Errata.

Page 12, v, for fig. 31 read fig 34a.

Page 19, vii, for fig.4b read fig. 4

Page 48, x, for 1964 read 1963.

Page 100, xi, for wich read which.

Page 103, vii, for indicated by solid bands in figure read indicated in figure 19.

xx, for (solid black bands figure 26) read (see figure 19).

Page 151, i, for oOL read 10L.

Page 158, xiii, for (fig. 33) read (fig. 37).

Page 208, viii, read (see Huber-Pestalozzi,1962).

Page 211, read O.Lacustris Chod.

Page 213, read U.tenerrima Kutz.

Page 216, read N. baccata Hustedt.

Page 218, read L. versicolor (Wartm.) Gom.

Page 220, xii, read filamentous algae became dominant.

Thesis Submitted for the Degree of

Doctor of Philosophy.

1968.

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Ecological and Taxonomic studies of ~~the Algae of Slow Sand~~ Filter Beds. algae

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by


Edward George Bellinger, B.Sc.


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# Ecological and Taxonomic studies of filter bed algae.

## Abstract.

An investigation has been carried out on the algal flora of slow sand filter beds. Ecological field studies were made from August 1963 until December 1965 and experimental work was continued until June 1966. The filter beds studied were located at the Ashford Common Works of the Metropolitan Water Board, London. The major chemical nutrients, ($NH_4$-N, $NO_3$-N, $NO_2$-N, $PO_4$-P and $SiO_2$), and pH were determined throughout the period together with temperature and penetration of light into the water. Biomass, interpreted from calculated cell volumes, of the major species of algae encountered was used to express the results of cell concentrations.

The periodicity and distribution of the algae in the filter beds was investigated. The algal populations present were sub-divided into planktonic, epipelic and attached bottom living species and each one was found to have a distinct seasonal periodicity. Observations suggested that the filter bed algal populations were not often limited by nutrient concentrations but the algae on the sand surface may on several occasions have been limited by low light levels.

Experiments to determine the toxicity of copper sulphate to certain species of algae were carried out. The ability of algae to penetrate the sand and to survive was also investigated experimentally. These experiments showed that cells could stay alive in the filter bed sand after cleaning and act as population innocula when the bed was refilled. The flora of the sand surface was compared with that of

an in-situ polythene bag to ascertain the effects of nutrient limitation.

The variation in cell sizes of <u>Stephanodiscus astraea</u> and <u>Asterionella formosa</u> were investigated together with the relative growth rate, k', of <u>S. astraea</u>, and seasonal variations in both were found.

Ecological and Taxonomic studies of the Algae of Slow Sand Filter Beds.

Contents

# PART ONE. INTRODUCTION.

# I. Introduction.

Slow sand filter beds of waterworks are small man-made bodies of water with a continuous inflow, usually at one end, and a continuous outflow by percolation through a layer of sand covering the bottom. In area and depth they are very much like ponds except that they have vertical sides.

Although there has been much work on the algae of larger bodies of water such as lakes (reviewed in Lund, 1965 and Hutchinson, 1967) and reservoirs (Reynolds, 1950; Taylor, 1954; Holsinger, 1955 and Ridley, 1964) and on rivers (Butcher, 1946; Rice, 1938), there has been little detailed work on bodies of water the size of ponds. Fritsch (1906), Fritsch & Rich (1913), Hodgetts (1921) and Howland (1931) studied the periodicity of algae in small lakes and ponds but mainly with reference to meteorological conditions, little work being done on the chemistry of the water. Atkins (1926-7) and later Lind (1938) extended the algal periodicity studies to include parallel chemical studies of the water. More recently Rao (1953-55) studied the algae of six ponds and attempted to correlate seasonal changes in algae with chemical changes in the water. All of these investigations on ponds concentrated upon phytoplankton algae. The only detailed studies of the unattached bottom living algal flora of fresh waters are those of Lund (1942) and Brook (1954). The work of the latter author is the only other detailed study of the algae of slow sand filter beds of waterworks. The work of Brook (1952-54-55) was concerned mainly with the bottom living attached and unattached floras of filter beds which had been in operation for varying lengths of time, with relatively little attention being paid to the chemistry of the water. No account was taken of the algae in the supernatant water. Brook (1954) stated that the environmental conditions in the slow sand filter beds were in many respects like those of a river and he compared his results with those of Butcher (1932, 1940, 1946).

In the present investigation an attempt has been made to relate both physical and chemical changes in the water to the seasonal periodicity of the algal flora of slow sand filter beds. Chemical factors such as pH, silica, ammoniacal and nitrate nitrogen and phosphate phosphorus together with physical factors such as light and temperature were investigated. Ecological field work was started in August 1963 and continued until December 1965 although certain experiments were continued until June 1966. Observations are thus available for more than two complete annual growth cycles. Rather than sample from various filter beds which had been in use for the same length of time (see Chapter II), the same four filter beds were examined continuously throughout the period of study. Local variation from one filter bed to another was thus taken into account. In conjunction with the field work certain laboratory studies were carried out to elucidate some of the field results. Reference has also been made to species of algae which are of taxonomic interest.

It was hoped that this study might help explain some of the differences, observed by waterworks authorities, in the performance of slow sand filter beds within individual works. This could in turn lead to a more efficient use of slow sand filter beds.

## II. The supply of water to the Slow Sand Filters and their operation

The slow sand filter beds studied are situated in the Thames valley west of London at the Ashford Common Works of the Metropolitan Water Board (O.S. map reference TQ 087/700). The position of the Ashford Common Works in the relation to the main storage reservoirs supplying the works and to the River Thames is shown in Figure 1.

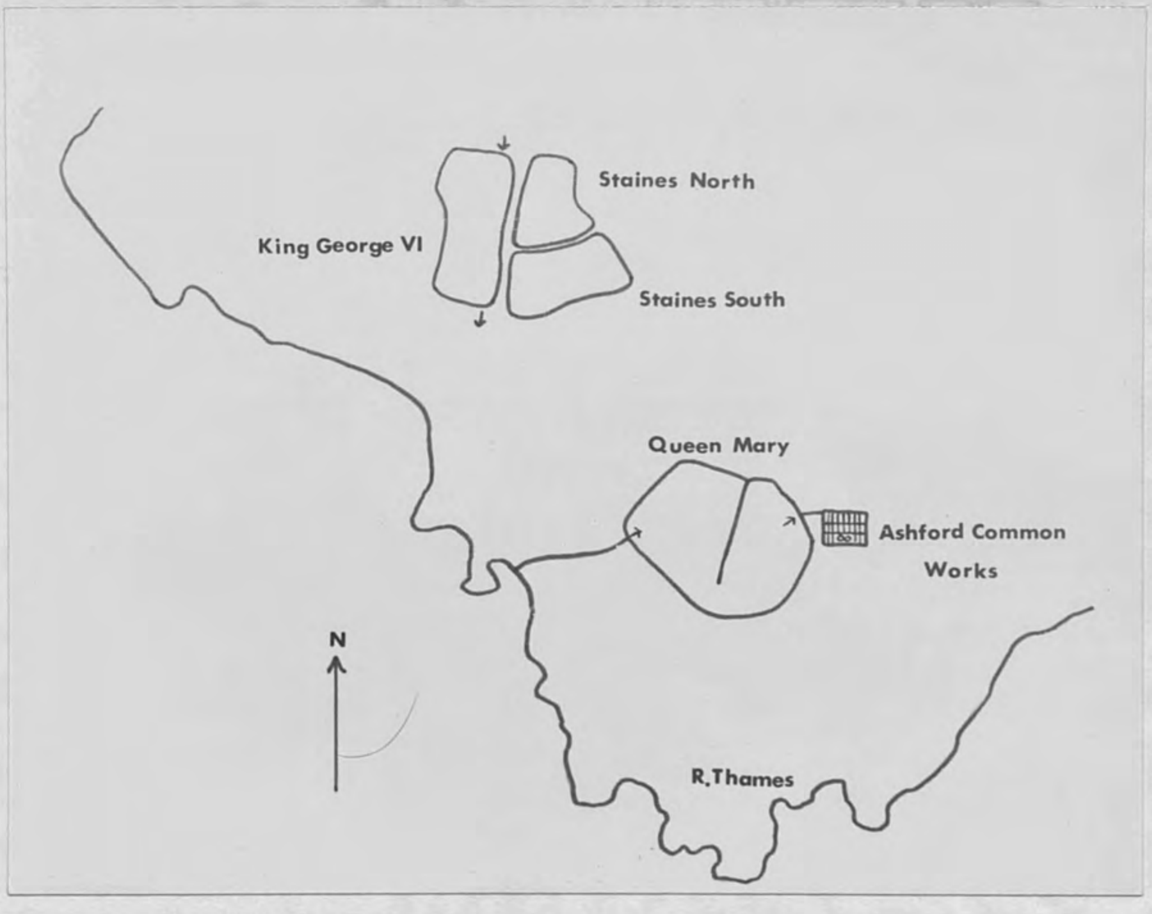


Figure 1. The location of the Ashford Common Works and the supply reservoirs in the Thames Valley.

All of these reservoirs receive their water via intake channels and puming stations from the River Thames. Unlike many reservoirs in other parts of the country, those in the Thames Valley are entirely

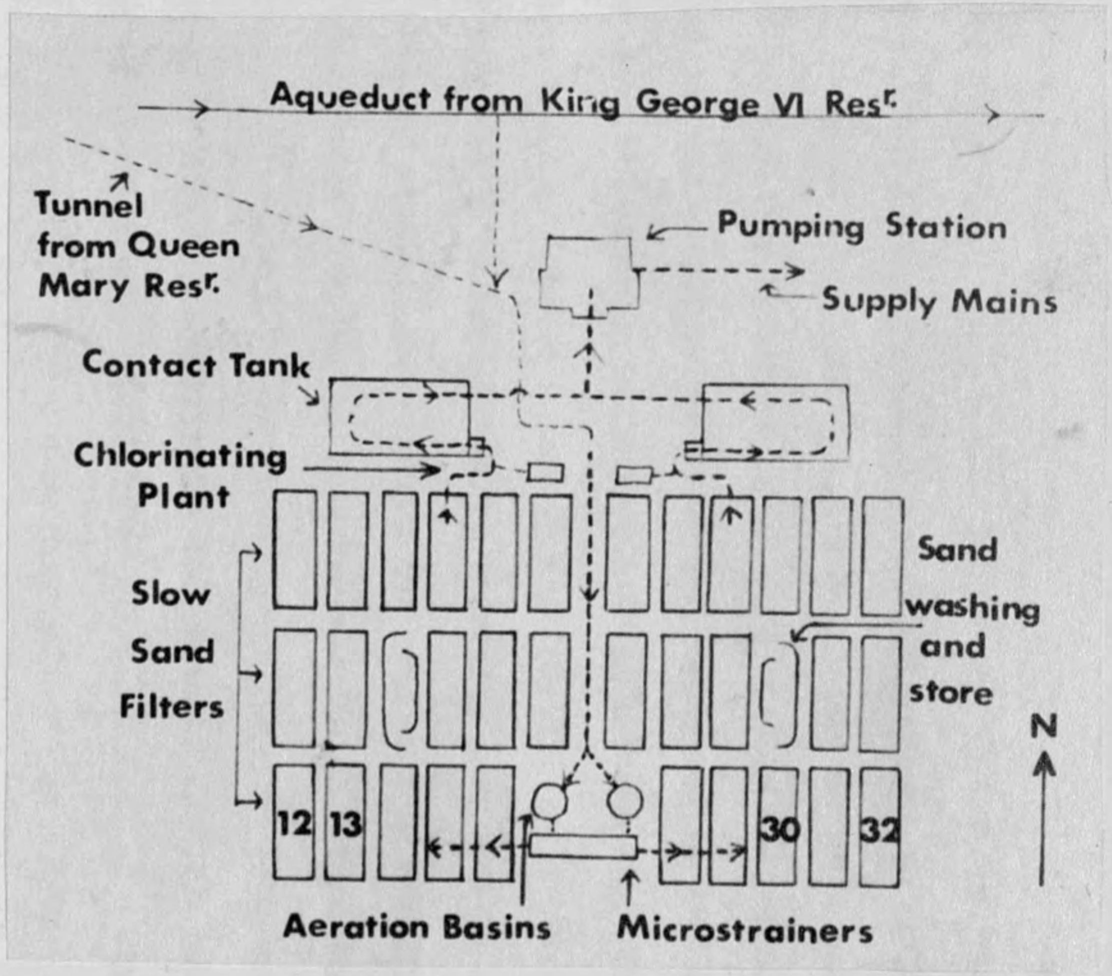


Figure 2. A plan of the Ashford common purification works showing the passage of water (indicated by arrows) through the works.

man-made basins, not just dammed valleys. The construction of these reservoirs is described by Ridley (1964). The water is generally alkaline and rich in dissolved organic and inorganic substances.

The water from the reservoirs passes, by means of an underground tunnel, to the purification works (Figure 2). As the reservoir water surface is well above the surrounding ground level, the flow along the tunnel is entirely due to the natural pressure head of the reservoir water. For mechanical reasons this pressure head has to be reduced before the purification treatment begins. To reduce the pressure head, and also to mix and oxygenate the water thoroughly, it is passed from the tunnel into two aeration basin fountains (See Plate 1). The purification works at Ashford Common is divided into two parts, each one being supplied by an aeration basin fountain. It must be stressed, however, that the original water supply for each half is exactly the same. The division into two was made for reasons of waterworks engineering and management. The water passes from the aeration basins into a series of rotary microstrainers (two sets of twelve). These consist of revolving drums, the walls of which are made of a fine stainless steel wire mesh with a mean pore size of 38µ x 45µ. The water passes in through the center of the drum end and out through the drum sides. Full descriptions of the detailed action of rotary microstrainers and their removal of algae are given by Bellinger (1968) and Taylor (1963 & 1965). The object of rotary microstrainers is to reduce the filtration load on the secondary slow sand filter beds onto which the water next passes, by removing particles exceeding a linear

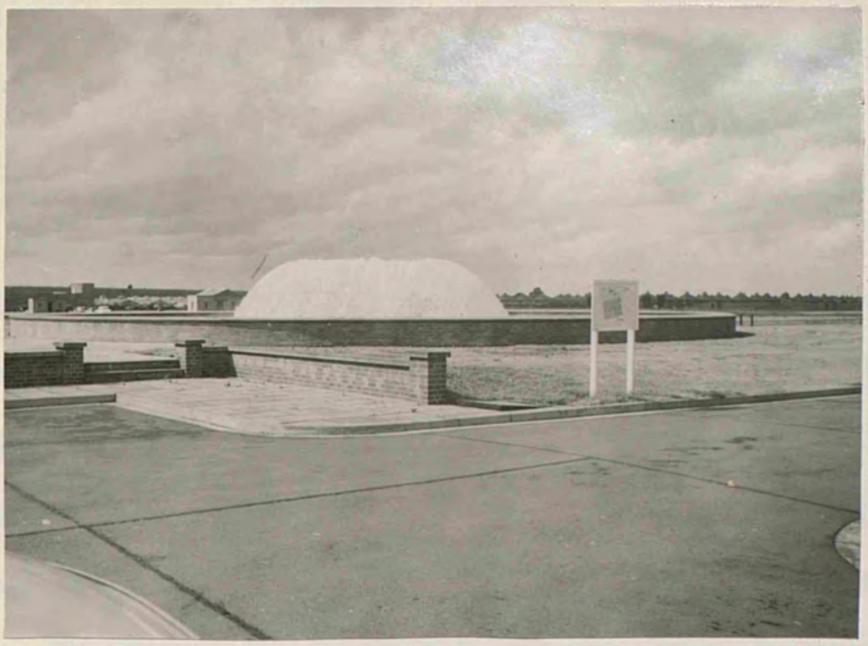

Plate I. An aeration basin fountain.

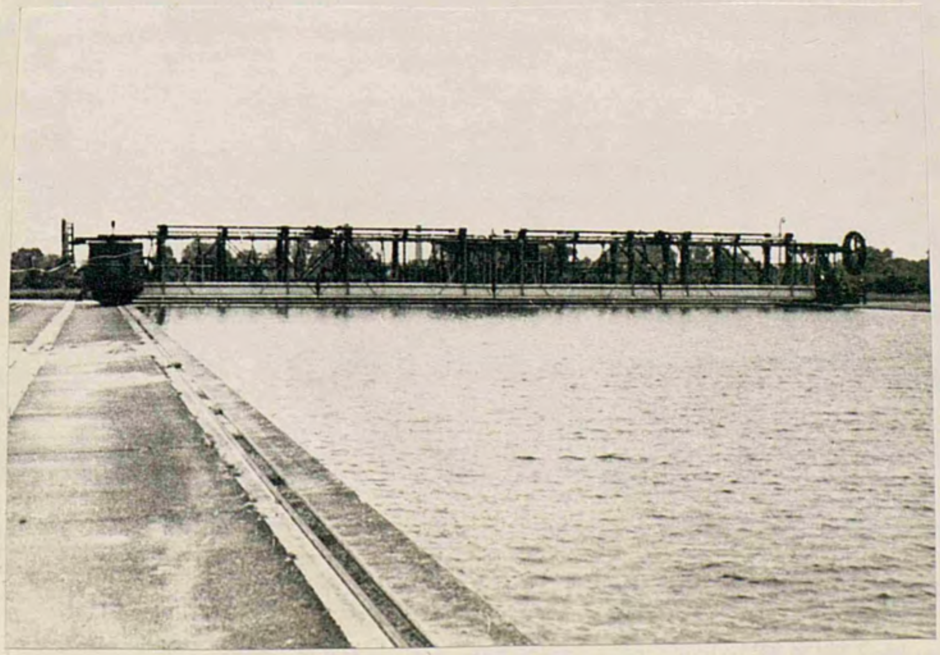

Plate II. The in-situ hydraulic sand washer ('hydra').

dimension of approximately 35µ.

