	-0.040
	26/8/75	19.5	14	0.050	0.057	-0.044	0.154	0.163	------	-0.032	0.025	0.135	0.198	0.010	0.114	0.018
5	25/9/75	14.6	13	0.165	0.036	0.020	-0.026	-0.064	------	-0.009	0.110	------	0.043	0.085	0.073	0.033
	8/10/75	12.4	14	-0.025	-0.034	-0.019	------	------	------	-0.034	0.028	------	-0.011	0.062	0.036	0.011
7	15/1/76	4.8	22	0.109	0.012	0.004	------	------	------	------	------	------	------	0.046	0.035	0.015
	6/2/76	4.2	14	0.041	0.022	0	------	------	------	------	------	------	------	0.028	-0.023	-0.011
	20/2/76	5.0	14	-0.058	0.029	0.006	------	------	------	0.013	------	------	------	0.007	0.065	0.048

Figure 6.22 INSTANTANEOUS RATES OF INCREASE (r) OF MEIOFAUNA, RELATED TO TEMPERATURE



the filter had been put back into operation. These initial hatching rates would seem to be very temperature dependent with maximum r values obtained at 20°C. This situation is not so obvious with other meiofaunal groups and it seems likely that these organisms may have recolonised the area in less than the 14 days taken by the enchytraeid.

Values of r obtained from E. buchholzi worms do not seem to show any relationship with temperature although higher values of r were found between 15 and 20°C for cocoon production and compared with Learner's (1972) laboratory hatched worms which showed higher cocoon production per individual at 20°C. As far as one can judge from values of r obtained for empty cocoons, hatching rates would also seem to be higher at these temperatures. Ova production per adult per day has been shown by Learner (1972) to vary from 0·73 (8°C), 0·61 (15°C) to 1·13 (20°C). The mean positive r (excluding the initial value of each run) for E. buchholzi worms was 0.089 and for full cocoons 0.050, which suggests 1.780 ova per cocoon, assuming 100% survival of ova. However, the mean ova per cocoon in the field was 1.188 and under laboratory conditions 3.7. Learner (1972) demonstrated hatching success rates of 67% (8°C), 89% (15°C) and 64% (20°C) under laboratory conditions.

No clear trends can be seen by Aelosoma, P. idrensis or the harpacticoids. In the latter two cases this may be due to the fact that few data points were available. Muus (1967) has quoted rough values of r for benthic harpacticoids in culture of between 0·25 and 0·32. In this study slightly lower positive values of r were determined between 0·071 and 0·135. Aelosoma did not appear to show any temperature dependency between 10 and 20°C.

The nematodes did seem to demonstrate higher rates of r between 15 and 20°C. Positive values of r varied between 0·013 and 0·115 which compare with those found by Nielsen (1949) 0·2 and Hopper and Meyers (1966) which varied between 0·019 and 0·098 (mean = 0·068) under laboratory conditions. Flatworms also seemed to show some temperature dependence with peaks of r at 20°C.

Apart from the nematodes, chironomid larvae and flatworms, not much temperature dependence can be seen. This fact would tend to agree with the overall lack of seasonality demonstrated earlier which would suggest that other environmental factors must be overriding the effect of these physiological rate processes.

CHAPTER 7 PILOT SLOW SAND FILTER - APPARATUS AND METHODS

7.1 Pilot filter design

The need for detailed information of the kind which could not be easily obtained from an operational filter, led to the realisation that it was necessary to build a small scale pilot filter which was capable of being easily manipulated by a research worker. Apparatus used in the construction of this pilot filter is listed in appendix 7.1.

7.1.1 Main unit (figure 7.1 and plate 4)

The necessity to reproduce characterstic flow conditions inside a pilot filter was discussed in 3.6.1. This meant that the minimum diameter of the filter should be 10cm. In this case clear perspex cast tubing of length 2m and inner diameter 13·9cm was used. Clear tubing was used in order that a visual check could be made, during preparation of the filter, to ensure that no sorting of sand grains took place and no air bubbles formed.

A perspex end plate was made for the base of the column. This was held in place by screws and was removable to facilitate easy cleaning of the column. The seal was water tight.

A perforated base plate was manufactured from perspex to support the sand and gravel layers. It was positioned 15cm above the end plate and was supported by a tripod of vertically placed perspex rods.

Several holes, 1·8cm in diameter, were bored at intervals throughout the depth of the column. At each chosen depth 2 holes were bored at right angles to one another. It was intended that the surface of the sand placed in the filter should lie just below the highest pair of holes, which were drilled 90cm above the end plate. The maximum depth of sand was intended to be 60cm supported on a 15cm layer of gravel which would lie on top of the base plate. There was a greater proportion of holes drilled in the topmost region of the column in order that more intense monitoring of the detritus rich region could be made.

A piece of perspex tubing with an outside screw thread was constructed (figure 7.2) and glued in position inside each hole and at right angles to the filter wall. Thus it was possible to attach

Figure 7.1
DIAGRAM OF PILOT FILTER

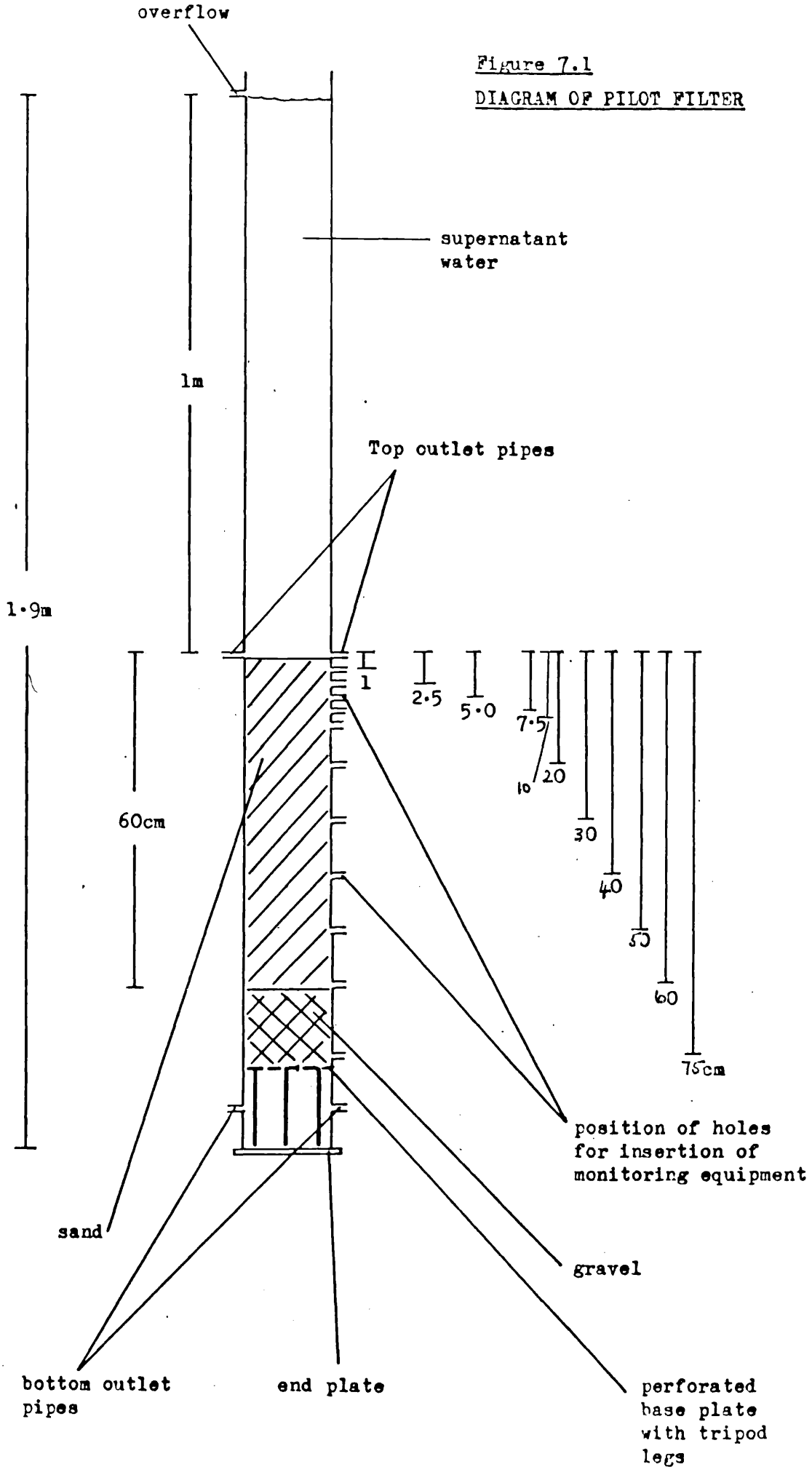


Plate 4. View of pilot filter from right side, with blackout boards removed.

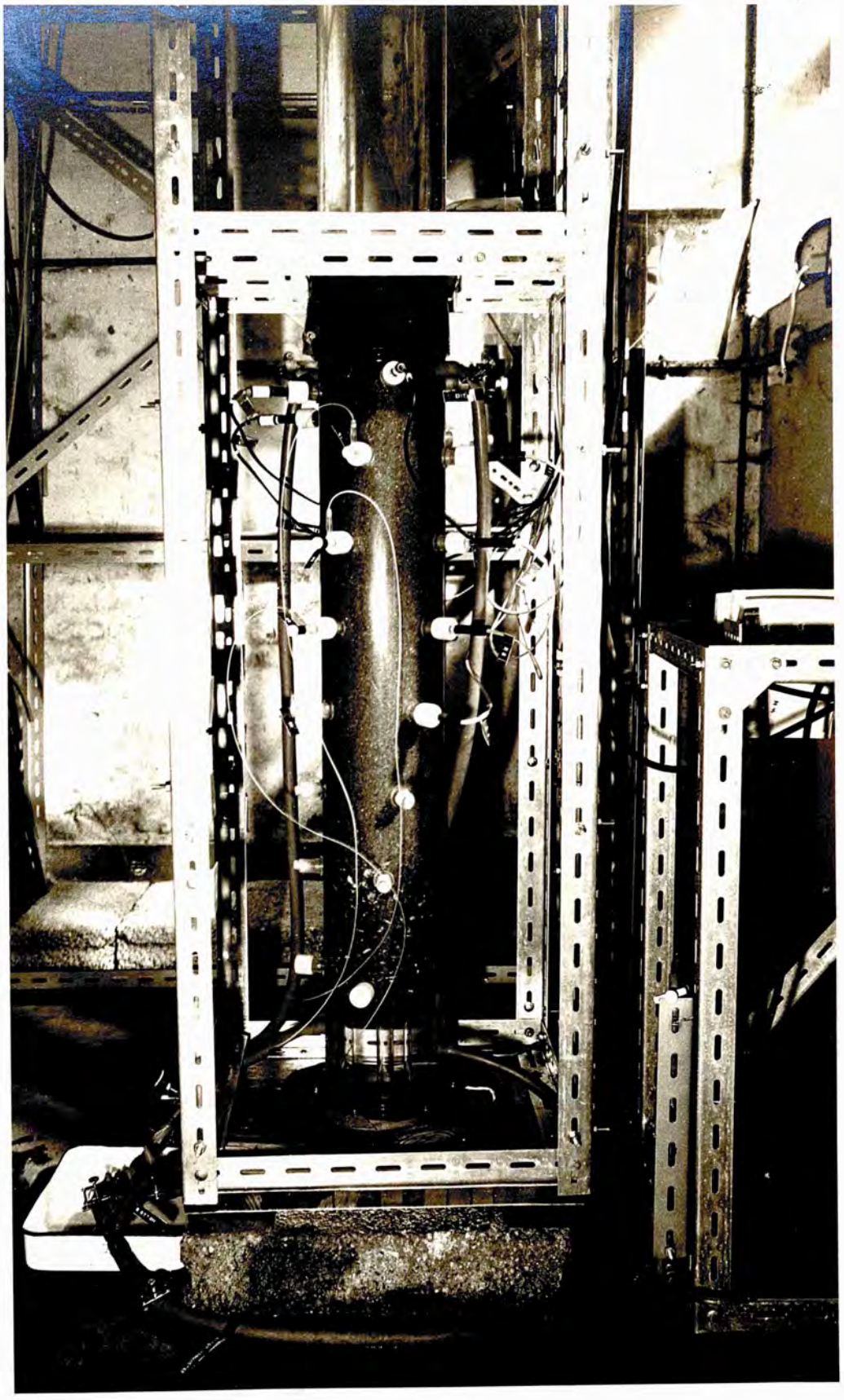


Figure 7.2 SCREW ATTACHMENT TO HOLES MADE IN PILOT FILTER WALL

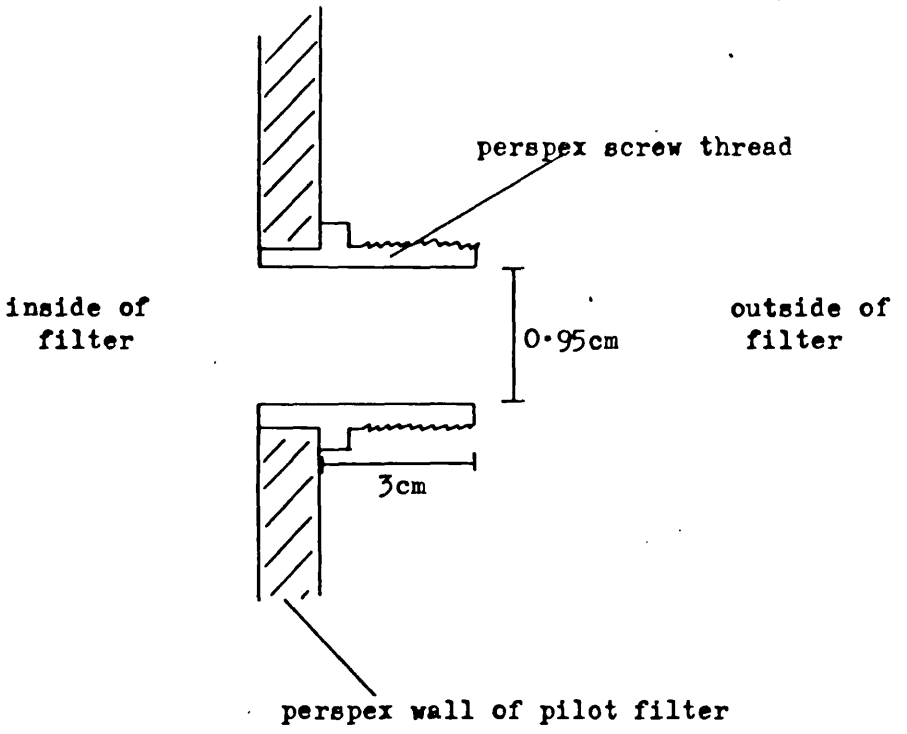
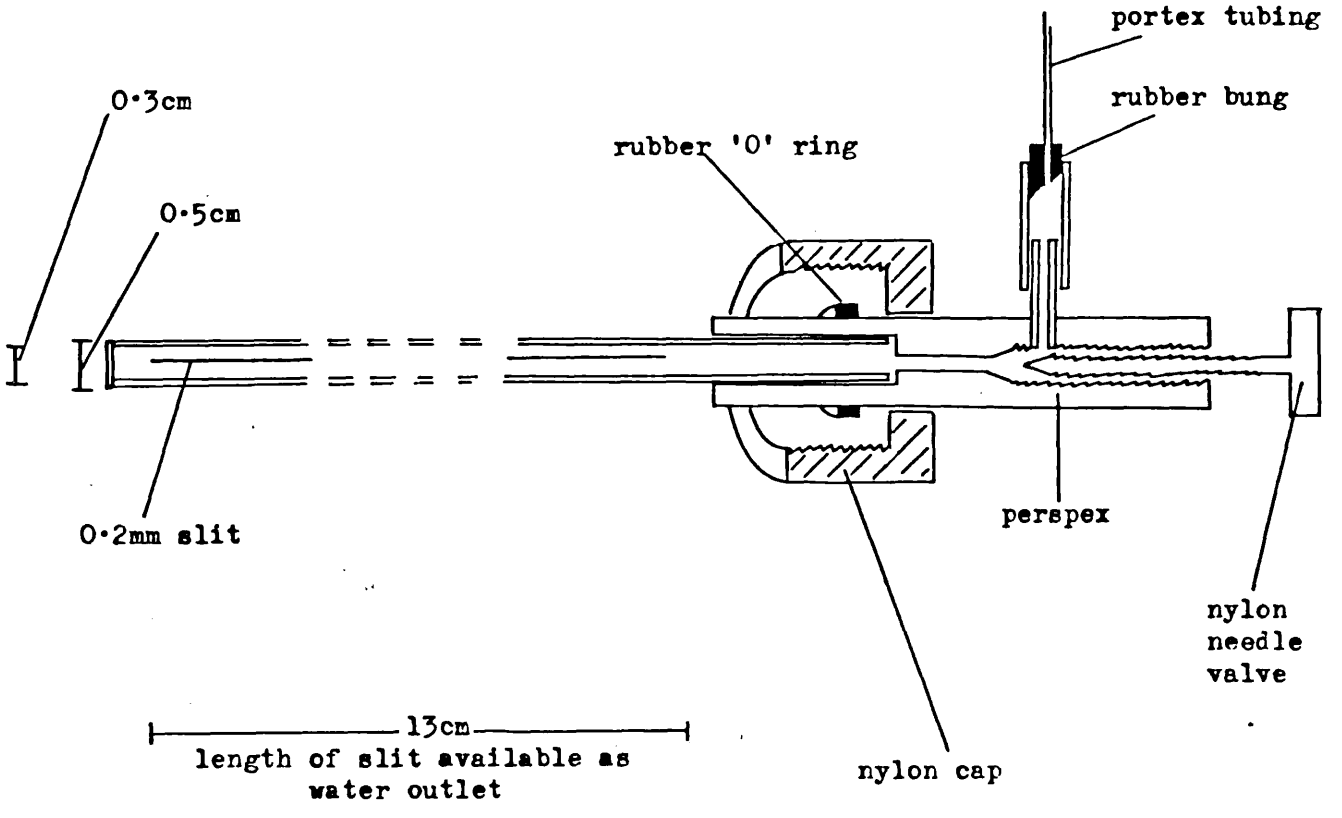


Figure 7.3 INTERSTITIAL WATER OUTLET APPARATUS



monitoring equipment at various depths in the columns with a water tight seal.

7.1.2 Outflow arrangements

The main outflow arrangement was made from one of the holes bored beneath the base plate. This was connected via rubber tubing to a flow meter which was kept permanently in position. A second outflow arrangement was made by connecting one of the uppermost pair of holes to a similar piece of rubber tubing. This outlet, however, was clamped shut throughout the running of the column and was only used to improve cleaning methods.

7.1.3 Manometric attachments

One manometric attachment was made by coupling tubing between the second of the two lowest holes in the column and a scaled manometer. The second manometric attachment was made between the second of the two uppermost holes in the column and a second scaled manometer. Both these manometers were constructed of clear p.v.c. tubing for easy reading and positioned inside an aluminium angle frame with a millimeter scale in between. It was possible to attach these manometers to the frame surrounding the column.

7.1.4 Frame and blackout arrangements

The column was placed on top of a square wooden base (45cm square, plate 4) to which was attached an aluminium angle frame. The use of the frame was for 2 main purposes; the first to provide a frame which was protective and also allowed the user to move the column around without damaging the column itself; the second purpose was to provide a frame to which pieces of blackout material and monitoring equipment could be attached.

To reproduce operational lighting conditions it was necessary to black-out the lower half of the filter which contained the sand. This was done by attaching to the frame pieces of hardboard on all 4 sides of the column in this region (plates 5 and 6). Two shaped pieces of hardboard were also positioned horizontally around the column above the sand surface. At the junction between hardboard and aluminium frame, strips of foam were stuck to the hardboard to ensure a light proof fit.

Plate 5. Pilot filter and water storage tanks.

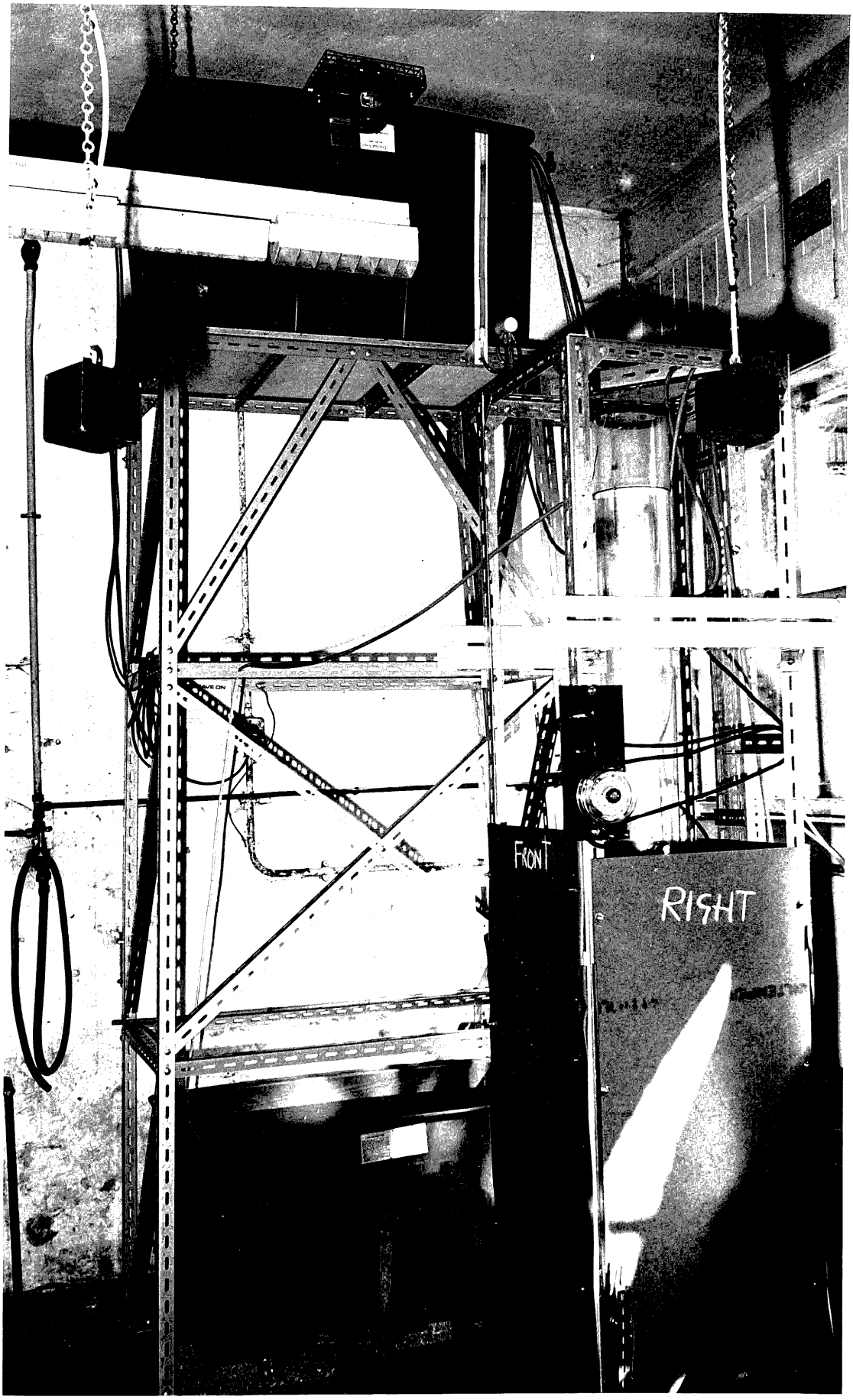
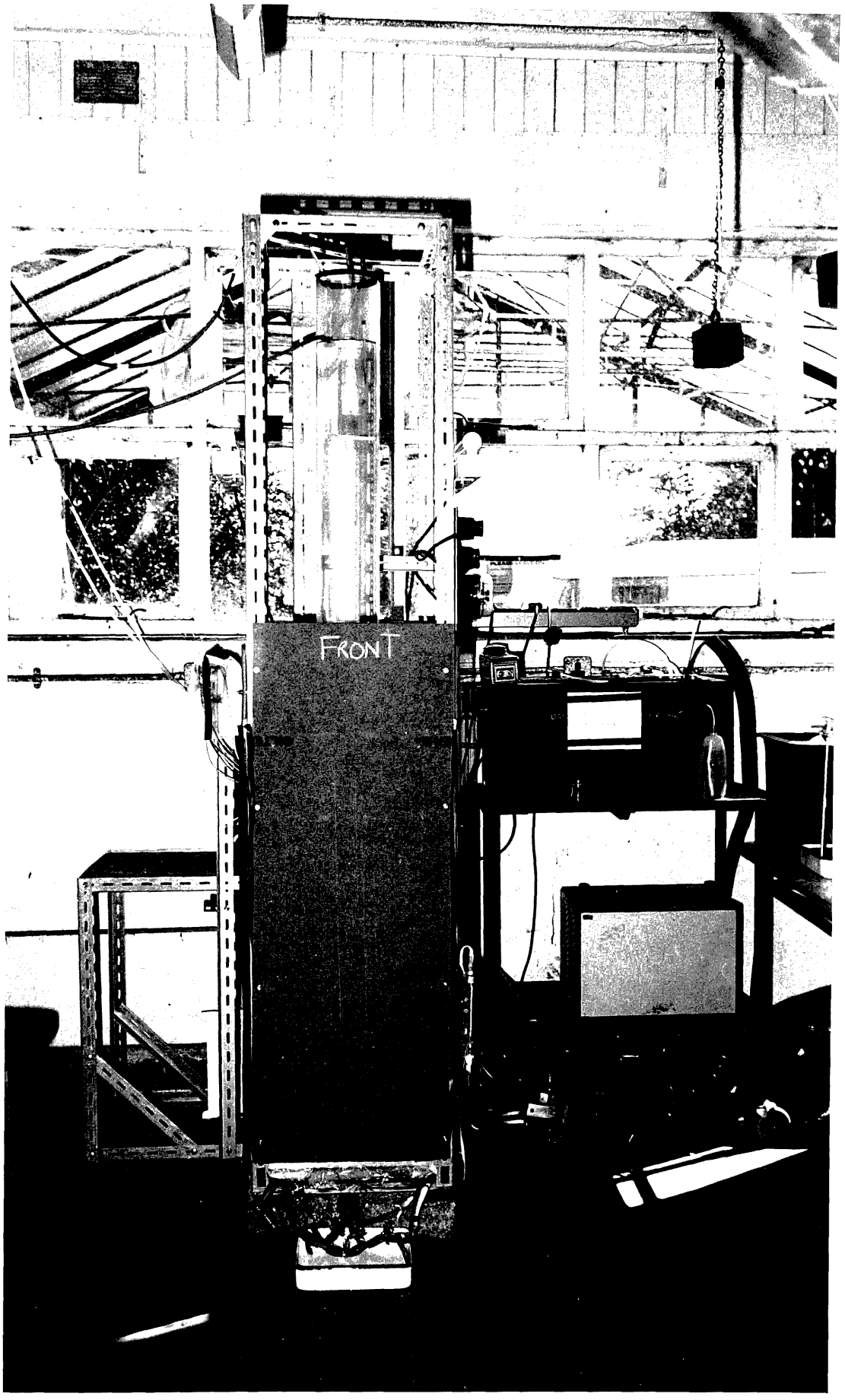


Plate 6. Pilot filter, view from front with blackout boards in position.



It was possible to ensure that all outflow pipes and manometric attachments exited from the frame at one point. This was at the bottom of the frame (plate 6) and it was possible to prevent entry of light at this point by using thick pieces of foam rubber which were packed around the tubing.

7.1.5 Lighting arrangements

For various reasons it was necessary to run the pilot filter indoors although adjacent to large windows. It was therefore necessary to supplement the amount of lighting around the upper portion of the filter and also to prevent lighting from becoming too one-sided. This was done by attaching two fluorescent grow-lux-lamps (plate 5) to the frame. These were controlled to night and day conditions by a dial switch also attached to the frame and set to switch on and off at the appropriate sunrise and sunset times.

7.1.6 Interstitial water outlet apparatus

The apparatus used consisted of a perspex tube placed horizontally in the filter through one of the holes available at each depth (figure 7.3). This tube had a 0·2mm slit cut along its length (Ives 1966) but which was closed off 0·5cm from the side of each filter wall, thus avoiding any boundary effects being measured.

It was necessary to arrange the flow rate through the tube to give a velocity at the inlet slit approximately equal to that in the pores of the filter. So it was necessary to attach a needle valve to the outflow in order that the flow rate could be controlled. The flow rate itself was determined with the use of a flow meter tube connected by tubing to the apparatus. The sampling points ran continuously as intermittent operation would cause local slowing of deposited material, thus giving unrepresentative results (Ives 1966).

If many sample points were operating throughout the depth of the filter, the quantity of filtrate may be considerably less than the influent. Ives (1966) suggested that the difference should not be more than 5%. As far as the present pilot filter is concerned it would be possible to remove interstitial water from all 11 sampling points and the total amount of water removed would be approximately 5%.

7.1.7 Velocity of water in the interstices

It was necessary to remove interstitial water for analysis at the same rate at which it was passing through the interstices. If water was removed at a faster rate then this water would be unrepresentative of water passing the extraction point at that time.

Water passing onto the surface of the filter at a known velocity will travel faster through the interstices of the filter due to its reduced porosity. In order to calculate the velocity in the interstices it is necessary to determine the porosity of the sand (section 7.1.8) which was found to be 37·02%.

If the velocity of water passing onto surface of filter be $x cm.hr^{-1}$

So, velocity in interstices will be $x.\frac{100}{37}$ $cm.hr^{-1}$

Surface area of slit in sampling unit = $13(0.02)$ cm^2

So, velocity of water passing out of sampling unit =

$$13\,(0{\cdot}02)(\tfrac{100}{37})x \quad cm^3.hr^{-1}$$

$$= 0{\cdot}0117x \text{ ml.min}^{-1}$$

It was planned to run this pilot filter at $20cm.hr^{-1}$ and so the rate at which water passed out of the interstitial water samplers was $20(0{\cdot}0117) = 0{\cdot}2340 ml.min^{-1}$.

7.1.8 Porosity of sand

In sand saturated with water, the porosity of the sand may be defined as the percentage of the total volume of the sand occupied by water. Determinations of this kind are usually carried out on dried sand and the porosity determined as the percentage of the total volume of the sand occupied by air. Assuming that in water saturated sand the water completely fills the interstices the porosity of the wet sand should be identical with the porosity of the dried sand.

Determination of the 'real specific gravity' of the sand, i.e. the specific gravity of the sand particles

A specific gravity bottle full of distilled water at room temperature ($20^{\circ}C$) was weighed (ag). Half of the water was poured out, the bottle was weighed, air dried sand added and the bottle reweighed. Weight of sand added was calculated (bg). This mixture was boiled gently to release trapped air, cooled to room temperature,

the bottle was filled with distilled water and weighed (cg). The increase in weight (c - a) is due to the difference between bg of soil and the weight of an equal volume of water. The specific gravity of the soil particles is, therefore, equal to

$$\text{R.S.G.} = \frac{b}{b - (c - a)} \quad g$$

Determination of the 'apparent specific gravity' of the sand, i.e. its specific gravity including trapped air

A specimen tube was weighed empty and filled to a predetermined mark (i) with air dried sand and (ii) with water. This was repeated twice and the apparent specific gravity was calculated :

$$\text{A.S.G.} = \frac{\text{weight of sand}}{\text{weight of same volume of water}}$$

Porosity was calculated as the percentage pore space

$$\% \text{ pore space} = 100 - \frac{\text{ASG}}{\text{RSG}} \cdot 100$$

and was determined to be 37·02% (table 7.1).

7.1.9 Water supply tanks

Water used to supply the pilot filter was collected once a week from Hampton Treatment Works. It was pump ed from the surface water of the bed with which it was being compared, into 24 x 25L polythene jerry cans. On return to the laboratory the contents of some of the containers were emptied into two supply tanks and the remainder were stored in a constant-temperature room at 8°C until required.

A system of two supply tanks was used to keep the pilot filter supplied with water (plate 5). Water was poured by hand into the bottom tank and from there it was pumped to the top tank. The top tank had an overflow pipe to pass water back to the bottom tank. A water level indicator tube was connected on the outside of the tank.

An outflow pipe from the top tank led to a peristaltic pump which ensured an even supply of water to the pilot filter. The pilot filter had an overflow pipe positioned 1m above the sand surface and any overflow was channelled back to the bottom tank. Water in the top tank was kept circulated by a motor driven propellor in order to prevent the sedimentation of particles in the water.

Table 7.1 Determination of the porosity of sand from Bed 45.

	Bottle no.		
	1	2	3
a (g)	141.5533	145.3645	141.5968
b (g)	5.0296	4.9949	4.9643
c (g)	144.7024	148.5277	144.7344
real specific gravity $\frac{b}{b-(c-a)}$	2.6746	2.7269	2.7176
apparent specific gravity	1.6699	1.6970	1.7463

Mean r.s.g. $=2.7064 \pm 0.0693$ (2.6% of $\bar{x}$), ($t=4.303$, $p=0.05$, d.f.=2)

Mean a.s.g. $=1.7044 \pm 0.0962$ (5.6% of $\bar{x}$), ($t=4.303$, $p=0.05$, d.f.=2)

percentage pore space , i.e. $100 - \frac{\text{a.s.g.}..100}{\text{r.s.g.}} = 37.02\%$

7.2 Dissolved oxygen analysis

7.2.1 Apparatus

Methods available for the analysis of dissolved oxygen in water include a modification of the standard Winkler (1888) method described in many textbooks (e.g. Golterman 1969, H.M.S.O. 1972). An alternative is the use of an oxygen electrode of the Mackereth (1964) type, employing a galvanic cell covered in a polythene membrane. The disadvantage with the latter method is that it consumes small quantities of oxygen in operation and so is only of use where large quantities of water or fast rates of flow are available. As has been pointed out in 7.1.7, the flow rate of water from the interstitial water samples is very low and so the use of a Mackereth electrode would not be appropriate in this case. The use of the syringe technique of Fox and Wingfield (1938) could not be employed in this situation where the rate at which water is removed from the interstices is of primary importance. It was obvious that a different technique should be employed in order to sample interstitial water for oxygen analysis.

The method developed involved a series of bottles into which water leaving the interstitial water samplers was directed (figure 7.4 and plate 7.1). The bottles each consisted of a lower and upper half. The lower half was manufactured from Quickfit socket joints, cut off and sealed approximately 6cm (smaller bottle) and 14cm (larger bottle) from the socket opening. The upper half was made from Quickfit conical joints cut off approximately 5cm from the cone opening. This upper half was sealed with a small rubber bung through which 2 holes had been bored and contained fine polythene tubing. The lower end of the bung had been cut at 45^{o} and the tubing arranged so that the tube which carried water leaving the bottle was positioned at the higher level of the slant, in order to channel any air bubbles out of the bottle.