The slow sand filter beds are concrete basins with vertical sides and area of ¾ acre (approximately 3000 $m^2$). The bottoms of these basins are covered with either porous tiles or a layer of porous concrete (see Figure 31). This is overlaid with coarse, then fine gravel. These in turn are overlaid with coarse and fine sand. The fine sand layer is usually between 1 and 3 feet (0.3 - 1.0m) deep depending upon the number of times the bed has been cleaned. (See page 13). Above the fine layer of sand is a water layer between 4 (1.3m) and 6 feet (2.0m) in depth. The water to be filtered enters the filter bed at one end just above sand level. It then percolates slowly through the sand at a rate of about 4 inches (10cms.) an hour (48 gals/sq.ft./day, 240mls./$cm^2$./day), and is collected in the tiles or porous concrete to be piped away for chlorination.

Suspended particles in the water which have not already been removed by the rotary microstrainers are trapped on the sand surface firstly by purely physical or mechanical means and then later, after the filter bed has been in use for a week or two, by means of a "zoogleal" film (Pearsall _et al_., 1946). This trapping of suspended particles on the film is more efficient at removing suspended matter from the water than the sand alone. As the amount of trapped material increases, and growth and reproduction of algae takes place, the resistance to the passage of water through the zoogleal layer and the sand column also increases. If a manometer tube were connected to the outlet pipe of the filter bed and the height to which the water rose in that tube were measured, the longer a bed had been in use, the greater would be the discrepancy between the height to which the water would rise in the manometer and the level of the water in the filter bed. This difference is of course a measure of the resistance to flow in the filter bed and might be as much as four feet. The increase in the resistance to the passage of water through the sand arises from an accumulation of particles in the zoogleal film at the sand surface. When the pressure difference (known as head loss) has reached a level of between two and four feet the filter beds are drained down for cleaning. Two methods of cleaning have been used on slow sand filter beds. The first method, which is still employed, involves mechanical skimming. In this method the top ½-1 inch of sand and detritus are skimmed off by means of a mechanical skimmer (see Plate III), and removed for cleaning. The surface of the sand is then raked smooth and the bed refilled from underneath with water. This prevents any air from being trapped in the sand. The bed is then put back into use. Whenever the depth of sand is reduced to about 12 inches (30cms.) the bed is resanded, i.e. it is cleaned and then an appropriate depth of sand placed over the surface to bring it back to its original level.

The second method of cleaning, which was not used after January 1964 on the filter beds studied, is by means of an in-situ hydraulic sand washing plant. Full descriptions of the plant and its method of operation are given by Laval (1952) and Burman & Lewin (1960). The actual machine is shown in Plate II. Briefly the method is as follows: a washing caisson is lowered into the sand to a depth of

Plate III. A mechanical skimmer removing the surface sand and silt.

PlateIV. Close up view of skimming mechanism.

6 inches (15 cms.). The upper part of the caisson consists of a tube of thistle shaped cross section connected by means of flexible hoses to a suction pump. Entering the top of the caisson are many guide tubes through each of which passes a lance with a radial jet orifice at its end. The machine was originally operated by lowering the caisson onto the sand surface. The lances are lowered through the guides into the sand and water is forced through them. The water may be removed through the caisson tube and the flexible hoses by means of the suction pump. After about one minute the lances and caisson are lifted clear of the sand and moved to the next position in the filter bed and the operation is repeated. The main object of this method of cleaning is to wash the sand in-situ, i.e. to remove the algae and accumulated organic matter from the sand without having to remove the sand from the filter bed. It has, however, one great disadvantage in practice. Unless the suction velocities are precisely set, algae tend to be trapped by sand particles falling back onto the sand surface after being washed into suspension by the lance jets. This results in a re-innoculation of the bed with algae which then grow vigorously due to reduced competition, hence the abandonment of this method.

The hydraulic in-situ sand washer (henceforth referred to as the "hydra") was used to clean beds 30 and 32 up to the end of January 1964 (see Figure 3.) The mechanical skimming was used throughout the period of study in beds 12 and 13 and after January 1964 in beds 30 and 32.

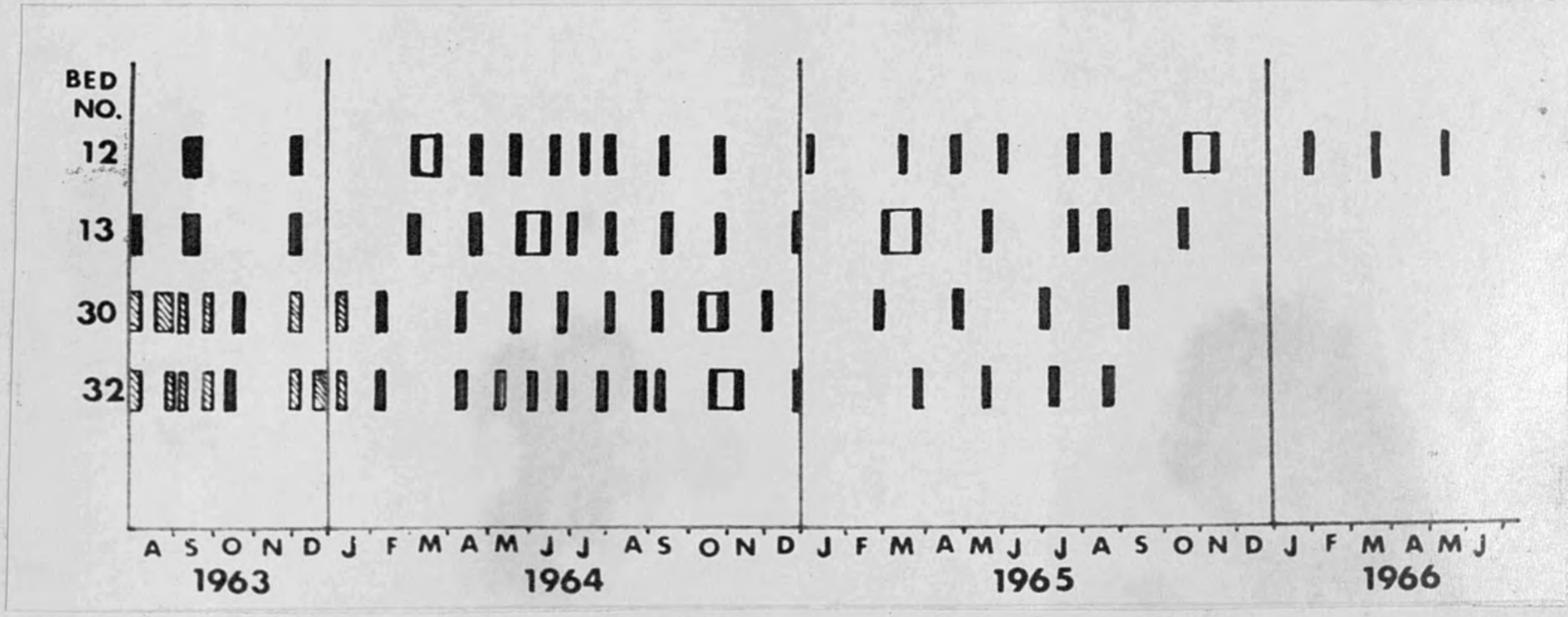


Figure 3. The periods of cleaning of the four filter beds studied. ▮ = mechanical skimming, ▯ = hydra cleaning, ▯ = resanding.

# PART TWO. METHODS.

## III. Methods.

### A. Collecting.

Samples were collected at weekly or fortnightly intervals throughout the period August 1963 to December 1965. Water for chemical analysis was collected from the water passing out of the rotary microstrainers (see page 10), and occasionally from the supernatant and the filtered water of the filter beds themselves. Water samples for oxygen determinations were collected from the reservoir outlet before treatment by filtration. This avoided any changes in the oxygen concentration due to treatment.

Filter bed supernatant water for phytoplankton determinations was collected by means of a rubber hose (Lund, 1949) from at least ten stations around each filter bed (see Appendix 2). Samples from the various stations were thoroughly mixed and an aliquot removed for counting. Occasionally samples throughout the depth of the supernatant water were obtained. These were collected at vertical intervals of 1 ft. (30.5 cms.) by clipping a rubber tube to various points on a pole placed vertically at different stations in the filter bed. Samples were obtained by applying suction to the shore end of the tube. To minimise contamination of the sample by materials remaining in the tube, a double bottle technique was used (Figure 4.) In this method the valve was used to maintain the reduced pressure developed by the hand suction pump. The water passed first into the small bottle (B) and then, when this was full, in to the larger bottle (A). Bottle B was allowed to refill approximately twice over before the bung was removed and the sample preserved for counting.

Samples of the unattached bottom living flora were obtained using a modification of the suction pump technique of Brook (1954). Two bottles were used (Figure 4.), the larger one (A) acting as vacuum reserve. A twelve foot length of rubber tubing was attached to the small bottle (B) at one end and to a 7.5 cm. diameter filter funnel which was weighted by a lead ring (Figure 4.b.) at the other end. The funnel was lowered onto the sand surface and left in place. Water containing the unattached algae was then sucked up into bottle B. When this was full the bung was removed and the sample poured into a 5L. flask. Samples were collected from several stations to take into account local aggregations as it had been found (see Chapter VII and also Appendix 2, page 226 ) that the sand surface did not support an even growth of algae as was suggested by Brook (1954). Care was taken to operate, as nearly as possible, a constant rate of pumping at each sampling. The separate samples from each station were thoroughly shaken together in the 5L. flask and an aliquot preserved for counting.

The attached algal flora was present mainly on the sand surface. The concrete walls of the filter beds, which were regularly painted with copper sulphate/lime paint, rarely supported appreciable growths of algae and were usually ignored in these investigations. Sand surface samples could only be collected when the filter bed had been drained down for cleaning. To investigate the attached algal flora during the filter bed runs a horizontal slide technique was used involving two main methods. In the first, derived from Butcher (1931),

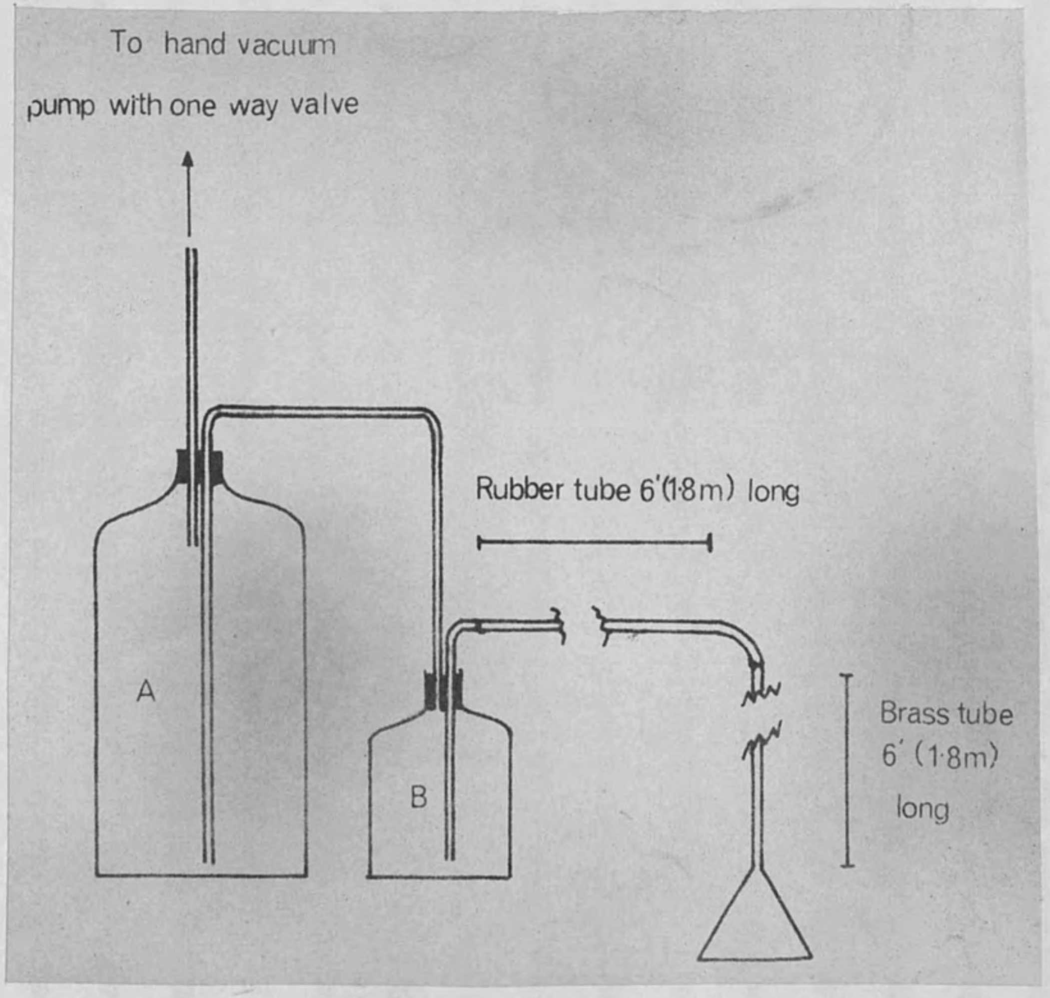


**Figure 4.** The double bottle collecting apparatus.

twelve glass microscope slides (7.5cm. x 2.5cm.) were fitted into weighted wooden frames and lowered through the water onto the sand surface. At the end of each subsequent week two slides were removed from the frome for examination. In the second method groups of four slides were clipped at intervals of 30cm. from the water surface down the length of a vertical pole standing in the filter bed about one metre from the edge. At fortnightly intervals one slide was removed from each depth for examination. In all these experiments the slides were carefully prepared by chemically cleaning and flame sterilising. Algae were removed from one half of the slide by scraping with a razor blade. The removed algae were carefully washed into a measuring cylinder. They were then preserved in Lugol's iodine for counting. A coverslip was then placed over the slide and the surface observed microscopically. The scraped area was observed to check that most of the algae had been removed and the unscraped area observed to ascertain the distribution and species of algae present.

On occasions, when the filter beds had just been drained for cleaning, core samples, or samples from different depths, of sand were taken to determine the distribution of algae through the sand.

B. Chemistry.

Phosphates, nitrates and silicates were determined absorbtiometrically as in Mackereth (1961). Dissolved oxygen was determined using the Pomeroy-Kirchman-Alsterberg modification of the Winkler technique (Pomeroy and Kirchman 1945). Ammonia was determined spectrophotometrically using sodium phenate reagent (Riley, 1953). Hydrogen ion concentration was determined using an E.I.L. model 23A pH meter and following the procedure outlined in the Standard Methods (American Public Health Association 1960). The dry weight of the suspended matter and its loss on ignition were determined by filtration through a Whatman GF/C 7.0cm. glass fibre filter paper (Nusbaum, 1958). The pads were prepared by drying in an oven at 105°C and storing in a dessicator. The pad was weighed before use and an appropriate volume (one to four litres) of water filtered depending upon the numbers of algae present. The pad was sucked dry and washed with distilled water. It was then oven dried at 105°C to constant weight before reweighing and ashing at 405°C to determine the loss on ignition.

Total particulate carbon was determined by wet oxidation using acid dichromate. Glass fibre pads (Whatman GF/C) were cut into 25mm. discs and immersed in chromic-sulphuric acid mixture, to remove any traces of carbon, for at least twenty four hours. The pads were then washed free of the acid with distilled water. Between 25 and 500mls. of sample, depending upon the concentration of algae, were then filtered and the pad sucked dry. It was then placed in a 50ml. round bottomed flask together with the sulphuric acid-dichromate oxidant made up according to Strickland & Parsons (1960). To this was added 5ml. of mercuric chloride-silver sulphate catalyst. This was then refluxed for one hour using cold finger condensors over a boiling water bath. The excess dichromate was then titrated against ferrous ammonium sulphate solution using ferroin indicator as described in Standard transparency readings could only be taken in fairly calm conditions as the effect of surface ripples was very marked. Whenever possible measurements were taken at 0.1 metre intervals throughout the depth of the water. On some occasions only the 0.1 metre and the sand surface readings were taken. Computation of underwater light intensities were made using the Kew Observatory total radiation figures and following the method outlined by Talling (1957) and Jenkin (1937).

D. Enumeration.

Algal cells were enumerated using the inverted microscope technique of Utermohl (1931), (see also Lund 1951, and Lund, Kipling and LeCren 1958).

A modification of the basic method, involving the use of bipartite counting chambers, as developed for routine quantitative phytoplankton determinations in the Botany Department of Royal Holloway College by Dr. J.H. Evans (see also Lovegrove, 1960) was used. The top part of the modified counting chamber (Figure 5) consisted of a ⅛th inch (3.0mm.) thick perspex slide with a ⅜th inch (9.0mm.) hole passing through the centre. A perspex tube of internal diameter ⅜th inch (9.0mm.) was cemented vertically over the hole. The length of the tube depended upon the volume of liquid that it was desired to sediment. The bottom part consisted of a matching perspex slide without a tube cemented in place but with ⅛th inch (3.0mm.) perspex spacers cemented to the underside of each end.

The method of use was as follows. A ⅞th inch (22.0mm.) square

Methods (A. P. H. A., 1960).

Nitrites were determined using the Greiss-Ilosvay method (Mackereth, 1961). Albuminoid nitrogen was determined using the direct method of Kitto (1938).

C. Meteorological and Physical determinations.

The results for air temperature, rainfall, evaporation, sunshine and solar radiation were obtained from the Meteorological Observatory, Kew.

Water temperatures were obtained using a thermistor, readings being taken to 0.2 degrees C. Underwater illumination was measured using a model 123 selenium rectifier photoelectric cell manufactured by Sangamo-Weston Ltd. The waterproof photometer case was constructed according to Atkins et al (1938). The photometers were used in conjunction with red, green, blue and yellow filters. These were types OB1, OG1, OY2 and OR2 by Chance (England) having transmission maxima at 480, 530, 625 and 675 mμ respectively (see Appendix 1). The cells were wired into an electrical circuit so that they were back to back, i.e. the difference in reading between the submerged photocell and the surface reference cell was read directly on the microammeter (see Appendix 1 for circuit diagram). The transparency of the water was measured according to Atkins et al (1938). Readings were always taken between 11.00 a.m. and 2.00 p.m. and a note made of weather conditions. No corrections were made for changes in solar elevation. Since the filter bed supernatant water was relatively shallow, coverslip was smeared with vaseline at the edges and then stuck to the underside of the bottom perspex slide making sure that no vaseline entered the hole in the slide and that a complete seal was made between the slide and the coverslip. The upper slide and tube were then smeared on the underneath with vaseline and sealed onto the top surface of the bottom slide. Care was again taken to ensure that no vaseline

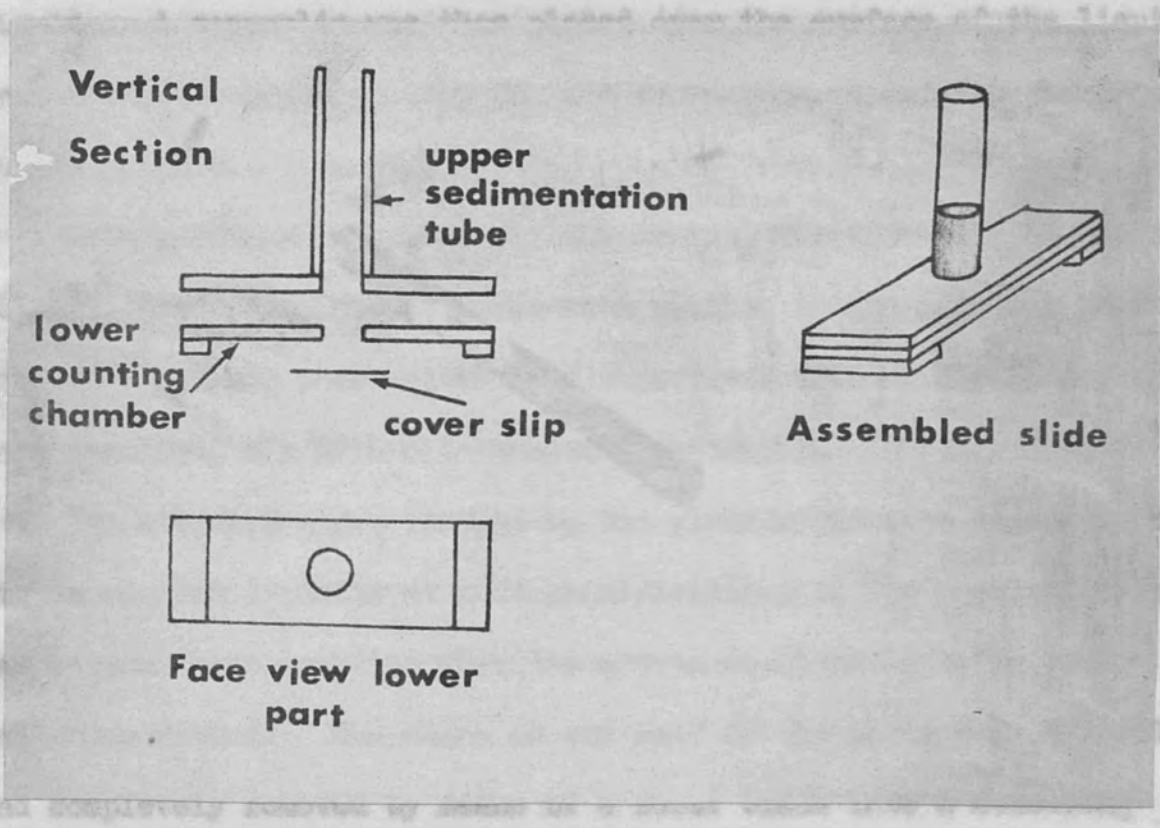


Figure 5. Split counting chamber for inverted microscope counting.

entered the hole and that the seal was complete. It was also imperative to make sure that the holes and the tube of the top and bottom halves were exactly lined up. If any vaseline did enter the hole it was carefully removed with a needle. The sample to be counted, already preserved in Lugol's iodine, was then run into the tube and the chamber placed in the shade on a horizontal surface to allow sedimentation to take place. One hour was allowed for each millilitre of sample to be sedimented, after which time the top half was slid gently sideways and the excess liquid pipetted off. The top was then completely removed leaving the lower chamber with only about 3.0mm. depth of liquid. A coverslip was then placed over the surface of the liquid, care being taken not to trap any air bubbles or to disturb the liquid in the chamber.

This system greatly reduces the optical disadvantages of the basic sedimentation technique. Using this chamber it was possible to use both high and low power microscope objectives without difficulty and, when required, a 1/12th oil immersion objective.

The attached algae growing on the glass microscope slides were either counted directly at x200 magnification, if the population density was low, or more usually, when the growth was luxuriant, by the following method. The algae on one half of the slide were carefully and completely removed by means of a razor blade into a measuring cylinder. Lugol's iodine was added to preserve and weight the algae and the volume made up to 1 litre with distilled water. The resulting suspension was thoroughly shaken to break up any climps or filaments and a volume for counting withdrawn (Butcher, 1940). A glass coverslip was placed on the specimen slide and both the cleared and the untouched halves observed at x200 magnification. This enabled a check to be kept on the efficiency of the clearing and on the species and natural distribution of algae on the slides.

For certain series of samples and individual experiments where only the major constituents of the algal populations were being determined the membrane filter technique described by McNabb (1960) was used. The samples were preserved in Lugol's iodine rather than formalin. When the algae had been concentrated on the membrane, it was placed in some immersion oil already on a microscope slide. This was to avoid any possibility of the algae being disturbed. A warming up step to accelerate the clearing of the membrance was then employed as recommended by Moore (1963). The temperature used was 65°C which cleared the membrance within 30 minutes.

Core and separate depth samples were either observed directly under a microscope or the sample was shaken with a known volume of water and algae poured off. This was repeated several times to ensure maximum removal of algae. Aliquots of the resulting suspension were then counted using the inverted microscope technique.

## IV. A consideration of the use of algal volumes in interpreting algal populations.

Although counting methods for determining the population sizes of algae have many advantages (Lund & Talling, 1957), expressing the results as numbers of cells per litre may be misleading. This is due to the wide range of cell or colony size which may exist even within a well-defined species. In addition, for some algae, especially for example in the coenobial Volvocales and Chlorococcales, there may be a very large difference in size between the young and mature stages. Partial compensation may be achieved by expressing the results in volumes of cells per litre (Holsinger, 1955; Verduin, 1951; Whipple, 1927). To do this one has to know, with reasonable accuracy, the average cell volume of each species and variety encountered. With some algae this is not too difficult as their volume may be adequately calculated by geometric formulae, e.g. $\pi r^2 h$ for *Stephanodiscus astraea.* For others, however, volume determinations must necessarily be only approximate due to their complex shape. Plasticine models were used to determine the volume by displacement of water, e.g. with *Staurastrum* species.

Whichever method is used to determine the volume, accurate measurements of cell dimensions are essential. At least one hundred cells for each species quoted were measured and in some cases, e.g. *Stephanodiscus astraea*, over one thousand were measured. The results are given in table 1. The results obtained during this study are compared with those of Nauwerk (1963).

With the exception of Stephanodiscus astraea and its varieties the volumes quoted were used in calculating the weight of algae present. A specific gravity of 1.0 was assumed for all algae, thus their weight = volume x 1.0. A closer seasonal study of the cell volume of S.astraea was made. Measurements of cell diameter and where possible cell heights were taken throughout three years on acid cleaned frustules. When the population density allowed, over fifty cells from each sample were measured. The arithmetic mean was calculated and the average cell volume determined for each sample. The results are given in Figure 6. A distinct seasonal variation can be seen with maximum average cell diameters occurring in the winter and minimum diameters in the summer periods.

The onset of the spring maximum corresponded to a rapid decrease in the average cell volume. The spring of 1966 initially had a similar trend but applications of copper sulphate reduced cell numbers considerably. By May the population density was again increasing and a second spring maximum occurred in mid-summer hence the less regular shape of the curve for 1966. The autumn-early winter period was marked by an increase in the average cell volume.

An attempt to ascertain whether the average cell diameter of a single population of S.astraea could vary as much as recorded in natural populations was made. For this purpose unialgal cultures were started in various media (Rodhe VIII; Rodhe, 1948; Chu, 10; Chu, 1942; and natural steam sterilized reservoir water) with or without soil extract added but without success. The cultures were able to

Table 1. The volumes of algae in cubic microns calculated from personal observations (column 1) compared with the results of Nauwerk,1963,(column 2).

| | 1 | 2 |
|---|---|---|
| Chlorophyta | | |
| Chlamydomonas angulosa | 1760 | - |
| Pandorina morum (col.) | 2000 | 3000 |
| Eudorina elegans (col.) | 4200 | 3000 |
| Volvox aureus (col.) | $2.71 \times 10^4$ | $3.0 \times 10^4$ |
| Ulothrix tenuissima | 1200 | - |
| U. zonata | 7000 | - |
| U. tenerrima | 400 | - |
| Pediastrum boryanum (col.) | $1.6 \times 10^4$ | 8000 |
| Coelastrum microporum (col.) | 6500 | 3000 |
| Chlorella sp. | 30 | 20 |
| Oocystis solitaria | 440 | 500 |
| O. crassa | 3050 | - |
| O. lacustris | - | 200 |
| Ankistrodesmus falcatus | 320 | 250 |
| A. falcatus v, mirabilis | 850 | - |
| Schroederia setigera | 660 | 500 |
| Tetraedron minimum | 200 | 30 |
| Scenedesmus acutus | - | 1000 |
| S. bijuga | 700 | - |

| | 1 | 2 |
|---|---|---|
| S. quadricauda | 1000 | 1000 |
| S. quadricauda v. maxima | 4450 | - |
| Actinastrum hantzschii (col.) | 350 | - |
| Crucigenia tetrapedia (col.) | 150 | - |
| Micractinium pusillum | 160 | - |
| Tetrastrum staurogeniaeforme (col.) | 160 | - |
| Bacillariophyta | | |
| Melosira varians | 2000 | 1800 |
| M. granulata | 1340 | 800 |
| M. granulata v. angustissima | 440 | 370 |
| Stephanodiscus astraea | 13700 | 25000 |
| S. astraea v. intermedia | 5650 | 5000 |
| S. hantzschii | 800 | 2500 |
| Tabellaria fenestrata | 3100 | 3000 |
| Diatoma vulgare | 1460 | - |
| Fragilaria crotonensis | 640 | 300 |
| F. capucina | 490 | 200 |
| Asterionella formosa | 756 | 800 |
| Synedra acus | 630 | 200 |
| S. ulna | 4220 | 5000 |
| Nitzschia acicularis | 320 | - |
| N. palea | 260 | - |
| N. sigmoidea | 6400 | 5000 |
| N. linearis | 3450 | - |
| Surirella ovata | 4000 | - |

| | 1 | 2 |
|---|---|---|
| Cryptophyta | | |
| Cryptomonas ovata | 1000 | - |
| Rhodomonas minuta | 400 | - |
| Cyanophyta | | |
| Microcystis aeruginosa | 32 per cell | $1 \times 10^5$ per 200µ colony |
| Oscillatoria limosa per cm. | $2 \times 10^6$ | - |
| O. tenuis .. | $2 \times 10^5$ | $1.5 \times 10^5$ |

survive for about six weeks but then died. Thus no information is at present available as to whether the variations in average cell diameters of this alga were due to changes within the same population or due to the development of different populations at different times of the year. The results obtained and their relationship to other environmental factors such as light and temperature are discussed fully in Chapters IX and XI.

Whatever the reasons for the seasonal variations in cell volumes it is important to take them into account when calculating the volumes of populations in periodicity studies. This variation also illustrates the need for more comprehensive records of all cell dimensions for use in surface area or volume calculations if errors are to be avoided.

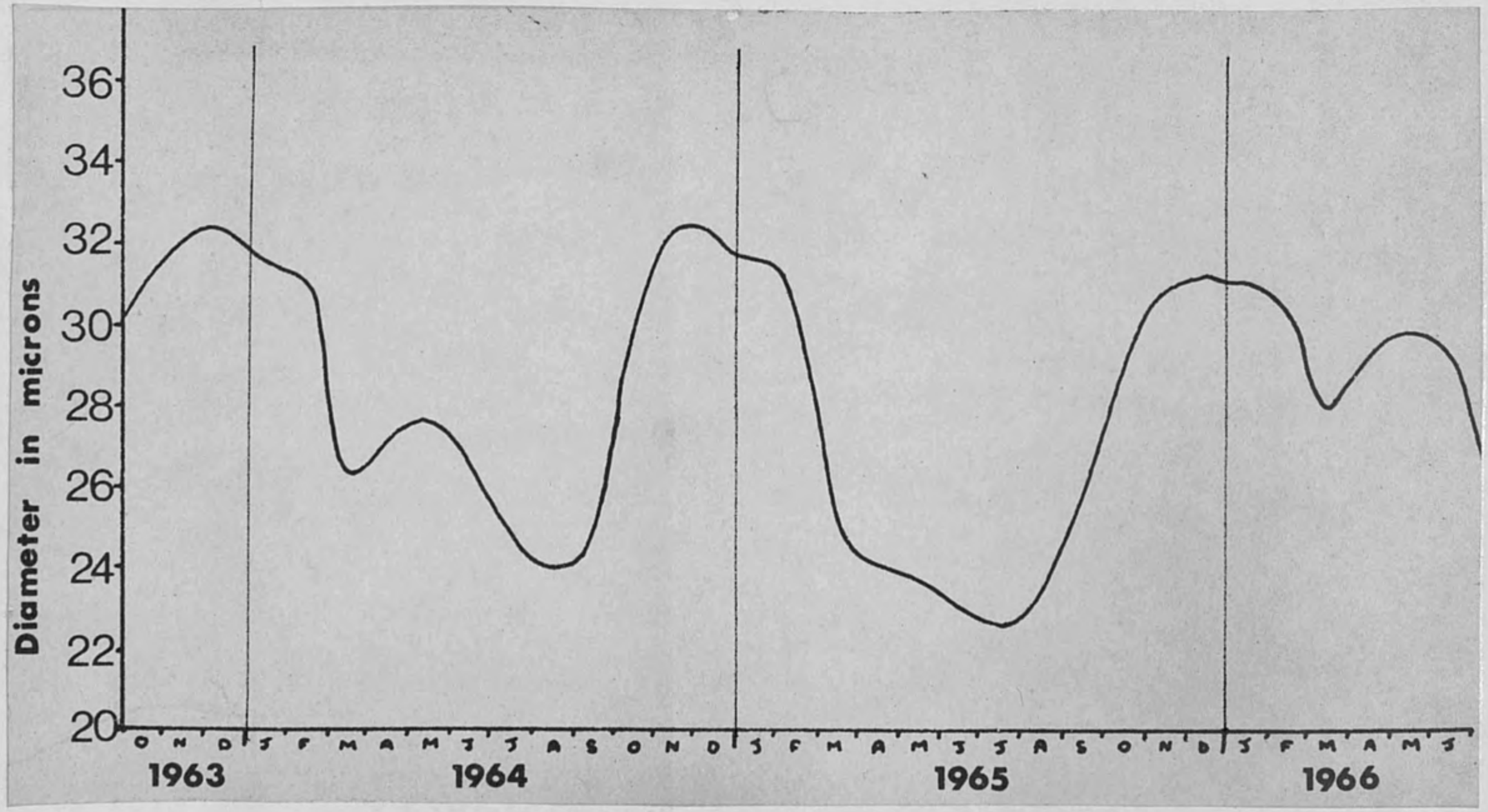


**Figure 6.** The seasonal variation in the average cell diameters of populations of *Stephanodiscus astraea*.

# PART THREE. RESULTS AND DISCUSSION.

V. Meteorological data. Physics and Chemistry of the inflow water.

Records of rainfall, evaporation, air temperature and radiation were obtained from the Meteorological Office, Kew. The natural flow of the River Thames over Teddington Weir throughout the period was obtained from the Thames Conservancy Board.