The smaller bottles contained approximately 2·7 - 3ml each, while the larger bottle contained about 10ml. The purpose of these series of bottles was to collect water as it travelled out of the interstices at the correct rate. The reason for the larger third bottle was to indicate that when this bottle was full, each of the smaller bottles had been flushed through three times. This is a standard precaution used when water for dissolved oxygen analysis is

Figure 7.4 APPARATUS USED TO COLLECT WATER FROM INTERSTITIAL SAMPLING UNITS

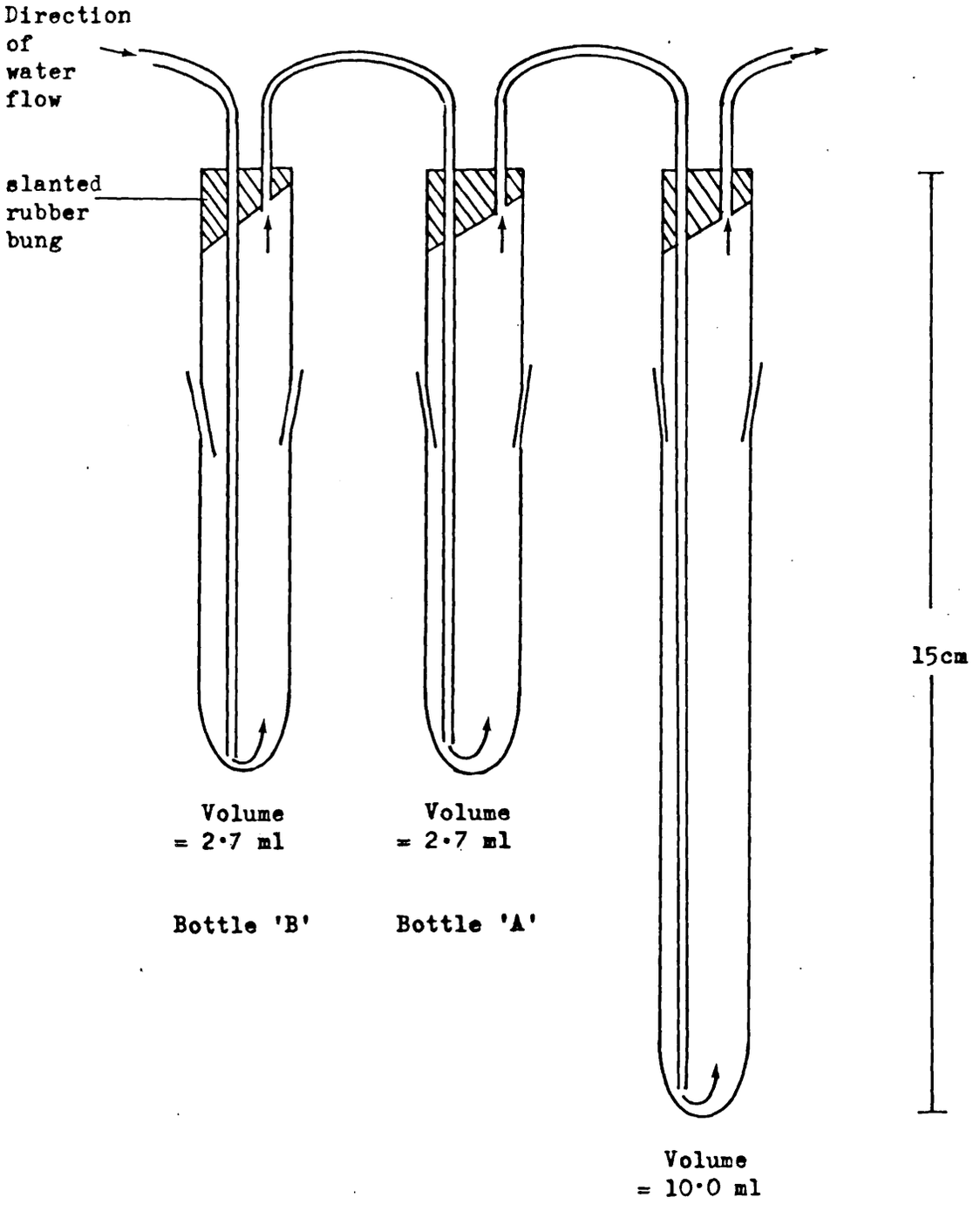


Plate 7.1 Top- interstitial water outlet apparatus.
Bottom- water collecting apparatus.

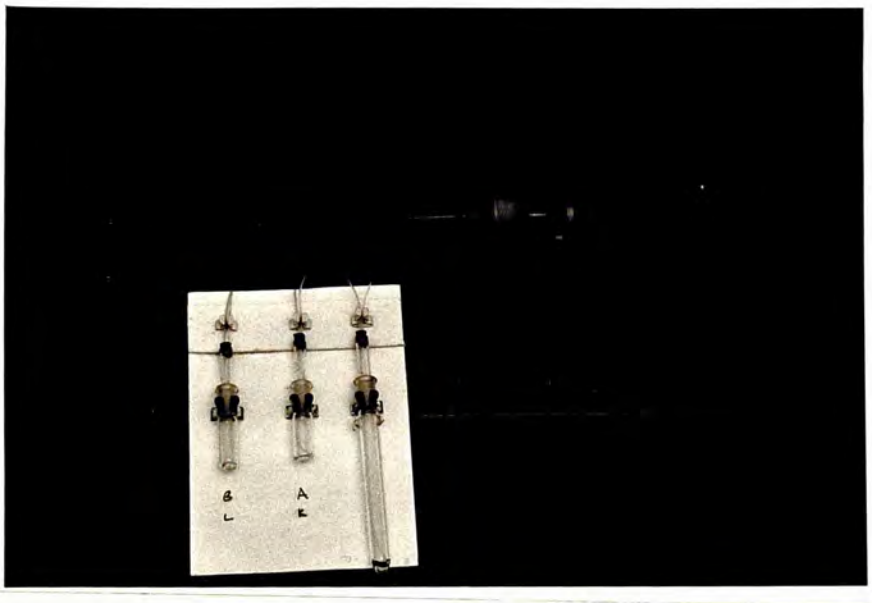
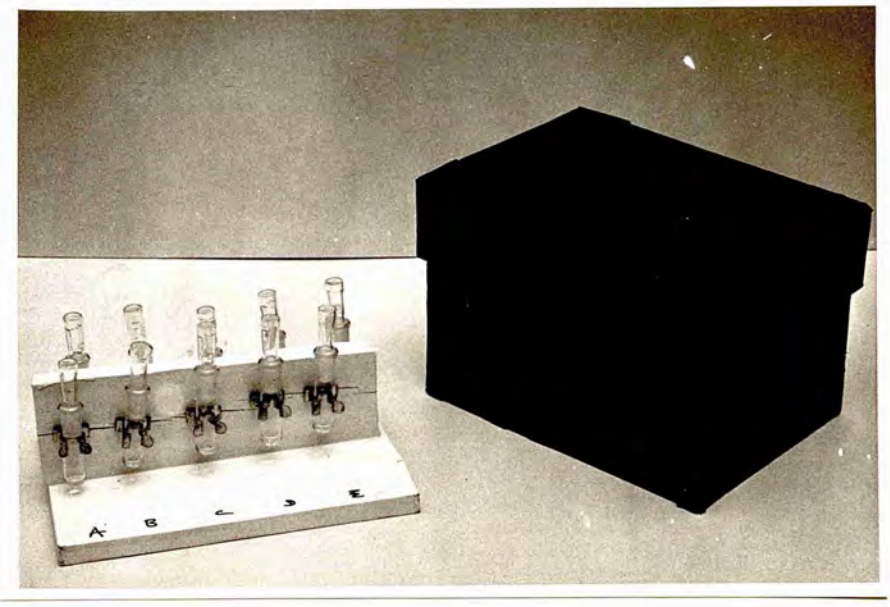


Plate 7.2 Water bottle holding frame and blackout box.



collected in a way in which it initially comes in contact with air (Kamler 1969, and recommended by Golterman 1969, H.M.S.O. 1972).

It was possible to mount these bottles on a wooden frame (plate 7.1) which was screwed to a piece of angle iron, which could then be attached inside the frame of the pilot filter (plate 8). A series of these frames were attached to the pilot filter frame so that it was possible to collect water from 5 of the interstitial water sampling units.

A light-proof curtain, made from a sheet of aluminium foil protected by a covering of polythene, hung between the racks of bottles and the pilot filter itself. This was to prevent entry of light into the filter when the bottles were being adjusted and the blackout board was removed. The blackout board was normally screwed into position (plate 6), thus sandwiching the bottles between the blackout board and blackout curtain.

7.2.2 Technique of obtaining samples of water

The apparatus was set up as in plate 7.1 and attached to the pilot filter as in plate 8. A check was made of the flow rate of each interstitial water sampling unit which was then connected to the appropriate water collecting apparatus. The blackout board was then replaced and the apparatus left for the bottles to fill with water. At the end of the appropriate period the blackout was removed.

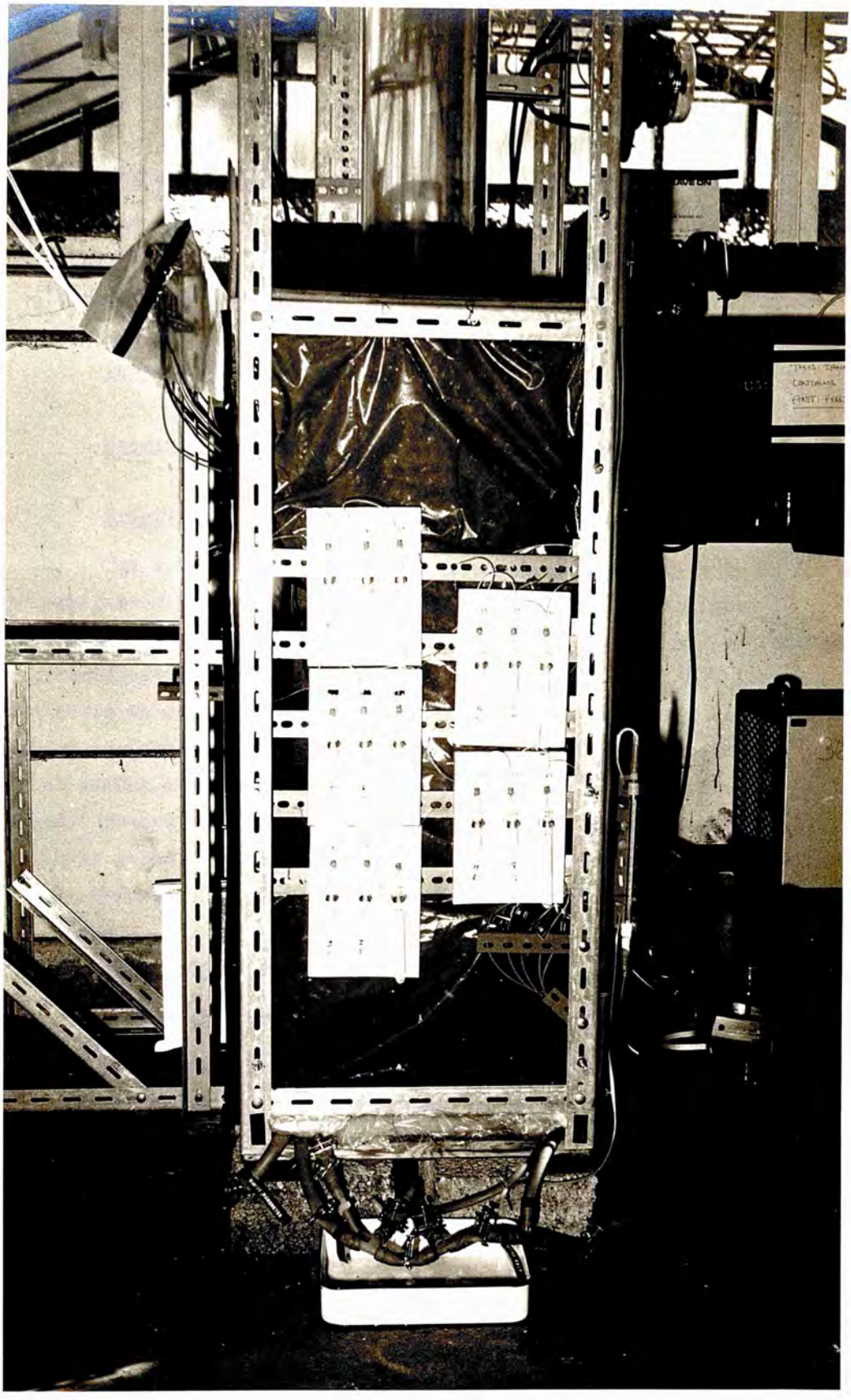
Before each of the smaller bottles was stoppered it was necessary to disconnect them from the apparatus in a precise series of moves to prevent entry of air.

Although the bottles (figure 7.4) filled in order of bottle 'B', bottle 'A', followed by the large bottle, bottle 'A' must first be disconnected by removing the cork. If bottle 'B' was first disconnected then a siphon effect caused by the large bottle would draw air into 'A'.

After the cork was removed the upper portion of the bottle was disconnected from the bottom half. This prevented any retention of water in the upper portion of the bottle and completely filled the neck of the lower half. The bottom half was held at a slight angle and a Quickfit stopper pushed in with a twisting motion and when done carefully no air bubbles formed. Each bottle was then placed in a rack (plate 7.2) contained inside a blackened light-proof box.

This procedure was repeated on bottle 'B' and then on the

Plate 8. Pilot filter, view from front with blackout boards removed illustrating interstitial water collecting apparatus.



remaining bottles in the series. When all bottles had been stoppered a second series of bottles was attached to the water collecting devices. So, in plate 7.1, bottle K replaced A and bottle L replaced B. The blackout was replaced and the bottles allowed to fill.

7.2.3 Method of chemical analysis

The method of analysing the water contained within the bottles is a scaled-down version of the basic Winkler method, as described by Golterman (1969) and H.M.S.O. (1972).

Apparatus (appendix 7.2)

Reagents (appendix 7.3)

Principle

The formation of a precipitate of manganous hydroxide is brought about by bringing together Winklers A and B solutions. Oxygen present in the water sample combines to form higher hydroxides. On acidification, in the presence of iodide, these react to liberate iodine in an amount chemically equivalent to the original dissolved oxygen content of the sample. The iodine is then determined by first adding excess sodium thiosulphate, which is then titrated against potassium biniodate solution. The end point is located by a simple potentiometric titration. Nitrate interference is eliminated by the use of sodium azide added to Winklers B solution.

Procedure (figure 7.5)

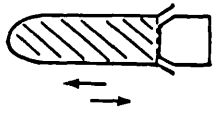
1. Remove stopper from bottle.
2. Add 70 µl each of Winklers A and B solutions, careful not to use blowout facility of Oxford pipettor as this will introduce air.
3. Replace stopper and shake.
4. Allow precipitate to settle to ⅓rd volume.
5. Shake bottle again.
6. Allow precipitate to settle to ⅓rd volume.
7. Add 140 µl acid.
8. Replace stopper and mix.

Figure 7.5 METHOD OF MICROWINKLER DISSOLVED OXYGEN ANALYSIS

1. Remove stopper

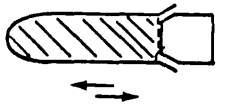
2. Add 70 µl each Winkler's A and B solutions

3. Replace stopper and shake

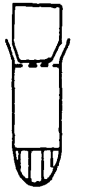


4. Allow precipitate to settle to ⅓rd volume

5. Shake



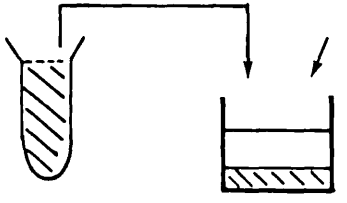
6. Allow precipitate to settle to ⅓rd volume



7. Add 140 µl acid

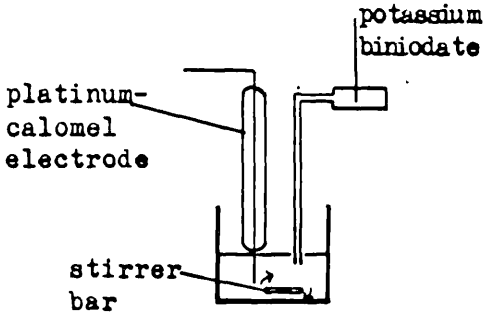
8. Shake

9. Remove 1000 µl aliquot to titration vessel

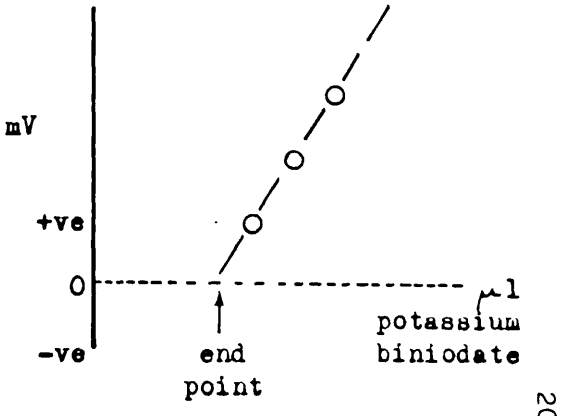


10. Add 4000 µl excess thiosulphate

11. Titrate against potassium biniodate



12. Extrapolate back to zero line to obtain end point



9. Remove 1000 µl aliquot with Oxford pipettor and place in titration vessel.

10. Add 4000 µl excess sodium thiosulphate.

11. Place vessel on magnetic stirrer, add stirrer bar and combination electrode and titrate against potassium biniodate to end point reached using a potentiometric circuit (figure 7.6).

12. Approach of the end point is indicated by "flickering" of the meter needle. Add more biniodate until a constant mV reading is obtained. Run in more biniodate until 2 more steady readings are recorded. Draw on graph.

13. Extrapolate back to zero line to obtain end point.

Potentiometric end point determination

The end point of the titration of potassium biniodate against excess sodium thiosulphate is located by a simple potentiometric circuit (figure 7.6)(Talling 1973). The combination electrode is lowered into the stirred solution containing excess thiosulphate, the potential recorded is zeroed using the variable back off potential or "buffer control" of the pH meter. Potassium biniodate is added from a Metrohm burette. A constant stirring speed is essential. After the end point is reached the volume of excess biniodate added is directly proportional to the potential difference, recorded in mV, up to a certain point where the relationship ceases to be linear (figure 7.7). The slope of the linear relationship is dependent on the resistance induced across the platinum and calomel reference electrode, the slope increases with increasing value of the resistance. In this case a resistance of 27KΩ was used. It is important to obtain all mV readings within the area where the relationship is linear, otherwise erroneously low values at the end point will be recorded in this case readings greater than 1·4mV were not accepted.

Just before the end point is reached, the mV recorder "flickers" indicating increases in potential, but after a period of time the meter settles down to zero again. This is due to incomplete mixing of the solution. The first steady potential reading should be recorded, together with the amount of biniodate added. By obtaining two more mV and biniodate volume readings it is possible to use the information to extrapolate back to the "zero potential" line, where the end point is located.

Figure 7.6 CIRCUIT DIAGRAM OF APPARATUS USED TO DETECT END POINT IN WINKLER-DISSOLVED OXYGEN TITRATIONS

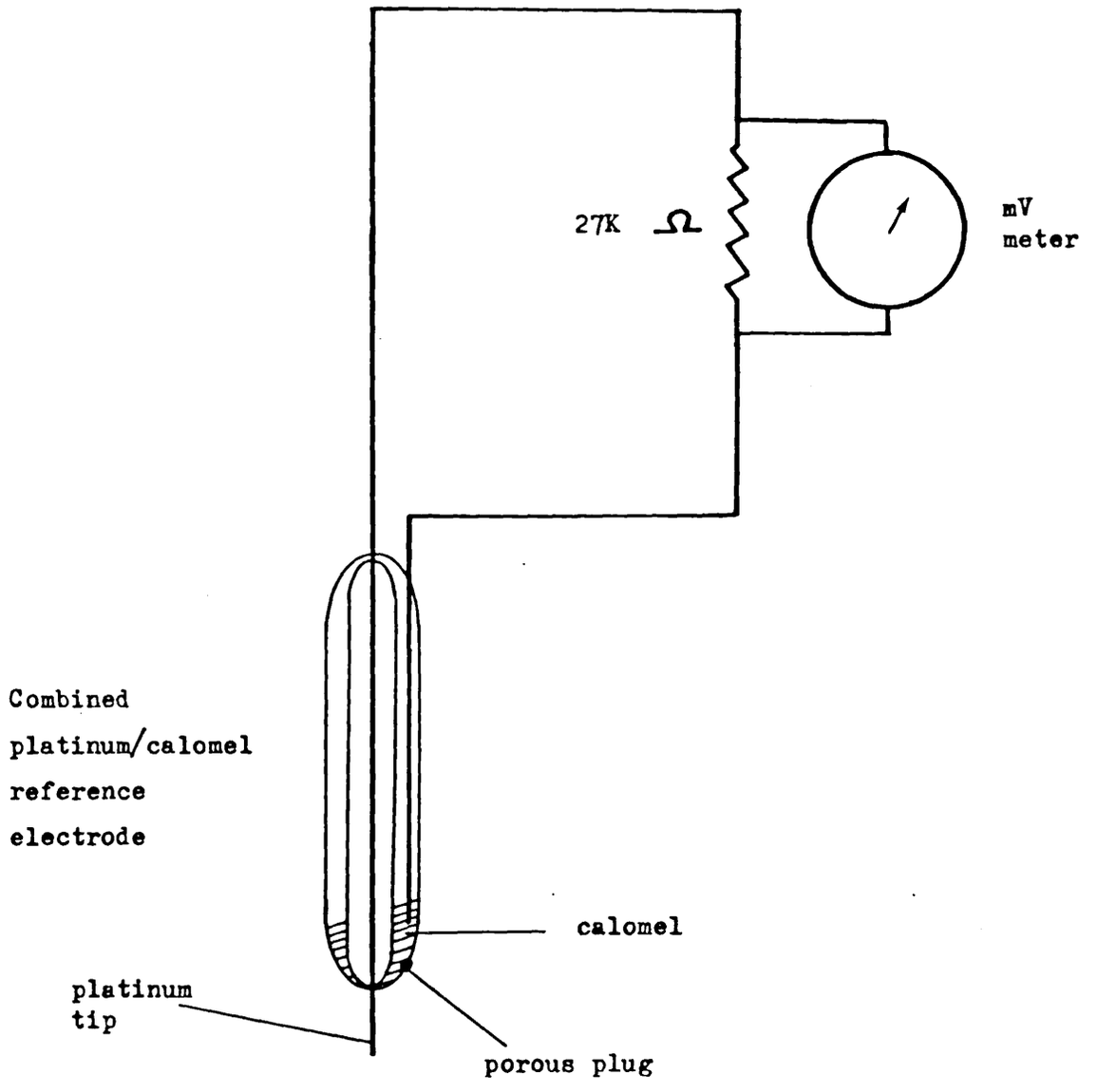
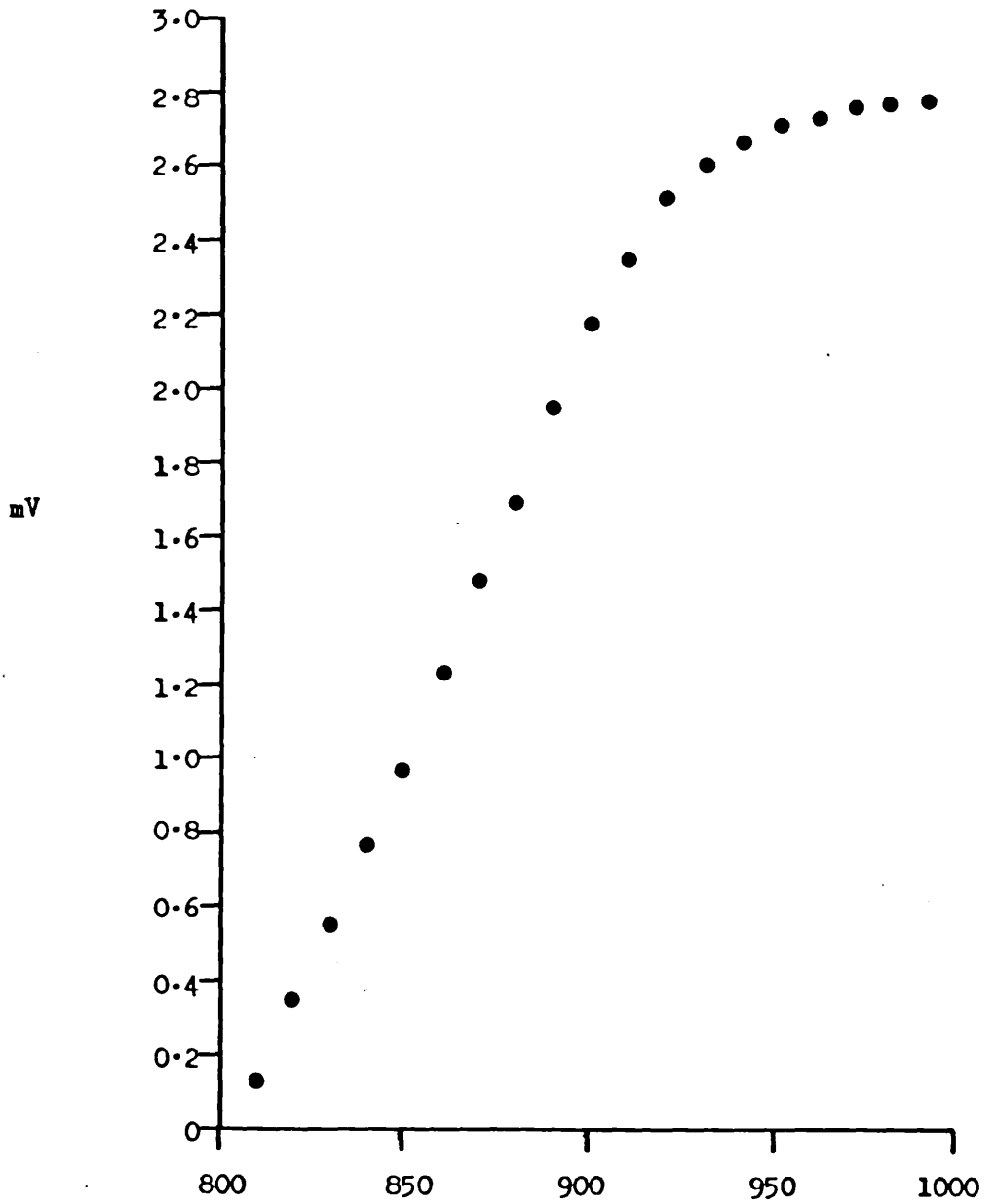


Figure 7.7 TITRATION OF EXCESS SODIUM THIOSULPHATE AGAINST POTASSIUM BINIODATE, PAST THE END POINT



For drifting and loss in sensitivity problems see section 4.3.

Calculation

Using the relationship 1ml 0·0125N thiosulphate is equivalent to 0·1mg O_2, the amount of dissolved oxygen in the sample can be determined.

$$\text{oxygen mg/L (uncorrected)} = \frac{8000}{A}(nT)(T - \frac{nB}{nT}.X)$$

where A = aliquot volume, μl
T = volume of sodium thiosulphate added (μl)
nB = normality of potassium biniodate
nT = normality of sodium thiosulphate solution (regularly determined by titration with potassium biniodate)
X = volume of potassium biniodate added at end point (μl)

It is necessary to correct this value in order to take account of the percentage displacement of Winkler reagents added. In order to do this each collecting bottle and stopper was labelled so that the same combination could be used each time. The volume of each bottle was calculated by filling with water, stoppering and weighing. The displacement volume caused by addition of reagent was then calculated for each bottle. In general, with a bottle of 2·7 to 3ml the percentage displacement was about 5%. In standard dissolved oxygen analyses of this kind the percentage displacement is usually 2%. In this case it was impossible to reduce the volume of reagents added, as it would have made it impossible to stopper the bottle, after the addition of reagents, without including some air bubbles in the bottle.

There is a possibility that dissolved oxygen in the Winkler reagents may be added to the water sample on addition of these reagents. To investigate this the salinity of the Winkler reagents was calculated. There is a relationship between salinity (measured as the total amount of solid material in grams contained in 1 litre of water, i.e. $^{0}/_{00}$), temperature and oxygen content of the water (Sverdrup et al, 1942). The salinity of the combined Winklers A and B solutions was then calculated to be $753\,^{0}/_{00}$. From the nomogram produced by Richards and Corwin (1956) it can be seen that even at

$0^{o}C$, with the salinity of Winklers A and B solutions being so high, the concentration of dissolved oxygen present can be taken as zero.

Precision of method

It was possible to obtain only two replicate dissolved oxygen measurements from one sample bottle. However, it was possible to obtain a measure of the precision of the method over a range of dissolved oxygen levels by analysing results obtained in the 24 hour run analyses (appendix 7.4). From 20 analyses it can be seen that the mean precision (standard deviation as a percentage of the mean) was 1·460%, with the 95% confidence limits being 0·981 - 1·939%. So the precision of the method is less than ± 2%, which agrees well with precision values published by Stainton et al (1977), of ± 2% using the standard (large volume) method and Fox and Wingfield (1938) of ± 2% using the syringe (small volume) method.

7.3 Redox potential measurements

7.3.1 Principles involved

Organisms obtain the energy they need for their vital processes through a series of chemical reactions involving the transfer of electrons from substances which serve as sources of energy to substances which may become products of metabolism. If the organisms are respiring aerobically the final electron acceptor is oxygen, in which case there is a reduction of molecular oxygen to water :

$$O_2 + 4H^+ + 4e^- \longrightarrow 2H_2O$$

Oxygen reaching the benthos and regions of high detrital organic matter is consumed by facultative and obligate aerobic organisms. As a result of the fact that they can use oxygen as an electron acceptor they can utilise electrons more effectively than anaerobic organisms and are able to reduce the oxygen content to a low level. Without the action of these organisms there would be no significant alteration in the oxygen content of deeper waters. So oxygen depletion requires the presence of both facultative and obligate aerobic organisms and oxidisable, usually organic, substrates.

When the oxygen supply is limited, the diffusion rate from another source may be insufficient to maintain the supply for aerobic organisms. (This is more marked in static water conditions, but

would be less in the slow sand filter situation where a constant influx of water is carried into the interstitial system). Consequently a proportion of organisms make use of electron acceptors other than oxygen for their respiratory oxidations and so a new population of anaerobic organisms will build up. The result is the conversion of numerous compounds into a state of chemical reduction and is reflected in a lowering of the oxidation - reduction (redox) potential of the system.

Some of these alternative electron acceptors contain oxygen, such as nitrates and sulphates :

$$NO_3^{2-} + 2H^+ + e^- \longrightarrow NO_2^- + H_2O$$

$$2NO_2^- + 8H^+ + 6e^- \longrightarrow N_2 + 4H_2O$$

$$SO_4^{2-} + 10H^+ + 8e^- \longrightarrow H_2S + 4H_2O$$

Some high valency cations will accept electrons and become reduced to a lower valency state :

$$Fe(OH)_3 + 3H^+ + e^- \longrightarrow Fe^{2+} + 3H_2O$$

$$MnO_2 + 4H^+ + 2e^- \longrightarrow Mn^{2+} + 2H_2O$$

Finally the hydrogen ion itself can accept an electron to become hydrogen gas :

$$2H^+ + 2e^- \longrightarrow H_2$$

A second consequence of the activity of organisms under conditions of oxygen deficiency is that organic nutrients are no longer fully oxidised to carbon dioxide and water. Instead, intermediate products are excreted or stored, such as simple fatty acids, alcohols and ketones. These organic compounds are then further decomposed by other organisms, with the production of carbon dioxide and water, methane and other hydrocarbons and sometimes hydrogen gas. The diffusion interface between reduced products and dissolved oxygen then becomes the site of chemosynthesis. With adequate light this interface may become a site of intense photosynthesis by reasons of increased nutrient diffusion, due to mineralisation and availability.

The term "detrital electron flux", used by Wetzel and Rich (1973) and Wetzel et al (1972), describes the fact that electrons, and not carbon, represent the actual continuity in this case and that they are the ultimate trophic medium of exchange following photosynthesis.

The redox potential of a system is a measure of its tendency to receive or supply electrons and is governed by the nature and proportions of the oxidising and reducing substances which it contains. The Nernst equation is used to describe the relationship between oxidised and reduced components of the system being measured :

$$Eh = E_o + \frac{R.T.}{nF} \ln \frac{ox}{red}$$

where Eh is the emf of an oxidation - reduction system referred to the hydrogen electrode

E_o is a constant for the system being measured, i.e. the standard redox potential of the couple being measured, at the ambient temperature and pH, and is equal to the mid point redox potential which is displayed when its oxidised and reduced forms are present in equal concentrations

R = gas constant

F = Faraday constant

T = absolute temperature

n = number of electrons taking part in the system

The tendency of a system to gain or supply electrons can be observed quantitatively by measuring the potential at an unattackable electrode, e.g. platinum, which when immersed in a system takes on the electrical potential of that system. If the half cell formed by the immersed platinum electrode is electrically coupled to a standard half cell, e.g. the hydrogen electrode, also in contact with the redox system, a cell is formed. In normal use the hydrogen electrode is replaced by other standard half cells, whose own potential, relative to the hydrogen electrode, is known. In this case the reference electrode used was the Ag/AgCl electrode.

Redox values are chiefly of use in characterising the 'aeration' of the system being measured, as it is possible to define the potentials at which a number of important chemical changes in equilibria occur. There are, however, variations in the literature

as to the precise potential at which these changes occur.

It is important to remember that redox potential is a measure of intensity level and not capacity. In this it resembles temperature and pH and just as temperature and pH give no information as to the heat capacity or buffering power, so redox potential is independent of the "poising effect", i.e. the capacity term in oxidation - reduction potentials.

It has been suggested by Pearsall and Mortimer (1938) that the organic matter, or some associated system, present in a system may exert a poising effect which assists the establishment of a more or less stable potential below +350mV. In clean river sand they found it difficult to get a steady potential above (E_5) + 350mV. They found that marked instability seemed to be associated with the potential range at which products of oxidation could be shown to exist and so appeared likely to be associated with the presence of atmospheric oxygen.

To summarise, oxygen is used as an electron acceptor by most aerobically respiring organisms. When the oxygen is used up and is not replaced by simple diffusion, facultative microorganisms move to nitrates, etc., as electron acceptors, reducing these to nitrites and ammonia. So a population of anaerobically respiring organisms builds up. The increase in reduced forms is manifest in changes in the redox potential (Eh) of the system, which is a measure of the relative oxidised and reduced forms present. The overall effect is a reduction in Eh, hence an increase in reducing conditions, changes in the population composition of microorganisms and consequently in their predators. Another effect is an increase in mineralisation which can cause greater productivity of algae, should these nutrients get back into photic zones, e.g. by water turbulence.

So redox potential measurements should therefore complement dissolved oxygen measurements and, in regions where dissolved oxygen measurements are very low or absent, will provide additional information about the overall state of reduction of the system.

7.3.2 Methods of obtaining redox measurements

Redox potential measurements were made with the use of a combination platinum Ag/AgCl electrode (Russel pH Ltd., Auchtermuchty CMM/ROD). These were placed through access holes into the pilot

filter and reached about 6cm into the body of the filter. A water tight seal was achieved with the use of a nylon cap as used with the interstitial water samplers (figure 7.3). Potential readings were read from a digital millivoltmeter (Solartron Electronics, digital multimeter No. 201013) and were recorded when the drift was less than $1mV.min^{-1}$ (Hargrave 1972c).

The measured redox potential recorded by the probe needed to be adjusted for two reasons.

1. Due to slight variations from probe to probe in values recored in the system. Prior to placing the redox probes in the pilot filter it is necessary to record their potential readings when placed in solutions where the theoretical potential differences between Pt electrode and the solution was known. This was done by saturating a known buffer solution with a little quinhydrone, immersing the combination electrode in it and recording the steady potential difference reached.

Values of Eh vary according to the prevailing pH conditions. The majority of authors (Pearsall and Mortimer 1939, Mortimer 1941, Hutchinson 1957) assume there is an increase of +58mV in e.m.f. for each unit of pH decrease. Edwards (1958) uses +60mV and Armstrong (1975) quoting Ponnamperuma et al (1966) uses +59mV. The method for testing redox probes suggested by Metrohm A.G. Co. also suggests +58mV and so this was adopted.