The results, expressed as monthly totals in millions of gallons, for the flow of the River Thames are given in Figure 7. The highest recorded flows were during the winters of 1963-4 and 1965-6. During the winter of 1964-5 however, the flow was fairly low being only a little above the summer levels. The flow of the river can be correlated to total rainfall but more closely to flood rainfall (Figure 8.)

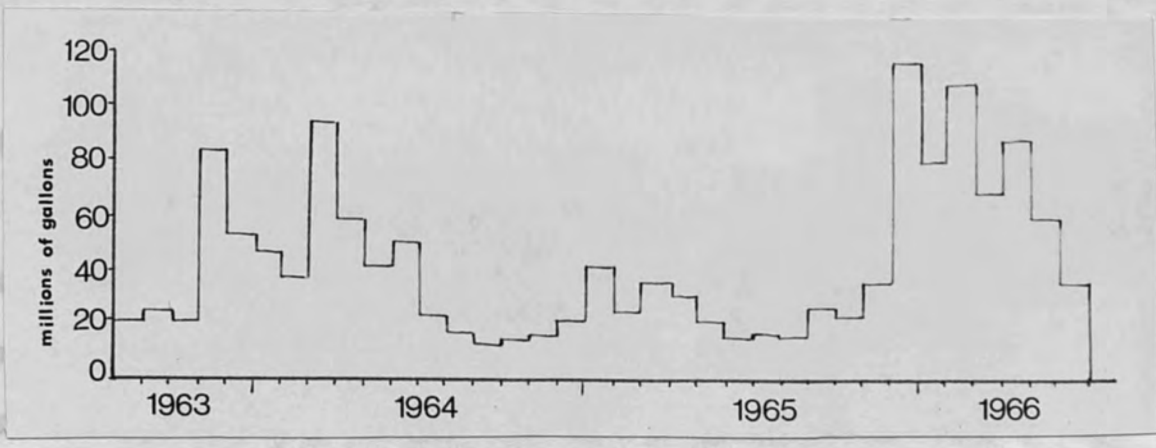


Figure 7. The flow of the River Thames at Teddington Weir.

Flood rainfall is the amount of water remaining after evaporation has taken place and is an indication of the amount of surface water run off into the river. The figures for flood rainfall were obtained by subtracting the total weekly evaporation in mm. from the total weekly rainfall in mm. The periods of highest flood rainfall, in November

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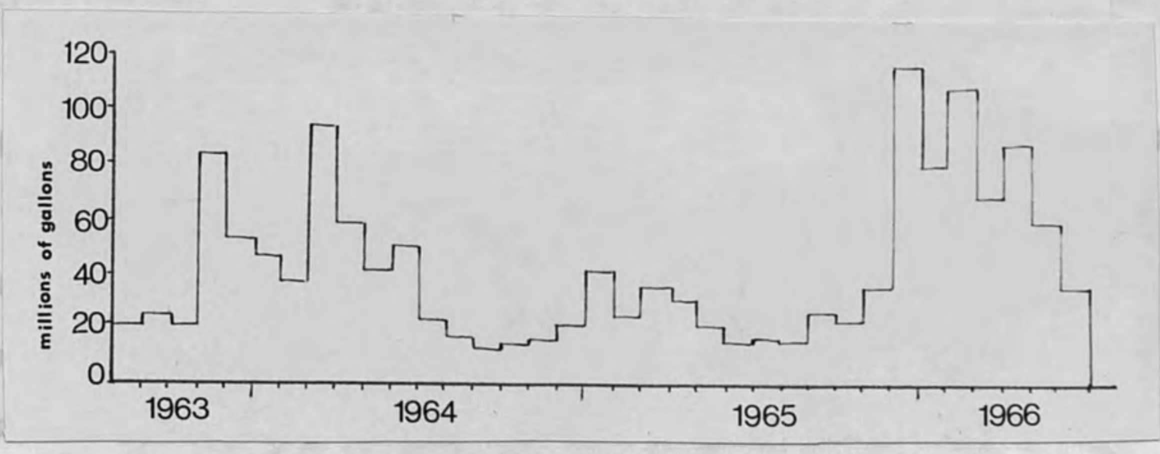


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Flood rainfall is the amount of water remaining after evaporation has taken place and is an indication of the amount of surface water run off into the river. The figures for flood rainfall were obtained by subtracting the total weekly evaporation in mm. from the total weekly rainfall in mm. The periods of highest flood rainfall, in November

1963, March and June 1964 and most of the winter and spring period of 1965-6, corresponded to periods of high river flow.

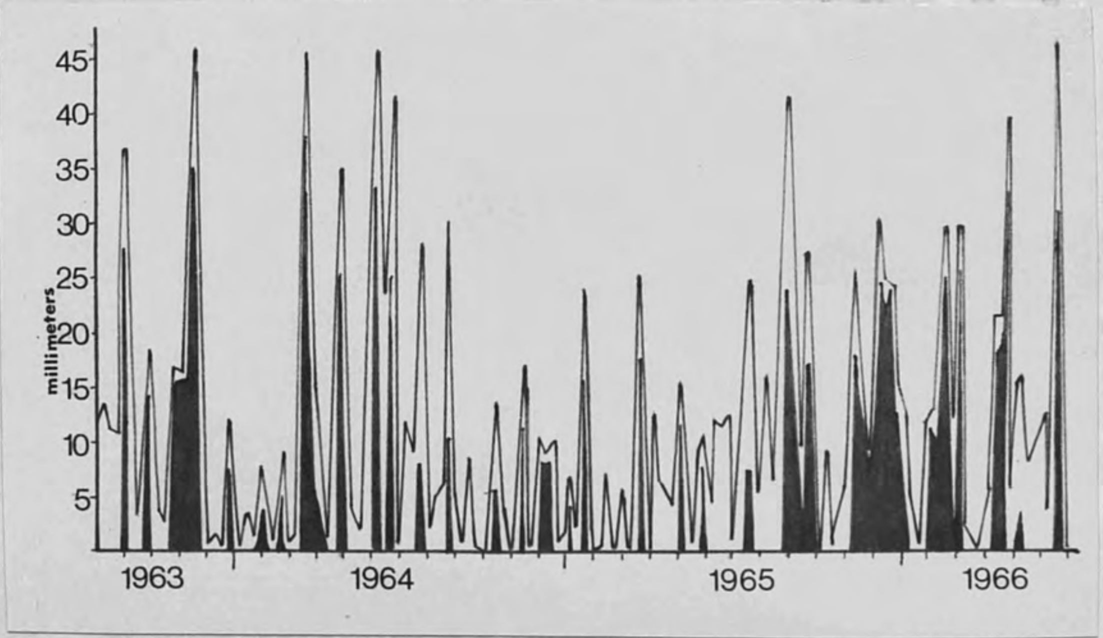
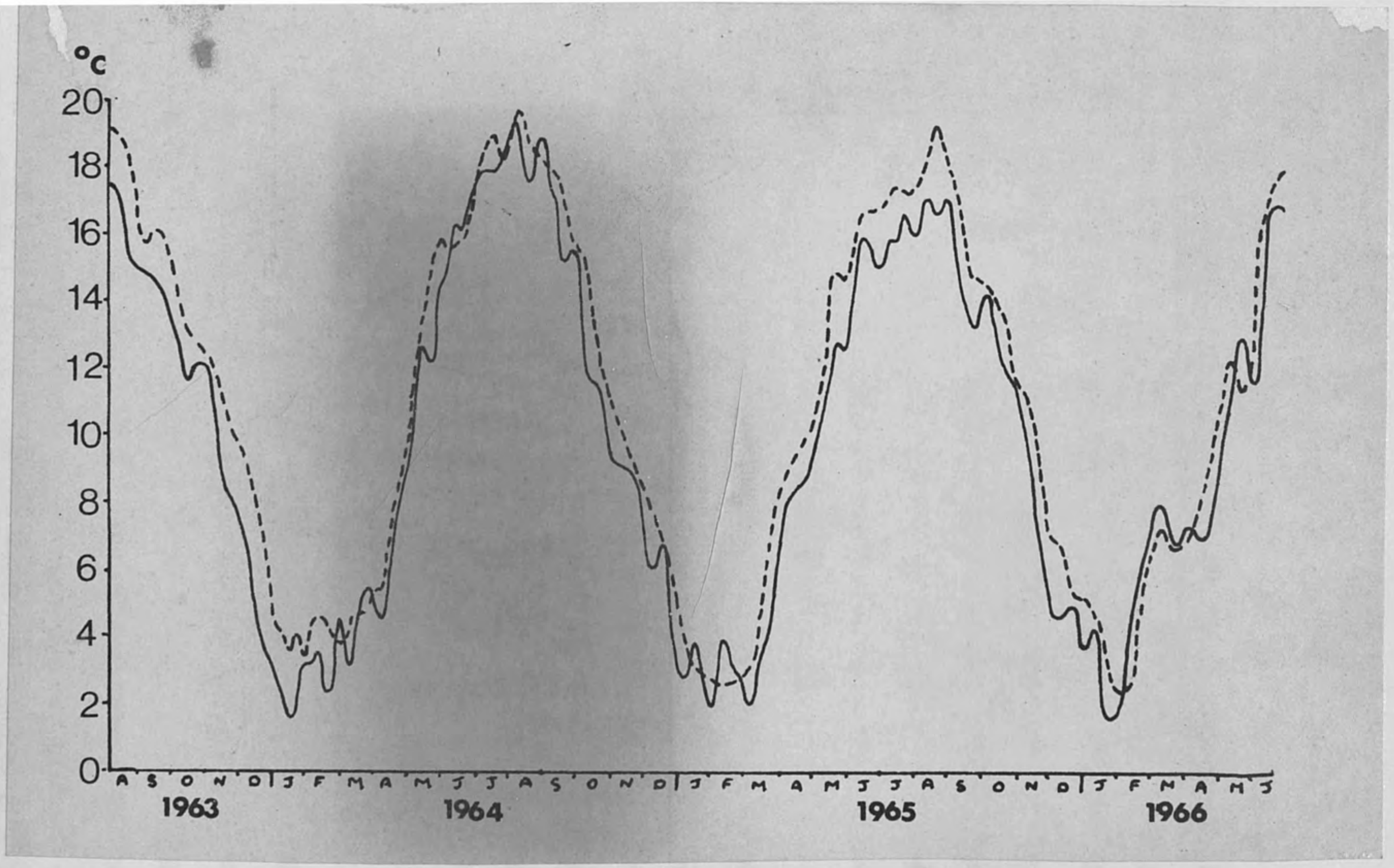



Figure 8. The total and flood rainfall (shaded areas represent flood rainfall).

There was a high recorded flood rainfall in September 1965 but only a small increase in river flow was recorded. Lowest flood rainfalls and smallest river flows usually occurred during the summer and autumn months. The magnitude of the River Thames flow did appear to affect the concentration of chemical nutrients in the water and thus affect their availability to algae in the filter beds (see pages 55 to 58 ).

The temperature of the water was taken in the aeration basin, (this gives the temperature of the water flowing out of the reservoir and entering the filter beds), and in the supernatant water of the filter beds near to the sand surface to avoid any surface heating




<u>Figure 9</u>. The water temperature in the reservoir and the filter bed supernatant water. ----- reservoir temperature, ——— filter bed temperature.

effects, (or in winter, cooling effects). The results are given in Figure 9. The temperature of the supernatant water in the filter beds was taken before midday and was usually a little below that of the reservoir, especially during the winter months when thin layers of ice were often observed to form on the surface. Air temperatures were usually below that of the water at night and above that of the water in the afternoon. Although turbulence and vertical transport usually maintained the supernatant water in the filter beds in an isothermal condition, on calm summer days and also when ice cover stopped any wind action, vertical thermal stratification of the water did occur. Up to 3.5°C difference was recorded between the water at the sand surface and that just below the ice in February 1965. Also surface water temperatures of 23.5°C were recorded in late July and early August 1965 during some afternoons. It is very difficult on many occasions to separate the effects of temperature and light on natural populations of algae as low temperatures are often coupled, in cold temperate climates, with low light intensities. There is some evidence, however, (discussed more fully in Chapters VI and VII), that many algae present in these waters will grow and reproduce at low temperatures if other conditions are suitable. In the winter and early spring periods of 1964 and 1966 Asterionella formosa and Stephanodiscus astraea both increased considerably in numbers while water temperatures were near to their lowest levels.

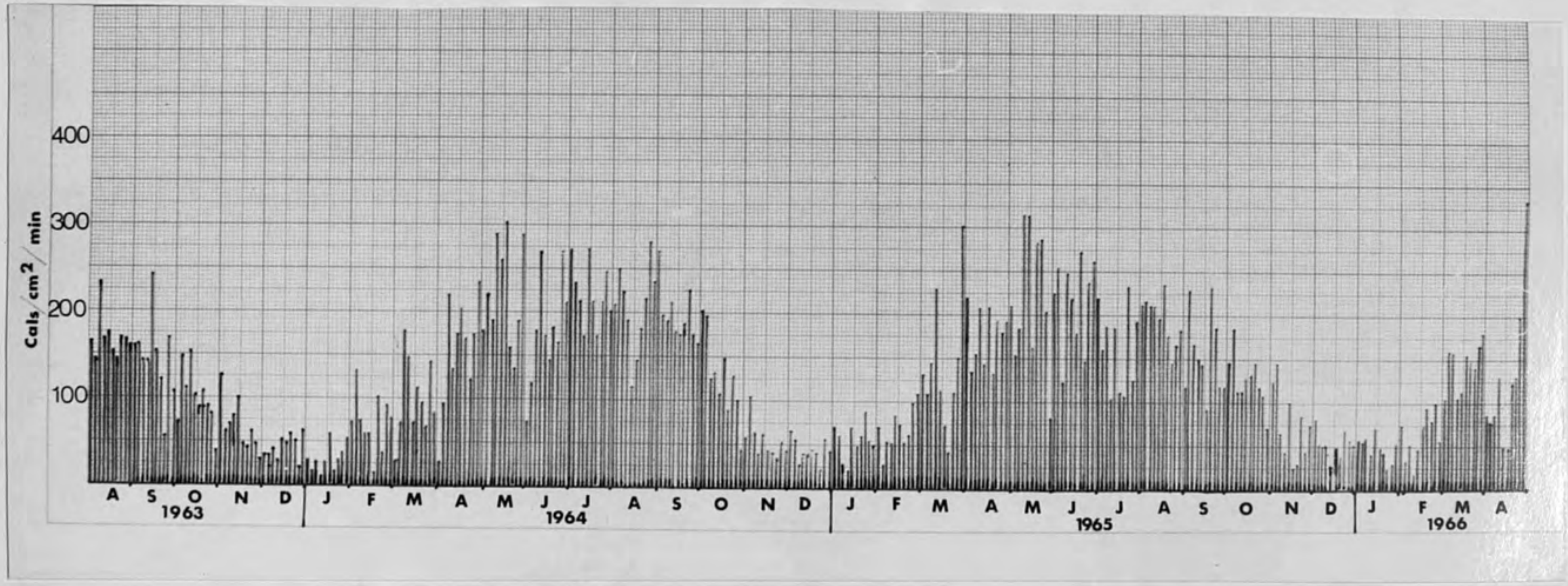
Solar Radiation

The figures for total radiation were obtained from the Meteorological Office, Kew, and were expressed in milliwatt hours per $cm^2$ per second ($mwhr/cm^2/sec.$). They were converted to photosynthetic radiation (i.e. that radiation falling in the range 3800Å to 7200Å) by multiplying the results by 0.5 as recommended by Edmondson (1956). They were then converted to calories per $cm^2$ per min. ($cals/cm^2/min.$) as these units rendered the results more easily comparable with other published data and these, or other closely related units, have been used by many other workers (Westlake, 1965).

From measurements using the submersible light meter, described in Appendix 1, over the period of investigation, an average extinction coefficient, k, of 0.68 was obtained. This is equivalent to a reduction in the incident radiation of 50% for every metre depth of water. In the slow sand filter beds the sand was overlaid with about 1.5 - 1.75 metres of water so that only 30% - 35% of the incident radiation reached the sand surface.

The results of the photosynthetic radiation incident on the water surface are given in Figure 10, each plot representing the average for three consecutive days. To obtain the figure for photosynthetic radiation incident on the sand surface 30% - 35% of the results given must be taken. On all but five occasions there was less than 100 cals/ $cm^2/min.$ of photosynthetic radiation reaching the sand surface.

Talling (1957) in photosynthetic experiments with <u>Asterionella formosa</u>, found that light saturation occurred at about 48 $cals/cm^2/min.$




<u>Figure 10.</u> The photosynthetic radiation incident on the water surface, each plot representing the average for three consecutive days.

at 5°C. Ryther (1956), working within the temperature range 18-23°C, found for marine diatoms that saturation occurred at about 25 cals/ $cm^2$/min. He also found that the saturation levels, in the same temperature range, for the Chlorophyta and some dinoflagellates were 10 and 60 cals/$cm^2$/min. respectively.

From October to March of each winter period studied the water temperatures were always below 14°C and often as low as 4°C. If it is assumed, on the basis of the results of Ryther (1956) and Talling (1957), that for diatoms generally light saturation occurs at about 35 cals/$cm^2$/min., then for this level of photosynthetic radiation to obtain at the sand surface a value of about 100 cals./$cm^2$/min. would be required at the water surface. Throughout most of the period November to February incident photosynthetic radiation levels were well below this figure and light limitation of photosynthesis for diatoms at the sand surface probably occurred.

The penetration of light into the supernatant water in filter bed No. 12 was measured throughout most of the period January 1965 to June 1966. Technical faults in the apparatus prevented measurements being made during July and August 1965 and March and April 1966. The results given in table 2 are expressed in terms of vertical extinction coefficient, k, calculated according to Talling (1960) using the equation:-

$$z5\% = 3/k$$

where z5% is the depth interval in which light is reduced to 5% of its initial value. Throughout the period green light showed the

**Table 2** . The vertical extinction coefficient k for red, blue and green light in the supernatant water of filter bed No. 12 during 1965 and 1966.

| Date | Red | Blue | Green |
|---|---|---|---|
| 15 . 1 . 65 | 0.735 | 1.510 | 0.635 |
| 22 . 1 . 65 | 0.430 | 0.572 | 0.338 |
| 9 . 2 . 65 | 1.200 | 1.335 | 0.560 |
| 12 . 3 . 65 | 0.660 | 0.668 | 0.255 |
| 15 . 4 . 65 | 0.514 | 0.555 | 0.316 |
| 19 . 4 . 65 | 0.440 | 0.565 | 0.240 |
| 4 . 5 . 65 | 0.550 | 1.850 | 0.470 |
| 11 . 5 . 65 | 0.294 | 0.314 | 0.200 |
| 11 . 6 . 65 | 0.575 | 0.800 | 0.271 |
| 20. 9 . 65 | 0.740 | 1.200 | 0.265 |
| 11 . 10. 65 | 0.925 | 1.130 | 0.272 |
| 3 . 1 . 66 | 0.730 | 1.620 | 0.224 |
| 12 . 1 . 66 | 1.430 | 1.540 | 0.230 |
| 2 . 2 . 66 | 0.835 | 1.500 | 0.170 |
| 9 . 5 . 66 | 1.090 | 1.850 | 0.340 |
| 20 . 5 . 66 | 0.260 | 0.340 | 0.222 |
| 11 . 6 . 66 | 0.800 | 0.860 | 0.224 |

greatest and blue light the least penetration. The amount of penetration of each wave band varied on each occasion. The penetration of green light was at a minimum in January, March and May 1965. Blue light penetration was at a maximum in March, April and May 1965 and May 1966. Several factors can operate to prevent light penetration into the water. Colouring of the water reduces the penetration of light at the shorter wave lengths, indeed the reduced penetration of blue light in these waters was almost certainly due to this. Suspended particulate matter also prevents light penetration. Sauberer and Ruttner (1941) and Talling (1960) have reported reduced penetration of light due to large phytoplankton populations.

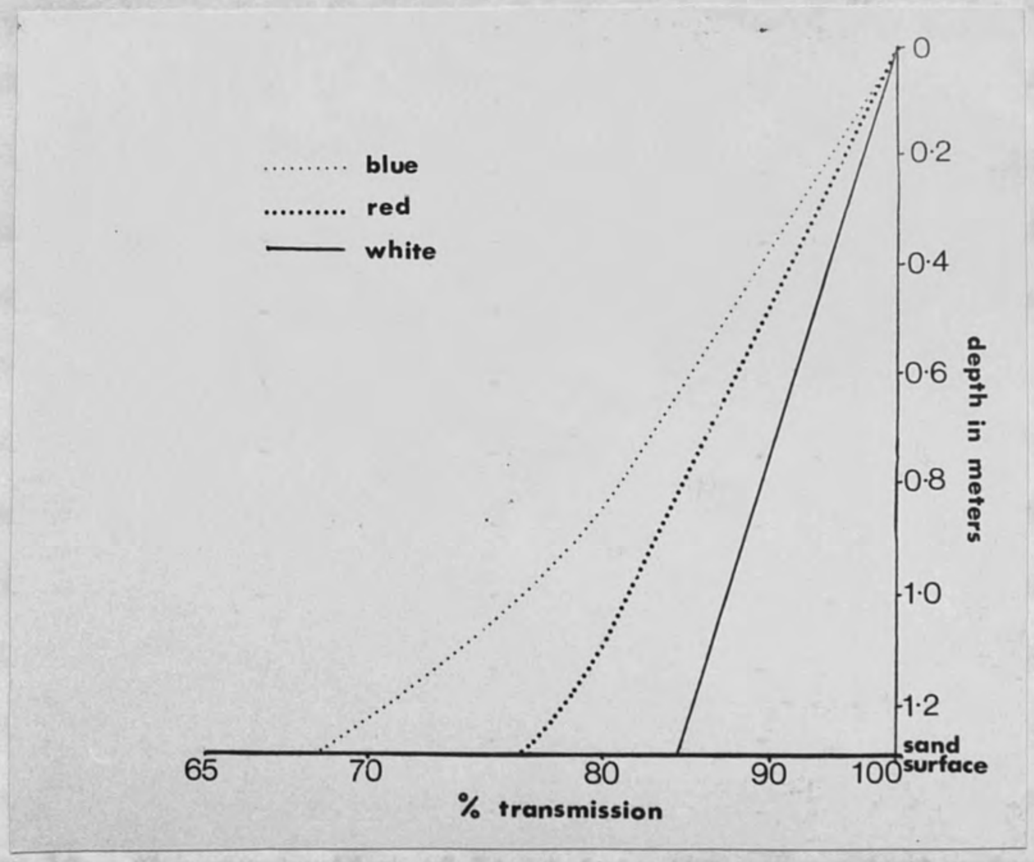



Figure 11. The penetration of light into the supernatant water of bed 12 on April 19th, 1965.

In February 1965 there was a large amount of silt and organic debris in the water together with large numbers of Stephanodiscus hantzschii. These caused a reduced penetration of light at all wave lengths. In June 1965 the water was coloured green by large numbers of Chlamydomonas sp.. On this occasion the penetration of both red and blue light was greatly reduced but the green was only slightly affected. In April 1966 reduced penetration at all wave lengths occurred when high numbers of Stephanodiscus astraea and Asterionella formosa were present in the water.

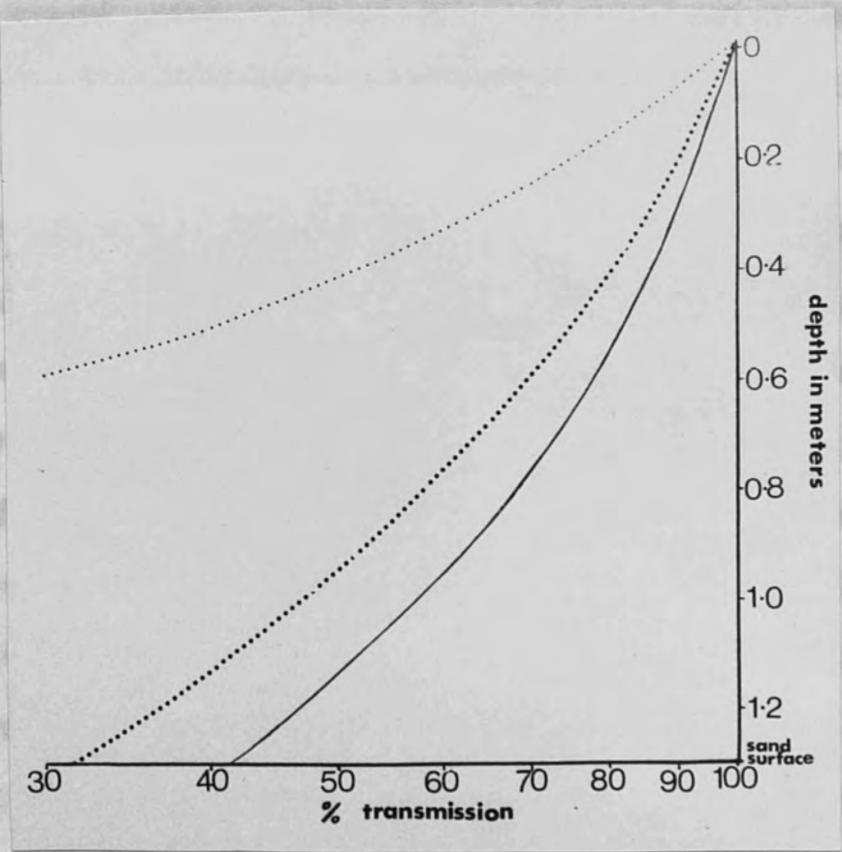



Figure 12. The penetration of light into the supernatant water of bed 12 on June 1st, 1965. ......... = blue, .... = red and ____ = white light.

On two occasions in 1965 the penetration of light into the supernatant water was measured at 0.2 metre intervals. The first occasion was on April 19th when measurements were taken at 1.30 p.m. The results, expressed as percentage transmission of the incident light, are given in Figure 11. Only small numbers of algae were present in the water which was also free from silt and organic debris. Over 68% of the light at each wave band measured reached the sand surface. The percentage transmission decreased more rapidly near to the sand surface with blue and red wave bands. This was possibly due to larger numbers of algae being present in the water at this depth (see Chapter VII). The second occasion was on June 1st when large numbers of <u>Chlamydomonas sp.</u> were present in the water (see Chapter VII). The results are given in Figure 12. On this occasion less than 4% of the blue light reached the sand surface. Only 34% of the red and 42% of the green light reached the sand surface. At the levels of incident radiation recorded on June 1st, this would certainly have caused the algae on the sand surface to be light limited (Ryther, 1956).

Although the filter beds are but shallow basins of water, occasions could occur when there was insufficient light for maximum algal growth at the sand surface. One example of such an occasion is given above. Large numbers of green flagellates were not uncommon in the supernatant water. Another occasion when light would have been limiting was when large amounts of filamentous green algae, e.g. <u>Ulothrix spp.</u> developed on the sand surface. These would most certainly have caused local shading and perhaps modified the distribution of other algae on the sand surface (see Chapter VII).

Chemical Results and Discussion.

Concentrations of the various chemicals are expressed in micro-gramme atoms per litre (µg. at./L) or micro-gramme molecules per litre (µg. mol./L). These units are obtained by dividing the concentration in micro-grammes per litre of the atom or molecules (i.e. $NO_3$-N or $SiO_2$) by their appropriate atomic or molecular weight.

The forms of nitrogen usually available for the growth of fresh-water organisms are well known (Hutchinson, 1957). Of these, the concentration of molecular nitrogen was not determined and that of nitrite nitrogen only infrequently determined as they were considered to be of lesser importance in these waters. Ammoniacal nitrogen, nitrate nitrogen and albuminoid (organic) nitrogen were determined regularly, usually at weekly intervals, during the period of study.

Only small quantities of albuminoid nitrogen were detected, the concentrations ranging between 7 µg. at./L and 26 µg. at./L. No correlation between the levels of albuminoid nitrogen and phytoplankton growth could be found. The method used (Kitto, 1938), however, is probably not sufficiently sensitive to detect small differences in concentrations and has been assumed to provide only an approximation of the proteinaceous nitrogen present (Standard Methods, A.P.H.A. 1960).

Ammoniacal nitrogen may be present in the water in two forms, $NH_4^+$ and $NH_4OH$. The relative proportions of these two forms depends upon the pH. This balance may be of ecological significance according to Cooper (1938). At 18°C, for example, the ratio of $NH_4^+$ to $NH_4OH$ at pH7 is 300 to 1; at pH8, 30 to 1 and at pH9.5, 1 to 1 (Hutchinson, 1957). Thus at high pH values toxic concentrations could develop of $NH_4OH$. Rodhe (1948) in his work on the requirements of plankton algae found that toxic amounts of $NH_4OH$ could be formed in culture media having ammonium compounds as a source of nitrogen and a high pH value. Concentrations of about 24 µg. at./L were thought to be toxic. The pH of the water passing onto the filter beds ranged between 7.5 and 8 (Figure 13). The ratios of $NH_4^+$ to $NH_4OH$ at these pH limits are about 90 to 1 and 30 to 1 respectively and the maximum amount of $NH_4OH$ nitrogen present would only have been 10 µg. at./L. Local photosynthetic activity in the filter beds could have raised the pH to above 9.0 (personal observation) and the $NH_4OH$ nitrogen concentration to above 10 µg. at./L. These levels may have impaired the growth of

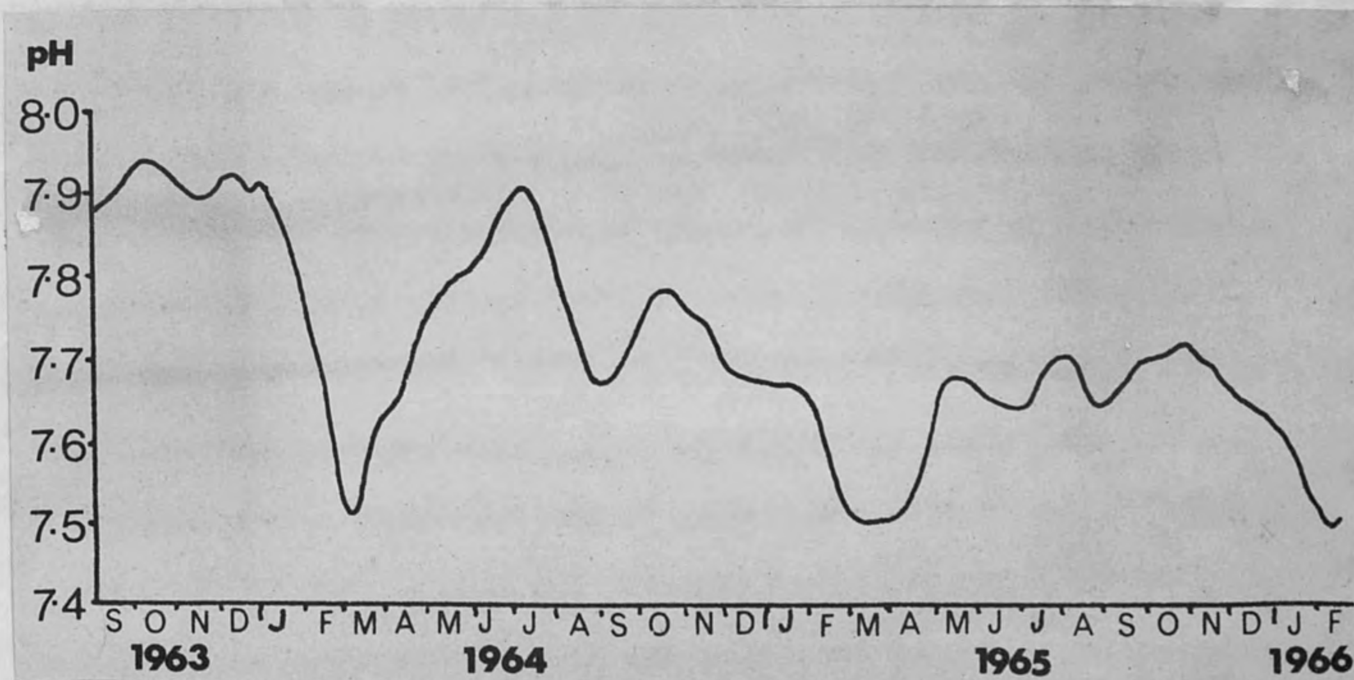



Figure 13. The pH of the water passing onto the filter beds.

certain algae present in the filter beds.

Concentrations of ammoniacal nitrogen (Figure 17) were usually lowest in the summer and autumn and highest in the winter and spring. Hutchinson (1957) states that ammoniacal nitrogen concentrations are generally at a maximum during periods of circulation and that minima occur during the winter. There were increases in concentration in the early winter and late autumn of 1963 and 1964 and during the winters of these two years distinct maxima in concentrations occurred. The winter of 1965 to 1966, however, differed from the previous two winters and was indeed unusual (Taylor, 1964) as the concentrations of ammoniacal nitrogen remained low throughout. This can be accounted for by the unusually high natural flow of the River Thames during the winter of 1965 to 1966 (Figure 7). This increased flow was associated with a reduction in ammoniacal nitrogen concentrations in the river water and this was reflected in the water passing into and out of the reservoirs. Menzel and Spaeth (1962) found, in the Sargasso Sea, that the highest concentrations of ammoniacal nitrogen occurred during and after periods of maximum phytoplankton development. This relationship did not seem to hold for the water passing out of Queen Mary Reservoir possibly because of the relatively high, compared with the Sargasso Sea, concentrations of particulate organic matter present throughout the year (Figure 14) but more probably because of the overriding influence of the River Thames ammoniacal nitrogen concentrations on the reservoir and subsequently the water passing onto the filter beds.