So a buffer solution saturated with quinhydrone solution produces a potential difference between platinum probe and solution of +267mV at pH 4·0 and +93mV at pH 7·0. The deviation from theoretical e.m.f. which each probe produced when immersed in these solutions was then recorded and used to adjust the values of e.m.f. recorded by the probes in the pilot filter. In each case the deviation from theoretical value was only a few mV.

2. Values of redox potential recorded are usually eventually quoted as Eh, i.e. the e.m.f. of an oxidation - reduction system referred to the hydrogen electrode. This is because the recorded potential of a system varies according to the type of reference electrode used. In this case a Ag/AgCl reference electrode was used. This type of electrode is useful in that it has a negligible e.m.f. - temperature hysteresis (Mattock 1963), i.e. there is a more or less direct relationship between e.m.f. and temperature. The standard e.m.f. of the cell, H_2/HCl/AgCl/Ag as a function of temperature varies slightly from author

to author. Ives and Janz (1961) quote values from 3 authors :

T °C	Harned and Ehlers	Bates and Bower	Harned and Paxton	Recommended values
0	236·42	236·55	236·52	237mV
5	234·00	234·13	234·05	234
10	231·34	231·42	231·37	231
15	228·54	228·87	228·49	228
20	225·58	225·57	225·49	225
25	222·46	222·34	222·39	222

Values used in this investigation are quoted as the 'recommended value' above. It can be seen that the relationship between e.m.f. and temperature is linear $E = 237 - 0{\cdot}600T$, where E = recorded e.m.f. of cell and T is the temperature in degrees centigrade.

So, it is necessary to calculate the standard e.m.f. of the Ag/AgCl - hydrogen cell at the known temperature, using the equation and to add this on to recorded e.m.f. using the Pt - Ag/AgCl probe to obtain the final Eh value.

As was shown above, the measurements of redox potential are affected by pH. It is sometimes, therefore, recommended to correct Eh values to a stated pH, often this is pH7, in which case the e.m.f. is referred to as E_7. This is done by using the relationship that Eh increases by +58mV for each unit of pH decrease. However, some authors (Hutchinson 1957, Edwards 1958, Ponnamperuma 1972, Armstrong 1975, Golterman 1975) question the validity of this correction and consider that it may be introducing an error as the correction factor is variable according to the nature of the redox couple involved. Also the adjusted potential is not the potential which actually operates in the system, so adjustments of this kind are best used when comparisons need to be made from one system to another. In this study the Eh recorded by the probes was used and not corrected to pH7·0.

7.4 Preparation of the pilot filter prior to experimental use

1. The filter was cleaned by soaking in RBS 25 solution over a period of 24 hours .
2. It was then emptied and thoroughly washed with tap water.
3. All access holes were stoppered.

4. Manometers were connected.

5. Column was backcharged with water from the top tank until it just covered the base plate. This was to prevent the formation of air bubbles below the plate.

6. Gravel was added to the filter to a depth of 15cm.

7. Sand was then added to a depth of 65cm.

8. Backcharging continued until the surface of the sand was covered to a depth of 10cm.

9. Backcharging then ceased and the surface of the sand was made flat with the use of a flat piece of perspex attached to the end of a rod introduced into the body of the filter from above. This smoothed over the surface of the sand. By this time the surface of the sand had settled to a total depth of 62cm or so.

10. Water was introduced into the top of the filter and the pilot filter was allowed to filter for a period of 4 hours to allow sand to compact. It was then about 60cm deep. It was very important to allow the sand to settle before any monitoring equipment was added as the combined weights of water and settling sand could very easily break any probes placed in the sand before it had fully settled.

11. The filter was then drained to just above the gravel level.

12. The holes in the side of the filter were then unstoppered. The perspex interstitial water samplers were gently pushed into position across the whole width at the chosen depth and the seals made water tight. Previous to this the interstitial water samplers had been immersed in water and any bubbles inside them removed and the needle valve closed.

13. A glass rod was used to make a hole in the sand filter where the redox probes would be positioned. Great care was needed here to remove any sand grains from the area of the access hole, or the glass probe would be easily shattered. The probe was then placed in the hole and the seal made water tight.

14. The pilot filter was backcharged as before, this was stopped when the water reached 10cm above the surface of the sand, then water was let into the pilot filter from above.

15. Blackout boards were added, the lights switched on and the clock set to the correct time.

16. The filter was allowed to run overnight to stabilise before the interstitial water samplers were put into action. Sections 1 - 15 would take a whole working day.

17. The following morning the outlets of the interstitial water

samplers were connected to the flow meter and the needle valves of each adjusted to provide the correct flow rate. The samplers were run continuously at these rates. At this stage the water was run to waste.

7.5 Methods for removal of sand samples for carbon and meiofaunal analyses

At the end of a pilot filter run it was possible to remove sand samples for analyses by two methods. First it was necessary to drain the filter of water and remove the monitoring equipment.

1. It was possible to push a corer into the holes vacated by the monitoring equipment and to extract small quantities of sand. As it was possible to remove only small volumes of sand these samples were used for carbon analysis. It was possible to measure the volume of sand removed here and thus determine $\mu g.C.cm^{-3}$ present at the various depths.

2. By carefully tilting the pilot filter it was possible to push a peat borer into the sand. This would penetrate a depth of 10cm and could then be removed. It was possible then to section the sand for meiofaunal analysis. The emptied borer could be used to remove more sand by pushing into the same hole and so on until a total depth of 30cm was examined.

7.6 Choice of working filter used for comparative purposes

It would have been desirable to compare the results of the pilot filter with data obtained from bed 45 run at the same rate. However, this was not possible as at the time the pilot filter became available to be used, the flow regime on bed 45 had been considerably altered and faster filtration rates had been used. This meant that the state of bed 45 in August, 1978, could not be comparable with the bed in 1975/76. To overcome this problem an alternative working bed, number 5, was used for comparative purposes. This had a history of being run at lower rates and so was more suitable for the purpose. It was intended that bed 5 should be run at $20cm.hr^{-1}$ and so arrangements were made for the pilot filter to operate at this rate. In the event, however, the operational bed was run at between 12 and $16cm.hr^{-1}$ (figure 8.1), a fact which could have caused some disagreement in results obtained from the two sites.

It was necessary to fill the pilot filter using sand which had

been removed from bed 5. After the bed had been drained and cleaned a hole was dug down to the level of the gravel layer. A representative sample of all depths was removed, together with a sample of gravel, and returned to the laboratory. The remaining hole was filled with washed gravel and sand.

Several inner and outer sampling cores were placed in holes dug in the bed (as in section 3). This time, however, sand removed from bed 5 was used to fill the cores. It was possible to use bed 5 sand on this occasion, as opposed to washed sand from the storage bays, as only one run was planned and fewer cores were placed in the bed. Thus it was possible for more direct comparisons to be made between data obtained from the pilot filter and bed 5 itself. The sampling program was as follows :

DAY	BED 5	PILOT FILTER	COMMENT
0	9/8/77	9/8/77	Filtration began
1	10/8/77		Cores removed from bed
3		12-13/8/77	1st 24 hour run on pilot filter
10		19-20/8/88	2nd 24 hour run on pilot filter
36		14-15/9/77	3rd 24 hour run on pilot filter
38		16/9/77	Pilot filter drained, sand samples removed for analysis
42	20/9/77		Cores removed from bed for analysis

CHAPTER 8 PILOT FILTER - RESULTS AND DISCUSSION

8.1 Head loss and flow rate data

Head loss and flow rate data for the pilot filter is available in appendix 8.6 and figure 8.1, the flow rate was maintained at 20cm.hr^{-1}. Equivalent Thames Water Authority data for bed 5 is plotted in figure 8.1. Unfortunately bed 5 was not run at 20cm.hr^{-1}, but at a slightly lower rate of between 12 and 16cm.hr^{-1}. It can be seen that the pilot filter was operating at head losses lower than those of the bed, although from Darcy's law one would expect higher head losses at higher filtration rates. The reasons for these lower head losses experienced in the pilot filter are more probably related to the lower accumulation of organic matter in the pilot filter affecting the permeability of the filter.

8.2 Meiofaunal and carbon analyses

It was not possible to sample the pilot filter for carbon or meiofauna either at the start or during the course of the filter run, or this would obviously have destroyed the construction of the filter. It was, however, possible to gain some insight into the structure of the carbon and meiofaunal profiles of the sand taken from the same sample as that used to fill the pilot filter. This was placed in sampling cores in the operational bed and removed at intervals. Problems were, however, encountered with sampling these cores from the bed and although six cores had been placed in the bed prior to the start of the run, only three could actually be pulled out when sampling. The result is only one sample removed at the start of the run and only two at the end. Samples were removed from the pilot filter at the end of the run. Results are presented in appendix 8.7 and figure 8.2. It is clear from both carbon and meiofaunal data that at the start of the filter run there would appear to be no obvious patterns of vertical distribution to be seen. Any distributional zonations remaining in the bed after drainage for cleaning would be destroyed when sand samples were removed from the bed before being placed in sampling cores or in the pilot filter.

Carbon

At the end of the run it can be seen that the amount of carbon in the top 1cm of the sampling cores has increased from a

Figure 8.1 FLOW RATE AND HEAD LOSS DATA FROM PILOT FILTER AND BED 5

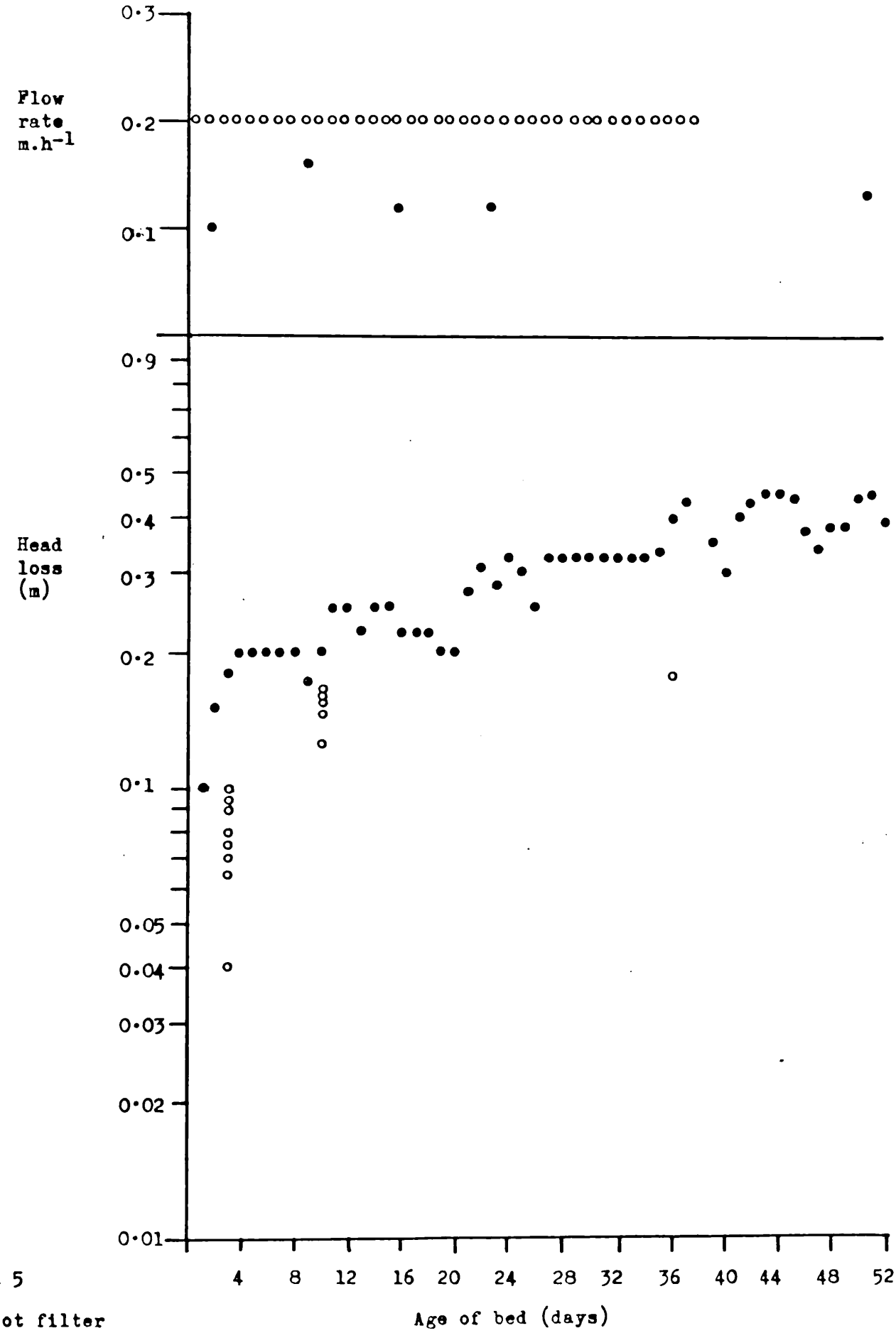
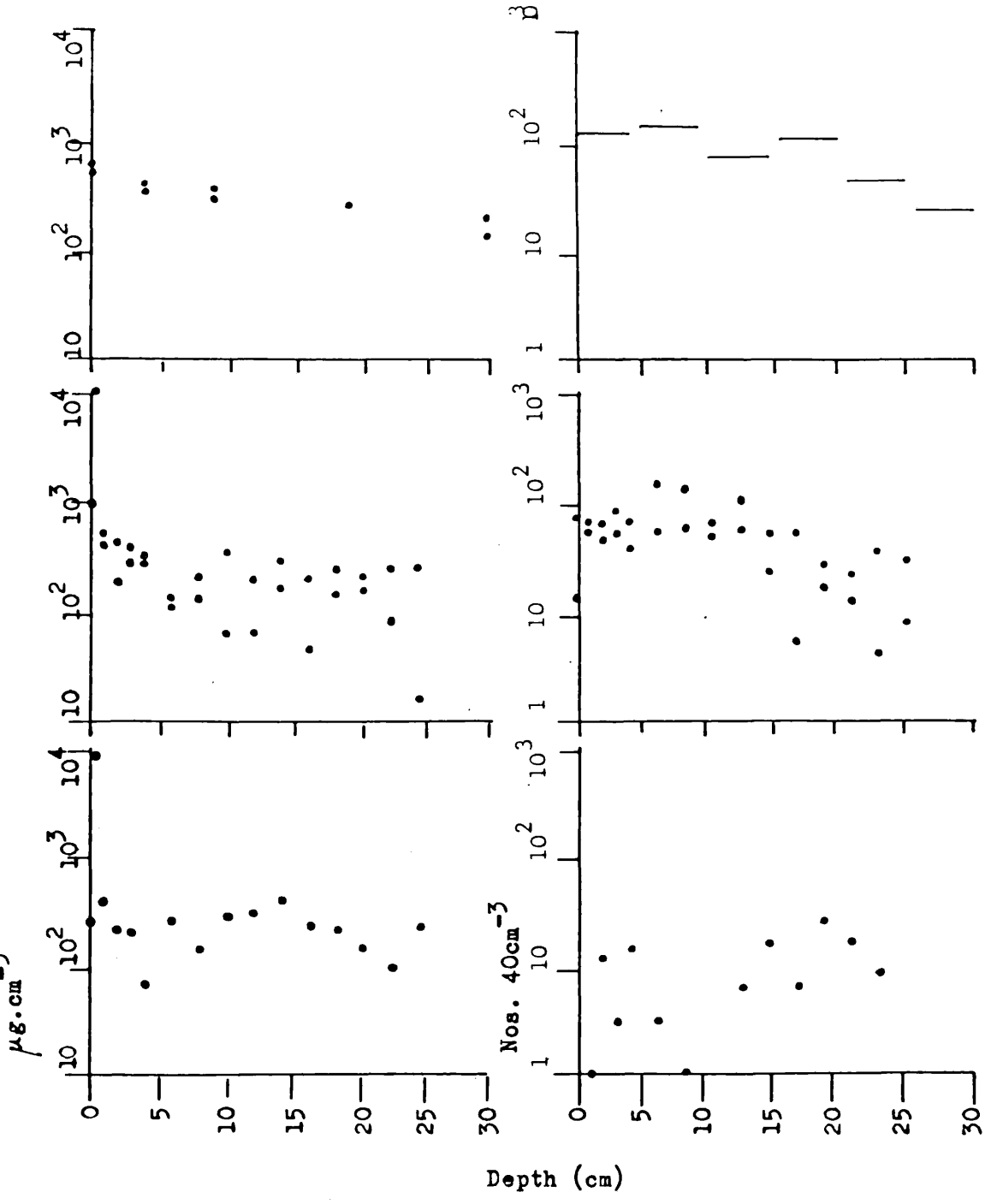


Figure 8.2 DEPTH PROFILES OF CARBON AND MEIOFAUNA IN THE PILOT FILTER AND BED 5



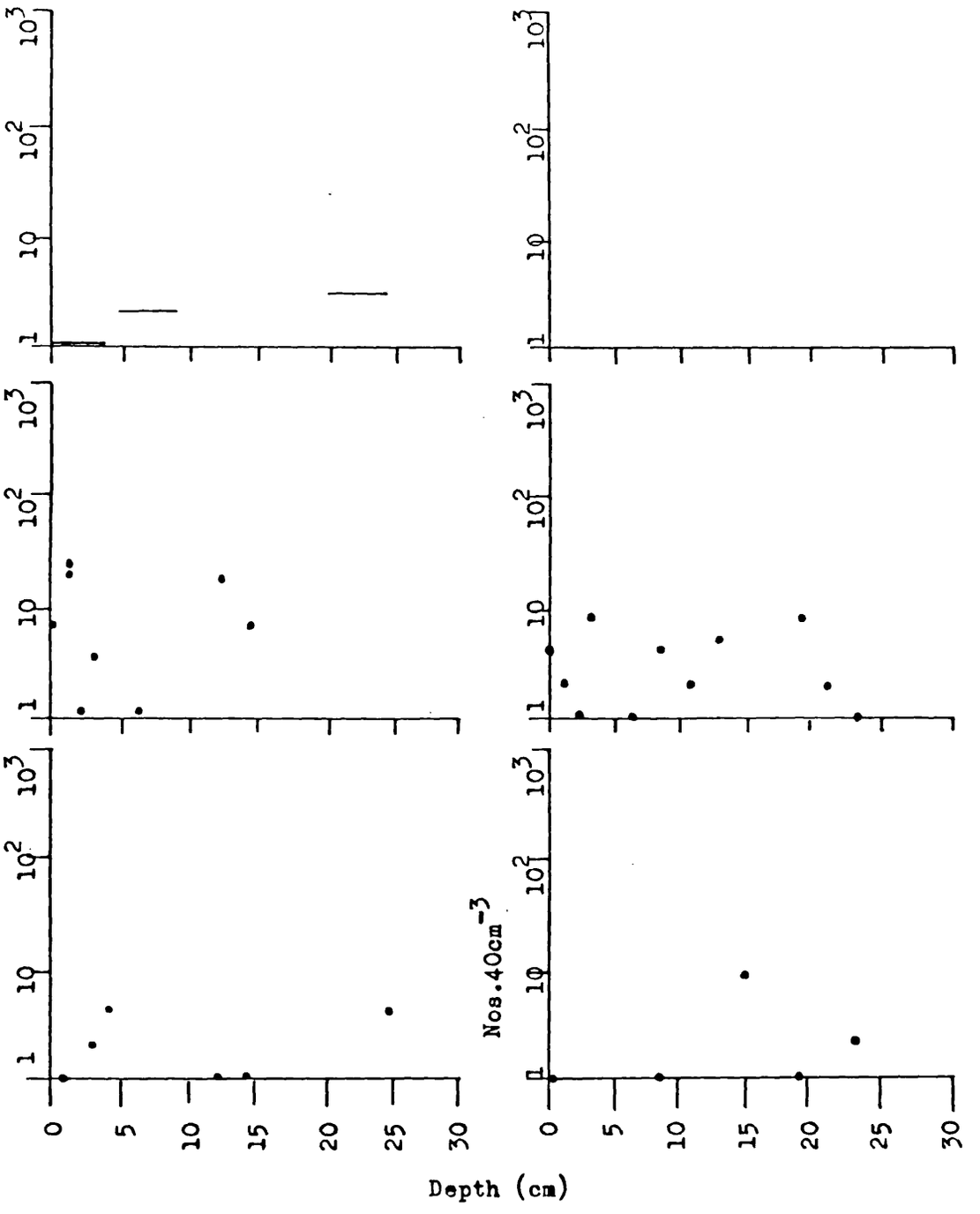
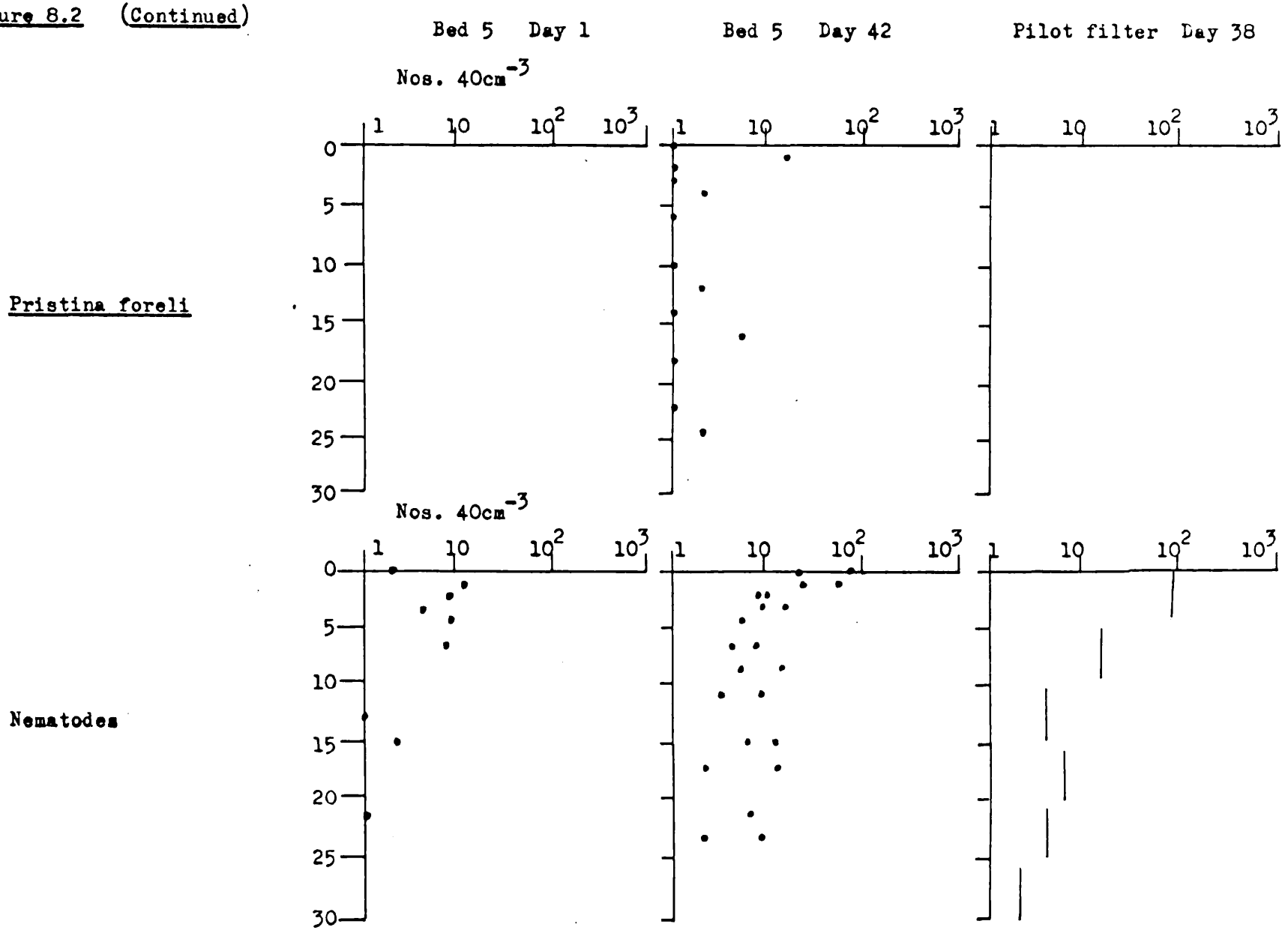


Figure 8.2 (continued)

Figure 8.2 (Continued)



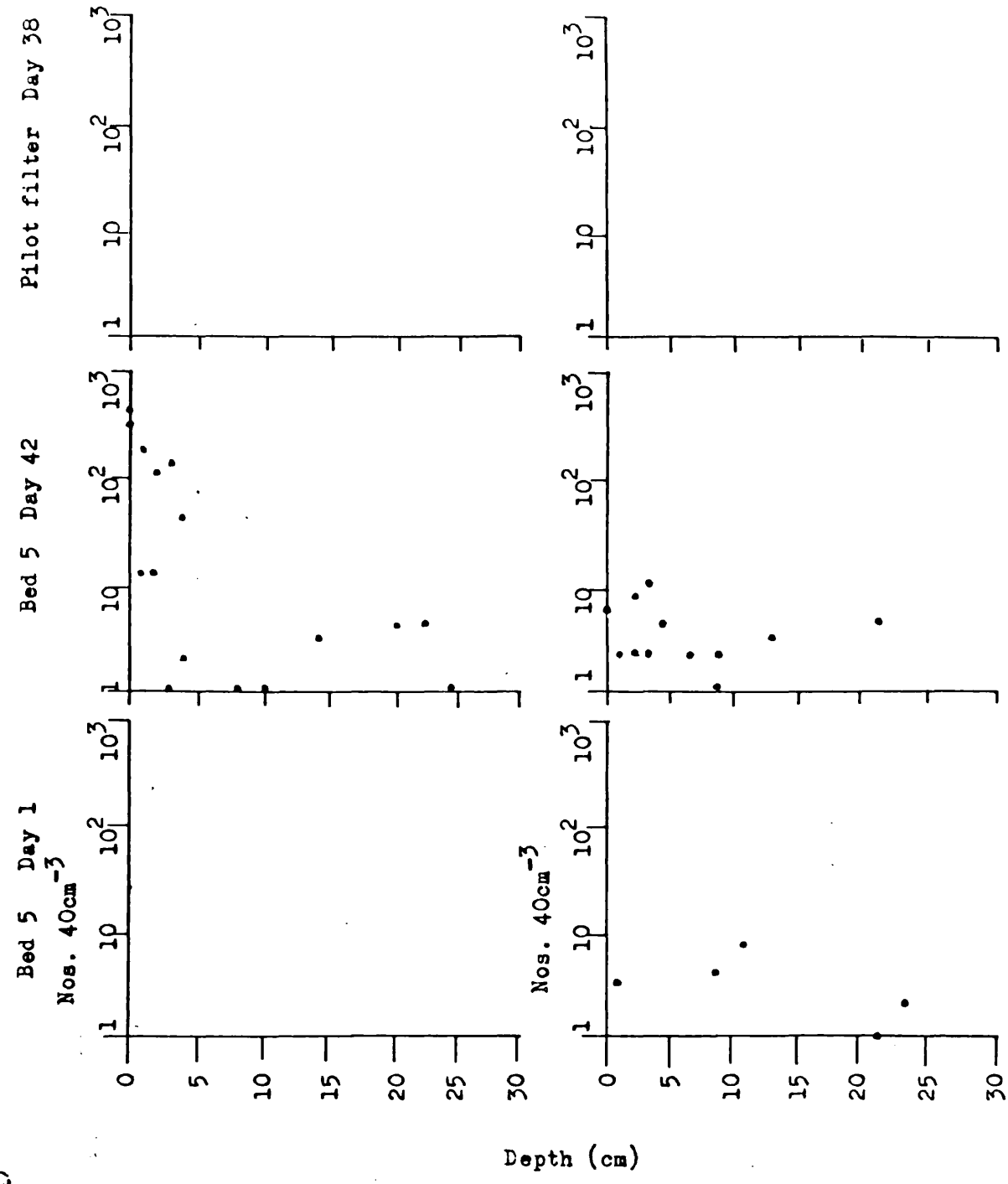


Figure 8.2 (Continued)

mean of 220 $\mu g.cm^{-3}$ to approximately 1000 $\mu g.cm^{-3}$, but only to 650 $\mu g.cm^{-3}$ in the pilot filter. This indicates that the top 1cm of the pilot filter has accumulated only about one half of the carbon present in the operational filter. The data does suggest that just below the top 1cm there has been a lower accumulation rate. This is more obvious in the bed than the pilot filter, but in the latter case no sample was taken at 2cm. As far as one can see there would appear to be no apparent changes in carbon level at either site below 5cm depth.

E. buchholzi

There would appear to be little difference in the numbers of E. buchholzi at the end of the run in the bed and pilot filter, although the more characteristic zonation patterns are not so obvious in the pilot filter. It would be misleading to deduce that the increases in numbers at the start of the run is due to reproductive activities of the worms. The fact that very few cocoons are present, especially in the pilot filter, would perhaps suggest that the increase in numbers is due to migrations from below.

P. idrensis and P. foreli

Samples removed from the pilot filter did not produce any individuals of these 2 species, although they were found present at the end of the run in the bed.

Nematodes

Numbers of nematodes in the pilot filter appear similar to those in the bed at the end of the run and in both cases they show highest numbers at the top of the filter, as was shown in chapter 6.

Chironomid larvae and harpacticoids

No chironomid larvae were found in the samples removed from the pilot filter at the end of the run, whereas approximately 350 individuals.$40cm^{-3}$ were found in the bed. Similarly, no harpacticoids were found in the pilot filter when low numbers were found present in the bed.

It would appear that there is a slower accumulation rate of carbon in the pilot filter compared with the bed. This may be due

to several reasons. The first could be settlement of suspended detritus in the water collected from the surface of the bed and used to feed the filter. Attempts were made, however, to keep this material in suspension by constant stirring of water in the header tank. The second possibility could be due to low primary production in the surface water and on the surface of the sand. Conditions in the laboratory could not reproduce the extremes of sunlight intensity which were possible out of doors, although supplementary lighting was in use. This possibility of low primary productivity is also indicated in the lack of obvious diurnal fluctuations in the dissolved oxygen content of the surface water (figures, 8.7, 8.8, 8.9). A third possibility is that the lower carbon levels may be related to the lower numbers of organisms present in the pilot filter compared with the bed. This is most obviously demonstrated with the chironomid larvae and to a lesser extent with both Pristina spp and also the harpacticoids.

It would seem, therefore, that the functioning of the pilot filter in a laboratory away from direct sunlight conditions and some distance from the source of influent water caused the filter to behave less realistically than in the field. On the other hand, the numerical abundance of the enchytraeid and nematode worms does not seem drastically altered compared with the field situation.

8.3 Dissolved oxygen

Data available from dissolved oxygen determinations are available in appendices 8.1, 8.2, 8.3. It can be seen that two values have been determined for each collecting bottle and that the dissolved oxygen of two bottles has been determined at each chosen depth, so four values are available for any one depth. From figure 7.4 it can be seen that the 'b' bottle was consistently filled before the 'a' bottle. At this stage it is necessary to examine the data in order to decide how to treat these results. The precision of the dissolved oxygen analysis method has already been determined to be ± 2% (section 7.2.3). It is necessary here to determine whether there was any significant difference between the means calculated from bottle 'a' and bottle 'b'. This was done by calculating a value for 't' (students' t test) for each pair of bottles examined (appendices 8.1, 8.2, 8.3). In 27% of cases (25 out of 92) there is a significant difference ($p < .05$) between the means of values obtained in bottles 'a' and 'b'.

So it is clear that in 27% of cases the four dissolved oxygen values for one depth cannot be used to produce one mean value.

In order to adopt an overall strategy one must choose whether to accept data from bottle 'a' or 'b'. It is necessary to further examine the data to see whether or not the dissolved oxygen values from one bottle are consistently higher or lower than the other bottle in the pair. This was done with the use of a two-way analysis of variance in which the means of bottle 'a' and bottle 'b' (appendix 8.4) were paired (table 8.1). On the basis of 92 pairs of results it can be seen that there is no overall significant difference ($p > \cdot 5$) between the dissolved oxygen values determined in bottle 'a' and bottle 'b'. In other words, the levels of dissolved oxygen in bottle 'a' is not consistently higher or lower than that determined in bottle 'b'. The significant difference ($p < \cdot 001$) demonstrated between values of dissolved oxygen determined at different depths is only to be expected.

On the basis of these statistics it is clear that one cannot use all 4 values of dissolved oxygen to determine a mean for any one depth. Secondly, it has been shown that there is no consistent difference between the levels of dissolved oxygen determined in either bottle of the pair. It would thus seem that there is no basis, on the results of these statistics, on which to decide the choice of bottle. Biologically and chemically it would seem a better choice would be to choose the bottle which had been collecting the water which has most recently left the column, i.e. bottle 'b'. In fact it would seem that there is really no reason to use the second bottle at all, as far as the collection of water for dissolved oxygen analysis is concerned. However, in some situations due to the inclusion of air bubbles at some stage in the analysis, no dissolved oxygen value for bottle 'b' was available, in which case it would seem perfectly reasonable to accept data from bottle 'a'. Results are available in appendix 8.5 and figures 8.3 - 8.9.

The temperature of the inflow and outflow water was monitored at each sampling time (appendix 8.6). It can be seen that on any one occasion the variation between inflow and outflow temperatures did not exceed $1^{o}C$. Percentage oxygen saturation was calculated, results are available in appendix 8.5 and figures 8.7, 8.8, 8.9 presenting diurnal variations.