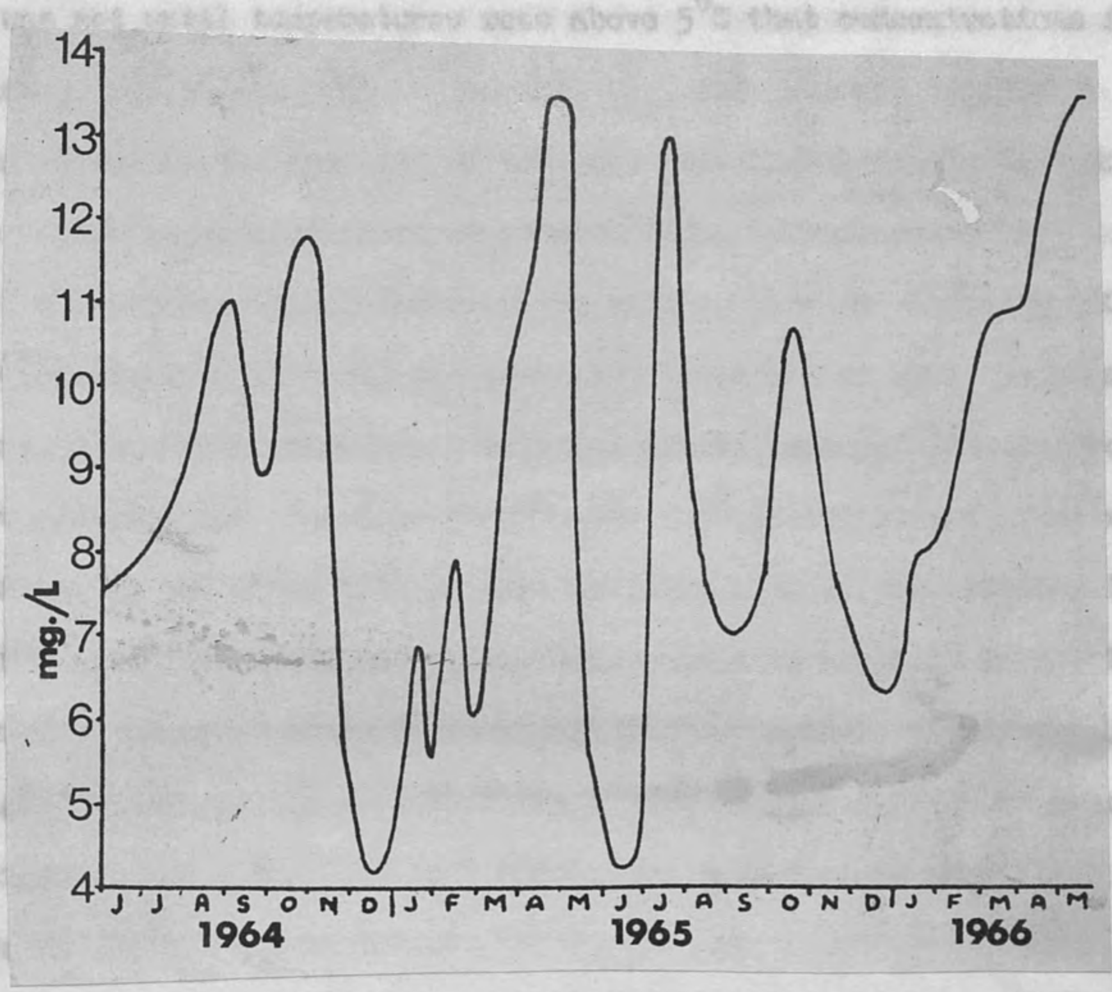



Figure 14. Concentrations of particulate organic matter present in the water passing onto the filter beds.

Nitrification in the surface waters of Lake Mendota was found to be at a maximum in April, May and October (Domogalla, Fred and Peterson, 1926). If the same seasonal changes in nitrification occurred in the filter bed water this could possibly account for the low concentrations of $NH_4$-N observed during the late spring and autumn periods. The highest concentrations of ammoniacal nitrogen occurred during the late winter and early spring of 1963 to 1964 and 1964 to 1965 when water temperatures were at their lowest (Figure 9, p. 37).

It was not until temperatures rose above 5°C that concentrations fell below 40 µg. at./L. It is possible that temperatures below 5°C greatly reduce the activity of nitrogen oxidising bacteria thus during the winter ammoniacal nitrogen remained in a reduced state.

Nitrate nitrogen concentrations were usually in the range 100 to 400 µg. at./L (Figure 17) and were many times higher than the ammoniacal nitrogen concentrations. As both the filter beds and the reservoirs have a continuous flow through of water the concentrations of nitrate nitrogen in the water will be greatly influenced by the concentrations of nitrate nitrogen in the River Thames where it was high during the period of study. Maximum concentrations occurred in the spring, slightly later in the year than the maximum concentrations of ammoniacal nitrogen. This suggests that ammonia oxidation might contribute towards the nitrate maxima.

Silicon dioxide (Silica) was present in highest concentrations during the winter and summer periods and in lowest concentrations during the spring and autumn periods. Concentrations ranged from 6 µg. mol./L. to over 300 µg. mol./L (Figure 17). Maximum concentrations did not always correspond with either high or flood rainfalls (Figure 8) as was suggested in the theory of diatom periodicity (Pearsall, 1923). In such a complex and highly eutrophic system as the River Thames -reservoirs-filter beds it is difficult to demonstrate such a simple relationship as that proposed by Pearsall (1923). Silica concentrations were indeed high during the winter when rainfall was high but increases in summer concentrations did not correspond with either high or flood rainfall. Much silica is contributed to the upper layers of Queen Mary Reservoir by partial turnovers, i.e. intermediate breakdowns of thermal stratification, in the late summer. Lowest concentrations of silica were recorded during or soon after the occurrence of maximum numbers of diatoms (see Chapter VI). As soon as these diatom populations subsided, either naturally or because of copper sulphate applications, silica concentrations increased probably due to replenishment by the inflowing water.

Although the concentrations of phosphate phosphorus were the lowest of the recorded major inorganic nutrients compared with the recorded concentrations in Lake Windermere, not more than 2 µg. P/L (0.0645 µg. at./L), Mackereth (1953), they were relatively high throughout the year, the minimum concentration being 0.8 µg. at. P/L. As it has been recorded that 1.0 µg. P/L (0.0322 µg. at./L) can produce a population of 16 x $10^6$ cells per litre of <u>Asterionella formosa</u> (Mackereth, 1953) phosphorus would not seem to have limited algal growth in Thames Valley waters. Concentrations were usually above 20 µg. at./L and ranged between 0.8 to 61.8 µg. at./L (Figure 17). Maximum concentrations were recorded during the autumns and winters of 1963 to 1964 and 1964 to 1965. Minimum concentrations were recorded in the late summer periods. The winter of 1965 to 1966 was again unusual in that high concentrations of phosphate phosphorus were not recorded (as with ammoniacal nitrogen), and levels never exceeded 30 µg. at./L. This may again be explained by the high flow of the River Thames causing considerable dilution of nutrients. Although rapid fluctuations in concentration did not occur as much as with nitrate nitrogen or silica this did not necessarily indicate a less rapid turn over of phosphorus. Rigler (1964), using radioactive $P_{35}$, found that, although gross weekly concentrations of phosphorus were more or less constant, the inorganic phosphorus was being re-cycled in less than ten minutes in many lakes. Thus even though weekly records of concentrations did not indicate it, rapid utilisation and re-cycling of phosphorus might have been taking place.

At the end of September 1964, after phosphate phosphorus was at a minimum, a fifty-fold increase in concentration was recorded, (Figure 17). This increase was observed to occur within two weeks and about a month after the autumn recirculation in the reservoir (Taylor, 1963). The recorded increase in concentration was probably not entirely due to this autumn recirculation, no such rapid increase was observed in 1963 and only a slight increase was observed in late August 1965 (from 18 - 28 µg. at./L). A small increase in phosphorus did occur in the River Thames in 1964 just before this period but this would not have contributed more than 10 µg. at./L to the reservoir increase in concentration. There are many records of high phosphorus release during vigorous phytoplankton growth or in dying populations (Antia et al. 1963; Watt & Hayes 1963). During the period of phosphate phosphorus increase there were large populations of several algae actively growing in the water, e.g. Rhodomonas minuta, Scenedesmus spp. and Oocystis spp.. One population of Stephanodiscus astraea, previously present in high numbers but then declining, was also present.

Johannes (1964) found that Acnanthes subhyalina could release 80% of the dissolved organic phosphorus found in solution during sixteen days in culture. He did not state whether this release was from living or dead cells. If this organic phosphate release was coupled with mineralisation by phosphatases as indicated by Johannes (1964) the algal populations present could have contributed to the observed increase although the amount involved would only have represented a small proportion of the total. Rigler (1961) also found that phosphate replenishment by algal decomposition and remineralisation were negligible in lake water. Heron (1961) found a marked correlation between rainfall and surface water phosphorus. No such marked correlation was observed during this study in either the river, the reservoir or the filter bed waters.

Determinations were made on the phosphate phosphorus content of rain water. The mean was found to be 2.15 µg. at./L P which would have had a diluting effect rather than increasing the phosphorus concentration in the Thames Valley waters. If a similar level of rainfall-phosphate had been obtained in the Lake District the reverse effect would be expected.

Concentrations of phosphorus were observed to fall in the spring at about the same time as did those of silica. That phosphorus removal did not occur to any great extent before decreases in silica concentration agrees with the results of Heron (1961) for the English Lake District. There is a correlation between the increase in diatoms and the decrease in phosphorus concentrations in the spring (see

Chapter VI). This is in agreement with Mackereth (1953) who suggested that in phosphorus-rich waters, cells of *Asterionella formosa* remove large amounts of phosphorus from the water to maintain a high cell phosphorus content.

The results for the dissolved oxygen percentage saturation are given in Figure 15. The most obvious regular feature was the marked depletion of oxygen in the autumn months and throughout the winter periods. The onset of each of these periods of depletion coincides with the autumn turnover times (indicated by arrows in Figure 15) in the reservoir. The destratification of the reservoir could cause a redistribution of dissolved and particulate matter from the hypolimnion and mud surface. This could have a two fold effect on the water. Firstly there would be a redistribution of organic and inorganic nutrients possibly leading to an increase in the algal and bacterial populations. Secondly reduced sustances such as ammoniacal nitrogen and hydrogen sulphide would be redistributed throughout the water depth. These would exhibit a marked chemical oxygen demand on the water. If the growth of mainly non-photosynthetic bacteria greatly exceeded algal growth, or if the chemical demand on the water were high enough, oxygen consumption would have been in excess of production resulting in an overall undersaturation. Figure 16 gives the colony counts (*E.coli* and 37 Agar plate colony counts per ml. obtained from the Metropolitan Water Board) in the outlet water of Queen Mary Reservoir. Higher numbers were present throughout the autumn and winter periods of each year corresponding to periods of undersaturation

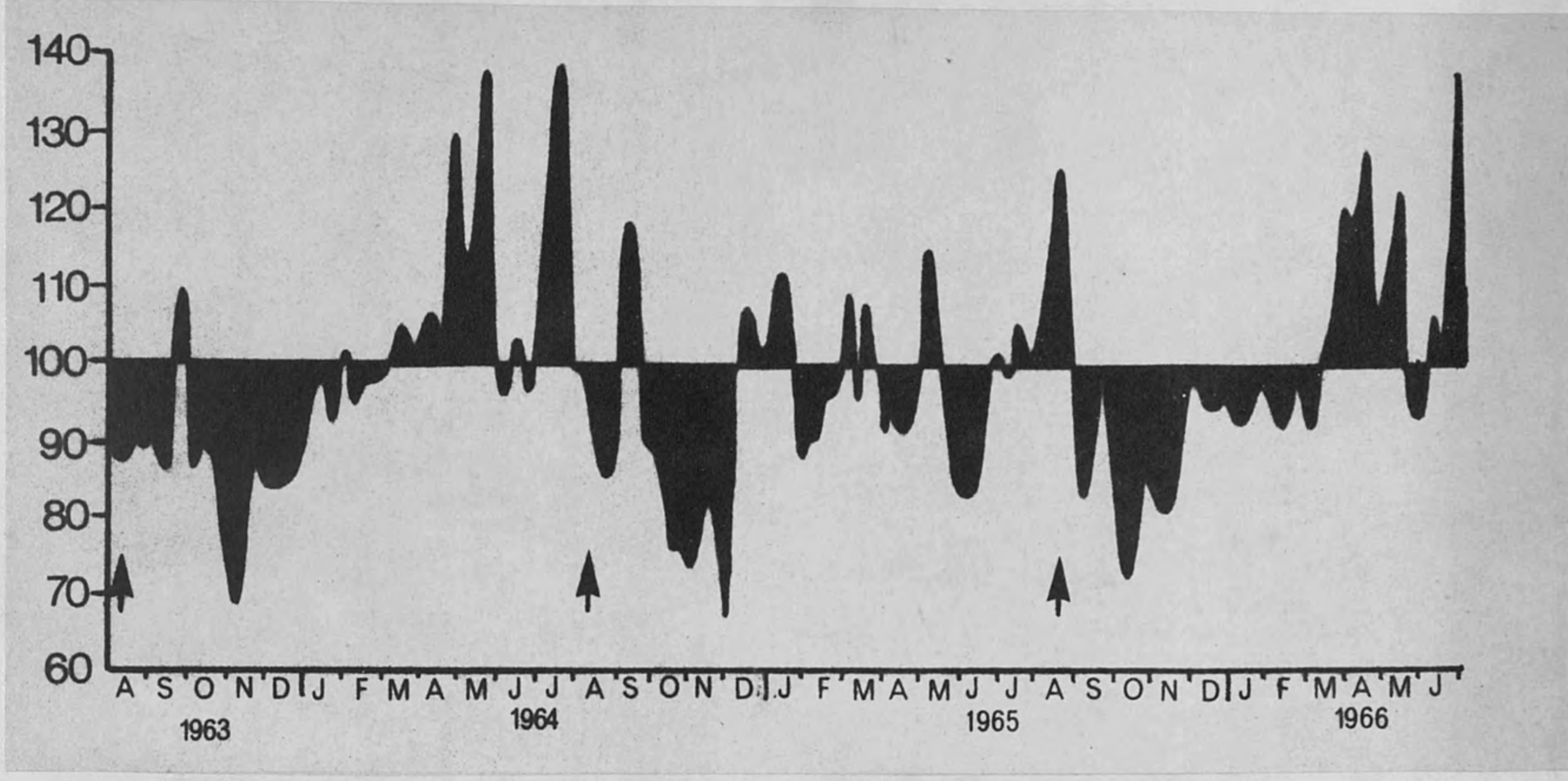



**Figure 15.** The percentage saturation of dissolved oxygen in the water leaving Queen Mary reservoir during the period of study. The vertical arrows represent the times of the autumn turnover in the reservoir.

in the water. There were also large numbers of bacteria present in June 1964. This was also reflected by a reduction in the dissolved oxygen. Kuznetzow & Karsinkin (1931) working on Lake Glubokoye estimated that a bacterial population of $2 \times 10^6$ cells/L would consume 0.24 mg./L oxygen per day. This was considered to be a conservative estimate by these authors as populations of up to $4.5 \times 10^6$ cells/L had been found in the hypolimnion. This rate of consumption, were it to have occurred, would seem to be adequate to explain the observed reduced concentrations of oxygen. Periods of supersaturation followed a less regular pattern. Supersaturation of the water did occur when large numbers of algae were present (see Chapter VI) although the two occurrences could not always be correlated. In April, May and September 1964 large numbers of algae were present in the water (mainly Stephanodiscus astraea, Asterionella formosa, Rhodomonas minuta and Oocystis spp.) and the water was supersaturated with oxygen. In June 1964 Oocystis spp. and Ankistrodesmus falcatus were present in large numbers byt the oxygen content of the water was between 4% above and 5% below saturation. Either the algae present were not photosynthesising at a maximum rate or the oxygen consumption of other matter in the water exceeded the production by the algae. No marked correlation between the percentage saturation of dissolved oxygen and rainfall was observed.

The nutrient concentrations in the water passing out of the reservoir onto the filter beds did bear some relation to the populations of algae present in the reservoir. It was, however, difficult to show

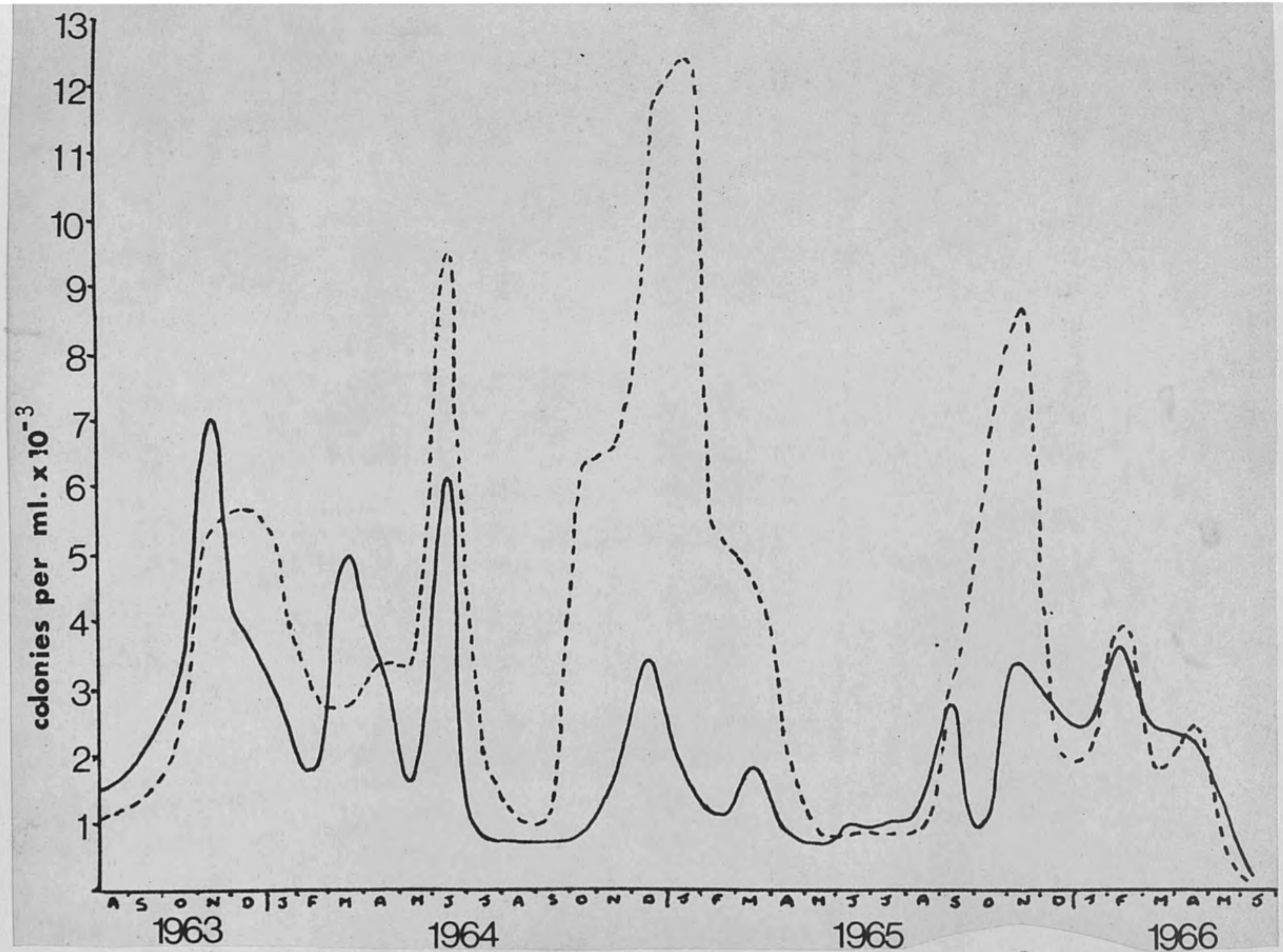
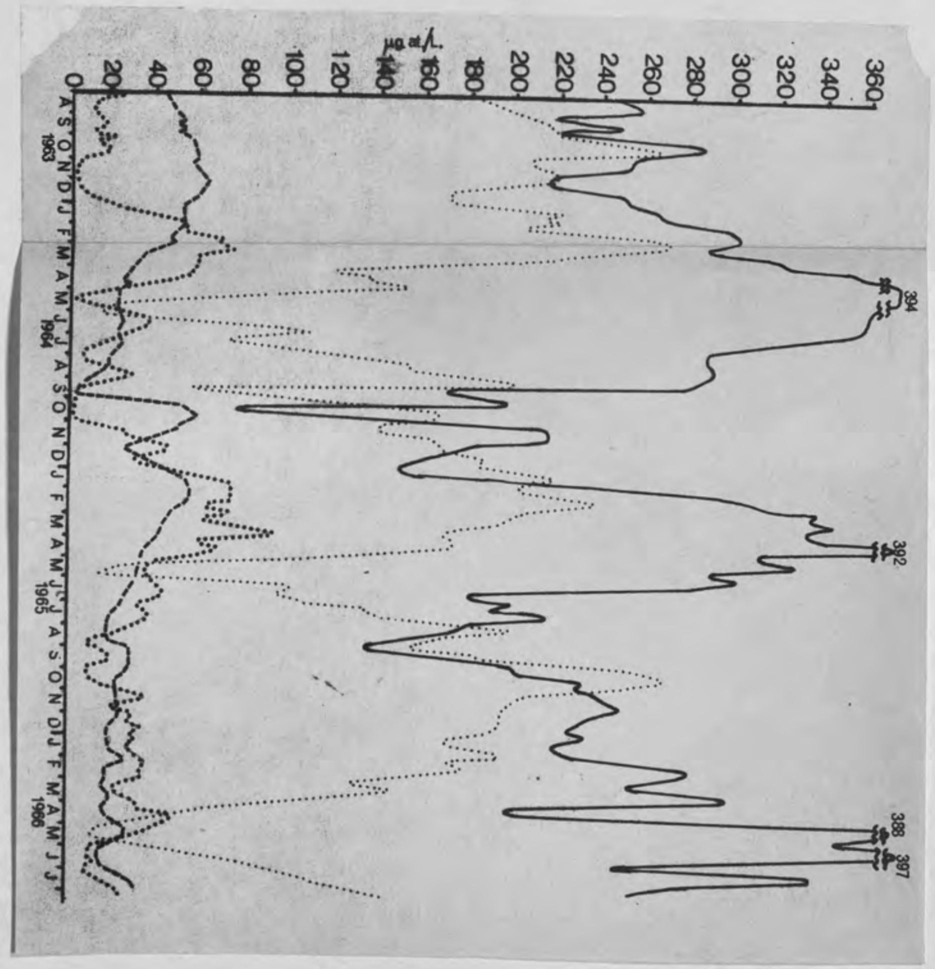



**Figure 16.** The colony counts for E. coli ( —— ) and the 37°C Agar plate ( - - - - - ) /ml. in the water leaving Queen Mary reservoir.

any such relationship with the algal populations within individual filter beds. There are two main possible effects of these nutrient concentrations on the algae. Firstly they could affect the physiological state of the incoming algae and possibly determine whether or not they reproduce in the filter beds. Secondly, nutrient concentrations in the inflowing water may have been low enough to limit the growth of algae in the filter beds. It has been found, for example, that the minimum requirements of <u>Asterionella formosa</u> for phosphorus is 0.002 µg. at./L at a concentration of $10^6$ cells/L. (Mackereth, 1953) and that 0.5 µg.mol./L silicate was the lower limit for growth (Jorgensen, 1957). Filter beds have a constant flow of water through them. At an average rate of flow through the sand of four inches an hour, each square centimetre of sand surface would have 240mils. of water passing it per day, i.e. approximately 1L of water passes every 4 $cms.^2$ of sand surface per day. At the minimum concentrations recorded in the filter bed supernatant waters (see Figure 17) this would have made available 0.8 µg.at./L P and 6.0 µg.Mol./L SiO. This concentration of PO4-P is capable of supporting a population of 100 x $10^6$ $cells/cm.^2$ of <u>A.formosa</u> (Mackereth, 1953) and the concentration of SiO2 was twleve times the recorded minimum concentration required for growth. Total population densities on the sand surface reached 2 x $10^6$ $cells/cm.^2$ at the end of a filter bed run (see Chapter VII). It seems unlikely, then, that nutrient concentrations were limiting except perhaps towards the end of a filter bed run when local cell concentrations may have exceeded the recorded averages for the sand surface (see Chapter VII D). Nutrient concentrations were measured on occasions in the water passing out of the filter beds but due to re-solution of chemicals on the way through the sand column these were found to bear no relationship to the concentrations of nutrients in the supernatant water or of algae on the sand surface.

Figure 17. The concentrations in ug.at./L. of $NO_3$-N —— , $NH_4$-N ···· , $SiO_2$ ····· , and $PO_4$-P ----
in the water entering the filter beds,



## VI. The Algae of the inflow water.

Observations were made throughout the period August 1963 to June 1966 on the numbers of algae in the aeration basin water. The water passing out of the reservoir goes through the aeration basins before entering the rotary microstrainers. (See Chapter II). Although the concentrations of algae present in the aeration basin water broadly reflected the concentrations of those present in the reservoir they were not always precisely similar. This was because water was abstracted from the reservoir at one or more different depths at different times of the year depending upon the quality of the water for waterworks purposes. The changes in the abstraction depth was under the control of the waterworks engineers and often no prior knowledge of such changes was obtained. The sample from the aeration basin did not, therefore, always form a representative sample of the reservoir and changes in the abstraction level could cause a change in the phytoplankton composition of the aeration basin water. The results obtained are expressed as total volumes of cells/ml. x $10^{-3}\mu$ (or weights in $\mu g \times 10^{3}$) and are given in Figures 18 and 19.

There was a fairly well marked seasonal sequence of diatoms in the spring and autumn and Chlorophyta and Xanthophyta in the summer and autumn. This sequence was often modified by applications of copper sulphate to the reservoir and, depending upon environmental conditions, the growth of different species often overlapped. Copper sulphate was usually applied at the tip of the baffle or to the area east of the baffle (see Chapter II) at a final concentration of between 0.5 and 1.0 mg./L $CuSO_4$. The times of applications are shown as vertical arrows on Figures 18 and 19. Of the algae recorded in the samples, only thirteen genera or species occurred in high concentrations (calculated volume = 100 x $10^3 \mu$ /ml.) and only these will be considered in detail.

Stephanodiscus astraea was the most abundant diatom recorded, being present throughout the entire period of study. It was not reported as a major constituent of the River Thames phytoplankton by Rice in 1938 and is possibly mainly a lacustrine form. Although applications of copper sulphate often modified the extent of its growth, it developed a spring and an autumn maximum each year, the spring being larger than the autumn maximum. Of the major dissolved inorganic nutrients only silica concentrations showed a close relationship to the amounts of S. astraea present. The decrease in cell numbers, after the spring maximum, started as silica concentrations fell to a minimum (Figure 17). It is probable that the true growth picture had been masked by copper sulphate applications and this, not nutrient depletion, caused the decline of the spring maximum. Pearsall et al. (1946) reported that S. astraea could increase even when the concentration of silica was below 0.5 p.p.m. (8.35 µg. mol./L). The relationship between autumn concentrations of silica and algal biomass was not so distinct and might have been masked at times by recirculation of silica during the autumn overturn.

Stephanodiscus hantzschii has been recorded in the River Thames and other rivers as one of the major constituents of the phytoplankton

(Rice, 1938; Swale, 1962). Its occurrence in the Queen Mary Reservoir coincided with large populations being present in the River Thames. Rice (1938) reported that it produced only a summer maximum. Although there was a summer maximum in 1964, and it was absent for the rest of the year, there was a spring and an autumn maximum in 1965 with no real summer maximum. Cell concentrations decreased after September 1965 but small numbers persisted throughout the winter, increasing in 1966 to produce a spring maximum in March. Although there was a correlation between silica concentrations and the spring maximum of S. hantzschii, this was not so apparent for the summer and autumn populations. There was, for example, an increase in cell numbers in May 1964 when silica concentrations were as low as 8 µg. mol./L (0.5 mg./L.) The winter population of 1965 was most probably limited by low light levels and low temperatures.

Asterionella formosa was present in the River Thames during the winter and spring periods from August 1963 to December 1965. It has been recorded as a common constituent in larger standing bodies of water (Lund, 1949; Reynolds & Taylor, 1950; Taylor, 1954.) It developed a spring maximum in each of the years studied and also a small maximum in the late autumn of 1965 in the Queen Mary Reservoir. Lund (1964) states that the growth of Asterionella is limited by chemical conditions in the summer and by physical conditions in the winter. It was found that the decline of the spring population of 1964 to 1965 occurred while silica concentrations were decreasing but before they were near to their reported limiting levels. This decrease in numbers before silica limitation was also reported by Pearsall et al. (1946). The spring maximum in 1966 did not decrease until silica concentrations were below 20 µg. mol./L (1.3 mg./L). There was then a catastrophic decrease in numbers. The autumn population of 1965 did not completely die out during the winter, indeed growth increased from December onwards to produce a larger spring maximum than in previous years. Light and temperature were not significantly higher during the winter of 1965 to 1966 but did increase more rapidly in February 1966 than in previous years. The larger spring maximum in 1966 can be attributed to the persistance of larger numbers of cells through the previous winter and, to some extent, to slightly more favourable physical conditions towards the end of that winter. Throughout the period of study some chytrid infected cells were observed but these formed only a small proportion of the total population. It is doubtful that parasitism was an important factor during this period in limiting Asterionella populations.

Fragilaria crotonensis was present during the autumn of 1965 and 1965 and during the spring of 1966 having persisted through the previous winter. The onset of the autumn maxima corresponded with higher silica and lower nitrate nitrogen concentrations. Coincident with the autumn maximum of 1964 was a rapid rise in phosphate phosphorus concentrations but these did not fluctuate greatly during the growths of autumn 1965 and spring 1966. Lund (1964) pointed out that, as the growth rates of F. crotonensis and Asterionella formosa were very similar in the spring period, one would expect these two diatoms to compete very closely. In the autumn of 1965 such competition did occur and _F. crotonensis_ produced a larger, earlier, maximum than did _A. formosa_. ( Lund (1964) also stated that the reason that they did not usually compete in the spring is that the numbers of _Fragilaria_ cells usually decreased greatly during the winter whereas cell numbers of _Asterionella_ did not. This means that _Asterionella_ would have a better start in the subsequent spring. Although such a decrease in numbers of _Fragilaria_ did occur in Queen Mary Reservoir during the winters of 1963 to 1964 and 1964 to 1965 the autumn population persisted throughout the winter of 1965 to 1966. In the spring of 1966 _Fragilaria_ was able to compete very closely with, and indeed reached its maximum before, _Asterionella_. The spring maximum produced by Asterionella was, however, larger and longer lasting. The early decline of the _Fragilaria_ populations was probably due to the repeated copper sulphate applications during March 1966. This also retarded the growth of _Asterionella_ but it later recovered.

The only other diatom to occur regularly in relatively high concentrations in the reservoir phytoplankton during the period of study was _Synedra ulna_. It produced regular spring and autumn maxima. The occurrence of regular double maxima was reported by Rice (1938) in the River Thames. Taylor (1964) reported that it was abundant in the spring diatom growth in Saddington Reservoir, Leicestershire. It was, however, less abundant than the other diatoms mentioned from Queen Mary Reservoir and only contributed at a maximum $220 \times 10^3$ $\mu^3$/ml. to the total calculated algal volume.

Tribonema bombycinum (see Taxonomic Notes) occurred during the summer and autumn periods. It produced only a small maximum in 1964 but in 1965 it was present from May until October reaching concentrations of over 3,000 x $10^3$ $\mu^3$/ml. Its maxima corresponded to periods of lower nitrate nitrogen and phosphate phosphorus concentrations and it might have been able to utilise or take up these elements more efficiently than other algae present at the same time. T. bombycinum was present at times of high solar radiation and high temperatures but these do not seem to be requirements for its growth as it has been frequently observed as abundant in winter populations in other local reservoirs (personal observation).

Rhodomonas minuta and several species of Cryptomonas, which have been grouped together for convenience, were the main members of the Cryptophyceae present. Of these R. minuta was the most abundant and was present throughout most of the period. Lund (1962) reported that this alga was common in many bodies of water and recorded its periodicity in certain lakes of the English Lake District. He reported that it was least common in the winter period. This was true in Queen Mary Reservoir although a relatively large population persisted throughout the winter of 1964 to 1965. Maximum concentrations generally occurred in April and May (occasionally reaching 6.0 x $10^3$ cells per ml.). This was followed by a marked decrease in numbers in early summer, as in Blelham Tarn and Esthwaite Water (Lund, 1962). After this fall there was an increase to give a smaller autumn maximum. Some of the fluctuations in its growth could be attributed to applications of copper sulphate but these effects did not seem to be long lasting and the populations soon recovered. There did not appear to be any correlation between the fluctuations and dissolved nitrate nitrogen, ammoniacal nitrogen, phosphate phosphorus and silica concentrations. Although light might seem to be a limiting factor during the winter period (Lund, 1962), there was no more light during the winter of 1964 to 1965 when the population persisted than in 1965 to 1966 when it died out. Temperatures were a little higher in the winter of 1964 to 1965.

The numbers of Cryptomonas spp. fluctuated greatly throughout the period. These fluctuations did not seem to have any direct correlation with changes in concentrations of major dissolved nutrients or with physical conditions. Little can be said about the possible reasons for these fluctuations and as was said by Lund (1962) the ecology of Cryptomonads is a mystery.

Five members of the Chlorophyceae, three Chlorococcales and two Volvocales, occurred as important constituents of the phytoplankton. The two volvocaleans were Carteria quadrangulata and several species of Chlamydomonas which, as they occurred as mixed populations, have been grouped together.

Carteria quadrangulata occurred in large numbers on two occasions in 1964. In September a small population developed which died out by the end of the month. In November and December a larger population developed and formed the major part of the non-diatom phytoplankton at that time. There does not seem to be any correlation between these growths and dissolved nutrients. The growth of C. quadrangulata does not seem to be dependent on light and temperature levels.

Chlamydomonas spp. were present mainly in the spring and autumn. The spring maxima decreased rapidly in April and May. In 1964 the population recovered and produced a second smaller maximum in July. An autumn maximum developed in 1963 and 1964 but not in 1965 possibly because of repeated copper sulphate applications in the summer of 1965, although on other occasions (e.g. June, 1964 and March, 1966) this did not appear to affect the populations. As with some other algae, cell numbers of Chlamydomonas spp. were lower during periods of lowest light and temperature. They were also lower or completely absent during periods of lowest nitrate nitrogen concentrations.

Three chlorococcalean genera were present, each being represented by at least two species which, as they occurred together have been grouped under the generic name.