It should be noted here that Eh and dissolved oxygen results

Table 8.1 Two way analysis of variance of data in appendix 8.4

Source of variance	d.f.	F	p
Between bottles 'a' and 'b'	1,91	0.239	0.75
Between depths	91,91	26.740	0.001

Figure 8.3 REDOX POTENTIAL AND DISSOLVED OXYGEN PROFILES IN PILOT FILTER, DAY 3

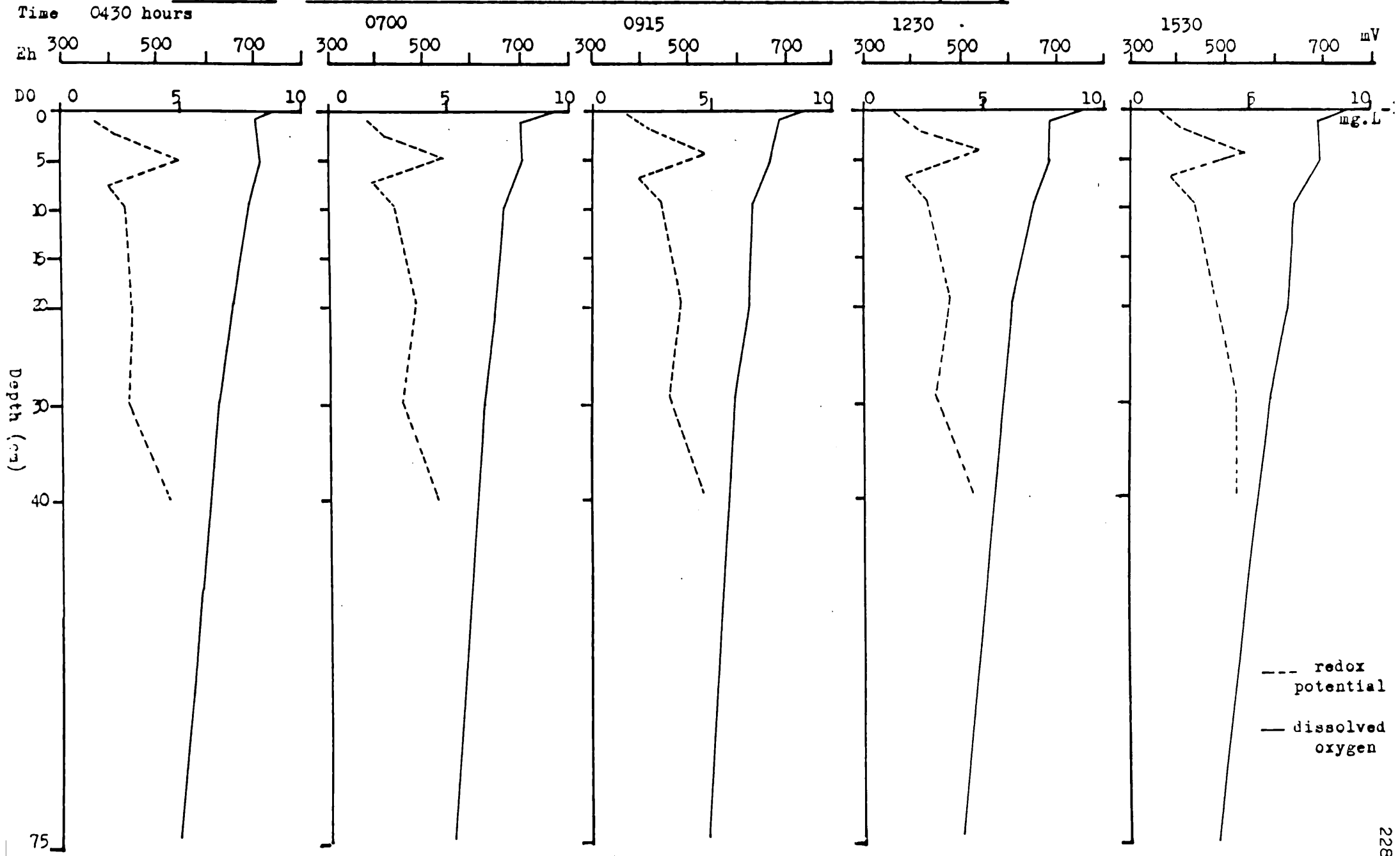
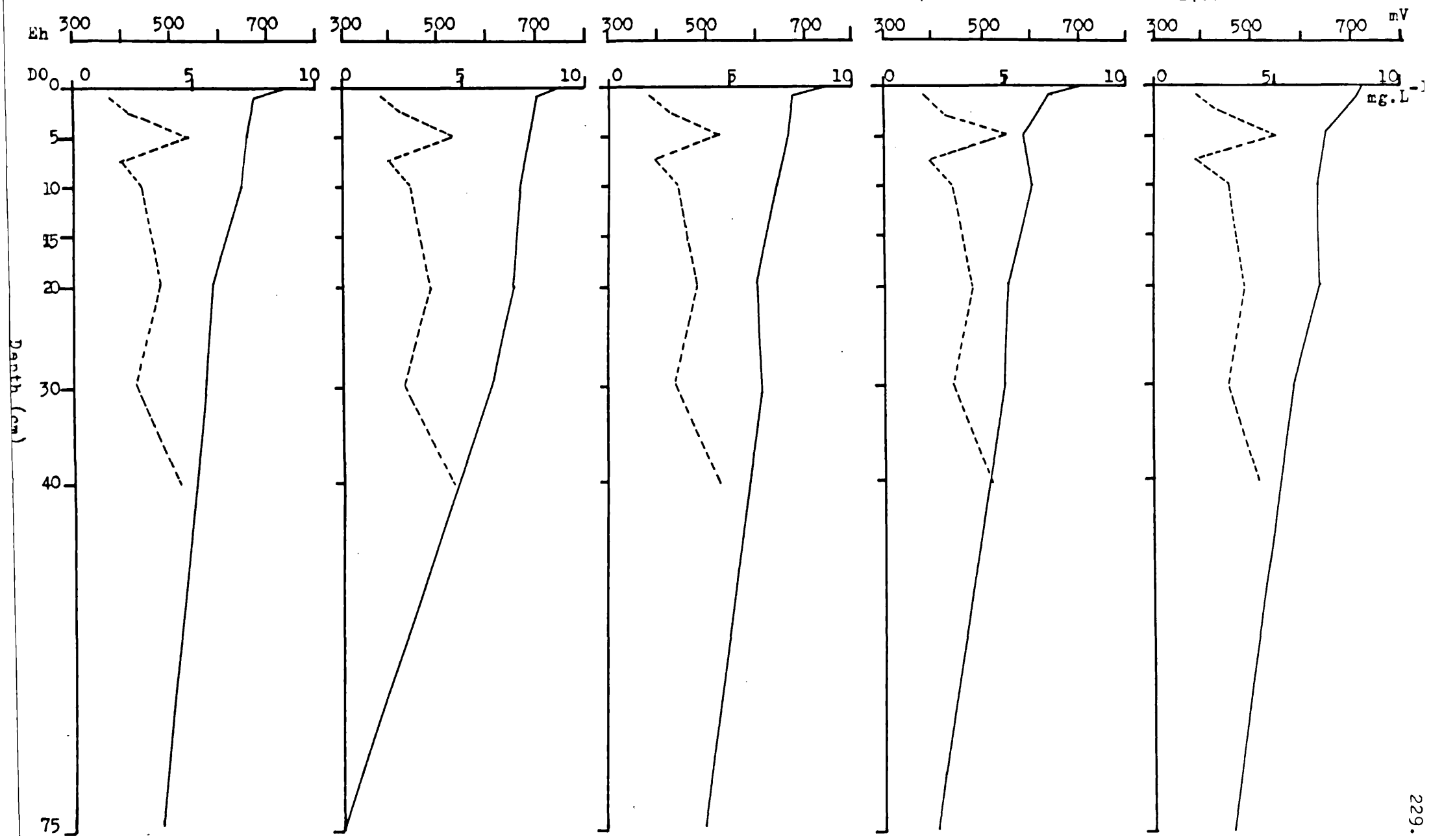


Figure 8.3 (Continued)



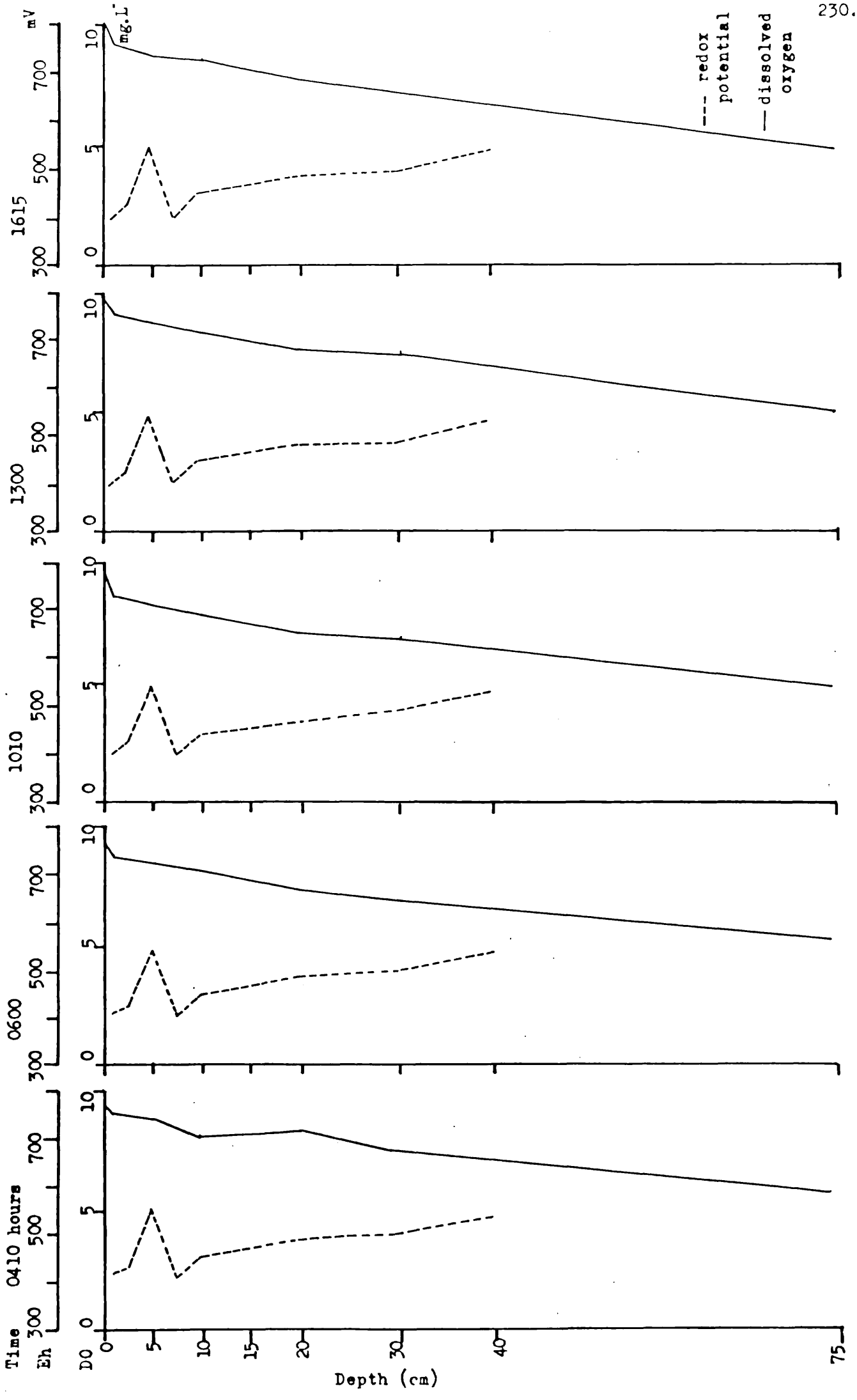


Figure 8.4 REDOX POTENTIAL AND DISSOLVED OXYGEN PROFILES IN PILOT FILTER, DAY 10

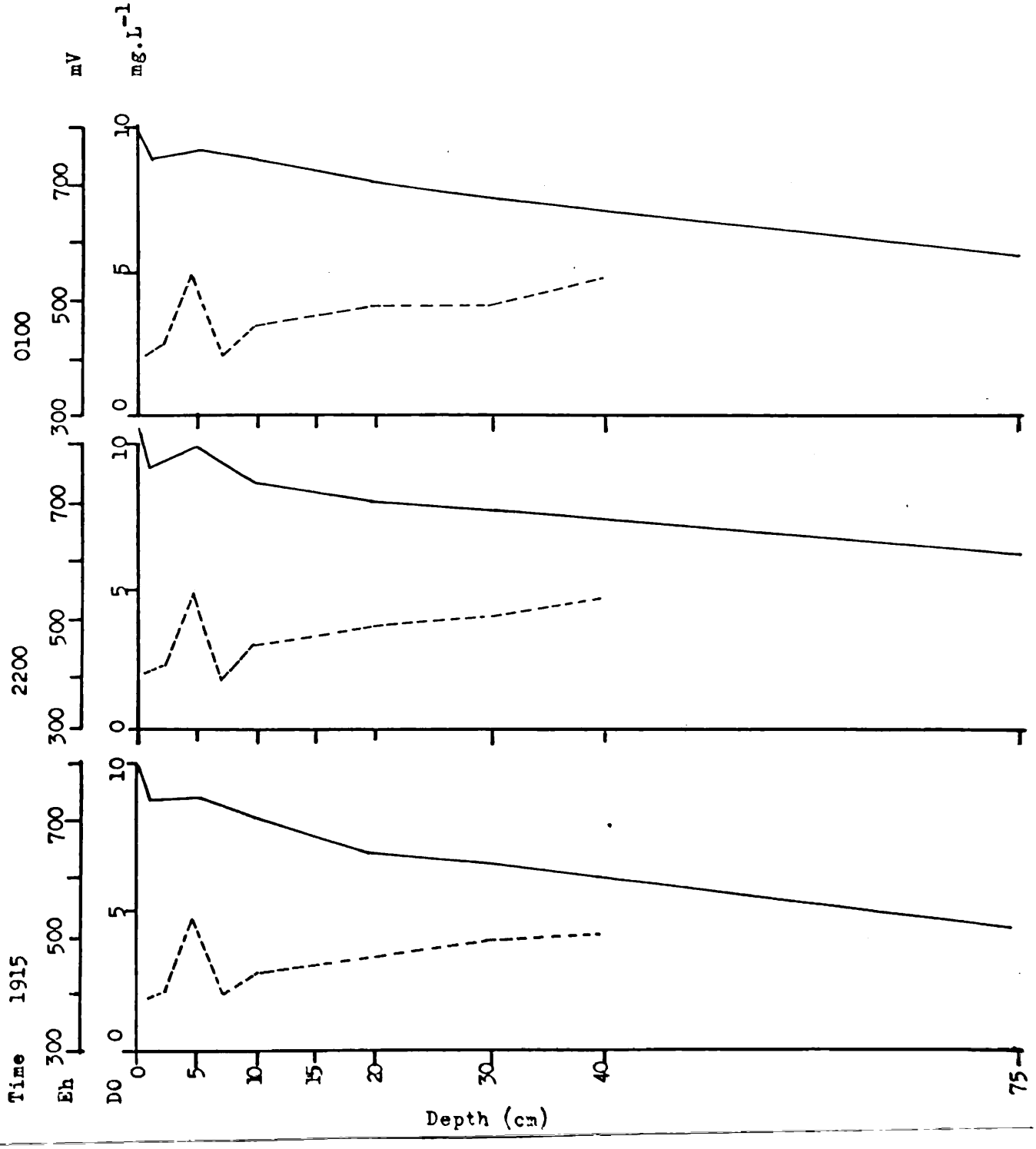
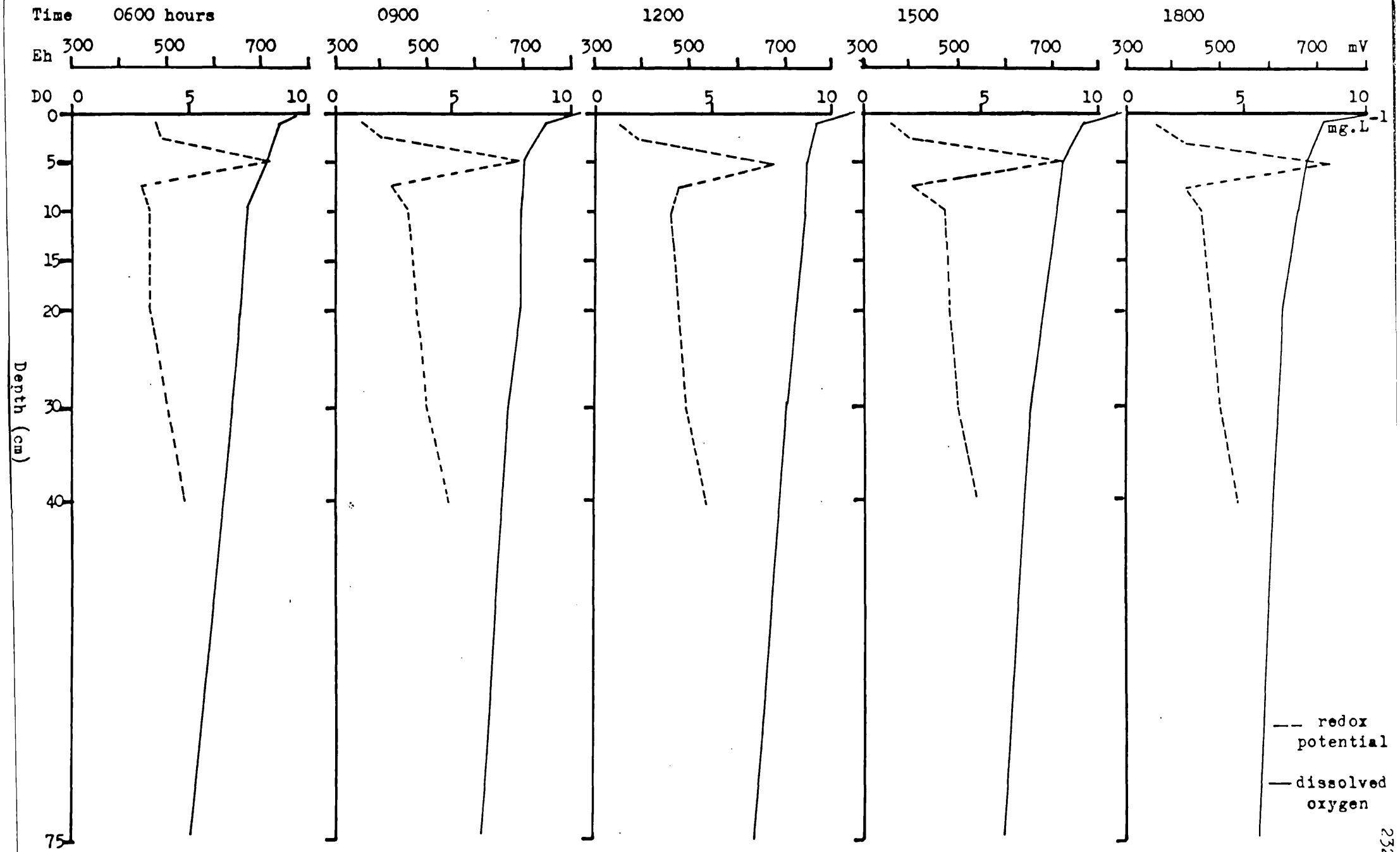


Figure 8.4 (Continued)

Figure 8.5 REDOX POTENTIAL AND DISSOLVED OXYGEN PROFILES IN PILOT FILTER, DAY 36



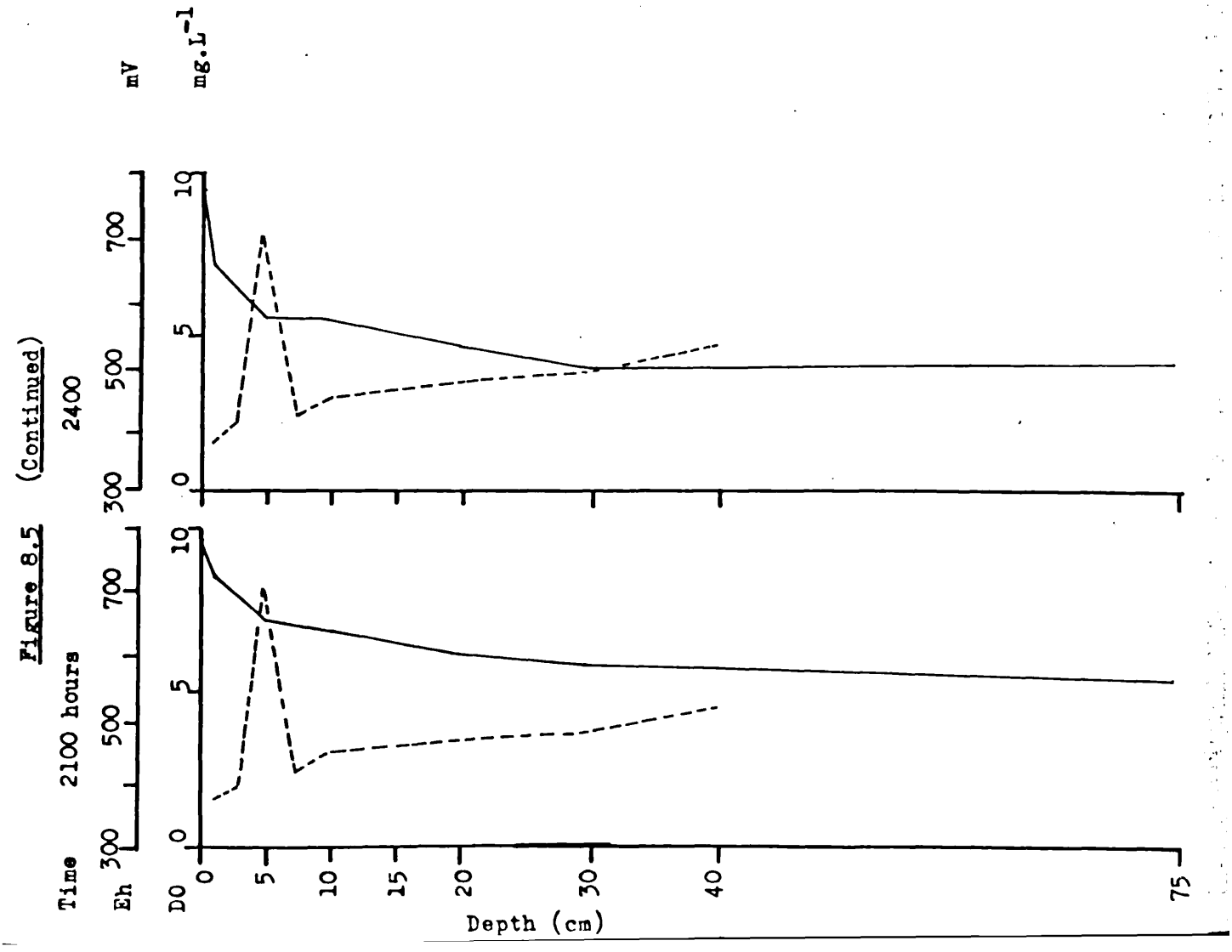


Figure 8.5 (Continued)

Figure 8.7 DIURNAL DISSOLVED OXYGEN (PERCENTAGE SATURATION) FLUCTUATIONS IN PILOT FILTER, DAY 3

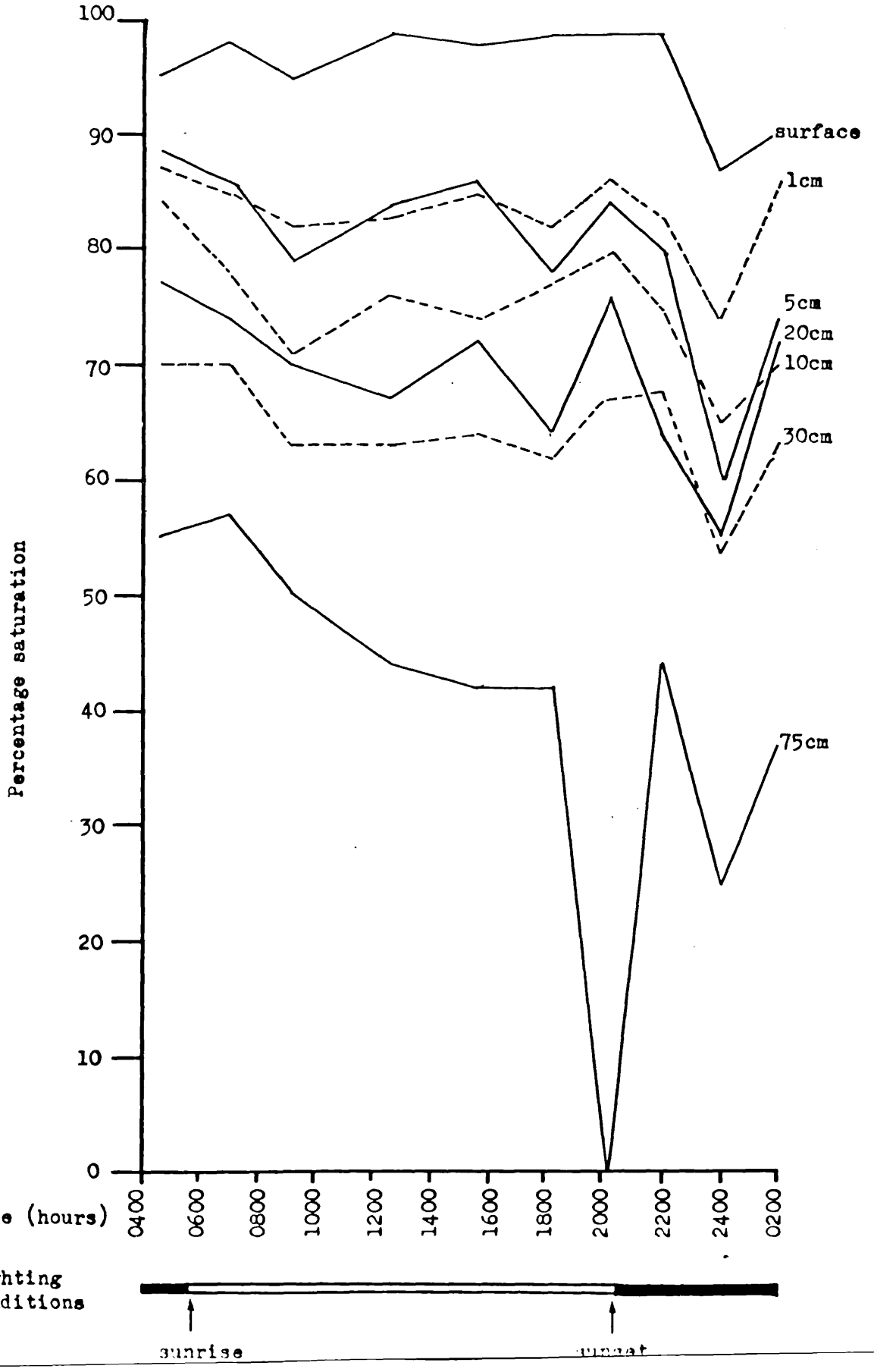


Figure 8.8 DIURNAL DISSOLVED OXYGEN (PERCENTAGE SATURATION) FLUCTUATIONS IN PILOT FILTER, DAY 10

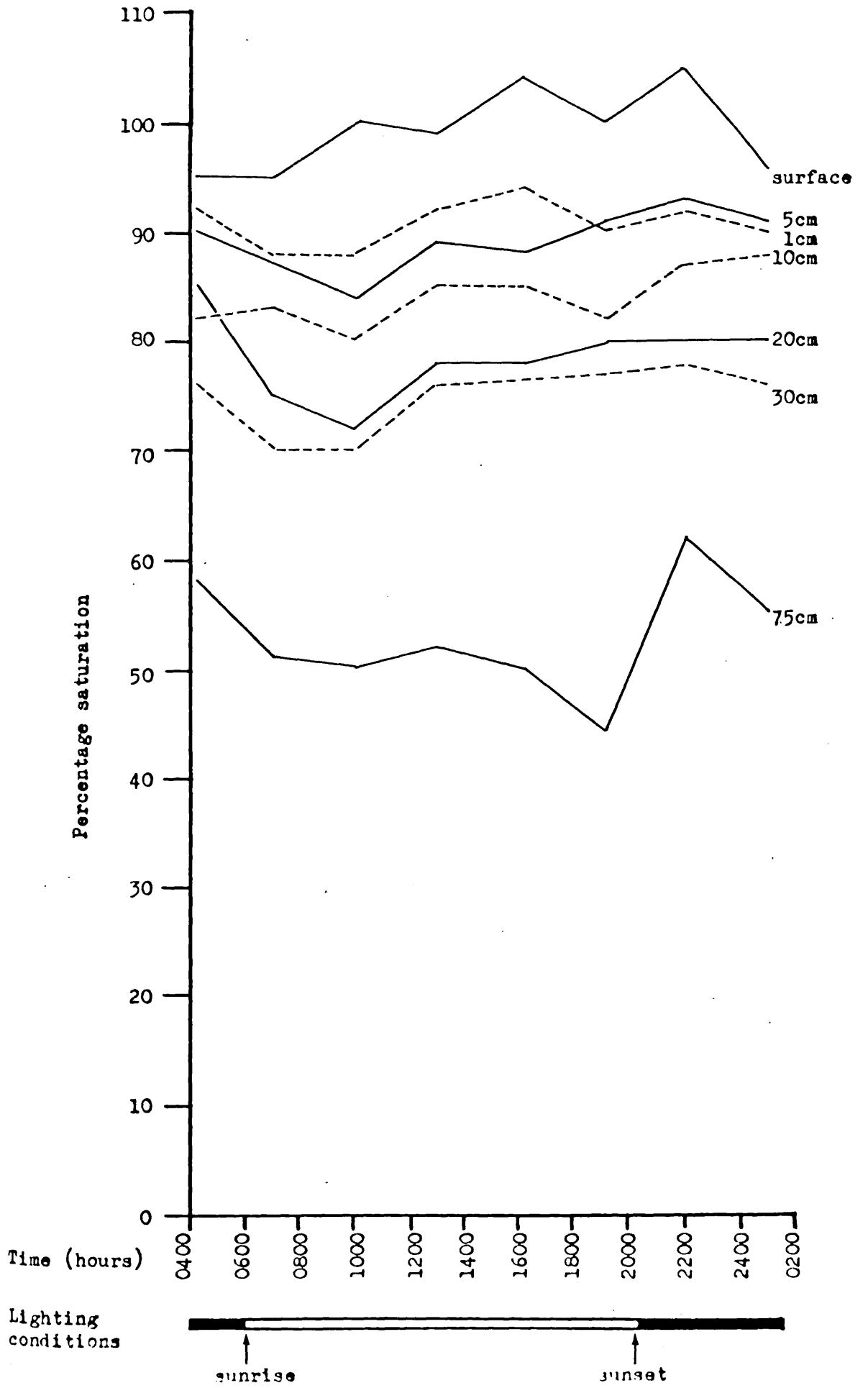
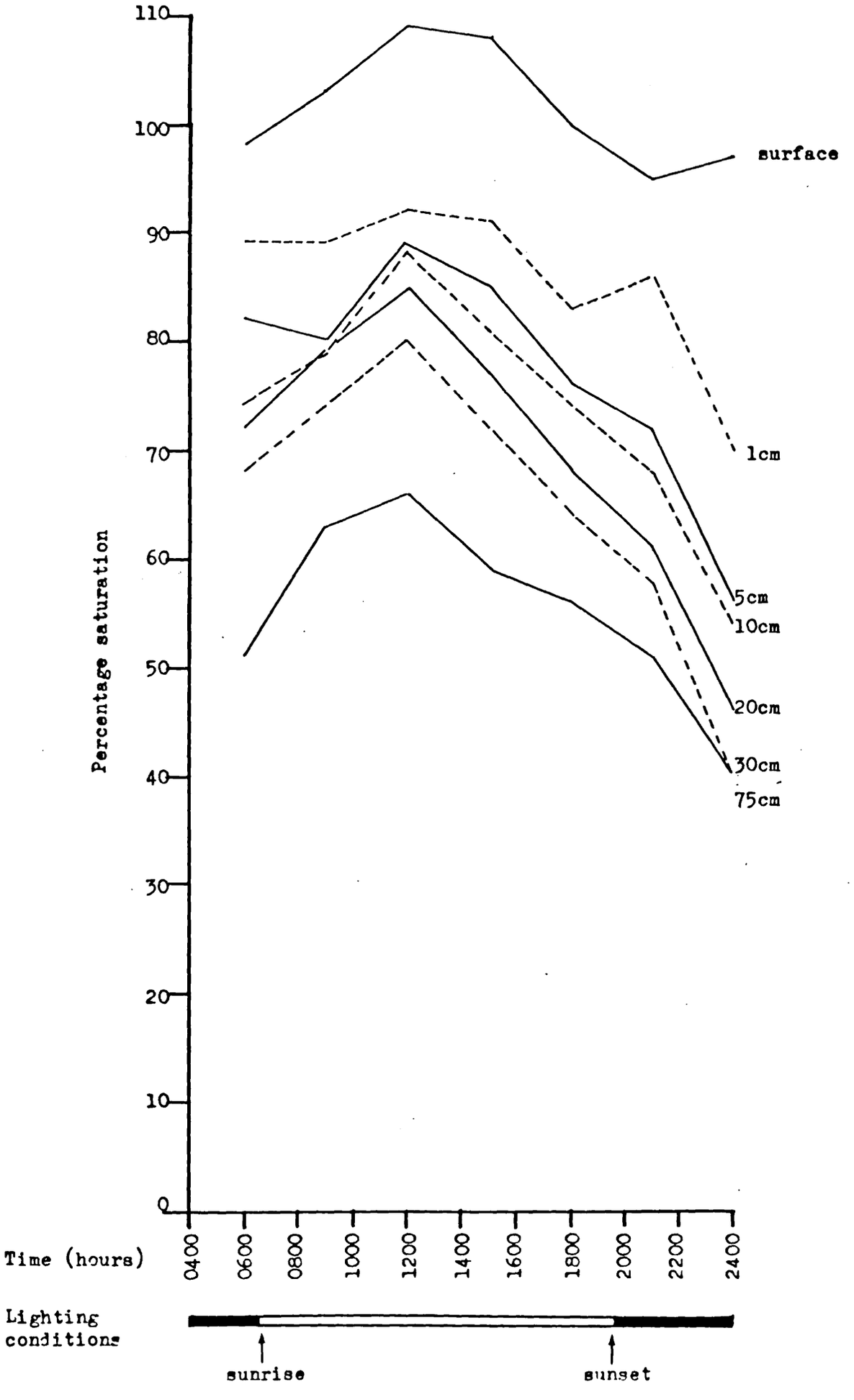


Figure 8.9 DIURNAL DISSOLVED OXYGEN (PERCENTAGE SATURATION) FLUCTUATIONS IN PILOT FILTER, DAY 36



were recorded at approximately 2 or 3 hourly intervals. The amount of time taken for the water to pass through the whole length of the sand column would be about 80 minutes, which is within the time intervals set for monitoring. Because of this it was decided not to correct the data with regard to this time factor, but to present it at the times it was actually recorded.

From figures 8.3 - 8.5 it can be seen that on all three days examined there is a distinctly higher removal rate of oxygen in the top 1cm and that on day 36 there would appear to be an intermediate removal rate between 2 and 5cm. This was investigated by determining the slopes of the relationships at the three depths: 1cm., 2 - 5cm, 6 - 25cm. For depths 6 - 25 cm a linear regression analysis was performed. Results are available in table 8.2. In the regression analysis, depth 6 - 25cm, only in one case (day 36, 2400 hrs) was there no significant relationship ($p > .05$) between oxygen concentration and depth, suggesting that oxygen consumption processes below 5cm are at a constant rate.

In order to compare the rates of oxygen consumption, the reciprocal of the slopes of these regression lines should be calculated. This figure ($1/b$) would represent the amount of oxygen ($mg.L^{-1}$) consumed by passage through 1cm of the filter at the three different depths. In order to find the oxygen consumption per cm^3 of sand, it was necessary to multiply $1/b$ by 0·37 (the porosity of the sand), that is, determining the uptake of oxygen in μg by the quantity of water contained in $1cm^3$ of sand. In fact the amount of time taken for a front of water to pass through 1cm of filter sand would be $\frac{60}{55 \cdot 34} = 1 \cdot 084$ minutes (filtration rate in sand = 55·34 $cm.hr^{-1}$). So the amount of oxygen consumed by $1cm^3$ of sand could be expressed approximately in terms of $\mu g O_2 . cm^{-3} . min^{-1}$ (although it would not be appropriate to correct the oxygen consumption rates to a rate per minute as it is not possible to predict that oxygen consumption processes would function in exactly the same way at a slightly different filtration rate).

The oxygen consumption rate was plotted in figure 8.10 with respect to time. It can be seen that there appears to be some separation of the oxygen consumption rates at the three depths in the filter. This was further investigated by an analysis of variance test (table 8.3). On any one date there is a significant difference ($p < .05$) between the recorded oxygen consumption rates at the three

Table 8.2 Oxygen consumption rates of sand at various depths in the pilot filter.

Day	Time hrs	Abscissa	Ordinate	b			linear regression stats. for depth 6-75 cm			Oxygen consumption rates 1/b(0.37), $\mu g\ O_2.cm^{-3}$ sand ($\mu g\ O_2.cm^{-3}.min^{-1}$)		
				0-1cm	2-5cm	6-75cm	d.f.	F	p	0-1 cm	2-5 cm	6-75 cm
3	0430	D.O.mg/L	Depth cm	1.198		21.973	1,3	84.900	0.005	0.3090		0.0170
	0700	"	"	0.814		25.985	1,3	37.179	0.01	0.4547		0.0141
	0915	"	"	0.792	12.422	27.215	1,3	32.376	0.025	0.4673	0.0300	0.0137
	1230	"	"	0.718		19.227	1,3	51.737	0.01	0.5154		0.0192
	1530	"	"	0.803		17.783	1,3	63.268	0.005	0.4607		0.0207
	1810	"	"	0.677	14.599	20.170	1,3	55.151	0.01	0.5465	0.0252	0.0185
	2010	"	"	0.899	12.384	8.581	1,3	89.320	0.005	0.4114	0.0300	0.0433
	2200	"	"	0.637	23.952	20.846	1,3	72.685	0.005	0.5809	0.0155	0.0178
	0400	"	"	0.702	3.742	18.031	1,3	92.722	0.005	0.5273	0.0988	0.0204
	0140	"	"	4.505	3.118	19.289	1,3	96.614	0.005	0.0821	0.1188	0.0192
10	0410	"	"	3.165	16.589	22.846	1,3	69.194	0.005	0.1169	0.0222	0.0163
	0700	"	"	1.543		22.415	1,2	113.403	0.01	0.2398		0.0167
	1010	"	"	0.997	9.368	20.574	1,3	208.088	0.001	0.3711	0.0396	0.0181
	1300	"	"	1.698	10.204	20.200	1,3	397.295	0.001	0.2179	0.0363	0.0185
	1615	"	"	1.001	7.859	17.894	1,2	554.926	0.005	0.3696	0.0470	0.0207
	1915	"	"	0.805		15.404	1,3	54.449	0.01	0.4595		0.0241
	2200	"	"	0.763		23.467	1,3	68.926	0.005	0.4851		0.0159
	0100	"	"	1.105		19.067	1,3	258.564	0.001	0.3349		0.0192

Table 8.2 continued.

Day	Time hrs	Abscissa	Ordinate	b			linear regression stats. for depth 6-75 cm			Oxygen consumption rates 1/b(0.37) $\mu g\ O_2.cm^{-3}$ sand ($\mu g\ O_2.cm^{-3}.min^{-1}$)		
				0-1cm	2-5cm	6-75cm	d.f.	F	p	0-1 cm	2-5 cm	6-75 cm
36	0600	D.O.mg/L	Depth cm	1.004	7.813	23.861	1,3	68.110	0.005	0.3685	0.0474	0.0155
	0900	"	"	0.726	4.287	38.474	1,3	112.026	0.005	0.5095	0.0862	0.0096
	1200	"	"	0.577	9.639	32.699	1,3	621.949	0.001	0.6412	0.0385	0.0115
	1500	"	"	0.638	4.667	27.662	1,3	78.422	0.005	0.5798	0.0792	0.0133
	1800	"	"	0.549	6.202	33.847	1,3	23.826	0.025	0.6738	0.0596	0.0111
	2100	"	"	0.932	2.827	30.822	1,3	15.085	0.05	0.3970	0.1310	0.0118
	2400	"	"	0.367	2.505	28.069	1,3	5.206	0.25	1.0083	0.1476	0.0133

Figure 8.10 OXYGEN CONSUMPTION RATES OF SAND IN PILOT FILTER AT THREE SEPARATE DEPTHS IN THE FILTER

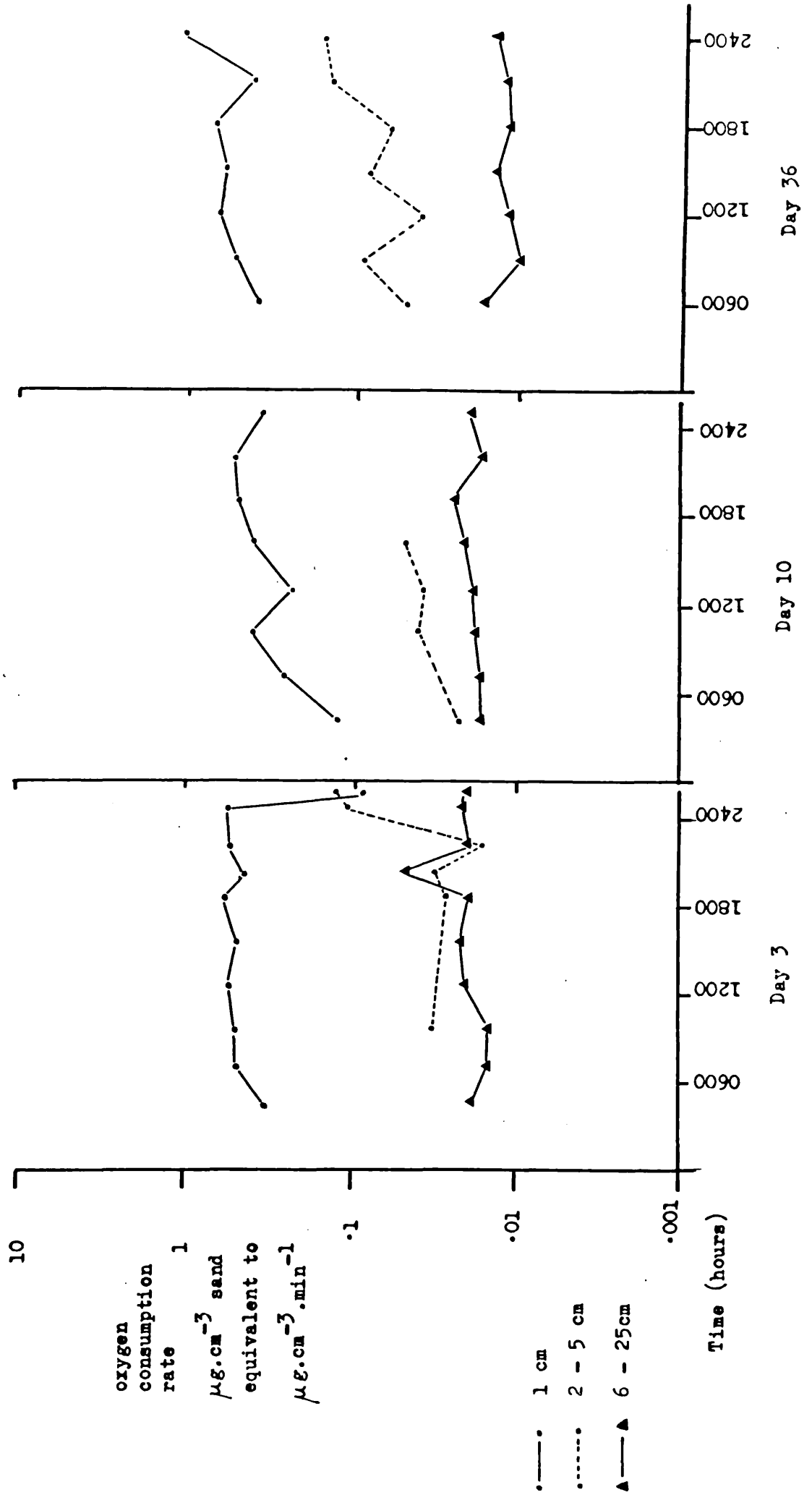


Table 8.3 Analysis of variance of Oxygen consumption data in table 8.2.

Day	Source of variance	d.f.	F	p
3	Between depths 0-1 & 2-5cm	1,14	38.129	0.001
	" " 2-5 & 6-75	1,14	5.438	0.05
	" " 0-1 & 6-75	1,18	80.618	0.001
10	" " 0-1 & 2-5	1,10	20.065	0.005
	" " 2-5 & 6-75	1,10	21.961	0.001
	" " 0-1 & 6-75	1,14	47.556	0.001
36	" " 0-1 & 2-5	1,12	38.423	0.001
	" " 2-5 & 6-75	1,12	21.071	0.001
	" " 0-1 & 6-75	1,12	51.775	0.001

Table 8.4 Mean rates of Oxygen consumption ($\mu g.cm^{-3} sand.min^{-1}$)

Day	Depth cm	$\bar{x}$	S	n	SE	Upper limit	Lower limit
3	0-1	0.4355	0.1460	10	0.1044	0.5399	0.3311
	2-5	0.0531	0.0440	6	0.0461	0.0992	0.0069
	6-75	0.0203	0.0084	10	0.0060	0.0264	0.0143
10	0-1	0.3244	0.1253	8	0.1048	0.4292	0.2095
	2-5	0.0363	0.0104	4	0.0165	0.0528	0.0197
	6-75	0.0187	0.0027	8	0.0023	0.0210	0.0164
36	0-1	0.5969	0.2149	7	0.1988	0.7957	0.3981
	2-5	0.0842	0.0414	7	0.0383	0.1225	0.0459
	6-75	0.0123	0.0019	7	0.0018	0.0141	0.0105

depths. With time, over the 36 day period, the separation of these three rates becomes more apparent as the oxygen consumption rate at depth (2 - 5cm) increases. It would not seem unreasonable to compare the three rates of oxygen uptake with the three regions of carbon concentration at 1cm, 2cm and below 2cm, discussed in chapter 6. The mean rates of oxygen consumption are presented in table 8.4. On day 36, where the greatest separation of the three consumption rates can be seen, the mean rates are 0·5969, 0·0842, 0·0123 $\mu gO_2.cm^{-3}$ sand at 0 - 1, 2 - 5, 6 - 25cm respectively.

It was possible to relate oxygen consumption rates on day 36 with the amounts of carbon present at that depth (figure 8.11). The carbon values were the geometric means from log transformed data; at depths 1cm and 5cm each value was the mean of two figures and below 5cm, the mean value was taken from six figures. It can be seen that on a double logarithmic scale there is a significant relationship ($p < .001$) between oxygen consumption rate and the amount of carbon present on that date. The relationship was expressed by $Y = 1{\cdot}007.10^{-12}.X^{4{\cdot}204}$, where Y = oxygen consumption rate ($\mu gO_2.cm^{-3}$) and X = μ gC.cm^{-3} present in the sand. The relationship thus obeys a power function, b = 4·204.

It was possible to calculate a mean oxygen uptake rate for these three levels in the filter (table 8·5). Using this information it was further possible to find the oxygen uptake rates per mg of C on day 36 at these three levels in the filter. For comparative purposes it was possible to convert the uptake rate per g of carbon to a crude uptake rate on a dryweight basis. This was done using the following conversion statistics :

Calorific content of aquatic detritus = 4422 cals.g^{-1} dryweight (Cummins and Wuycheck 1971) and 1gC = 10K cals (Winberg 1971b). So it is possible to say that 1gC is equivalent to 2·26g dry weight of aquatic detritus. To summarise, the theoretical oxygen consumption rates of the sand filter at the three depths are :
1cm, 24·761mg$O_2.g^{-1}.hr^{-1}$ (dry weight); 2 - 5cm, 6·209 ;
6 - 25cm, 1·248. This data and the evidence of the power function relationship indicates that oxygen consumption processes are not linearly related to the amount of carbon present throughout the depth of the filter. It would seem that organic matter in the surface layers is much more amenable to degradation (biological and chemical - see below) than in the lower layers. It is well known that the

Figure 8.11 OXYGEN CONSUMPTION RATES OF SAND AT THREE DEPTHS IN THE PILOT FILTER, DAY 36, IN RELATION TO ORGANIC CARBON CONCENTRATION.

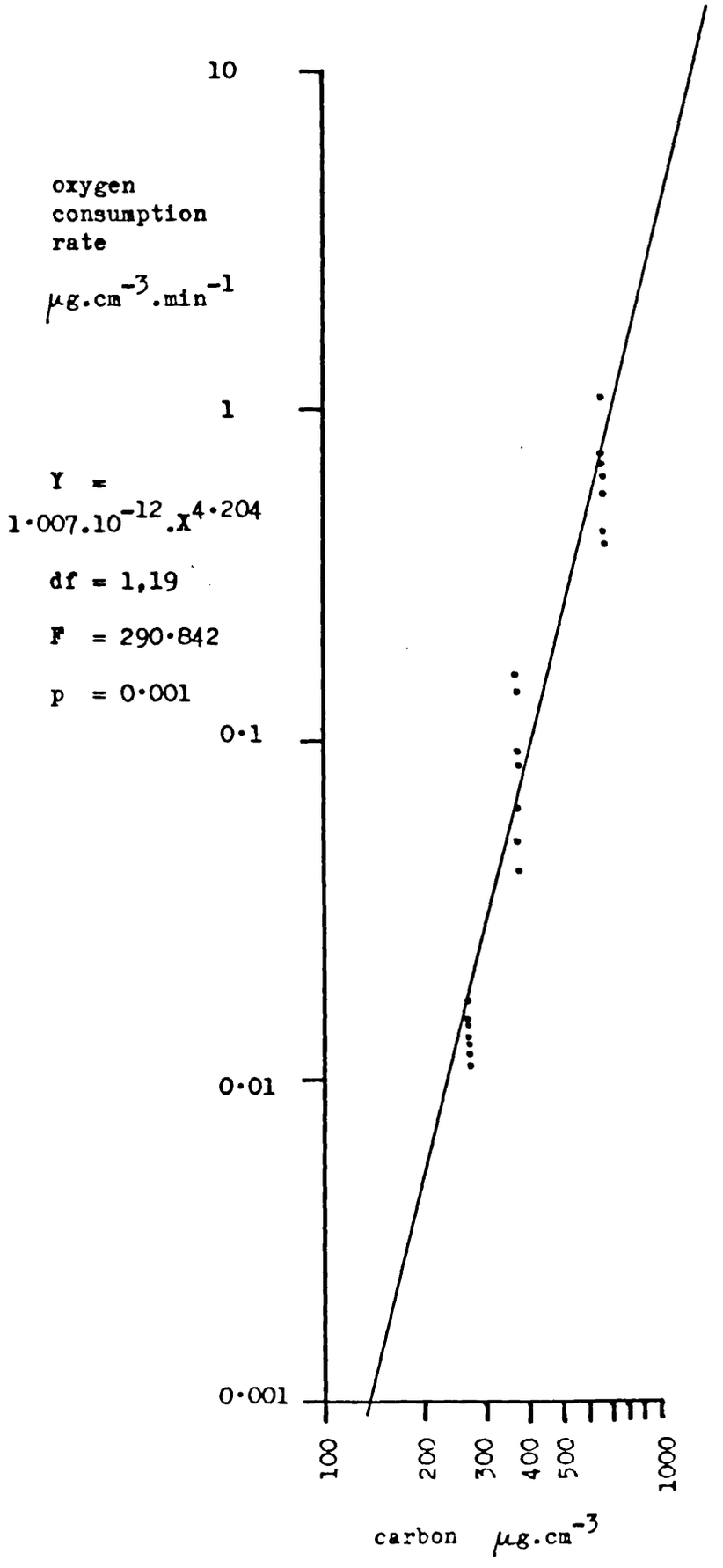


Table 8.5 Oxygen consumption rates of sand on day 36.

		Depth in pilot filter day 36		
		0-1cm	2-5cm	6-75cm
Mean C $\mu g.cm^{-3}$		640	360	262
Oxygen consumption rates	$\mu g\ O_2.cm^{-3}$ sand = $\mu g\ O_2.cm^{-3}.min^{-1}$	0.5969	0.0842	0.0123
	$\mu g\ O_2.mg\ C^{-1}.min^{-1}$ = $mg\ O_2.g\ C^{-1}.min^{-1}$	0.933	0.234	0.047
	$mg\ O_2.g\ C^{-1}.hr^{-1}$	55.955	14.033	2.820
	$mg\ O_2.g\ DW^{-1}.hr^{-1}$	24.761	6.209	1.248

amenability of detritus particles to decomposition is very much related to the age of the detritus particle concerned and to the amount of bacteria, algae and microfauna colonisation that has taken place (Kaushik and Hynes 1971, Olah 1972, Hargrave 1972, Bardgate et al 1974, Bärlocher and Kendrick 1975, Fenchel and Harrison 1976, Godshalk and Wetzel 1976, Short and Maslin 1977). The majority of detritus present in the surface layers of the sand is obviously of more recent origin than that in the depths below and so would have a greater potential for degradative processes (both biological and chemical) than would the older detritus below. It could be argued, therefore, that the organic carbon measured in the depths of the filter is more refractory in nature than that at the surface, which would explain the very much higher rates of oxygen consumption evidenced at the surface of the filter bed.

Hargrave (1972a) has demonstrated oxygen consumption rates of aquatic sediments varying from 0·1 μg - 100mg$O_2.g^{-1}.hr^{-1}$ on a dry weight basis. Higher uptake rates are demonstrated (table 8.6) for habitats containing high levels of bacteria. It would seem that the three rates of oxygen consumption fall within the ranges quoted from other sources. The lower rates found in this study are comparable with uptake rates demonstrated for habitats containing detritus particles. The higher uptake rate is more comparable with rates found in sewage effluent or in bacterial colonies removed from sediments.

The high uptake rates demonstrated in the surface of the filter may also be related to the fact that the sand receives a constant influx of oxygenated water. This maintains high oxygen concentrations in the surface of the sand and also prevents a build-up of unwanted by-products of metabolism by physically removing them in its path through the sand. Thus the aerobically respiring organisms present are in a much more favoured situation than those which inhabit lake sediments and so may lead to the maintenance of a larger aerobically respiring bacterial population.

As far as diurnal variation in percentage saturations are concerned, it can be seen (figures 8.7, 8.8, 8.9) that on days 3 and 10 there is little diurnal variation corresponding with the lit and unlit periods. This would suggest low photosynthetic activity at this time. On day 36 more definite fluctuations in percentage saturation can be seen to be operating at all depths. Data available at this time from bed 45 (P. Toms, T.W.A., pers. comm.) would indicate the presence of

Table 8.6 Oxygen consumption rates of various aquatic sediments.

Source		Consumtion mg O_2.g DW^{-1}.hr^{-1}	Author
Pebbles	L. Esrom	0.0011	Hargrave 1972a
Sand	L. Esrom beach	0.0058	"
	Helsingor beach	0.0046	"
	Juleback beach	0.0086	"
Mud	L Esrom	0.86	"
	Store Grib	0.74	"
	Frederiksborg castle	1.34	"
Detritus	L. Esrom shore	0.33	"
	Phragmites	0.24	"
	Limnaea feaces	0.55-3.50	"
	Spartina	0.63-1.81	Odum and de la Cruz 1967
	Thalassia	0.40-1.05	Fenchel 1970
Sediment	community respiration, Marion Lake	3.70	Hargrave 1969
Bacteria	Pseudomonas bacteria, L. Esrom sediment	100	Hargrave 1972a
	Marine bacteria	650-1400	Johnson 1936
	Multiplying marine bacteria	250-450	Zobell and Stadler 1940
Sewage	Suspended solids in treated sewage effluent	150	Wat.Pollut.Res.Rep. 1968 (value taken from Hargrave 1975)
Slow sand filters		1.248	This study
		6.209	"
		24.761	"

diurnal fluctuations in dissolved oxygen content of surface and filtrate water varying from 6 - 8mg/L at the surface and from 4 - 8 mg/L in the filtrate water over a 24 hour period. It must be pointed out that bed 45 was subject to strong diurnal sunlight variations and was also being operated at a rate of approximately 40cm.hr^{-1} and so the two situations are not directly comparable.

Apart from the general increase in oxygen consumption rate at depth (2 - 5cm)(figure 8.10), there would appear to be little change over the 36 day period at the two other depths. As far as the lower depths are concerned (6 - 25cm) this would seem to indicate that at this depth there is little increase in respiratory processes due to either autochthonous increases in bacterial and meiofaunal activity or due to influxes of allochthonous respiratory material. This would closely correspond with the little change with time in carbon concentrations at this depth presented in chapter 6.

In the top 1cm of sand there would appear to be a higher removal rate of dissolved oxygen even on day 3, when little time has been available for the sedimentation of incoming detritus and bacteria. It could be that the pilot filter started off at the beginning of the run with a relatively higher load of respiratory material present in the top 1cm. Alternatively, the influent water may have had a higher load of potential respiratory material in the first few days than it did later on. It was difficult to replicate outdoor lighting conditions in the laboratory in which the pilot filter was run and the result may have been a lower phytoplankton production than would normally develop in an operational filter at that time of year.

At this stage it would seem reasonable to consider the relative contributions of chemical and biological oxygen consuming processes in the overall removal of oxygen. No attempt has been made in this study to determine the contribution of chemical processes to the total oxygen removal and so it is only possible to speculate about this phenomenon with regard to sand filters. According to the literature different sediments vary in the proportion of chemical and biological oxidation which takes place. Brewer et al (1977) have shown that inorganic chemical oxidations can account for 9 - 16% of the total oxygen consumption of a sediment. Hargrave (1975) has shown that chemical oxidations can represent 25% (Lake Esrom) and 39% (Bedford Basin) of the total. The same author (1972c) has demonstrated in Lake Esrom sediment that the proportion of chemical oxidations can

can increase with depth: at 5mm it accounted for 20%, but below 1cm all oxidations were chemical in nature. Presumably this is due to a decrease in the amount of biological oxidations taking place. In sand and beaches Hargrave(1972a) demonstrated that all oxygen consumption was completely stopped by the addition of 2% formalin, suggesting that here no chemical uptake processes are going on.

It would seem, therefore, to depend very much on the nature of the sediment concerned, as to exactly what proportion of total removal chemical oxidations are responsible for. However, it is likely that some chemical oxidations proceed in a slow sand filter although their actual contribution is not known. From the previous discussions it would not appear unreasonable to deduce that the decreases in oxygen consumption rate with depth would indicate a corresponding decrease in respiratory activity of aerobic meiofaunal and bacterial activities with depth.

8.4 Redox potential

Values of Eh recorded are presented in appendix 8.6 and figures 8.3, 8.4, 8.5, daily changes in Eh are shown in figures 8.12, 8.13, 8.14. A discontinuity in Eh measurements can be seen in all cases at 5cm depth, where the recorded Eh is very much higher than in adjacent situations. This may be due to some malfunctioning of the probe or may indeed be a true reflection of the redox conditions at that point. It was found that this probe behaved normally when tested in quinhydrone both before and after the pilot filter run. General experience (e.g. Hayes et al 1958, Whitfield 1969, Ponnamperuma 1972, Giere 1973) has shown that it is very difficult to get good reproducability of Eh measurements between adjacent sites in the environment. This can be due to a variety of reasons, some of which are chemical and affect the nature of the platinum probe (Whitfield 1969), others are due to the heterogeneity of the environment. It is apparent that the platinum probe can only measure conditions in the immediate vicinity of the platinum tip, the result being that Eh measurements reflect the redox potential of the local environment and not the system as a whole. The reason for the very high Eh value at 5cm depth could well be due to leakage of air into the area around the probe and it would seem unreasonable to deduce anything of significance from this one high value.

The relationship between Eh and depth in the pilot filter was

Figure 8.12 DIURNAL REDOX POTENTIAL FLUCTUATIONS, DAY 3

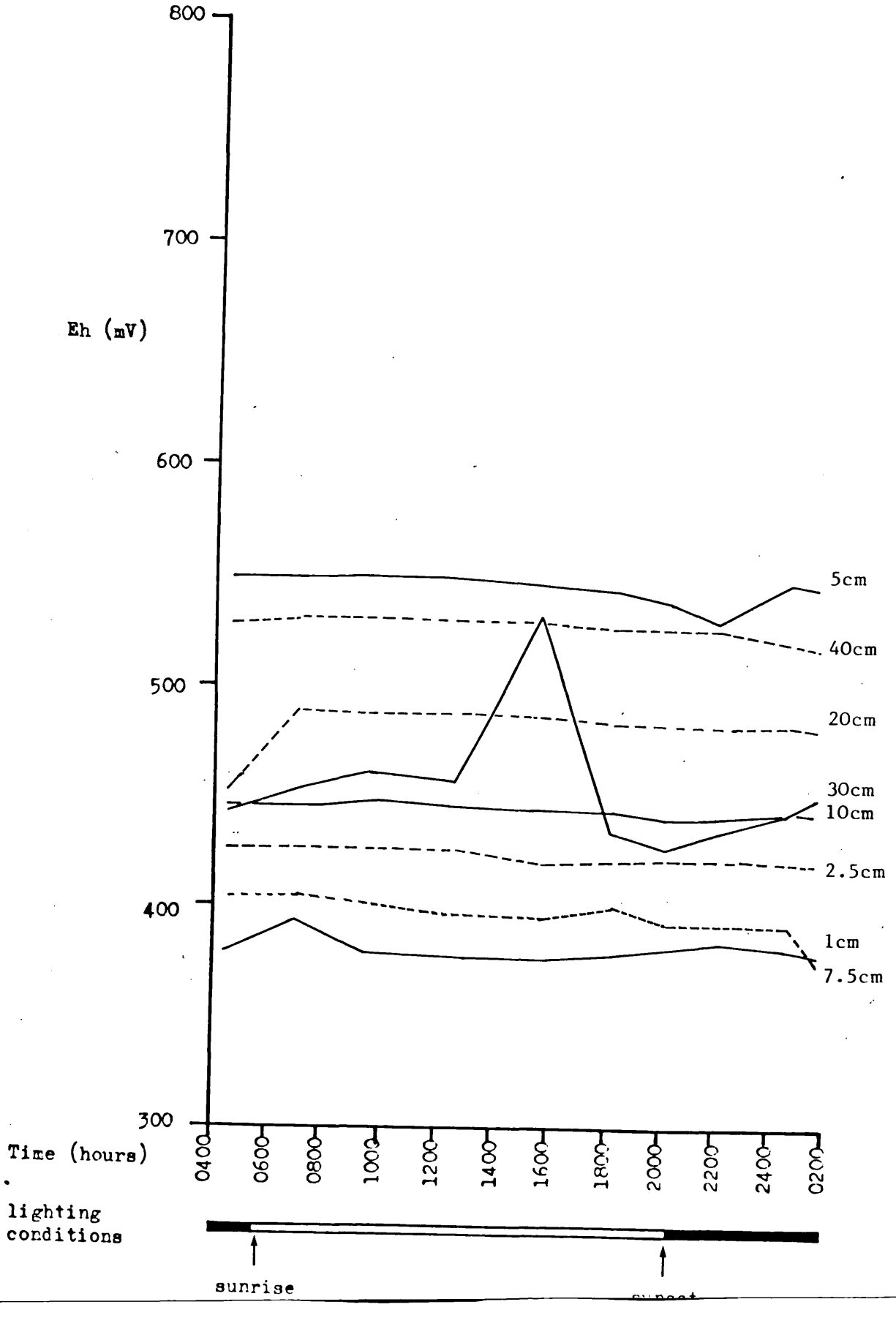


Figure 8.13 DIURNAL REDOX POTENTIAL FLUCTUATIONS, DAY 10

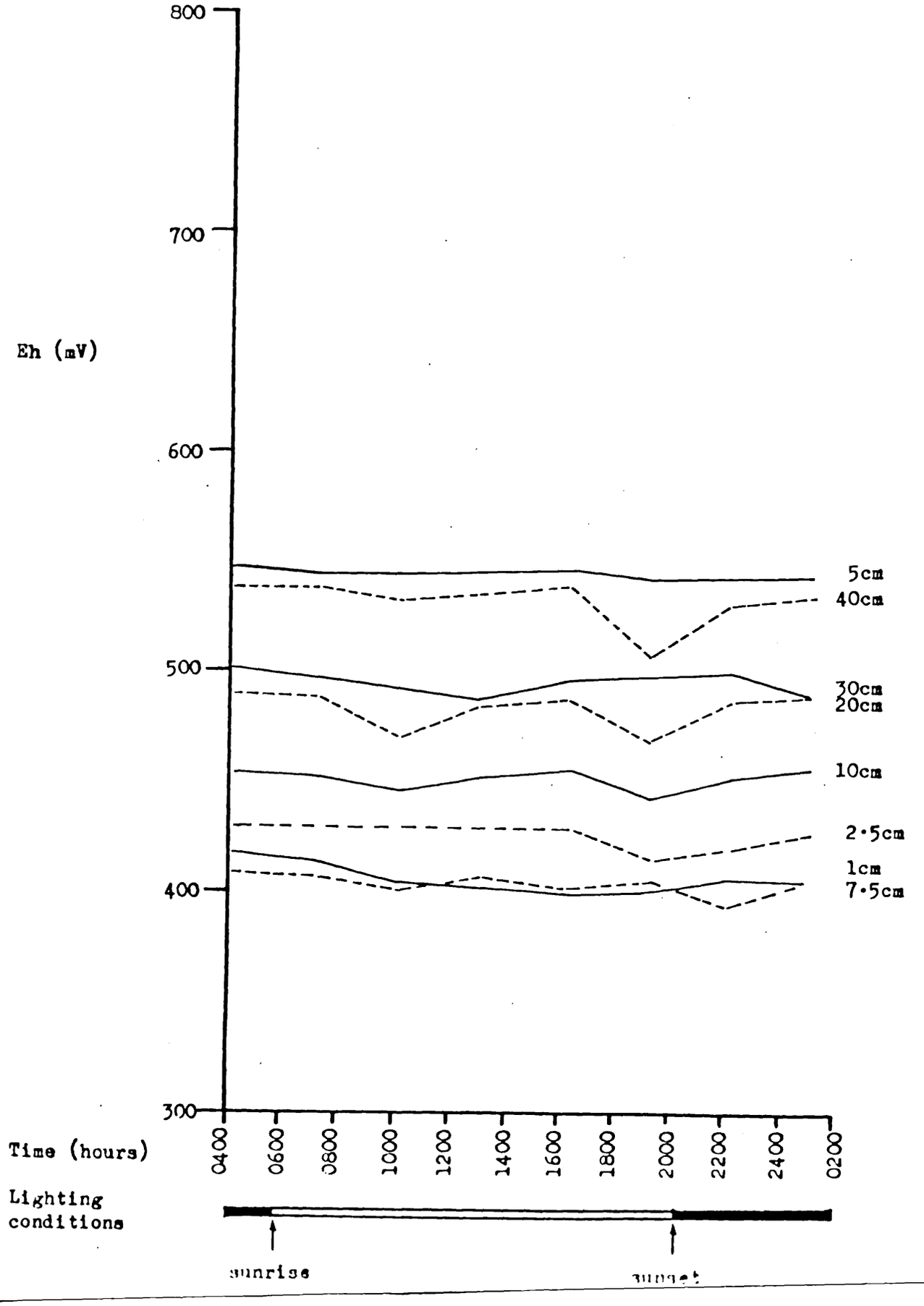
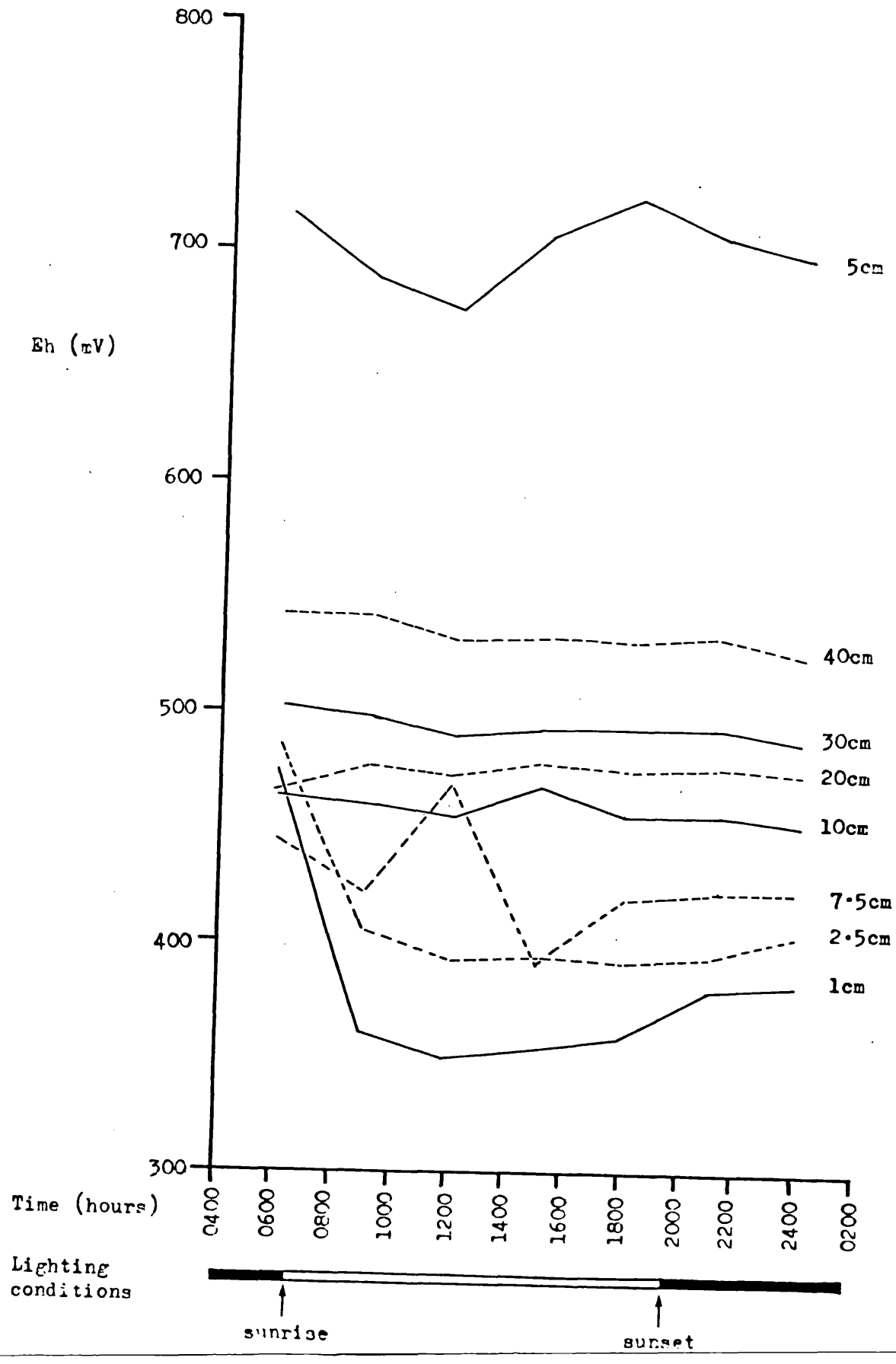


Figure 8.14 DIURNAL REDOX POTENTIAL FLUCTUATIONS, DAY 36



investigated by linear regression analysis (table 8.7). It can be seen that in all cases the relationship was significant ($p < \cdot025$) and demonstrates an increase in Eh with depth. This result would appear to be inconsistent with the decreasing oxygen concentrations with depth and so suggests that the Eh and dissolved oxygen systems recorded here are not linked. This would indicate that the Eh values recorded here are truly representative of the redox system prevailing in the sand filter and not due to chemical interferences. In some situations (as discussed in Hutchinson 1957, Ponnamperuma 1972, Golterman 1975) it has been shown that the Eh recorded in an oxygen saturated environment is lower than its theoretical value. This is due to chemical factors affecting the nature of the platinum probe and has resulted in the measurements of Eh values of approximately 520mV instead of the theoretical potential of 800mV. However, this would not appear to be the situation here.

In this study the range of Eh recorded is between 375 and 535mV and would suggest that conditions are suitable for nitrate reduction to be proceeding. (Although at lower depths (below 30cm) these procedures would seem to have ceased, unlike aerobic respiratory processes which appear to occur at all depths). Most values of Eh quoted for this procedure vary between 350 and 450mV (Mortimer 1941 , Ponnamperuma 1972). The possibility that the Eh changes were a response to pH fluctuations within the column is unlikely as an overall increase of 350 to 550mV would indicate a pH decrease of 4pH units, which is not evidenced from Thames Water Authority data. The maximum fluctuations in pH experienced in passage through a treatment works would be 0·4pH units and are more usually in the region of 0·1 to 0·2pH units (I.P. Toms, T.W.A., pers. comm.).

Denitrification has been defined (Nicholas 1963) as a specific case of nitrate respiration in which nitrate, nitrite or some intermediate is converted to nitrogen or its oxides. The ability to bring about denitrification is characteristic of a wide variety of common facultative bacteria including the genera Pseudomonas, Achromobacter, Micrococcus and Bacillus. Some species, however, can reduce nitrate to nitrite only, some reduce nitrite to nitrogen gas only and some can bring about the reduction of nitrate to nitrite and nitrogen gas. The bacteria which can partake in nitrate reduction respire aerobically in the presence of oxygen and it is not usual for them to use nitrates as the terminal electron acceptors except in the absence of oxygen.

Table 8.7 Linear regression analysis of redox data in appendix 8.6.

Day	Time	Abscissa	Ordinate	d.f.	F	p	b
3	0430	Eh(mv)	Depth(cm)	1,5	14.848	0.005	-0.273
	0700	"	"	"	16,362	0.005	-0.270
	0930	"	"	"	18.104	0.005	-0.253
	1230	"	"	"	16.314	0.005	-0.247
	1530	"	"	"	40.108	0.001	-0.223
	1810	"	"	"	10.617	0.01	-0.243
	2010	"	"	"	8.678	0.025	-0.233
	2200	"	"	"	10.638	0.01	-0.246
	0040	"	"	"	12.782	0.01	-0.256
	0140	"	"	"	12.787	0.01	-0.242
10	0410	"	"	"	52.491	0.001	-0.293
	0710	"	"	"	51.247	0.001	-0.290
	1010	"	"	"	52.124	0.001	-0.295
	1300	"	"	"	43.858	0.001	-0.289
	1615	"	"	"	40.158	0.001	-0.266
	1915	"	"	"	68.727	0.001	-0.324
	2200	"	"	"	39.469	0.001	-0.269
	0100	"	"	"	38.677	0.001	-0.283
36	0600	"	"	"	6.936	0.025	-0.350
	0900	"	"	"	46.079	0.001	-0.229
	1200	"	"	"	15.447	0.005	-0.209
	1500	"	"	"	27.007	0.001	-0.208
	1800	"	"	"	44.685	0.001	-0.234
	2100	"	"	"	70.264	0.001	-0.257
	2400	"	"	"	73.889	0.001	-0.285

This is because the enzymes needed for nitrate reduction to take place are not synthesised under aerobic conditions. There is, however, evidence available in the literature that nitrate reduction does take place in environments which appear to be aerobic. Russel (1961) found denitrification to be proceeding in aerobic soils due to the presence of anaerobic pockets. Hallberg (1968) has reported the presence of microniches of reduced sediment within oxidised layers. In a well aerated open water situation Jannasch and Pritchard (1972) found denitrification procedures to be taking place due to the presence of Pseudomonas bacteria which were attached to suspended particulate matter. They concluded this was possible due to the production of a microanaerobic zone which had developed around each individual particle. Similar results were reported by Cavari and Phelps (1977). In 1941 Mortimer reported the occurrence of nitrate reduction in sediments within an Eh range of 400 - 450mV and a dissolved oxygen concentration of $4mg.L^{-1}$. This was probably due to the presence of microanaerobic zones as reported above.

It seems likely therefore, that in this study the dissolved oxygen results present an over-simplification of conditions in the pilot filter. This was due to the fact that interstitial water samplers remove water from across the whole width of the filter and so the resulting dissolved oxygen figures will represent a mean of all dissolved oxygen concentrations across this width. The Eh results, however, have emphasised the heterogeneity of the pilot filter and so it seems likely that there are regions in the filter which are anaerobic and could therefore be the sites for localised nitrate reduction by facultative bacteria. This heterogeneity in oxidation conditions in the pilot filter could be compared with the over-dispersed distribution of both organic carbon and organisms shown in chapter 6.

The steady increase in Eh with depth could possibly be indicating that there is a corresponding reduction in the activity of these organisms. This would tend to produce a relative increase in the proportion of oxidised products and hence the increase in recorded Eh. Limiting substrate concentrations could produce such a reduction in the activity of the bacteria and although no information about dissolved organic carbon concentrations in the bed is available, it has been shown (chapter 6) that there is little penetration of particulate organic carbon below 2cm depth. Also, selective cropping and digestion of these bacteria by the meiofauna and protozoa could possibly reduce numbers of these organisms, although it must be equally forcefully

stated that meiofaunal activities could be stimulating to bacterial growth, (i.e. Hargrave 1972a and b, Fenchal 1970, 1972, Johannes and Santomi 1966).

From table 8.8 covariance analysis has shown that there are no significant changes ($p > .25$) in the slope of the Eh line over the three 24 hour periods. Similarly, from figures 8.12, 8.13, 8.14 it can be seen that there is little fluctuation in Eh recorded at each probe over these three 24 hour periods. The few wide fluctuations which can be seen are most probably caused by insufficient time being allowed for drift to stop after the probe has been reconnected to the mV recorder. There also appears to be no overall change in level of Eh recorded over the 36 day period. This would seem to indicate that there is little change in the activity of these nitrate reducing organisms, both over a 24 hour period and over the 36 day duration of the pilot filter run. Limiting substrate conditions or cropping by meiofauna could be causing this.

Nitrate analyses by Rittersbusch (1978) in an experimental system comparable with a slow sand filter used for ground water recharge, have shown that with depth there is a general increase in the level of nitrates present. This suggests that nitrification procedures are going on, although no information about the degree of aeration or bacterial content of the bed is given. Inoculation of nematodes and rotifers at various depths into the filter could be correlated several days later with a decrease in nitrate levels at the inoculation points. However, these results do tend to be somewhat inconclusive as no information on the degree of survival of these organisms or predation of these organisms on microbes was presented and no attempt was made to follow any migration of the fauna. Husmann (1968) has also presented data on the vertical distribution of nitrates in a slow sand filter (filtration rate $5cm.hr^{-1}$), but these results must be treated with caution (see later). There would seem to be no obvious correlation with depth or the distribution of bacteria (plate counts) present.

In truly benthic situations the replacement of dissolved oxygen, consumed by biological and chemical processes, is mainly due to slow diffusion processes (although the effectiveness of some faunal species in reaeration has been demonstrated (Edwards 1958). The result being an overall reduction in redox potential with depth, frequently reaching negative values at lower depths, often with a

Table 8.8 Covariance analysis of redox data in table 8.7

Day	Analysis	d.f.		p
3	Homogeneity of mean squares	9	X^2= 1.696	0.995
	Comparison of slopes	9,30	F= 0.171	0.75
	Comparison of elevations	9,39	F= 1.066	0.5
10	Homogeneity of mean squares	7	X^2= 5.380	0.9
	Comparison of slopes	7,22	F= 0.046	0.75
	Comparison of elevations	7,29	F= 6.634	0.001
36	Homogeneity of mean squares	6	X^2= 12.963	0.05
	Comparison of slopes			
	Comparison of elevations			

characteristic discontinuity layer a few cm below the surface, as demonstrated by Mortimer (1941 and 1942), Schiemer and Farahat (1966), Hargrave (1972c), Edwards (1958), Hayes et al (1958), Sarkka and Paarsivirta (1972). Redox profiles in sandy beaches tend not to follow the general pattern, depending on the state of aeration of the interstitial water entering the sand. It would appear that the absence of particulate organic matter and the maintenance of a vertical water flow system (Boucher and Chamroux 1976) tends to prevent the development of Eh and dissolved oxygen discontinuity layers. This can also be seen in sandy beaches which are subject to strong tidal currents, have low particulate carbon levels and good drainage (Maguire 1977, Williams and Hynes 1974, Giere 1973, Hulings and Gray 1971). In beaches where replacement of interstitial water is poor, obvious Eh and dissolved oxygen discontinuity layers develop (Wieser et al 1974, Fenchal and Jansson 1966, Fenchel 1969).

As far as the depth distribution of organisms is concerned, these are often shown to correlate with both the degree of oxygenation and Eh conditions (Williams and Hynes 1974, Giere 1973, Jansson 1967, Wieser et al 1974, Enkell 1968, Fenchel and Jansson 1966, Fenchel 1969) but this relationship may be one determined by the distribution of food materials. In situations which are well oxygenated throughout the depths, Giere (1973) has shown good correlations between numbers of oligochaetes and food distribution. This type of relationship has been further investigated by Fenchel and Jansson (1966) who considered that the depth distribution of the fauna could well be related to the depth distributions of certain types of bacteria which are characteristic of different redox potentials (as demonstrated by Baas Becking et al, 1960). Fenchel (1969) found that the high numbers of ciliates concentrated in the region of the Eh discontinuity layer were correlated with higher concentrations of end products of anaerobic metabolism and higher numbers of bacteria in that region. He recognised three groups of meiofauna : 1) Those living in well oxygenated areas; 2) those living in the vicinity of the Eh discontinuity layer; and 3) those living in the reduced anaerobic and sulphide containing areas beneath this. These three zones were also recognised by Maguire (1977) in an examination of the meiofauna in a marine beach which, although it did not go anaerobic, did show an obvious Eh discontinuity layer. Maguire also described a fourth group which was adaptable and was found in all of these three regions. It could be possible to consider the first of these zonations of Fenchel and Maguire to be compar-

able with the distribution of nematodes in this study, which are seen to inhabit the well oxygenated upper zones of the slow sand filter. Similarly the fourth "adaptable" zonation shown by Maguire could be considered similar to that demonstrated in this study by P. idrensis, P. foreli, the harpacticoids and flatworms.

Depth distributions of oxygen in a slow sand filter operating at $5cm.hr^{-1}$ are available from Husmann (1968). Unfortunately the sampling method used allowed water to be removed from the interstices at a rate which was more than 20 times faster than the rate at which it would have been passing through the interstices. Also, the water was sampled intermittently for periods of 2 hours on and 2 hours off. Both these factors mean that any results obtained should be regarded with caution. Although there is an overall decrease in dissolved oxygen with depth the data does not exhibit the three clear uptake rates demonstrated in this study. No obvious correlations are apparent between bacterial numbers and both depth and organic carbon content. The more obvious correlations demonstrated are between the depth distributions of the nematodes and oligochaetes and the organic carbon distribution.

It would seem, therefore, that no one factor is responsible for the distribution of organisms in the environment. The degree of aeration of a sand system is dependent on the amount of exchange of interstitial water and drainage of the system and also the level of oxygen consuming organic matter present, both chemical and biological. All of these factors may affect the type and distribution of bacteria and their predators which are present.

CHAPTER 9 SUMMARY

Slow sand filters are primarily used to treat water prior to domestic and industrial supply. They are sometimes also used to treat water before ground water recharge and more rarely in the tertiary treatment of sewage. This investigation centred on bed No. 45 at Hampton Treatment Works of the Thames Water Authority.

Problems associated with sampling this medium have been discussed in chapter 3. The method used was successful in that it allowed samples of sand to be removed from the bed during the course of a filter run and without altering the nature of the sand in the process. Also the method satisfied the requirements of the Thames Water Authority in preventing the contamination of deeper sand by surface detritus. Thus permission was given from the Thames Water Authority to sample from full scale operational beds. The main disadvantage of the method was that it did not allow horizontal migration of organisms into the apparatus from adjacent areas, although it was possible for vertical migration to take place.

The types of meiofaunal organisms found in this slow sand filter are characteristic of those found in other interstitial environments. In general they are relatively small, either in length or cross section and fairly mobile, with the largest truly interstitial organism, E. buchholzi, reaching maximum lengths of 4 - 5cm. Of the oligochaetes present, members of the Enchytraeidae, Naididae and Aelosomatidae were represented. Other meiofaunal organisms included nematodes, chironomid larvae, harpacticoids and microturbellaria. Similar species compositions have been reported by Lloyd (1974) in slow sand filters at Ashford Common Treatment Works (Thames Water Authority). Workers in Germany (Rittersbusch 1974, Husmann 1958) list similar taxonomic groups, although there are slight differences in the exact species present.

Sand grain size analysis indicated that there was some heterogeneity in composition from core to core, probably due to the nature of the procedure used by the Thames Water Authority to wash the sand before it was placed in the sampling cores. Though as a whole this sand was not significantly different from that already present in the sand bed or in other parts of the works. Backwashing procedures and the method used to fill the sand cores did not cause the sand to become vertically sorted in any way in either the sand bed or sampling cores.

The material present in a slow sand filter which may be available as food for detritus and filter feeding organisms includes bacteria, algae and dead and decaying organic matter. To separate and measure the individual components of the detritus was considered to be too time-consuming, especially in a situation where the food requirements of the meiofauna were not known. Instead, measurements of the total particulate organic carbon present in the sand were made. It must be realised that measurements of this kind will give an overall "blanket figure" indicating the total amount of organic carbon available to the organisms, but exactly how much of it is capable of being utilised is not known.

A method was devised to determine the amount of organic carbon present in the sand. This involved a wet oxidation procedure and potentiometric end point titration and was designed to cope with the wide range of organic carbon present in a sampling core. Precision of the method was good, at concentrations greater than 20 $\mu gC.ml^{-1}$ precision was ± 3% which compared well with the literature. The minimum detectable level was 5 $\mu gC.ml^{-1}$ and, the maximum carbon value which could be accurately determined was 400 $\mu gC.ml^{-1}$. The method was at least 96% accurate, again comparing well with published literature.

From statistical analyses it would appear that the horizontal distribution of organic carbon within a sampling core is random, but that from core to core in the bed the distribution is overdispersed and tending towards the lognormal. At the start of a filter run there are no significant vertical differences in the amount of carbon present, but with time three distinct zonations can be seen, in order of decreasing carbon concentration at 1cm, 2cm and below 2cm. It was possible to make comparisons on two occasions with carbon data compiled in this study and Thames Water Authority carbon analyses obtained at the end of a filter run. At the end of run 5 there would not appear to be any significant differences observed, but at the end of run 7 there would appear to be differences. This may be due to the fact that sand in the bed itself is continually accumulating organic carbon, although below 2cm this is at a very low level, whereas in the sampling cores washed sand was used at the start of each filter run. Alternatively this discrepancy may be due to the difficulties of statistically treating data with widely differing samples (this study n = 45, Thames Water Authority data n = 7).

Depth analysis of the carbon data in the cores would indicate

that the main cause of head loss in a filter is due to accumulation of material in the top 1cm, and to a lesser extent at 2cm. It has also been suggested that large numbers of chironomid larvae in the surface layers of the sand can also be a contributory factor.

The horizontal distribution of meiofauna indicates that when numbers are low at the start of a filter run then distributions tend to be random. However, with time the distribution of most organisms tends to become lognormal. These lognormal distributions have been reported from many types of habitat. Various authors have considered that the lognormal distribution is characteristic of stable environments, whereas random distributions are more indicative of changing or perturbed habitats. In this situation the random distributions are more obvious at the start of a filter run when few organisms have migrated into the sampling cores from below and could thus represent an environment in a state of flux where organisms are still responding to changing environmental conditions. With E. buchholzi this situation is most marked and it has been suggested that it may take fifteen days or so for this population to reach stability.

Several authors have reported that distributions of organisms can be correlated with particle size characteristics. However, in the absence of major particle size discontinuities in the filter bed it would seem that the reasons for these overdispersed distributions may be related to some factor regarding the nature of the food available. An obvious relationship to consider here is the lognormal distributions demonstrated by both the majority of meiofaunal populations and also by the organic carbon in the sampling cores. This correlation was not found to be significant, however, and may well be due to the fact that the carbon determinations measured total organic carbon present, but did not measure the amount of material present which could be utilised by each species.

In order to obtain more basic information regarding the oxygen and redox conditions within a slow sand filter bed it was necessary to construct a small scale pilot filter, as measurements of this nature would be difficult on an operational bed. The pilot filter was run at a slightly lower filtration rate ($20cm.hr^{-1}$) than bed 45 had previously been subjected to ($20 - 35cm.hr^{-1}$) and was compared with a slow sand filter bed which for various reasons was run at filtration rates between 10 and $16cm.hr^{-1}$. Some discrepancies were found in the number of organisms inhabiting the pilot filter and the

operational bed. This was mainly due to the chironomid larvae, none of which were found in the pilot filter, whereas they were present in in the bed. Lower levels of carbon were found in the surface layers of the pilot filter than in the bed and this may well have been due to a lower primary productivity occurring in the pilot filter, as it was not subjected to the extremes of sunlight to which the bed was. This was further evidenced by the lack of diurnal oxygen fluctuations in the pilot filter which were recorded in an operational bed during the same experimental period.

A method was developed to collect interstitial water samples from the pilot filter at the same rate at which the water was passing through the interstices. This was essential for oxygen measurements to be representative of the regions from which they were removed. This technique involved the development of a microWinkler oxygen determination method to analyse the small (2ml) volume of water removed in this way. The method was found to have a precision of ± 2% which compares well with standard techniques.

Monitoring of dissolved oxygen concentrations in the pilot filter has revealed three regions of oxygen uptake in the bed at 1cm, 2 - 5cm and below 5cm of 0·59, 0·08 and 0·01 $\mu gO_2.cm^{-3}$ sand.min^{-1} respectively. The rate below 5cm was constant with respect to depth. It would seem reasonable to relate these three uptake rates to the three regions of carbon concentration demonstrated earlier at 1, 2 and below 2cm in the bed. Oxygen consumption rates were related to organic matter concentrations on a dry weight basis at the appropriate depths and shown to be (1cm) 24·761, (2 - 5cm) 6·209, (below 5cm) 1·248 $mgO_2.g^{-1}.hr^{-1}$ (dryweight). This data was shown to obey a power function relationship $Y = 1{\cdot}007.10^{-12}.X^{4{\cdot}204}$ where Y = oxygen consumption rate in $\mu g.cm^{-3}$ sand.min^{-1} and X = carbon concentration in $\mu g.cm^{-3}$ sand, indicating that oxygen consumption processes are not linearly related to the amount of carbon present throughout the depth of the filter. It would, therefore, seem that organic matter in the surface layers is much more amenable to biological and chemical degradation than in the lower layers. This could well be due to the relative ages of the detritus at the various depths with the majority of detritus in the surface layers of the sand being of more recent origin than that below. It could be argued then that the organic carbon measured in the depths of the filter is more refractory than that at the surface.

Results of the redox potential investigations have indicated the presence of microanaerobic zones in an otherwise well-oxygenated environment (percentage saturation of the outflow water rarely fell below 40%). The presence of these zones emphasises the heterogeneity of the slow sand filter previously demonstrated by organic carbon and meiofaunal distributions. These anaerobic zones have also been reported from other habitats which are similarly relatively well oxygenated. The level of Eh recorded (375 - 535mV) suggested that nitrate reducing processes are proceeding. These would appear to cease at about 30cm depth, where the Eh value goes above 450mV, which could be caused by limiting substrate conditions at this depth for the nitrate reducing bacteria.

Analyses of the vertical distribution of meiofauna in the sampling cores has revealed three types of distribution. The first includes the nematodes and chironomid larvae which shown maximum abundance at the surface and decrease in abundance with depth. Reasons for this type of distribution may be due to a high demand for food, corresponding with the high levels of organic carbon in the top few cms or due to a high demand for dissolved oxygen. Alternatively, as is most likely the case with the chironomid larvae, it may be related to the nature of the life cycle.

The second type of distribution was demonstrated by E. buchholzi which showed avoidance of the top few cms of the filter with maximum abundance just below this region. This could have been due to the physical effect of high levels of detritus in the top few cms of the filter causing the environment to become less penetrable to this relatively large organism. This distribution may also indicate that the enchytraeid was able to tolerate lower oxygen levels than the nematodes and chironomid larvae.

The third type of distribution was shown by the naidids, Aelosoma, microturbellaria and harpacticoids and demonstrated more or less no zonation with depth. These organisms are relatively small and hence may have a greater mobility in the interstitial system and thus may not be so limited by the distribution of food. They must also be able to tolerate lower oxygen levels occurring in the depths of the filter.

Similar distribution patterns have been reported by other authors (Fenchel 1969 and Maguire 1977) who related meiofaunal distributions to the oxygen and redox conditions in the sand.

Factors controlling horizontal and vertical distributions of organisms can thus be seen to be very complex. The degree of aeration of the sand system is dependent on the degree of exchange of interstitial water, drainage of the system, the amount of oxygen consuming organic matter present and limiting substrate concentrations for bacteria and algae. All of these factors may affect the type and distribution of bacteria and hence their predators in the system.

The length - dry weight relationship of the only sexually reproducing oligochaete E. buchholzi was found to be significant ($p < \cdot001$) and described by the relationship $W = 1\cdot37.10^{-5}.L^{1\cdot70}$ where W = dry weight in μg and L = body length in μm. The slope of the relationship, b = 1·70 indicated the very linear mode of growth in oligochaetes which compared with published regression coefficients of other oligochaetes. A useful relationship between segment number and dry weight was also investigated and found to be significant ($p < \cdot001$). This relationship was expressed by $W = 2\cdot24.10^{-4}.S^{3\cdot19}$ where S = segment number of the worm. The mean dry weight of mature worms (i.e. with eggs) was found to be 13·1 μg, of immature worms (no eggs) 6·4 μg and of worms recently hatched from cocoon to be 1·2 μg. The mean segment number of worms on hatching from cocoons was 14·5 with a range of 12 - 16. A significant relationship ($p < \cdot001$) was demonstrated between the number of ova per cocoon and cocoon length (field data). The mean number of ova per cocoon was much less (1·188) than the number of juveniles hatching from cocoons under laboratory conditions (3·7). This suggested mortalities of ova in cocoons in the field of 68%, although this result may have been due to the different filtration conditions which the laboratory hatched cocoon had originally been subjected to in the field.

This enchytraeid worm was much more abundant than the other oligochaete worms present. It may owe its success to the fact that it is the only actively sexually reproducing worm present. Thus it produces resistant eggs which may survive better than the adult stages of other oligochaetes, during the washing and storage procedures involved in cleaning the sand, thus getting replaced into a bed in resanding operations and thus forming the basis of a new population.

There would appear to be little seasonal fluctuations in abundance of E. buchholzi, although there may be a suggestion of slightly higher numbers in July/August followed by a decline in

September to October. It would appear that egg laying and hatching procedures occur at all times of the year, though numbers of cocoons containing eggs indicate slightly higher numbers in June, July and August, followed by an increase in the number of recently hatched worms in August/September. There was a relative decrease in the proportion of mature (worms with eggs) worms in the population between July and October which may be due to degeneration of the sexual organs following the increased egg laying in the spring, a phenomenon noted by other authors.

The chironomid larvae exhibit two peaksof abundance in June/July and in October, whilst the nematodes show higher numbers in August/September followed by a decline in the winter. On the whole there would appear to be little obvious seasonality in numerical abundance of the meiofauna, a phenomenon reported from other interstitial habitats which receive large amounts of depositing organic matter. It would seem that the all year round high availability of food tends to mask growth responses to temperature fluctuations which are normally made apparent in population size fluctuations.

Thethree naidid worms and A. hemprichi were never found to be sexually active. Varying proportions of the populations were found to be asexually budding. It would appear from the literature that naidids often resort to sexual reproduction during the winter months, possibly to produce resistant "overwintering" eggs which will survive poor conditions and provide the basis of a new population in the spring when improved conditions return. The absence of these sexual forms might indicate relatively favourable conditions in the slow sand filter so that there is no need to produce these resistant stages.

An investigation of the relationships of instantaneous rates of change of the population (r) and ambient temperatures, was made. Although many other factors will affect rates of change of the population, it was possible to demonstrate temperature dependence, but in only two groups - the nematodes and flatworms. The initial values of r for each sampling run of the chironomid larvae were much higher than later on the run and also shown to be temperature dependent and would appear to demonstrate high rates of hatching at the start of each filer run. This overall lack of temperature dependence on rates of change of the populations would tend to agree with the overall lack of seasonality demonstrated earlier and could suggest that other environmental factors must be overriding the effect of their physiological rate processes.

This study has provided much hitherto unknown information regarding the spatial and temporal distribution of meiofaunal populations in slow sand filters and has also pointed to the difficulties involved with sampling such a system. Detailed information concerning the state of oxidation of the system has been compiled with the use of a small scale pilot filter. It is clear that this investigation has pointed to many gaps in the knowledge of these interstitial systems and has emphasised the need for more research into the biology of slow sand filters. Areas which need attention include the food requirements and energetic relationships of the various groups; the effect of meiofaunal populations on the allochthonous and autochthonous organic carbon production and hence their effect on head loss rates; more research needs to be directed into the substrate and respiratory requirements of the algae and bacteria on which browsing meiofaunal populations may be dependent. It is hoped that this investigation has provided some of the background information from which future studies can be initiated.

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Appendix 3.1 Materials used in the construction of sampling apparatus.

Material

1. uPVC drainpiping I.D.= 100mm, wall= 3.2mm. 4x2m lengths.
2. " " I.D.=150mm, wall= 4.1mm. 4x2m lengths.
3. Perspex plate 6mm thick.
4. 9-11cm ajustable jubilee clips.
5. 20cm diameter plastic filter funnels
6. Spherical polystyrene floats, 8cm diameter.
7. 1m lemgths of bamboo cane.
8. Nylon string.

25 sets of apparatus were manufactured.

Appendix 4.1 Efficiency of extraction of meiofauna procedure.

	Replicates	Number of decantations						
		1	2	3	4	5	6	7
Total % organisms removed	1	62	83	91	96	98	99	100
	2	66	85	96	99	100	100	100
	3	49	79	93	95	100	100	100
	4	69	86	95	98	99	100	100
	5	71	89	97	99	100	100	100
	6	88	97	100	100	100	100	100
	7	44	82	91	97	100	100	100
	8	65	87	94	97	99	99	100
	9	67	85	94	99	100	100	100
	10	56	83	94	97	99	99	100
n		10	10	10	10	10	10	10
$\bar{x}$		63.7	85.6	94.5	97.7	99.5	99.7	100
S^2		150.68	23.82	7.39	2.46	0.50	0.23	0
± 95% confidence limits		8.78	3.49	1.94	1.12	0.51	0.15	0

Appendix 4.2 Efficiency of enumeration procedure.

		Number of scans					
	Replicates	1	2	3	4	5	6
Total % organisms removed	1	33	70	89	100	100	100
	2	41	68	87	96	100	100
	3	50	72	92	98	100	100
	4	45	75	95	100	100	100
	5	42	82	94	100	100	100
	6	72	85	95	98	100	100
	7	49	72	88	95	100	100
	8	55	79	86	95	100	100
	9	33	92	100	100	100	100
	10	39	67	82	95	99	100
n		10	10	10	10	10	10
$\bar{x}$		45.9	76.2	90.8	97.7	99.9	100
S^2		134.54	66.18	28.62	5.12	0.1	0
± 95% confidence limits		8.30	5.82	3.83	1.62	0.23	0

Appendix 4.3 Apparatus and reagents used in the wet oxidation digestion of organic carbon.

Apparatus

1. Glass vials (10ml volume) with snap fastening plastic lids, for storage and shaking.
2. Oxford pipettors, 1ml amd 4ml.
3. Reaction vessels, 'Monax" pyrex glass, 35ml volume.
4. Magnetic stirrer with small bar and good speed control.
5. Piston type burette for ferrous ammonium sulphate, Metrohm E457, 5ml volume.
6. Combined platinum-calomel reference electrode, Metrohm EA234.
7. Meter with a high internal resistance which can be used to record potential difference, preferably one with a variable back-off potentiometer control ('buffer control' of a pH meter) to eliminate the small current present before the end point is reached. Pye Dynacap pH meter.
8. A potential source to keep the platinum electrode at +1 volt, 1.5 volt Nife cell used in conjunction with a tap off potentiometer.

Reagents

1. 0.2N potassium dichromate
2. Concentrated sulphuric acid.
3. 0.1N ferrous ammonium sulphate. Store in the cool in a brown bottle. Standardise daily before use.

Appendix 4.4 Analysis of length of shaking period required to bring dried organic carbon particles into suspension.

Length of shaking period	Amount of C present in each 0.5ml aliquot removed from bottle (µg)			Total C removed from 1.5ml (µg)	Cumulative total removed (µg)	Amount of C in a 5ml vol. µg				Cumulative total (µg) in 5ml.
	Replicates					Replicates			$\bar{x}$	
5 mins	117.79	111.67	130.05	359.51	359.51	1177.9	1116.7	1300.5	1198.37	1198.37
30 mins	96.04	98.46	111.54	306.04	665.55	960.4	984.6	1115.4	1020.13	1379.64
60 mins	83.27	96.04	85.57	264.88	930.43	832.7	960.4	855.7	882.93	1548.48
2 hrs	80.63	72.94	76.79	230.36	1160.79	806.3	729.4	767.9	767.87	1698.30
3 hrs	66.23	52.14	49.42	167.79	1328.58	662.3	521.4	494.2	559.3	1720.09
4 hrs	40.04	42.58	38.91	121.55	1450.11	400.4	421.8	389.1	405.10	1733.68

Appendix 5.1 Precision of weighing a platinum pan on a Cahn Gram Electrobalance.

Replicate weighings of pan µg				
659.6	693.2	693.0	694.8	694.8
694.2	695.0	694.0	693.6	694.2
694.4	694.2	694.2	695.0	694.0
694.8	694.2	694.2	694.6	694.4
694.2	694.8	695.0	694.6	694.6
695.6	694.8	694.8	694.4	694.4
694.6	694.8	694.8	694.6	694.6
694.8	694.8	694.8	695.4	695.6
695.2	695.0	694.6	695.6	694.8
695.2	695.2	695.4	695.4	695.4

error ?

n= 50

$\bar{x}$=694.7

S= 0.570 (0.082% of $\bar{x}$)

SE= $\pm$ 0.163 (0.023% of $\bar{x}$)

Appendix 5.2 E. buchholzi, percentage frequency of egg number per cocoon, 1. field data, this study; 2. Learner's 1972 laboratory hatched data; 3. laboratory hatched juveniles, this study.

1. Field data, this study.

Run	Day	% frequency, egg no. per cocoon						Mean egg no. per cocoon
		1	2	3	4	5	>5	
2	1	80	20					1.149
	15	68	10				19	1.091
	29	84	15	1				1.117
	46	38	63					1.542
3	4	58	37	3	2			1.368
	19	100						1.000
4	1	86	14					1.104
	12	96	3	1		1		1.046
	26	94	2	4				1.061
	41	79	21					1.157
	55	92	8					1.059
5	3	100						1.000
	16	56	39		3		3	1.430
	30	100						1.000
7	1	100						1.000
	23	49	43	5	3			1.487
	37	64	33	3				1.296
	51	75	22	2				1.193
Total		78	19	2	0.4	0.1	0.7	1.188

2. Learner's 1972 laboratory hatched data.

Temp	% frequency of egg no. per cocoon									Mean egg no. per cocoon using log x
	1	2	3	4	5	6	7	8	9	
8°C	0	23.8	23.8	23.8	17.1	3.8	7.6	0	0	3.5
15°C	5.9	26.5	23.5	17.6	11.8	7.1	4.7	2.9	0	3.3
20°C	6.7	30.9	26.4	16.9	4.5	2.2	4.5	2.8	0.6	2.9

3. Laboratory hatched juveniles, this study.

Temp	% frequency of juveniles per cocoon									Mean no. of juveniles per cocoon
	1	2	3	4	5	6	7	8	9	
15°C	6.7	13.3	20	20	13.3	13.3	10	0	3.3	3.7

Appendix 5.3 E. buchholzi, dimensions of cocoons and no. of juveniles hatched per cocoon, under laboratory conditions 15°C.

length µm	width µm	volume $mm^3.10^3$	total hatched	segment nos.
387.0	206.4	8.632	1	14
335.4	232.2	9.469	1	13
309.6	245.1	9.738	3	15 14 14
332.5	258.0	11.240	3	15 14 15
335.4	232.2	9.469	2	15 14
516.0	258.0	17.984	6	15 15 15 16 15 15
464.4	258.0	16.186	3	15 15 15
464.4	361.2	24.312	7	15 14 15 14 15 15 14
309.6	309.6	15.538	2	15 15
309.6	283.8	13.056	3	15 15 15
412.8	335.4	24.314	5	15 15 15 15 15
335.4	283.8	14.144	4	14 14 14 14
464.4	361.2	31.724	5	15 15 15 15 15
399.9	283.8	16.865	4	14 14 14 14
516.0	387.0	40.464	9	14 14 14 14 14 14 14 14 14
464.4	387.0	36.418	6	13 13 13 13 12 13
451.5	322.5	24.588	4	15 15 15 15
516.0	322.5	28.100	6	13 14 14 14 14 14
387.0	270.9	14.871	4	14 14 14 14
399.9	270.9	15.366	4	15 15 15 15
387.0	322.5	21.075	3	13 13 13
516.0	335.4	30.393	5	16 15 15 15 15
322.5	245.1	10.144	2	14 15
567.6	425.7	53.858	6	15 15 15 15 15 15
387.0	258.0	13.488	3	16 15 15
438.6	322.5	23.885	7	14 14 14 14 14 14 14
258.0	232.2	7.284	2	15 15
451.5	335.4	26.594	7	14 14 14 14 14 14 14
438.6	283.8	18.497	4	15 15 15 15
438.6	335.4	25.834	5	16 15 15 15 15

mean length= 410.6 µm, 95% confidence limits= 381.7-439.4

mean width= 298.9 µm, " " " =278.8-318.9

Appendix 6.1 Sampling dates and temperatures.

Run	Starting dates and finishing dates	Sampling dates	Age of run (days)	Length of run (days)	Temp. °C
1	13/3/75				
	28/4/75			46	
2	1/5/75	2/5/75	1		11.0
		16/5/75	15		12.4
		30/5/75	29		14.0
		16/6/75	46		15.5
	17/6/75			47	
3	20/6/75				
		24/6/75	4		17.5
		9/7/75	19		18.3
	14/7/75			24	
4	16/7/75				
		17/7/75	1		19.0
		28/7/75	12		19.5
		11/8/75	26		20.5
		26/8/75	41		20.5
		9/9/75	55		18.5
	17/9/75			63	
5	22/9/75				
		25/9/75	3		15.5
		8/10/75	16		13.6
		22/10/75	30		11.2
	27/10/75			35	
6	31/10/75				
		6/11/75	6		11.0
	7/1/75			68	
7	14/1/75				
		15/1/76	1		5.8
		6/2/76	23		3.8
		20/2/76	37		4.5
		5/3/76	51		5.5
	8/3/76			54	

Appendix 6.2 Carbon data from 3 separate cores removed on same sampling trip showing 3 replicates per depth.

Depth cm	Carbon conc. $\mu g.cm^{-3}$								
	Core 1			Core 2			Core 3		
	Replicates			Replicates			Replicates		
1	1228	1472	1061	1768	1295	1117	2132	1934	2075
2	91	85	100	979	430	252	223	215	343
3	206	174	207	318	219	420	138	208	122
4	141	201	130	197	171	164	196	189	225
5	72	99	116	200	221	252	278	82	155
7	122	310	91	132	137	71	139	172	101
9	92	55	110	93	56	208	210	96	187
11	281	267	231	175	247	99	233	222	150
13	690	274	144	260	209	224	252	152	284
15	84	162	177	234	137	228	152	57	237
17	197	172	206	196	160	160	83	217	234
19	124	97	316	84	97	172	24	553	158
21	184	176	594	230	168	116	184	73	154
23	153	184	246	398	323	324	220	228	261
25	90	258	158	360	405	365	20	142	91

Appendix 6.3 Carbon data from sampling cores collected over the sampling period, (μg C. cm^{-3})

Run 4

Day	1			12			26			41			55		
Depth															
1 cm	169	*	*	617	677	403	694	547	1480	258	3803	4929	4986	*	*
2	157	*	*	138	151	221	365	147	525	101	76	3802	2660	*	*
3	108	*	*	150	84	255	107	231	455	78	129	148	200	*	*
4	147	*	*	192	180	257	126	90	501	157	172	217	221	*	*
5	142	*	*	37	35	275	97	163	454	56	56	174	190	*	*
6	173	*	*	193	181	251	82	340	450	45	117	314	218	*	*
7	126	*	*	225	226	277	181	378	336	79	164	343	212	*	*
8	90	*	*	177	174	286	196	412	797	53	23	216	154	*	*
9	12	*	*	60	111	276	191	327	619	69	27	160	165	*	*
10	*	*	*	82	179	335	69	272	791	51	184	156	196	*	*
Mean C below 2cm	107	*	*	126	131	239	122	244	470	67	89	198	170	*	*

Run 5

Day	3			16			30		
1 cm	162	133	187	607	423	437	1167	1367	939
2	98	158	165	394	303	374	508	576	691
3	148	*	134	185	214	232	260	430	346
5	138	133	184	216	242	245	261	355	213
7	146	138	191	205	238	276	258	274	233
9	*	127	175	199	225	267	285	257	250
11	144	158	197	187	252	204	291	277	236
13	143	165	178	213	232	245	227	284	246
15	147	168	117	216	117	230	259	164	177
Mean C below 2cm	133	114	163	188	211	226	243	269	220

Appendix 6.3 continued.

Run 7

Day	1			23			37			51		
1 cm	274	266	215	1567	519	412	1351	950	840	631	1029	2205
2	133	96	186	978	141	169	307	118	241	301	511	858
3	133	152	225	302	97	157	169	54	179	199	389	595
4	168	67	150	268	90	113	183	51	195	330	397	418
5	85	58	203	366	205	157	143	79	178	207	411	328
7	104	86	122	132	100	102	121	69	180	222	317	268
9	65	131	113	71	103	105	149	136	176	205	309	346
11	87	109	87	179	93	142	69	79	156	222	252	305
13	56	129	78	141	61	132	77	154	111	146	360	200
15	83	90	91	266	63	104	71	129	166	125	242	250
17	81	131	140	224	89	97	99	164	117	129	233	258
19	78	104	78	347	74	72	157	156	42	88	185	195
21	89	66	72	111	213	71	110	234	77	87	411	153
23	55	125	*	182	204	100	8	49	135	131	127	152
25	105	110	60	129	137	82	55	35	106	320	201	129
Mean C below 2cm	80	99	102	192	113	103	97	112	130	162	271	242

* = no sample

Appendix 6.4 Carbon data from bed 45 , T.W.A. analyses. ($\mu g\ C.cm^{-3}$)

Run	1	2	3	5	6	7
Depth						
0-2.5 cm	703	1820	1262	1120	1919	2277
2.5-5	640	678	1028	559	890	1548
5-7.5	345	309	630	386	879	1273
7.5-10	308	891	594	248	920	951
10-12.5	364	635	712	171	748	575
21.5-22.5	278	280	302	123	441	698
31.5-32.5	233	226	161	106	111	163
Bottom	322	144	505	992	440	763
(cm)	66	63.5	61	58.5	56	53.5

All samples taken at the end of each filter run.

Appendix 6.5 Counts of E. buchholzi removed from sand samples. nos.40cm^{-3}

Run 2

Day	1			15			29			46		
Depth												
1 cm	0	0	2	0	0	0	1	0	0	70	0	141
2	0	0	0	1	45	8	0	0	0	0	48	1
3	0	0	0	0	1	0	0	0	3	0	12	7
5	0	0	1	1	5	0	3	4	0	22	206	1
7	1	0	1	2	59	0	0	0	1	12	55	1
9	1	0	0	0	57	1	1	0	0	0	1	5

Run 3

Day	4			19		
Depth						
1 cm	0	5	0	0	0	0
2	9	3	1	0	95	6
3	0	8	1	0	195	170
5	0	27	5	1	30	67
7	8	0	0	2	17	72
9	14	1	2	1	15	36

Run 4

Day	1			12			26			41			55		
Depth															
1 cm	0	*	*	0	26	1	276	336	9	182	0	0	30	*	*
2	0	*	*	0	1	71	14	206	93	40	0	180	258	*	*
3	2	*	*	5	4	20	0	138	34	18	88	98	44	*	*
4	0	*	*	0	1	18	94	2	82	0	16	46	60	*	*
5	0	*	*	3	0	31	26	11	14	62	14	42	20	*	*
6	4	*	*	2	1	15	30	5	38	0	21	0	20	*	*
7	1	*	*	11	15	16	2	1	42	0	9	0	17	*	*
8	0	*	*	6	5	80	70	6	5	32	6	34	26	*	*
9	0	*	*	11	2	22	14	11	1	14	13	14	20	*	*
10	0	*	*	61	14	24	48	0	5	24	19	26	34	*	*

Appendix 6.5 continued.

Run 5

Day	3			16			30		
Depth									
1 cm	0	1	0	5	0	30	1	2	*
2	0	1	1	9	1	12	5	8	*
3	0	5	0	18	4	12	6	2	*
4	0	4	1	8	4	4	5	1	*
5	0	3	0	5	7	0	4	8	*
6	0	6	0	9	0	8	3	8	*
7	0	4	0	11	4	13	12	9	*
8	0	1	0	7	12	6	14	8	*
9	1	2	1	16	16	8	11	13	*
10	1	5	0	22	39	20	22	0	*
11	0	0	1	16	43	111	24	28	*
12	2	3	3	22	29	84	6	12	*
13	0	2	1	25	7	40	1	34	*
14	0	0	1	18	11	31	3	0	*
15	1	1	6	20	4	14	3	0	*

Run 7

Day	1			23			37			51		
Depth												
1 cm	0	0	0	2	0	0	1	1	2	3	11	1
2	3	0	0	12	1	6	1	3	1	11	20	1
3	0	0	0	39	0	28	2	8	3	22	41	1
4	2	0	0	72	0	29	3	13	5	80	87	0
5	0	0	0	71	0	4	11	49	15	86	128	0
7	0	0	0	40	0	30	46	53	20	124	70	1
9	0	1	0	16	2	23	29	42	18	123	81	0
11	0	0	0	11	0	12	13	68	18	41	30	0
13	0	0	0	43	0	20	31	78	19	50	29	1
15	0	1	0	27	0	32	59	80	4	35	17	0
17	0	0	1	73	1	16	48	68	7	43	20	0
19	0	1	0	96	0	15	93	96	6	45	6	0
21	0	0	0	17	1	24	71	103	12	25	25	0
23	0	0	2	13	2	25	20	23	7	17	6	2
25	0	0	0	4	3	1	6	8	0	4	2	1

*= no sample.

Appendix 6.6a Counts of E. buchholzi, cocoons with eggs, removed from sand samples. nos.$40cm^{-3}$

Run 2

Day	1			15			29			46		
Depth												
1 cm	0	5	41	1	65	1	0	0	0	10	65	9
2	0	0	5	0	4	1	0	1	1	12	15	18
3	0	0	1	0	0	1	1	0	5	7	0	8
5	1	1	4	1	2	1	0	0	0	4	3	1
7	1	0	2	0	0	0	0	0	0	9	1	6
9	1	0	0	0	0	2	0	0	0	0	5	7

Run 3

Day	4			19		
Depth						
1 cm	60	12	6	2	75	60
2	8	1	0	0	7	40
3	5	5	2	2	3	2
5	3	0	1	3	0	4
7	2	2	1	2	1	4
9	0	0	0	0	2	3

Run 4

Day	1			12			26			41			55		
Depth															
1 cm	2	*	*	0	0	48	0	18	18	21	10	0	5	*	*
2	0	*	*	18	3	0	6	0	24	4	0	0	12	*	*
3	0	*	*	7	6	7	8	2	0	2	4	4	8	*	*
4	0	*	*	15	8	1	4	0	2	2	2	0	0	*	*
5	4	*	*	0	0	3	8	0	0	0	0	0	4	*	*
6	1	*	*	7	1	10	2	2	0	0	0	0	2	*	*
7	0	*	*	7	4	7	2	0	2	0	0	0	0	*	*
8	0	*	*	2	0	0	0	0	6	2	0	2	2	*	*
9	0	*	*	5	2	8	4	2	0	2	1	0	0	*	*
10	0	*	*	1	0	4	2	0	0	2	0	0	0	*	*

Appendix 6.6a continued.

Run 5

Day	3			16			30		
Depth									
1 cm	0	0	0	2	0	0	0	0	*
2	0	0	0	4	1	0	0	0	*
3	0	0	0	8	0	0	0	0	*
4	0	0	0	1	0	0	0	0	*
5	0	0	0	0	0	2	0	0	*
6	0	0	0	0	0	0	0	0	*
7	0	0	1	0	0	0	0	0	*
8	0	0	0	3	0	0	0	0	*
9	0	0	0	2	0	0	0	1	*
10	0	0	0	2	1	0	0	0	*
11	0	0	0	3	0	0	0	0	*
12	0	0	0	0	0	2	0	0	*
13	0	0	0	2	0	0	0	0	*
14	0	0	0	1	0	0	0	0	*
15	0	0	0	1	0	0	0	0	*

Run 7

Day	1			23			37			51		
Depth												
1 cm	0	0	0	16	0	0	2	0	1	31	41	0
2	0	0	0	4	0	1	1	2	0	52	30	0
3	0	0	0	1	0	0	5	0	2	20	5	0
4	0	0	0	1	0	0	1	1	2	3	6	0
5	0	0	0	1	0	0	0	0	1	2	3	0
7	0	0	0	2	0	0	0	5	1	1	17	0
9	0	0	0	2	0	0	0	0	3	4	4	0
11	0	0	0	0	0	0	0	1	0	o	2	0
13	0	0	0	0	0	0	0	0	1	1	6	0
15	0	0	0	0	0	0	1	2	2	6	6	0
17	0	0	0	0	0	0	2	1	0	8	4	0
19	0	0	0	2	0	0	0	0	1	8	4	0
21	0	0	0	1	0	0	1	0	2	9	1	0
23	0	0	0	0	0	0	0	1	5	5	3	0
25	0	0	0	0	0	0	7	2	3	11	2	0

*= no sample.

Appendix 6.6b Counts of E. buchholzi, empty cocoons, removed from sand samples. nos.40cm^{-3}

Run 2

Day	1			15			29			46		
Depth												
1 cm	1	7	9	0	5	0	0	2	0	80	210	69
2	0	1	1	0	4	0	0	2	17	16	36	6
3	0	2	0	0	1	0	0	2	3	17	11	13
5	2	1	1	0	2	0	0	2	0	2	20	1
7	2	0	4	3	2	0	0	0	1	11	3	3
9	3	0	1	1	0	0	0	0	0	1	4	3

Run 3

Day	4			19		
Depth						
1 cm	0	5	0	0	0	0
2	2	0	2	1	0	6
3	0	0	3	4	0	2
5	0	0	0	6	8	2
7	1	0	3	1	0	1
9	0	0	2	0	0	1

Run 4

Day	1			12			26			41			55		
Depth															
1 cm	0	*	*	0	0	0	0	12	42	119	15	0	0	*	*
2	0	*	*	0	0	0	0	30	21	12	6	25	15	*	*
3	0	*	*	0	0	0	0	4	10	14	24	22	4	*	*
4	0	*	*	0	0	0	0	12	26	16	18	12	4	*	*
5	2	*	*	0	0	0	0	12	16	6	8	14	4	*	*
6	0	*	*	0	0	0	0	14	32	8	4	14	6	*	*
7	0	*	*	0	0	0	0	12	32	8	3	20	4	*	*
8	0	*	*	0	0	0	0	10	16	14	6	24	0	*	*
9	0	*	*	0	0	0	0	10	42	12	5	8	14	*	*
10	0	*	*	0	0	0	0	8	32	10	4	6	6	*	*

Appendix 6.6b continued.

Run 5

Day	3			16			30		
Depth									
1 cm	0	0	0	0	0	0	0	0	*
2	0	0	0	2	0	0	0	0	*
3	0	0	0	1	0	0	0	0	*
4	0	0	0	0	0	0	0	0	*
5	0	0	0	3	0	0	0	0	*
6	0	0	0	0	0	0	0	0	*
7	0	0	0	2	2	0	0	0	*
8	0	0	0	0	0	0	0	0	*
9	0	0	0	0	0	0	0	1	*
10	0	0	0	2	1	0	0	0	*
11	0	0	0	0	2	0	0	0	*
12	0	0	0	0	0	0	0	0	*
13	0	0	0	1	0	0	0	0	*
14	0	0	0	0	0	0	0	0	*
15	0	0	0	0	0	0	0	0	*

Run 7

Day	1			23			37			51		
Depth												
1 cm	0	0	0	0	0	0	0	2	0	2	1	8
2	0	0	0	0	0	1	0	0	0	0	0	1
3	0	0	0	0	0	0	0	0	0	0	0	0
4	0	0	0	0	0	0	0	0	0	0	0	0
5	0	0	0	0	0	0	0	0	0	0	0	0
7	1	0	0	0	1	0	0	2	0	0	0	0
9	0	0	0	0	1	0	0	0	0	0	0	0
11	0	0	0	0	0	0	0	0	0	0	0	2
13	0	0	0	0	0	0	0	1	1	0	0	0
15	0	0	0	0	0	0	0	0	0	0	0	0
17	0	0	0	0	0	2	0	0	0	0	0	0
19	0	0	0	0	0	2	0	0	0	1	0	0
21	0	0	0	1	0	0	0	0	0	0	0	0
23	0	0	0	0	0	0	1	0	0	1	1	1
25	1	0	0	0	0	0	1	0	0	0	0	0

*= no sample.

Appendix 6.7 Counts of A. hemprichi removed from sand samples, nos.40cm^{-3}

Run 2

Day	1			15			29			46		
Depth												
1 cm	0	0	0	0	0	1	18	0	0	25	0	6
2	0	0	0	0	3	47	0	0	0	0	0	0
3	0	0	0	6	0	0	18	0	0	0	0	0
5	0	0	0	7	0	0	0	0	0	213	123	1
7	0	0	1	5	0	44	0	0	0	33	16	3
9	0	0	0	1	1	18	0	0	0	0	0	4

Run 3

Day	4			19		
Depth						
1 cm	0	3	0	0	0	0
2	23	2	0	0	2	0
3	1	6	6	0	1	0
5	0	8	1	4	0	0
7	7	0	2	1	4	0
9	7	0	0	6	0	0

Run 4

Day	1			12			26			41			55		
Depth															
1 cm	0	*	*	0	1	0	4	1	0	0	0	0	0	*	*
2	0	*	*	0	0	63	0	0	11	4	0	0	87	*	*
3	0	*	*	14	2	6	0	0	1	0	2	1	0	*	*
4	3	*	*	0	0	0	9	0	4	0	0	0	0	*	*
5	0	*	*	0	7	0	4	0	18	0	1	0	0	*	*
6	0	*	*	0	8	2	0	0	2	0	0	0	0	*	*
7	0	*	*	1	12	7	0	0	8	0	0	0	1	*	*
8	0	*	*	1	4	22	37	0	7	0	1	0	2	*	*
9	2	*	*	1	0	0	6	0	0	0	0	0	1	*	*
10	1	*	*	0	1	6	1	2	0	0	0	0	1	*	*

Appendix 6.7 continued

Run 5

Day	3			16			30		
Depth									
1 cm	1	0	0	0	0	0	0	0	*
2	5	0	0	0	0	0	0	0	*
3	0	0	0	0	0	0	0	0	*
4	1	0	0	0	0	0	0	0	*
5	2	1	0	0	0	0	0	0	*
6	1	0	0	0	0	0	0	0	*
7	1	0	0	0	0	0	0	0	*
8	3	0	0	0	0	0	0	0	*
9	0	0	0	0	0	0	0	0	*
10	1	1	0	0	0	0	0	0	*
11	1	0	0	0	1	0	0	0	*
12	0	0	0	0	0	0	0	0	*
13	0	1	0	0	0	0	0	0	*
14	3	0	0	0	0	0	0	0	*
15	4	0	0	0	0	0	0	0	*

Run 7

Day	1			23			37			51		
Depth												
1 cm	0	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0	0
4	0	0	0	0	0	0	0	0	0	0	0	0
5	0	0	0	0	0	0	0	0	0	0	0	0
7	0	0	0	0	0	0	0	0	0	0	0	0
9	0	0	0	0	0	0	0	0	0	0	0	0
11	0	0	0	0	0	0	0	0	0	0	0	0
13	0	0	0	0	0	0	0	0	0	0	0	0
15	0	0	0	0	0	0	0	0	0	0	0	0
17	0	0	0	0	0	0	0	0	0	0	0	0
19	0	0	0	0	0	0	0	0	0	0	0	0
21	0	0	0	0	0	0	0	0	0	0	0	0
23	0	0	0	0	0	0	0	0	0	0	0	0
25	0	0	0	0	0	0	0	0	0	0	0	0

*= no sample.

Appendix 6.8 Counts of P. idrensis removed from sand samples, nos.40cm^{-3}

Run 2

Day	1			15			29			46		
Depth												
1 cm	0	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0	0
5	0	0	0	0	0	0	0	0	0	0	0	0
7	0	0	0	0	0	0	0	0	0	0	0	0
9	0	0	0	0	0	0	0	0	0	0	0	0

Run 3

Day	4			19		
Depth						
1 cm	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	0	0
5	0	0	0	0	0	0
7	0	0	0	0	0	0
9	0	0	0	0	0	0

Why so few?

Run 4

Day	1			12			26			41			55		
1	0	*	*	0	0	0	6	1	0	0	0	0	6	*	*
2	0	*	*	0	0	1	0	4	0	0	0	0	47	*	*
3	0	*	*	1	0	0	0	3	2	0	0	0	0	*	*
4	0	*	*	0	1	0	1	3	19	0	0	0	2	*	*
5	0	*	*	0	0	0	1	0	2	0	0	0	2	*	*
6	0	*	*	0	3	0	1	1	1	0	0	0	6	*	*
7	0	*	*	0	0	0	0	0	0	0	0	0	8	*	*
8	0	*	*	0	0	0	0	0	0	0	0	0	9	*	*
9	0	*	*	0	2	0	0	0	0	0	0	0	5	*	*
10	0	*	*	0	0	0	1	0	0	0	0	0	0	*	*

Appendix 6.8 continued.

Run 5

Day	3			16			30		
Depth									
1 cm	1	2	1	0	0	0	0	0	*
2	9	3	3	0	0	0	0	0	*
3	2	0	0	0	0	0	0	0	*
4	1	1	2	0	0	0	0	0	*
5	1	1	3	0	0	0	0	0	*
6	3	0	3	0	0	0	0	0	*
7	1	2	1	0	0	0	0	0	*
8	6	0	3	0	0	0	0	0	*
9	2	0	2	0	0	0	0	0	*
10	6	0	1	0	0	0	0	0	*
11	2	0	0	0	0	0	0	0	*
12	3	0	1	0	0	0	0	0	*
13	2	0	1	0	0	0	0	0	*
14	0	0	0	0	0	0	0	0	*
15	3	0	0	0	0	0	0	0	*

Run 7

Day	1			23			37			51		
Depth												
1 cm	0	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0	0
4	0	0	0	0	0	0	0	0	0	0	0	0
5	0	0	0	0	0	0	0	0	0	0	0	0
7	0	0	0	0	0	0	0	0	0	0	0	0
9	0	0	0	0	0	0	0	0	0	0	0	0
11	0	0	0	0	0	0	0	0	0	0	0	0
13	0	0	0	0	0	0	0	0	0	0	0	0
15	0	0	0	0	0	0	0	0	0	0	0	0
17	0	0	0	0	0	0	0	0	0	0	0	0
19	0	1	0	0	0	0	0	0	0	0	0	0
21	0	0	0	0	0	0	0	0	0	1	0	0
23	0	0	0	0	0	0	0	0	0	0	0	0
25	0	0	0	0	0	0	0	0	0	0	0	0

*= no sample.

Appendix 6.9 Counts of P. idrensis removed from sand samples, nos.40cm^{-3}

Run 2 -no individuals encountered.

Run 3 " " "

Run 4

Day	1			12			26			41			55		
Depth															
1	0	*	*	0	0	0	0	0	0	0	0	0	0	*	*
2	0	*	*	0	0	0	0	0	0	0	0	0	0	*	*
3	0	*	*	0	0	0	7	0	7	0	0	0	0	*	*
4	0	*	*	0	0	0	0	0	21	0	0	0	0	*	*
5	0	*	*	0	0	0	0	0	6	0	0	0	0	*	*
6	0	*	*	0	0	0	0	0	21	0	0	0	0	*	*
7	0	*	*	0	0	0	0	0	24	0	0	0	0	*	*
8	0	*	*	0	0	0	0	0	8	0	0	0	0	*	*
9	0	*	*	0	0	0	0	0	0	0	0	0	0	*	*
10	0	*	*	0	0	0	9	0	5	0	0	0	0	*	*

Run 5 -no individuals encountered.

Run 7 " " "

*= no sample.

Appendix 6. 10 Counts of N. elinguis removed from sand samples, nos.40cm^{-3}

Run 2

Day	1			15			29			46		
Depth												
1 cm	0	0	0	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0	0
5	0	0	0	0	0	0	0	0	0	2	1	0
7	0	0	0	0	0	0	0	0	0	0	0	0
9	0	0	0	0	0	0	0	0	0	0	0	0

Run 3

Day	4			19		
Depth						
1 cm	0	0	0	0	0	0
2	2	0	0	0	0	0
3	0	0	0	0	0	0
5	0	0	1	0	0	0
7	0	0	0	1	0	0
9	0	0	0	0	0	0

Run 4 -no individuals encountered.

Run 5 " " "

Run 7 " " "

Appendix 6.11 Counts of nematodes removed from sand samples, nos.40cm^{-3}

Run 2

Day	1			15			29			46		
Depth												
1 cm	1	0	1	0	0	1	2	0	116	30	o	363
2	0	0	0	0	0	0	3	0	0	1	24	0
3	0	0	0	2	0	0	0	0	5	0	0	0
5	0	1	0	0	0	0	1	0	0	17	24	0
7	0	0	0	0	0	0	1	0	0	0	1	0
9	0	0	0	0	0	0	0	0	1	0	0	0

Run 3

Day	4			19		
Depth						
1 cm	1	0	0	0	0	0
2	0	0	0	1	0	0
3	1	0	0	0	3	10
5	0	4	0	1	8	4
7	0	0	1	4	0	2
9	2	1	0	0	0	0

Run 4

Day	1			12			26			41			55		
Depth															
1 cm	1	*	*	0	72	1	114	174	744	385	0	5	40	*	*
2	0	*	*	1	4	11	2	0	0	42	6	30	39	*	*
3	0	*	*	4	0	21	2	30	6	18	56	48	16	*	*
4	0	*	*	2	0	1	30	0	48	4	26	56	32	*	*
5	2	*	*	1	0	5	12	6	22	20	28	42	12	*	*
6	0	*	*	0	0	3	0	2	14	0	23	28	10	*	*
7	0	*	*	4	2	1	0	0	6	4	23	30	10	*	*
8	0	*	*	0	0	12	32	8	4	10	12	30	14	*	*
9	0	*	*	0	0	0	12	8	2	4	12	42	4	*	*
10	0	*	*	0	0	0	2	4	4	8	11	42	2	*	*

Appendix 6.11 continued.

Run 5

Day	3			16			30		
Depth									
1 cm	5	0	0	0	1	0	0	0	*
2	0	0	1	8	8	0	0	0	*
3	4	0	0	1	6	0	0	0	*
4	5	2	0	1	2	0	0	0	*
5	0	0	0	0	0	0	0	0	*
6	0	0	0	3	0	0	0	0	*
7	4	0	2	1	1	0	0	0	*
8	1	1	2	3	1	0	0	0	*
9	2	1	2	0	1	0	0	1	*
10	2	1	0	0	1	0	0	0	*
11	2	0	0	0	1	0	0	0	*
12	0	0	1	0	0	0	0	0	*
13	0	0	1	0	0	0	0	0	*
14	1	2	0	0	0	0	0	0	*
15	1	1	0	0	0	0	0	0	*

Run 7

Day	1			23			37			51		
Depth												
1 cm	1	0	0	0	0	0	0	0	0	10	0	0
2	0	0	0	0	0	0	0	0	0	2	1	2
3	0	0	0	1	0	0	1	1	0	1	0	0
4	0	0	0	0	0	0	0	0	0	0	0	0
5	0	0	0	0	0	0	0	0	0	0	0	0
7	0	0	0	0	0	0	0	0	0	0	0	0
9	0	0	0	0	0	0	0	0	0	2	0	0
11	0	0	0	0	0	0	0	0	0	0	0	0
13	0	0	0	0	0	0	0	0	0	0	0	0
15	0	0	0	0	0	0	1	0	0	0	0	0
17	0	0	0	0	0	0	0	0	0	0	0	0
19	0	0	0	0	0	0	0	0	0	1	0	0
21	0	0	0	0	0	0	0	0	0	0	0	0
23	0	0	0	0	0	0	0	0	0	0	0	0
25	0	0	0	0	0	0	0	0	0	0	0	0

*= no sample.

Appendix 6.12 Counts of chironomid larvae removed from sand samples, nos.40cm^{-3}

Run 2

Day	1			15			29			46		
Depth												
1 cm	0	0	1	0	5	10	1	2	38	165	70	55
2	0	0	0	0	0	0	0	0	3	20	0	0
3	0	0	0	0	0	0	0	0	0	3	0	0
5	0	0	0	0	0	0	0	0	0	2	0	0
7	0	0	0	0	0	0	0	0	0	0	0	0
9	0	0	0	0	0	0	0	0	0	0	0	0

Run 3

Day	4			19		
Depth						
1 cm	30	23	57	1080	1870	710
2	0	0	0	28	15	190
3	0	0	0	7	0	0
5	0	0	0	4	0	0
7	0	0	0	4	0	0
9	0	0	0	0	0	0

Run 4

Day	1			12			26			41			55		
Depth															
1	0	*	*	0	0	0	21	3	0	0	20	0	10	*	*
2	0	*	*	0	1	0	2	0	0	0	0	0	0	*	*
3	0	*	*	0	0	0	2	0	0	0	0	0	0	*	*
4	0	*	*	0	0	0	0	0	0	0	0	0	0	*	*
5	0	*	*	0	0	0	0	0	0	0	0	0	0	*	*
6	0	*	*	0	0	0	0	0	0	0	0	0	0	*	*
7	0	*	*	0	0	0	0	0	0	0	0	0	0	*	*
8	0	*	*	0	0	0	0	0	0	0	0	0	0	*	*
9	0	*	*	0	0	0	0	0	0	0	0	0	0	*	*
10	0	*	*	0	0	0	0	0	0	0	0	0	0	*	*

Appendix 6.12 continued.

Run 4

Day	3			16			30		
Depth									
1 cm	0	0	0	176	57	13	122	64	*
2	0	0	0	18	6	14	4	8	*
3	0	0	0	1	2	4	2	7	*
4	0	0	0	4	0	1	0	2	*
5	0	0	0	0	2	1	1	4	*
6	0	0	0	0	0	1	0	2	*
7	0	0	0	0	0	0	0	2	*
8	0	0	0	1	0	0	0	1	*
9	0	0	0	0	0	0	0	7	*
10	0	0	0	1	0	0	0	32	*
11	0	0	0	0	0	0	2	4	*
12	0	0	0	0	0	0	0	34	*
13	0	0	0	1	0	0	0	2	*
14	0	0	0	0	0	0	0	0	*
15	0	0	0	0	0	2	0	4	*

Run 7

Day	1			23			37			51		
Depth												
1 cm	0	0	1	0	0	1	3	1	1	1	1	0
2	0	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0	0
4	1	0	0	0	0	0	0	0	0	0	0	0
5	0	0	0	0	0	0	0	0	0	0	0	0
7	0	0	0	0	0	0	0	0	0	0	0	0
9	0	0	0	0	0	0	0	0	0	0	0	0
11	0	0	0	0	0	0	0	0	0	0	0	0
13	0	0	0	0	0	0	0	0	0	0	0	0
15	0	0	0	0	0	0	0	0	0	0	0	0
17	0	0	0	0	0	0	0	0	0	0	0	0
19	0	0	0	0	0	0	0	0	0	0	0	0
21	0	0	0	0	0	0	0	0	0	0	0	0
23	0	0	0	0	0	0	0	0	0	0	0	0
25	0	0	0	0	0	0	0	0	0	0	0	0

*= no sample.

Appendix 6.13 Counts of harpacticoids removed from sand samples, nos.40cm^{-3}

Run 2

Day	1			15			29			46		
Depth												
1 cm	3	0	0	0	0	0	0	0	0	0	0	24
2	0	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0	24
5	0	0	0	0	0	0	0	0	0	47	7	9
7	0	0	0	0	0	0	0	0	0	32	1	16
9	0	0	0	0	0	0	0	0	0	0	0	13

Run 3

Day	4			19		
Depth						
1 cm	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	0	0
5	0	0	0	0	0	0
7	0	0	0	0	0	0
9	1	0	0	0	0	0

Run 4

Day	1			12			26			41			55		
Depth															
1 cm	0	*	*	0	3	0	0	0	0	0	0	0	0	*	*
2	1	*	*	3	2	0	0	0	0	0	0	5	21	*	*
3	0	*	*	5	3	1	0	10	0	2	4	4	0	*	*
4	0	*	*	6	0	0	10	0	8	0	2	0	24	*	*
5	1	*	*	4	1	0	0	2	4	2	10	6	0	*	*
6	0	*	*	0	6	0	4	4	6	16	3	0	44	*	*
7	2	*	*	2	4	1	0	0	56	6	1	0	10	*	*
8	0	*	*	2	1	2	30	8	14	0	3	2	30	*	*
9	1	*	*	2	0	1	16	2	0	0	1	0	48	*	*
10	0	*	*	0	0	1	18	2	8	6	2	0	72	*	*

Run 5 -no individuals encountered.

Run 7 -no individuals encountered.

*= no sample.

Appendix 6.14[a] Counts of the flatworm Catenula lemnae removed from sand samples, nos.40cm^{-3}

Run 2-no individuals encountered.

Run 3- " " "

Run 4

Day	1			12			26			41			55		
Depth															
1	0	*	*	0	0	0	0	0	0	0	0	0	0	*	*
2	0	*	*	0	0	0	0	2	3	0	0	0	15	*	*
3	0	*	*	0	0	0	0	8	4	0	0	6	0	*	*
4	0	*	*	0	0	0	2	0	28	0	0	0	8	*	*
5	0	*	*	0	0	0	0	0	18	0	0	0	4	*	*
6	0	*	*	0	0	0	0	0	34	0	0	0	2	*	*
7	0	*	*	0	0	0	0	4	68	0	0	0	0	*	*
8	0	*	*	0	0	0	5	0	10	0	0	0	0	*	*
9	0	*	*	0	0	0	6	0	0	0	0	0	4	*	*
10	0	*	*	0	0	0	6	2	6	0	0	0	0	*	*

Run 5

Day	3			16			30		
Depth									
1 cm	0	0	0	0	0	0	0	0	*
2	0	0	0	8	1	1	0	0	*
3	0	0	0	7	0	1	0	0	*
4	0	0	0	4	1	0	0	0	*
5	0	0	0	3	0	0	0	0	*
6	0	0	0	0	0	0	0	0	*
7	0	0	0	0	0	0	0	0	*
8	0	0	0	0	1	0	0	0	*
9	0	0	0	0	2	0	0	0	*
10	0	0	0	0	0	0	0	0	*
11	0	0	0	0	0	0	0	0	*
12	0	0	0	1	0	0	0	0	*
13	0	0	0	0	0	0	0	0	*
14	0	0	0	0	0	0	0	0	*
15	0	0	0	0	0	0	0	0	*

Run 7- no individuals encountered. *= no sample.

Appendix 6.14b Counts of flatworms, apartfrom C. lemnae, removed from sand samples. nos.$40cm^{-3}$

Run 2

Day	1			15			29			46		
Depth												
1 cm	0	0	0	0	0	0	1	0	0	10	0	6
2	0	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0	0
5	0	0	0	0	0	0	2	0	2	9	0	0
7	0	0	0	0	0	0	0	0	1	0	0	0
9	0	0	0	0	0	0	0	0	0	0	0	0

Run 3

Day	4			19		
Depth						
1 cm	0	0	0	0	0	0
2	0	0	0	0	0	0
3	0	0	0	0	2	0
5	0	0	0	0	0	1
7	0	0	0	0	0	0
9	0	0	0	0	0	0

Run 4

Day	1			12			26			41			55		
1 cm	0	*	*	0	0	0	0	0	0	0	0	0	0	*	*
2	0	*	*	0	0	0	0	0	0	0	0	0	9	*	*
3	0	*	*	0	0	0	0	4	0	0	0	0	12	*	*
4	0	*	*	2	1	0	2	0	0	0	0	0	28	*	*
5	0	*	*	0	1	0	0	0	0	0	4	0	56	*	*
6	0	*	*	0	0	0	0	10	6	0	0	0	16	*	*
7	0	*	*	0	0	0	0	0	0	0	0	0	8	*	*
8	0	*	*	0	0	0	1	4	4	0	0	0	10	*	*
9	0	*	*	0	0	0	4	14	0	0	0	0	18	*	*
10	0	*	*	0	0	0	2	2	0	0	0	0	8	*	*

Appendix 6.14b continued.

Run 5

Day	3			16			30		
Depth									
1 cm	0	0	0	0	0	0	0	0	*
2	0	0	0	22	0	0	0	0	*
3	0	0	0	2	0	0	4	2	*
4	1	0	0	0	0	2	0	1	*
5	2	0	0	0	0	0	2	1	*
6	0	0	0	1	0	1	0	1	*
7	2	0	0	1	1	0	2	0	*
8	0	0	0	0	0	1	1	0	*
9	0	0	0	0	0	1	0	0	*
10	1	0	0	0	0	0	0	0	*
11	1	0	0	0	0	0	2	0	*
12	0	0	0	0	0	0	1	0	*
13	1	0	0	0	0	0	1	0	*
14	0	0	0	0	0	0	2	0	*
15	0	0	0	0	0	0	0	0	*

Run 7

Day	1			23			37			51		
Depth												
1 cm	0	0	0	0	0	1	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	1	0	0	0	0	0	0	0	0
4	0	0	0	0	0	0	0	0	0	0	0	0
5	0	0	0	0	0	0	0	0	0	0	0	0
7	0	0	0	0	0	0	0	0	0	0	0	0
9	0	0	0	0	0	0	0	0	0	0	0	0
11	0	0	0	1	0	0	0	0	0	0	0	0
13	0	0	0	0	0	0	0	0	0	0	0	0
15	0	0	0	0	0	0	0	0	0	0	0	0
17	0	0	0	0	0	0	0	0	0	0	0	0
19	0	0	0	0	0	0	0	0	0	0	0	0
21	0	0	0	0	0	0	0	0	0	1	0	0
23	0	0	0	0	0	0	0	0	0	0	0	0
25	0	0	0	0	0	0	0	0	0	2	0	0

*= no sample.

Appendix 6.15 Numbers of organisms. $40cm^{-3}$ and organic carbon $\mu g.cm^{-3}$, from integrated totals.

Run 2

Day	1			15			29			46		
E. buchholzi	0	0	0	1	25	1	1	1	1	11	66	9
" full cocoons	0.4	1.3	4.2	0.3	4.0	1.1	0.2	0.2	1.2	6.1	6.0	6.0
" empty cocoons	1.3	1.0	1.9	0.9	1.8	0	0	1.1	2.4	11.9	21.2	5.2
A. hemprichi	0	0	0.1	4.0	0.3	17.2	3.9	0	0	56.8	31.3	1.7
P. idrensis	0	0	0	0	0	0	0	0	0	0	0	0
P. foreli	0	0	0	0	0	0	0	0	0	0	0	0
Nematodes	0.1	0.1	0.1	0.2	0	0.1	0.9	0	6.7	5.3	7.9	40.3
Chironomid larvae	0	0	0.1	0	0.6	1.1	0.1	0.2	4.6	10.9	7.8	6.1
Harpacticoids	0.3	0	0	0	0	0	0	0	0	19.3	2.0	12.7
Flatworms	0	0	0	0	0	0	0.6	0	0.8	2.8	0	0.7

Appendix 6.15 continued.

Run 3

Day	4			19		
E. buchholzi	4	8	2	2	57	68
" full cocoons	5.8	1.9	1.1	1.7	5.3	9.3
" empty cocoons	0.4	0.6	1.7	2.4	0.9	1.8
A. hemprichi	4.8	3.4	1.8	1.8	1.3	0
P.idrensis	0	0	0	0	0	0
P. foreli	0	0	0	0	0	0
Nematodes	0.4	1.1	0.1	1.3	2.6	3.3
Chironomid larvae	3.3	2.6	6.3	58.4	209.4	100
Harpacticoids	0.1	0	0	0	0	0
Flatworms	0	0	0	0	0.2	0.1

Appendix 6.15 continued.

Run 4

Day	1			12			26			41			55		
Carbon 1cm depth	169	*	*	617	677	403	694	547	1480	258	3803	4924	4986	*	*
" 2cm "	157	*	*	138	151	221	365	147	525	101	76	3802	2660	*	*
" below 2cm	107	*	*	126	131	239	122	244	470	67	89	198	170	*	*
E. buchholzi	1	*	*	6	1	29	38	54	33	25	17	45	53	*	*
" full cocoons	0.4	*	*	6.4	2.5	5.7	3.6	1.3	4.5	2.2	0.9	0.2	3.2	*	*
" empty cocoons	0.2	*	*	0	0	0	0	11.7	23.2	14.3	8.2	14.4	5.7	*	*
A. hemprichi	0.6	*	*	1.7	3.4	11.0	5.8	0.3	5.3	0.4	0.4	0.1	10.3	*	*
P.idrensis	0	*	*	0.1	0.5	0.1	0.6	1.2	2.4	0	0	0	8.8	*	*
P. foreli	0	*	*	0	0	0	0.9	0	8.9	0	0	0	0	*	*
Nematodes	0.2	*	*	1.2	3.5	5.7	13.7	12.4	39.7	26.2	19.1	33.2	15.9	*	*
Chironomid larvae	0	*	*	0	0.1	0	1.3	0.3	0	0	2.0	0	1.0	*	*
Harpacticoids	0.5	*	*	2.4	1.8	0.6	7.0	2.8	9.2	2.9	2.5	1.8	21.4	*	*
Flatworms	0	*	*	0.2	0.2	0	2.5	4.9	17.8	0	0.4	0.6	19.9	*	*

*= no sample.

Appendix 6.15 continued.

Run 5

Day	3			16			30		
Carbon 1cm depth	162	133	187	607	423	437	1167	1367	*
" 2cm "	98	158	165	394	303	374	508	576	*
" below 2cm	133	114	163	188	211	226	243	269	*
E. buchholzi	0	2	1	14	11	27	8	14	*
" full cocoons	0	0	0.1	1.8	0.1	0.3	0	0.1	*
" empty cocoons	0	0	0	0.7	0.3	0	0	0	*
A. hemprichi	1.1	0.2	0	0	0.1	0	0	0	*
P. idrensis	2.1	0.7	1.2	0	0	0	0	0	*
P. foreli	0	0	0	0	0	0	0	0	*
Nematodes	1.7	0.2	0.7	0.7	1.5	0	0	0.1	*
Chironomid larvae	0	0	0	6.5	2.5	1.9	4.5	5.9	*
Harpacticoids	0	0	0	0	0	0	0	0	*
Flatworms	0.9	0	0	3.5	0.4	0.5	1.4	0.4	*

*= no sample.

Appendix 6.15 continued.

Run 7

Day	1			23			37			51		
Carbon 1cm depth	274	266	215	1567	519	412	1351	950	840	631	1029	2205
" 2cm "	133	96	186	978	141	169	307	118	241	301	511	858
" below 2cm	80	99	102	192	113	103	97	112	130	162	271	242
E. buchholzi	0	0	0	36	1	19	34	53	10	50	37	0
" full cocoons	0	0	0	1.1	0	0	0.8	1.0	1.6	7.6	6.5	0
" empty cocoons	0.1	0	0	0	0.1	0.2	0.1	0.2	0	0.2	0.1	0.4
A. hemprichi	0	0	0	0	0	0	0	0	0	0	0	0
P. idrensis	0	0	0	0	0	0	0	0	0	0	0	0
P.foreli	0	0	0	0	0	0	0	0	0	0	0	0
Nematodes	0	0	0	0	0	0	0.1	0	0	0.5	0	0.1
Chironomid larvae	0	0	0	0	0	0	0	0	0	0	0	0
Harpacticoids	0	0	0	0	0	0	0	0	0	0	0	0
Flatworms	0	0	0	0.1	0	0	0	0	0	0.1	0	0

Appendix 6.16 Geometric means, meiofauna- nos.40cm^{-3}, carbon- µg.cm^{-3}; using log(x+1) for meiofauna , log x for carbon.

Run	2			3			4					5			7			
Day	1	15	29	46	4	19	1	12	26	41	55	3	16	30	1	23	37	51
Carbon 1cm depth							169	552	825	4327	4986	159	482	1144	250	695	1025	1127
" 2cm "							157	166	304	540	2660	137	354	587	133	286	206	509
" below 2cm							107	158	241	133	170	135	207	243	93	131	112	220
E. buchholzi	0	4	1	19	4	22	1	6	41	26	53	1	16	11	0	10	26	11
" full cocoons	1.6	1.4	0.5	6.0	2.5	4.6	0.4	4.6	2.9	0.9	3.2	0	0.6	0	0	0.3	1.1	3.0
" empty "	1.4	0.7	0.9	11.1	0.8	1.6	0.2	0	5.7	11.9	5.7	0	0.3	0	0	0.1	0.1	0.2
A. hemprichi	0	3.9	0.7	16.1	3.1	0.9	0.6	4.2	2.6	0.3	10.3	0.4	0	0	0	0	0	0
P. idrensis	0	0	0	0	0	0	0	0.2	1.3	0	8.8	1.3	0	0	0	0	0	0
P.foreli	0	0	0	0	0	0	0	0	1.7	0	0	0	0	0	0	0	0	0
Nematodes	0.1	0.1	1.4	12.2	0.5	2.3	0.2	3.0	19.0	25.5	15.9	0.8	0.6	0	0	0	0	0.2
Chironomid larvae	0	0.5	0.9	8.1	3.8	107.1	0	0	0.4	0.4	1.0	0	3.2	5.2	0	0	0	0
Harpacticoids	0.1	0	0	8.4	0	0	0.5	1.5	5.8	2.4	21.4	0	0	0	0	0	0	0
Flatworms	0	0	0.4	0.9	0	0.1	0	0.1	6.3	0.3	19.9	0.2	1.1	0.8	0	0	0	0

Appendix 6.17 Biomass of E. buchholzi.

Run	Day	Biomass µg C.40cm^{-3} geometric mean	E. buchholzi carbon as a % of total carbon in sand.
2	1	0.3	
	15	50.4	
	29	3.3	
	46	103.4	
3	4	17.6	
	19	178.5	
4	1	1.8	0.045
	12	52.5	0.588
	26	144.0	0.945
	41	84.0	0.391
	55	144.2	0.391
5	3	8.3	0.136
	16	108.4	1.020
	30	35.8	0.323
7	1	0.7	0.015
	23	112.1	1.332
	37	171.4	2.286
	51	153.9	1.143

Appendix 7.1 Apparatus used for construction of pilot filter.

Clear perspex cast tubing, 2m length, I.D.= 13.925 cm, wall= 1cm
Flexible clear PVC tubing, O.D. = 2.2 cm, I.D.= 1.6 cm
1 pack aluminium angle frame.
Oil tempered hardboard, 0.3 cm thick.
Fluorescent lamp units.
20 watt growlux fluorescent tubes.
Dial switch model S254, Sangamo Weston Ltd.
Rotameter flow meters.

Interstitial water samplers

Nylon rod, 2.54 cm diameter.
" " , 1.27 cm "
Perspex tubing, O.D.= 5 mm, I.D. = 3 mm.
" rod, 1cm diameter.
Rubber bungs, small diameter= 5 mm, large diameter = 7.5mm, height=1.6cm.
Portex surgical tubing, O.D.= 1.5 mm, I.D.= 1 mm.
PVC flexible clear tubing, O.D. = 7mm, I.D.= 5mm.

Water collecting bottles

Quickfit socket joints, socket size 10/19.
" conical " , cone size 10/19.
" stoppers, cone size 10/19, both ends closed
Portex surgical quality tubing, O.D.= 1.5 mm, I.D.= 1 mm.
Rubber bungs, large diameter= 7.5 mm, small diameter =5 mm, height= 1.6cm.

Appendix 7.2 Apparatus used for dissolved oxygen analysis method.

Glass vials (10ml volume) for titration.
Oxford pipettors, 20,50,1000 and 4000ul.
Magnetic stirrer with small bar and good speed control.
Piston type burette for potassium biniodate. Metrohm E457, 5ml volume.
Meter with a high internal resistance which can be used to record potential difference, preferably one with a variable back off potentiometer control ('buffer control ' of a pH meter) to eliminate the small current present before the end point is reached. Pye Dynacap pH meter.
Combined platinum - calomel reference electrode. Metrohm EA234.

Appendix 7.3 Reagents used in the dissolved oxygen analysis method.

Winkler's 'A' solution - 50g $MnSO_4.4H_2O$, dissolved in water and made up to 100ml.

Winkler's 'B' solution- 1. Alkaline iodide reagent.
40g NaOH dissolved in 56ml H_2O
90g pure NaI added, solution kept hot until iodide dissoves
solution then cooled and diluted to 100ml
solution filtered if necessary after standing overnight

2. Sodium azide solution.
2.5g NaN_3 added to 100ml
30ml of this solution was then added to 100ml of the alkaline iodide solution

Acid - 50% sulphuric acid.

Potassium biniodate solution - A stock solution of approximately 0.2N made by dissolving 1.6g in 100ml of distilled water. This was then diluted to 0.005N for use.

Sodium thiosulphate - A stock solution of 0.1N $Na_2S_2O_3$ made up and diluted to 0.001N for titration with biniodate.

Appendix 7.4 Precision of the microWinkler dissolved oxygen determination method.

n	Mean D.O. conc. $mg.L^{-1}$	S	Precision i.e. S as a % of mean.
2	8.318	0.072	0.867
2	8.041	0.030	0.378
2	8.105	0.186	2.295
2	8.233	0.127	1.537
2	7.603	0.006	0.084
2	7.762	0.039	0.501
2	7.157	0.109	1.522
2	6.543	0.100	1.524
2	8.141	0.147	1.807
2	7.999	0.093	1.158
2	7.537	0.263	3.491
2	8.075	0.226	2.794
2	7.540	0.057	0.750
2	7.299	0.039	0.533
2	6.645	0.081	1.224
2	6.951	0.236	3.398
2	7.457	0.175	2.342
2	7.712	0.006	0.073
2	7.390	0.060	0.813
2	6.568	0.139	2.110

mean of total vaues of precision= 1.460

S^2= 1.046

n = 20

SE= $\pm$ 0.479, p= 0.05, d.f.= 19

95% confidence limits = 0.981-1.939

Appendix 8.1 Dissolved oxygen concentrations ($mg.L^{-1}$), in interstitial water samples removed from pilot filter, day 3.

Time	Depth	Bottle 'a'		Bottle 'b'		t	p
0430	S	8.780	8.973				
	1cm	8.267	8.396	8.019	8.062	5.015	0.05
	5	7.973	8.236	8.322	8.143	0.805	0.9
	10	7.607	7.598	7.789	7.734	5.707	0.05
	20	7.080	7.234	8.345	8.182*		
	30	6.613	6.472	6.627			
	E	5.020	5.008				
0700	S	9.078	9.370				
	1	8.037	8.245	8.064	7.933	1.159	0.4
	5	7.723	7.351	8.234	7.915	2.194	0.2
	10	7.500	7.580	7.271	7.362	4.975	0.05
	20	6.587	6.702	7.118	6.784	1.735	0.4
	30	6.616	6.821	6.818	6.181	0.655	0.9
	E	5.403	5.162				
0915	S	9.055	8.895				
	1	7.333	7.580	7.716	7.708	2.068	0.2
	5	7.337		7.347	7.432		
	10	6.666	6.470	7.106	7.085*		
	20	6.081	6.392	6.411	6.543	1.424	0.4
	30	5.135	4.985	5.750	5.941	6.468	0.05
	E	4.838	4.655				
1230	S	8.944	9.227				
	1	7.877	7.954	7.599	7.787	2.191	0.2
	5	7.550	7.335	7.750	7.725	2.725	0.2
	10	7.149	6.962	7.020	7.045	0.244	0.9
	20	6.243	6.396	6.100	6.261	1.252	0.4
	30	5.611	5.684	5.811	5.768	3.354	0.05
	E	4.048	3.977				
1530	S	8.846	9.247				
	1	7.836	7.764	8.860	8.066*		
	5	7.859	7.642	7.892	7.922	1.429	0.4
	10	6.942	6.985	6.635	6.961	1.007	0.5
	20	6.687	6.721	6.560	6.616	3.540	0.05
	30	5.752	5.941	6.682	6.452*		
	E	3.874	3.660				

Time	Depth	Bottle 'a'		Bottle 'b'		t	p
1810	S	9.018	8.932				
	1cm	7.304	7.428	7.552	7.441	1.569	0.4
	5	7.347	7.538	6.866	7.580	0.594	0.9
	10	7.068	7.000	6.969	7.003	1.261	0.4
	20	6.341	5.955	5.805	5.881	1.551	0.4
	30	5.641	5.897*	5.930	5.220		
	E	3.861	3.811				
2010	S	8.870	9.432				
	1	8.342	7.982	8.079	7.973	0.725	0.9
	5	7.270	7.245	7.560	7.845	3.111	0.05
	10	7.670	7.751	7.349	7.311	8.506	0.02
	20	6.956	7.072	7.060	6.996	0.211	0.9
	30	6.330	6.121	6.222	6.141	0.393	0.9
	E	0.003	0				
2200	S	8.969	9.284				
	1	7.881	7.898	7.568	7.526	10.708	0.01
	5	7.796	7.838	7.457	7.321	6.015	0.05
	10	8.292	7.800*	7.084	6.735		
	20	6.646	6.553	6.109	6.070	10.119	0.01
	30	6.608	6.625	6.303	6.282	24.133	0.01
	E	3.986	3.940				
0040	S	8.327	8.159				
	1	6.853	6.870	6.877	6.761	0.725	0.9
	5	6.599	6.654	5.794	5.705	16.753	0.01
	10	6.220	6.152	6.164	6.067	1.189	0.2
	20	5.783	5.902	4.721	5.520	1.787	0.4
	30	6.696	6.914	5.053	4.912	14.040	0.01
	E	2.203	2.274				
0140	S	8.343	8.404				
	1	8.168	8.134	*	*		
	5	9.796	9.703*	6.851	6.885		
	10	8.263	8.390*	6.595	6.472		
	20	6.660	6.682	6.764	6.562	0.079	0.9
	30	5.681	5.685	5.200**	5.663		
	E	3.296	3.518				

S= surface sample
E= exit sample (75cm)
*= sample rejected air bubble present at Winkler stage
**= sample rejected
Comparasion of means, t-test d.f.=2+2-2=2

Appendix 8.2 Dissolved oxygen concentrations ($mg.L^{-1}$), in interstitial water samples removed from pilot filter, day 10.

Time	Depth	Bottle 'a'		Bottle 'b'		t	p
0410	S	9.157	9.611				
	1cm	8.244**	8.981	9.098	9.038		
	5	8.358	8.553	8.759	8.895	3.125	0.05
	10	7.763	8.167	8.102	8.187	0.870	0.5
	20	7.758	7.873	8.425	8.305	6.610	0.05
	30	7.430	7.383	7.562	7.473	2.207	0.4
	E	5.606	5.893				
0710	S	9.366	9.296				
	1	8.213	8.115	8.698	8.668	10.130	0.01
	5	*		*			
	10	7.964	7.892	8.295	7.929	0.986	0.5
	20	9.006	9.086*	7.307	7.370		
	30	6.926	7.079	6.830	6.940	1.262	0.4
	E	5.236	5.003				
1010	S	9.730	9.616				
	1	7.985	8.146	8.516	8.823	3.485	0.1
	5	7.997	8.056	8.296	8.189	3.536	0.1
	10	7.869	7.856	*			
	20	7.036	7.267	7.084	7.093	0.545	0.1
	30	*		6.868	6.808		
	E	3.810*	4.770				
1300	S	9.734	9.616				
	1	9.120	9.052	*			
	5	8.636	8.751	*			
	10	8.168	8.130	8.419	8.151	1.005	0.5
	20	7.626	7.541	7.682	7.628	1.419	0.4
	30	7.407	7.458	*			
	E	6.117*	5.142				

Time	Depth	Bottle 'a'		Bottle 'b'		t	p
1615	S	10.226	10.164				
	1cm	9.019	9.019*	9.196	8.274**		
	5	8.621	8.562	8.699	8.674	2.965	0.1
	10	8.257	8.278	8.667	8.425	2.293	0.2
	20	7.365	7.194	7.746	7.707	5.097	0.05
	30	*		*			
	E	4.916	4.724				
1915	S	10.017	10.005				
	1	8.414	8.388	8.753	8.783	18.466	0.01
	5	8.197	8.023	9.164	8.624	2.764	0.2
	10	8.811	8.316	7.955	8.372	1.236	0.4
	20	7.070	6.811	*			
	30	6.334	6.155	6.647	6.507	2.926	0.1
	E	4.321	4.246				
2200	S	10.390	10.455				
	1	9.139	8.977	9.145	9.077	0.603	0.9
	5	8.791	8.761	9.006	9.380	2.223	0.2
	10	8.384	8.167	8.654	8.607	3.198	0.1
	20	8.018	8.108	8.074	7.878	0.807	0.9
	30	7.760	7.610	7.613	7.724	0.177	0.9
	E	6.069	6.163				
0100	S	9.802	9.626				
	1	9.070	9.014	8.282	9.336	0.442	0.9
	5	*		9.064	9.238		
	10	8.564	8.699	8.782	8.804	2.360	0.2
	20	8.048	8.044	*			
	30	7.895	7.869	7.512	7.508	28.201	0.01
	E	5.296	5.591				

Key as in appendix 8.1

Appendix 8.3 Dissolved oxygen concentrations ($mg.L^{-1}$), in interstitial water samples removed from pilot filter, day 36.

Time	Depth	Bottle 'a'		Bottle 'b'		t	p
0600	S	9.799	9.729				
	1cm	8.456	8.422	8.866	8.670	3.308	0.1
	5	7.578	7.740	8.284	8.228	3.904	0.1
	10	7.432	7.565	7.416	7.463	0.837	0.5
	20	7.376	7.563	7.191	7.097	3.111	0.1
	30		6.872	6.846			
	E	5.131	5.052				
0900	S	10.282	10.327				
	1	9.833	9.849*	8.974	8.880		
	5	10.820	11.260*	8.126	7.862		
	10	7.925	7.913	7.979	7.813	0.276	0.9
	20	7.547	7.376	7.870	7.917	4.872	0.05
	30	7.442	7.374	7.331	7.419	0.593	0.9
	E	6.370	6.191				
1200	S	10.966	10.942				
	1	9.568	9.589	8.047*	9.221		
	5	9.106	9.047	8.778	8.833	6.720	0.05
	10	8.208	8.389	8.762	8.792	4.898	0.05
	20	8.434	8.600	8.483	8.373	0.089	0.5
	30	7.943	7.733	7.940	8.106	1.382	0.4
	E	6.752	6.723				
1500	S	10.802	10.765				
	1	9.434	9.579	9.256	9.174	3.499	0.1
	5	8.086	8.081	8.300	8.415	4.758	0.05
	10	8.082	8.192	7.920	8.226	0.394	0.1
	20	7.506	7.527	7.550	7.597	2.215	0.2
	30	6.984	7.036	7.024	*		
	E	5.837	5.871				

Time	Depth	Bottle 'a'		Bottle 'b'		t	p
1800	S	9.967	10.024				
	1cm	8.435	8.435	8.132	8.213	6.481	0.05
	5	7.423	7.186	7.600	7.455	1.605	0.5
	10	7.000	7.025	7.142	7.248	3.349	0.2
	20	6.670	6.740	6.549	6.528	4.548	0.05
	30	6.547	6.384	6.318	6.322	1.785	0.4
	E	5.559	5.513				
2100	S	9.709	9.619				
	1	8.332	8.314	8.697	8.484	2.503	0.2
	5	7.286	6.998	7.097	7.255	0.207	0.9
	10	7.057	7.074	6.877	6.873	22.145	0.1
	20	6.033	5.884	5.992	6.220	1.083	0.4
	30	6.244	6.116	5.774	5.728	6.305	0.05
	E	5.247	5.027				
2400	S	9.839	9.794				
	1	7.078	7.193	7.124	7.056	0.681	0.9
	5	5.164	5.822	4.406*	5.679		
	10		5.210	5.391	5.489**		
	20	4.429	4.433	4.614	4.550	4.710	0.05
	30	4.691	4.665*	3.775	3.988		
	E	2.822	3.093				

Key as in appendix 8.1

Appendix 8.4 Dissolved oxygen concentrations ($mg.L^{-1}$), summary of means for bottles 'a' and 'b'.

Day	Time	Bottle	Depth (cm) 1	5	10	20	30
3	0430	a	8.318	8.105	7.603		
		b	8.041	8.233	7.762		
	0700	a	8.141	7.537	7.540	6.645	6.719
		b	7.999	8.075	7.299	6.951	6.500
	0915	a	7.457			6.237	5.060
		b	7.712			6.477	5.846
	1230	a	7.916	7.443	7.056	6.320	5.648
		b	7.693	7.738	7.033	6.181	5.790
	1530	a		7.751	6.964	6.704	
		b		7.907	6.798	6.588	
	1810	a	7.366	7.443	7.034	6.148	
		b	7.497	7.223	6.986	5.843	
	2010	a	8.162	7.258	7.711	7.014	6.226
		b	8.026	7.703	7.330	7.028	6.182
	2200	a	7.890	7.817		6.600	6.617
		b	7.556	7.389		6.090	6.293
	0040	a	6.862	6.627	6.186	5.843	6.805
		b	6.819	5.750	6.116	5.121	4.983
	0140	a				6.671	
		b				6.663	

Day	Time	Bottle	Depth (cm) 1	5	10	20	30
10	0410	a		8.456	7.965	7.816	7.407
		b		8.827	8.145	8.365	7.518
	0700	a	8.164		7.928		7.003
		b	8.683		8.112		6.885
	1010	a	8.066	8.027		7.152	
		b	8.670	8.243		7.089	
	1300	a			8.149	7.584	
		b			8.285	7.655	
	1615	a		8.592	8.268	7.280	
		b		8.687	8.546	7.727	
	1915	a	8.401	8.110	8.564		6.245
		b	8.768	8.894	8.164		6.577
	2200	a	9.058	8.776	8.276	8.063	7.685
		b	9.111	9.193	8.631	7.976	7.669
	0100	a	9.042		8.632		7.882
		b	8.809		8.793		7.510
36	0600	a	8.439	7.659	7.499	7.470	
		b	8.768	8.256	7.440	7.144	
	0900	a			7.919	7.462	7.408
		b			7.896	7.894	7.375
	1200	a		9.077	8.304	8.517	7.838
		b		8.906	8.777	8.428	8.023
	1500	a	9.507	8.804	8.137	7.517	
		b	9.215	8.358	8.073	7.574	
	1800	a	8.435	7.305	7.013	6.705	6.466
		b	8.173	7.528	7.195	6.539	6.320
	2100	a	8.323	7.142	7.066	5.959	6.180
		b	8.591	7.176	6.875	6.106	5.751
	2400	a	7.086			4.431	
		b	7.090			4.582	

Appendix 8.5 Dissolved oxygen concentrations of interstitial water samples, means for each depth in the pilot filter.

Day	Time		Depth S	1	5	10	20	30	E
3	0430	c	8.876	8.041	8.233	7.762	7.157	6.543	5.014
		%	95	87	88	84	77	70	55
	0700	c	9.228	7.999	8.075	7.299	6.951	6.500	5.282
		%	98	85	86	78	74	70	57
	0915	c	8.975	7.712	7.390	6.568	6.477	5.846	4.746
		%	95	82	79	71	70	63	50
	1230	c	9.086	7.693	7.738	7.033	6.181	5.790	4.012
		%	99	83	84	76	67	63	43
	1530	c	9.046	7.800	7.907	6.798	6.588	5.847	3.770
		%	98	85	86	74	72	64	42
	1810	c	8.975	7.497	7.223	6.986	5.843	5.575	3.836
		%	99	82	78	77	64	62	42
	2010	c	9.151	8.026	7.703	7.330	7.028	6.182	0.0015
		%	99	86	84	80	76	67	0
	2200	c	9.126	7.556	7.389	6.910	6.090	6.293	3.963
		%	99	83	80	75	64	68	44
	0400	c	8.243	6.819	5.750	6.116	5.121	4.983	2.238
		%	87	74	60	65	55	54	25
	0140	c	8.373	8.151	6.868	6.534	6.663	5.683	3.407
		%	90	86	74	70	72	63	37
10	0410	c	9.384	9.068	8.827	8.145	8.365	7.518	5.749
		%	95	92	90	82	85	76	58
	0700	c	9.331	8.683		8.112	7.339	6.885	5.119
		%	95	88		83	75	70	51
	1010	c	9.673	8.670	8.243	7.863	7.089	6.838	4.770
		%	100	88	84	80	72	70	50
	1300	c	9.675	9.086	8.694	8.285	7.655	7.433	5.142
		%	99	92	89	85	78	76	52
	1615	c	10.195	9.196	8.687	8.546	7.727		4.820
		%	104	94	88	85	78		50
	1915	c	10.011	8.768	8.894	8.164	6.941	6.577	4.283
		%	100	90	91	82	80	77	44
	2200	c	10.422	9.111	9.193	8.631	7.976	7.669	6.116
		%	105	92	93	87	80	78	62
	0100	c	9.714	8.809	9.151	8.793	8.046	7.510	5.443
		%	96	90	91	88	80	76	55
36	0600	c	9.764	8.768	8.256	7.440	7.144	6.846	5.141
		%	98	89	82	74	72	68	51
	0900	c	10.304	8.927	7.994	7.896	7.894	7.375	6.280
		%	103	89	80	79	79	74	63
	1200	c	10.954	9.221	8.806	8.777	8.428	8.023	6.737
		%	109	92	89	88	85	80	66
	1500	c	10.783	9.215	8.358	8.073	7.574	7.010	5.854
		%	108	91	85	81	77	72	59
	1800	c	9.996	8.173	7.528	7.195	6.539	6.320	5.536
		%	100	83	76	74	68	64	56
	2100	c	9.664	8.591	7.176	6.875	6.106	5.751	5.137
		%	95	86	72	68	61	58	51
	2400	c	9.186	7.090	5.493	5.440	4.582	3.882	3.881
		%	97	70	56	54	46	40	40

S=surface water sample

E= exit sample (75cm)

c= D.O. conc. ($mg.L^{-1}$)

%= percentage saturation

Appendix 8.6 Redox potential measurements recorded in pilot filter.

Day 3

Time	0430	0700	0930	1230	1530	1810	2010	2200	0040	0140
Temp °C inflow	19.0	18.5	18.0	19.0	19.0	19.5	19.0	18.5	18.5	18.0
Temp °C outflow	19.0	18.5	18.5	19.0	19.5	19.5	19.5	19.5	19.0	18.5
Head loss cm	10	9.5	8.0	4.0	7.5	6.5	7.0	7.0	7.5	9.0
Flow rate ml.min^{-1}	52.5	50	50	50	50	45	50	50	40.5	50
Redox pot. mv										
Depth 1cm	378.8	393.4	378.7	376.6	376.1	378.5	381.8	384.3	381.1	378.6
2.5	425.8	426.6	426.1	425.6	419.4	420.3	421.4	421.2	419.6	418.6
5	547.3	547.9	548.4	547.9	544.8	542.5	537.2	528.2	545.6	543.3
7.5	404.1	405.3	400.7	396.9	395.1	400.9	393.1	392.8	392.0	375.5
10	444.2	445.1	447.4	444.3	443.1	442.4	439.3	440.1	442.5	444.1
20	452.0	488.4	486.8	487.4	485.8	483.1	482.5	481.5	481.8	479.9
30	442.2	453.1	459.7	455.7	530.4	433.1	425.8	433.6	442.0	449.3
40	527.8	530.5	530.1	529.2	529.1	525.8	525.7	525.2	519.8	516.9

Day 10

Time	0410	0710	1010	1300	1615	1915	2200	0100
Temp °C inflow	16.0	16.0	16.0	16.0	16.0	16.0	16.0	15.5
Temp °C outflow	"	"	"	"	"	"	"	15.3
Head loss cm	16.5	14.5	15.5	15.5	15.5	12.5	16.0	16.0
Flow rate ml.min^{-1}	52.5	52.5	40.5	50	50	36.5	50	48
Redox pot. mv								
Depth 1cm	420.0	416.2	406.6	404.1	401.0	402.6	408.3	406.2
2.5	432.8	432.3	432.0	431.8	432.5	417.8	422.8	429.5
5	549.4	546.5	546.5	548.4	548.9	544.7	544.8	546.2
7.5	410.6	409.1	403.0	409.2	404.1	408.0	396.3	407.1
10	455.9	453.8	447.5	454.1	458.1	444.3	453.5	457.6
20	491.9	490.7	472.3	486.9	490.3	471.2	489.1	491.4
30	503.2	499.5	493.9	489.6	498.6	500.1	502.2	491.7
40	539.9	539.8	534.0	537.0	541.5	509.3	532.5	536.0

Appendix 8.6 continued.

Day 36

Time	0600	0900	1200	1500	1800	2100	2400
Temp °C inflow	15.5	15.5	15.5	16.0	16.0	15.5	15.5
Temp °C outflow	"	"	"	"	"	"	"
Head loss cm	17.5	17.5	17.5	17.5	17.5	17.5	17.5
Flow rate ml.min^{-1}	50	50	50	50	50	50	50
Redox pot. mv							
Depth 1cm	474.0	360.7	349.4	354.3	359.7	380.6	383.0
2.5	486.1	405.1	392.7	394.9	392.6	395.7	405.6
5	715.5	688.1	674.6	707.8	723.9	708.0	699.1
7.5	444.5	420.9	468.8	391.5	419.6	423.1	422.8
10	462.5	458.8	454.2	468.1	455.0	456.2	451.4
20	465.6	476.7	471.9	478.1	475.3	477.3	474.0
30	501.8	497.8	489.9	492.8	493.4	494.2	487.8
40	542.6	542.3	532.0	533.7	531.4	535.1	526.1

Appendix 8.7 Meiofaunal (nos.$40cm^{-3}$) and organic carbon ($\mu g.cm^{-3}$) analyses of sand samples removed from Bed 5 and the pilot filter.

Location	Sand core, day 1														
Depth cm	1	2	3	4	5	7	9	11	13	15	17	19	21	23	25
E. buchholzi	0	1	11	3	14	3	1	0	6	15	6	24	15	8	0
" full cocoons	0	1	0	2	4	0	0	0	1	1	0	0	0	0	4
" empty cocoons	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
A. hemprichi	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
P.idrensis	1	0	0	1	0	0	1	0	0	8	0	1	0	2	0
P.foreli	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
N. elinguis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nematodes	2	11	8	4	8	7	0	0	1	2	0	0	1	0	0
Chironomid larvae	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Harpacticoids	1	3	0	0	0	0	4	7	0	0	0	0	1	2	0
Flatworms	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Carbon	257	388	210	196	67	251	138	273	291	375	221	205	141	93	213
Location	Sand core, 42 days														
Depth cm	1	2	3	4	5	7	9	11	13	15	17	19	21	23	25
E. buchholzi	75	62	43	47	36	51	55	63	55	22	5	17	12	4	8
" full cocoons	6	20	0	0	0	0	0	0	0	0	0	0	0	0	0
" empty cocoons	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
A. hemprichi	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
P. idrensis	4	2	1	8	0	1	4	2	5	0	0	8	2	1	1
P.foreli	1	15	1	1	0	0	0	0	0	1	5	0	0	0	2
N. elinguis	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nematodes	67	51	9	14	5	4	14	8	4	11	2	0	0	2	4
Chironomid larvae	325	170	100	130	40	0	0	0	0	0	0	0	4	0	1
Harpacticoids	0	0	7	9	4	0	2	0	0	0	0	0	4	0	0
Flatworms	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Carbon	1005	527	419	274	290	133	203	342	199	295	195	244	204	242	240

Appendix 8.7 continued.

Location	Sand core, 42 days														
Depth cm	1	2	3	4	5	7	9	11	13	15	17	19	21	23	25
E. buchholzi	13	56	60	83	62	140	125	48	101	50	48	24	20	34	28
" full cocoons	0	18	1	3	0	1	0	0	15	6	0	0	0	0	0
" empty cocoons	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
A. hemprichi	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
P.idrensis	0	0	1	0	0	1	0	0	0	0	0	0	0	0	0
P.foreli	0	0	0	0	2	1	0	1	2	0	0	1	0	1	0
N.elinguis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nematodes	20	22	8	8	0	7	5	3	0	6	12	0	6	8	0
Chironomid larvae	380	12	12	1	2	0	1	1	0	3	3	0	0	4	0
Harpacticiods	5	2	2	2	0	2	1	0	3	0	0	0	0	0	0
Flatworms	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Carbon	1019	413	183	375	310	111	130	64	63	172	45	144	158	82	15

Location	Pilot filter, 38 days					
Depth cm	1-5	6-10	11-15	16-20	21-25	26-30
E. buchholzi	115	134	69	103	40	22
" full cocoons	1	2	0	0	1	0
" empty cocoons	0	0	0	0	3	0
A. hemprichi	0	0	0	0	0	0
P. idrensis	0	0	0	0	0	0
P. foreli	0	0	0	0	0	0
N. elinguis	0	0	0	0	0	0
Nematodes	88	15	4	6	4	2
Chironomid larvae	0	0	0	0	0	0
Harpacticoids	0	0	0	0	0	0
Flatworms	0	0	0	0	0	0

Carbon					
Depth cm	1	5	10	20	30
	654	342	341	254	185
	627	379	294	270	135
mean carbon conc.	640	360	262*		

* = mean of six analyses.
10-30 cm.