Oocystis spp. (mainly O. elliptica and O. solitaria) were present throughout the early summer until the winter of each year studied. There was usually a maximum in July with a second later in the autumn. There were lower concentrations of cells present during 1965 and this may be attributed to repeated copper sulphate applications that year. The early summer populations arose after the decline of the main spring diatom growths and coincided with periods of high solar radiation and high water temperatures. The autumn maximum, however, coincided with periods of lower light and lower temperatures indicating that neither of these factors was limiting even at their lower autumn levels. The spring increase occurred after a period of copper sulphate application. Maloney and Palmer (1956) reported that over 2.0 mg./L was required to control certain species of Oocystis indicating that it may be resistant to smaller or single applications at the concentrations used. Repeated applications, however, may have had an adverse effect (e.g. low numbers in 1965).

Ankistrodesmus spp. (A. falcatus and A. pseudomirabilis) did not contribute as large a proportion of the phytoplankton volume as did Oocystis spp. They were present mainly during the spring period producing a maximum in March and April. In the winter of 1963 to 1964 a population arose which produced a maximum in January. This population persisted at low concentrations throughout February to produce the 1964 spring maximum in April. A smaller summer population then developed after the spring maximum had declined. As with Oocystis spp., Ankistrodesmus spp. have been reported to be able to tolerate higher concentrations of copper sulphate (8 mg./L) and their growth does not always seem to be affected by the concentrations applied. The growth period of Ankistrodesmus spp. coincided with the spring diatom growths but tended to be subsidiary to them. As it was smaller in size Ankistrodesmus was found to pass through the rotary microstrainers onto slow sand filters in large quantities.

Scenedesmus spp. were present during the autumn months of 1963 and 1964 and also during the winter of 1963 to 1964. The main autumn growth period occurred during periods of high light and temperature and population maxima corresponded to times of low nitrate nitrogen and ammoniacal nitrogen concentrations. S. quadricauda is able to grow under conditions of very low nitrogen and phosphorus concentrations by utilising its cell resources, (Rodhe, 1948). This would favour the growth of Scenedesmus during periods of intense competition for nutrients.

From the aeration basin the water passes through rotary microstrainers before flowing into the slow sand filter beds. The construction of rotary microstrainers has been explained in Chapter II and their effect on algal populations described by Ridley (1967), Bellinger (1968) and in the 41st annual Report of the Metropolitan Water Board. Larger algae (above 45u in their minimum dimension) are strained out from the water. The removal of smaller algae (below 45u in minimum dimension) depends upon their concentration in the water and whether or not they form colonies. Asterionella colonies are usually 100% removed as are Fragilaria and various filamentous species. Stephanodiscus astraea and Scenedesmus spp. are removed if cell concentrations are high enough to clog the straining fabric or if colonial forms produce a secondary filtering mesh inside the drum. Small algae such as Stephanodiscus hantzschii and Rhodomonas minuta are only removed if the straining fabric has been blocked by other algae. The overall effect of the rotary microstrainers is to allow most of the small algae to pass onto the slow sand filters but the larger algae may be removed in varying amounts. The actual concentrations of algae in the reservoir do not, therefore, necessarily have any bearing on the numbers passing onto the slow sand filters.

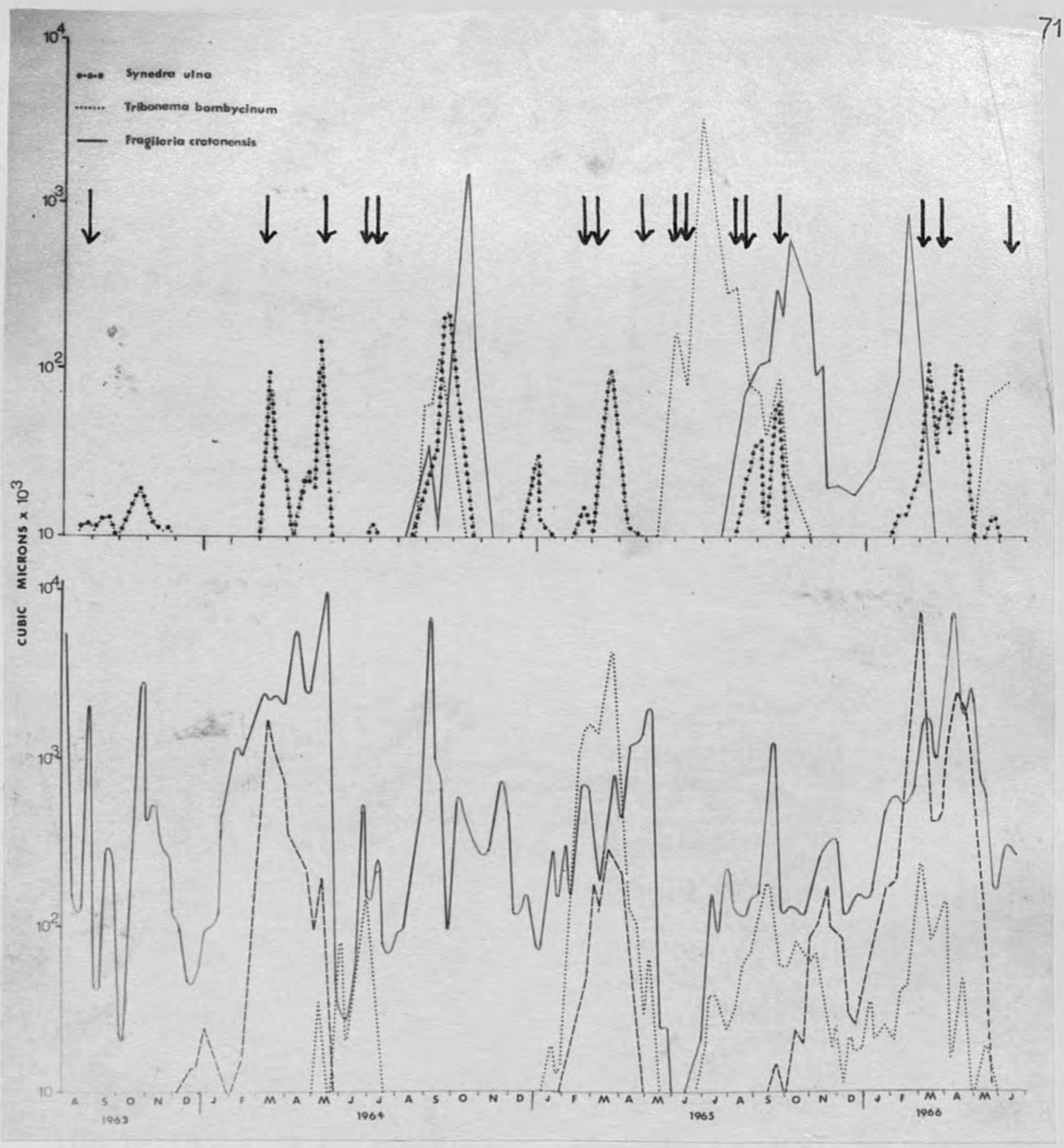



Figure 18. The periodicity of the algae passing out of the reservoir. The vertical arrows represent times of copper sulphate application to the reservoir. Chlorine was applied to the reservoir outlet pipe during the spring and late summer periods. Key to lower figure; —— = Stephanodiscus astraea, ..... = S, hantzschii, ----- = Asterionella formosa.

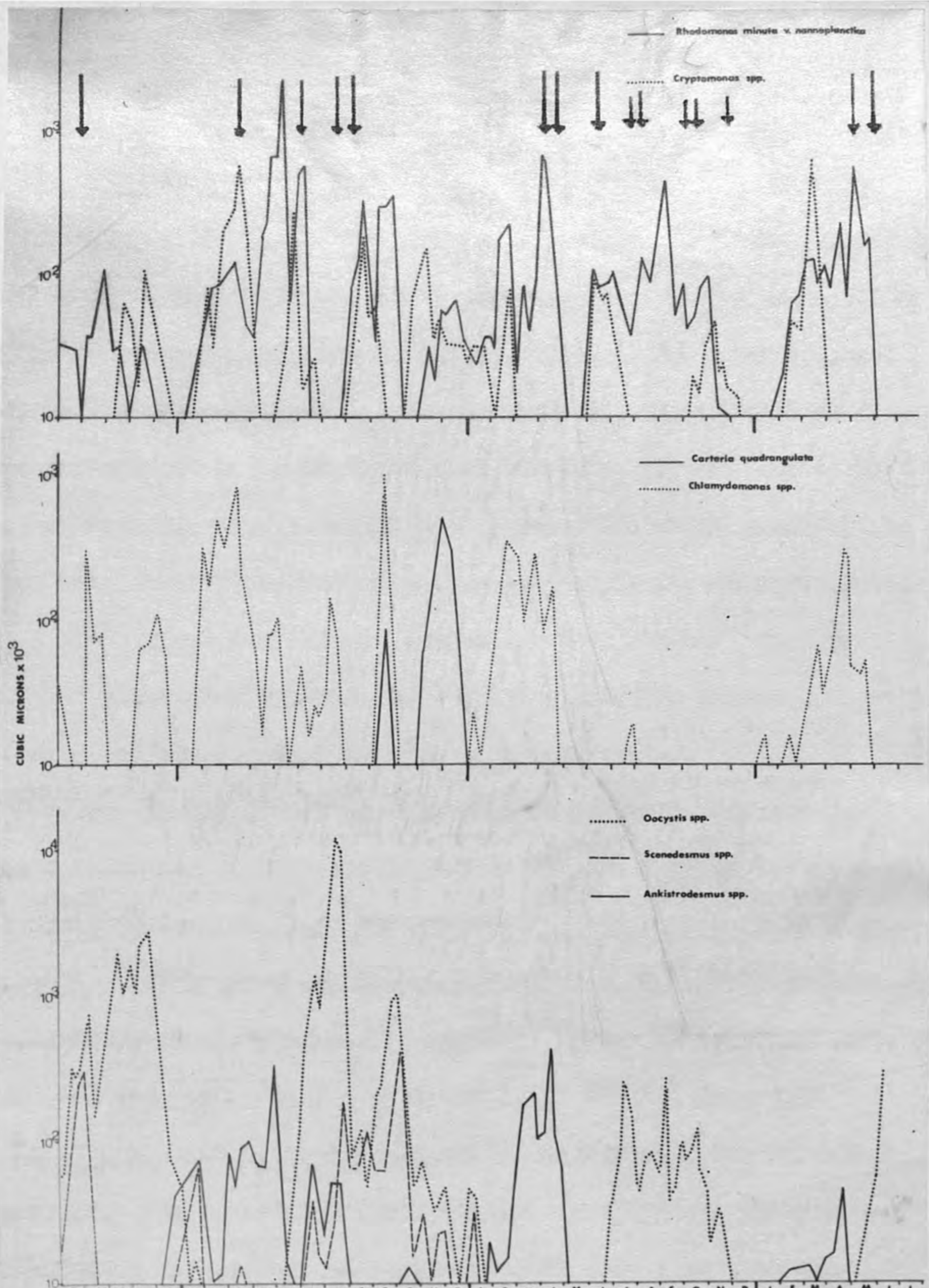



Figure 19. The periodicity of the algae passing out of the reservoir. The vertical arrows represent times of copper sulphate application to the reservoir. Chlorine was applied to the reservoir outlet pipe during the spring and late summer periods.

VII. A. The Algae of the supernatant water.

The algal flora leaving Queen Mary Reservoir was greatly modified by the rotary microstrainers before passing onto the slow sand filter beds (see Chapter II and also Bellinger, 1968). In spite of these modifications, however, for a large part of the year, the algal flora of the supernatant water of the filter beds was very similar to that of the reservoir. The rotary microstrainers did allow smaller species to pass onto the filter beds in preference to the larger ones (Bellinger, 1968). Throughout most of the period of study the cell numbers of all species of algae passing from the reservoir onto the filter beds were reduced by between 5% and 95%.

Certain species present in the filter beds were observed to increase in number in the supernatant water merely by accumulation and not by active division, e.g. Asterionella formosa and Stephanodiscus hantzschii. Other species, although present in but small numbers in the reservoir water, were observed to actively proliferate in the slow sand filter bed basins producing large local populations, e.g. Scenedesmus spp. and Chlamydomonas sp. The algae in the former category, i.e. those which did not actively proliferate but merely accumulated in number, are not dealt with in detail in this present discussion as their periodicity was very similar to that of the inflow water (see Chapter VI and Figures 18 & 19, pages 71 & 72 ). These species generally increased in number throughout the run of a filter bed at a rate depending upon cell concentrations in the inflow water. These species were also only recorded in the filter bed at times when they were also present in the reservoir. The species which proliferated in the filter beds will be dealt with in detail.

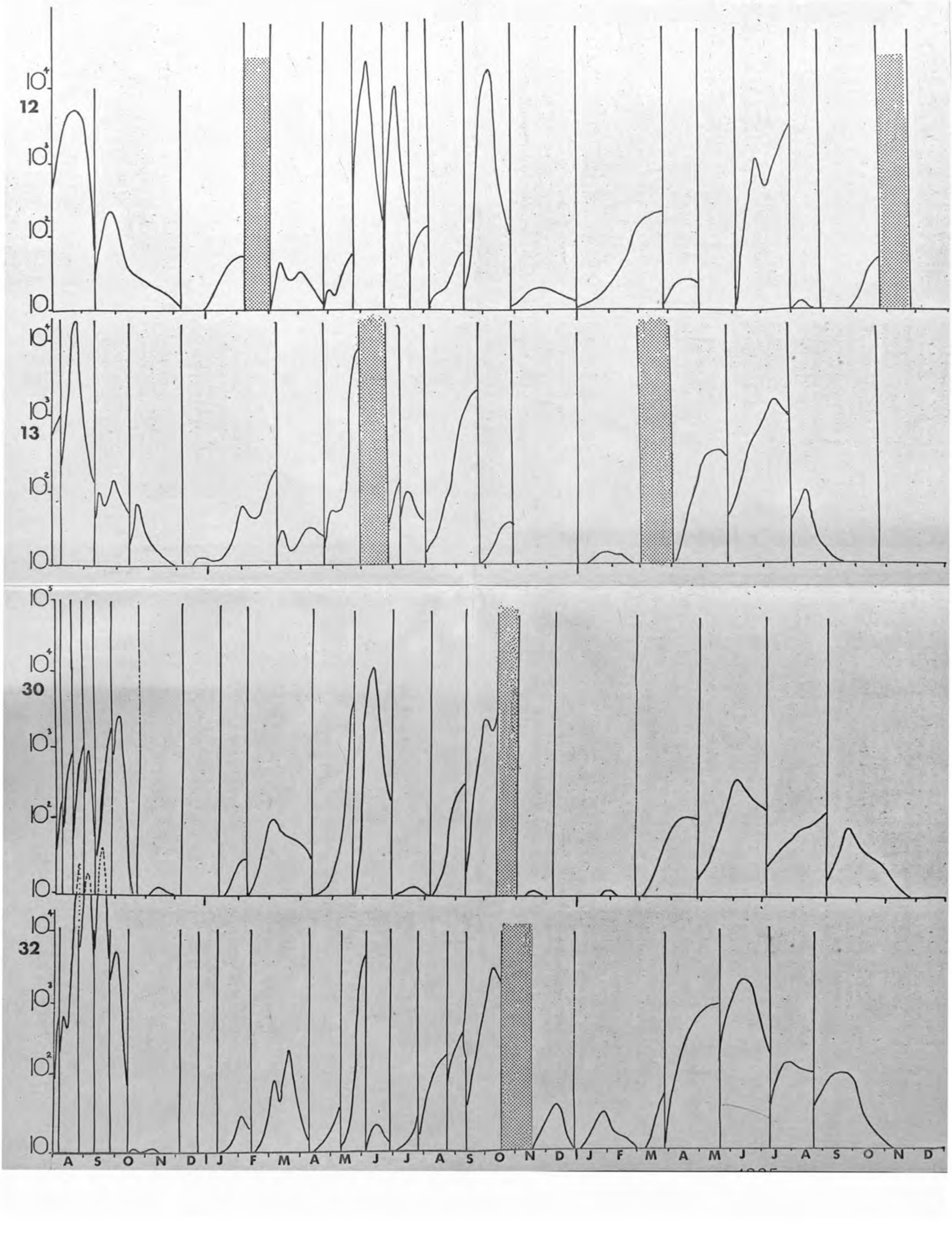
Species of Chlamydomonas and Carteria quadrangulata were present throughout the period of study in mixed populations. On most occasions Carteria quadrangulata formed only a small percentage of the mixed population and so, for the purposes of this study, it has been grouped together with Chlamydomonas.

Although present throughout most of the period of study, Chlamydomonas was most abundant during the late spring and summer of each year (see Figure 20). Its periodicity was similar in each of the four filter beds studied. The exception to this was in the autumn of 1963 when beds 30 and 32 were being cleaned by the "hydra". As this method of cleaning did not involve draining the filter bed down, the supernatant water was not completely replaced at any one time. The algae were thus able to grow undisturbed for a much longer period and, especially in bed 32, produce consistently high cell concentrations.

Rapid decreases in cell numbers were observed on several occasions, e.g. October 1963, June 1964 and June 1965 in bed 30. These decreases occurred after a period of calm warm weather when the algal populations in the supernatant water became stratified (see page 87 ). With the onset of more windy weather the stratification was broken down, the population dispersed and there was an overall decrease in cell numbers.

Oocystis spp. were common in the supernatant water of the filter

**Figure 20.** The periodicity of *Chlamydomonas spp.* in the filter bed supernatant water, The results are expressed as volumes of algae in $\mu^3 \times 10^3$.

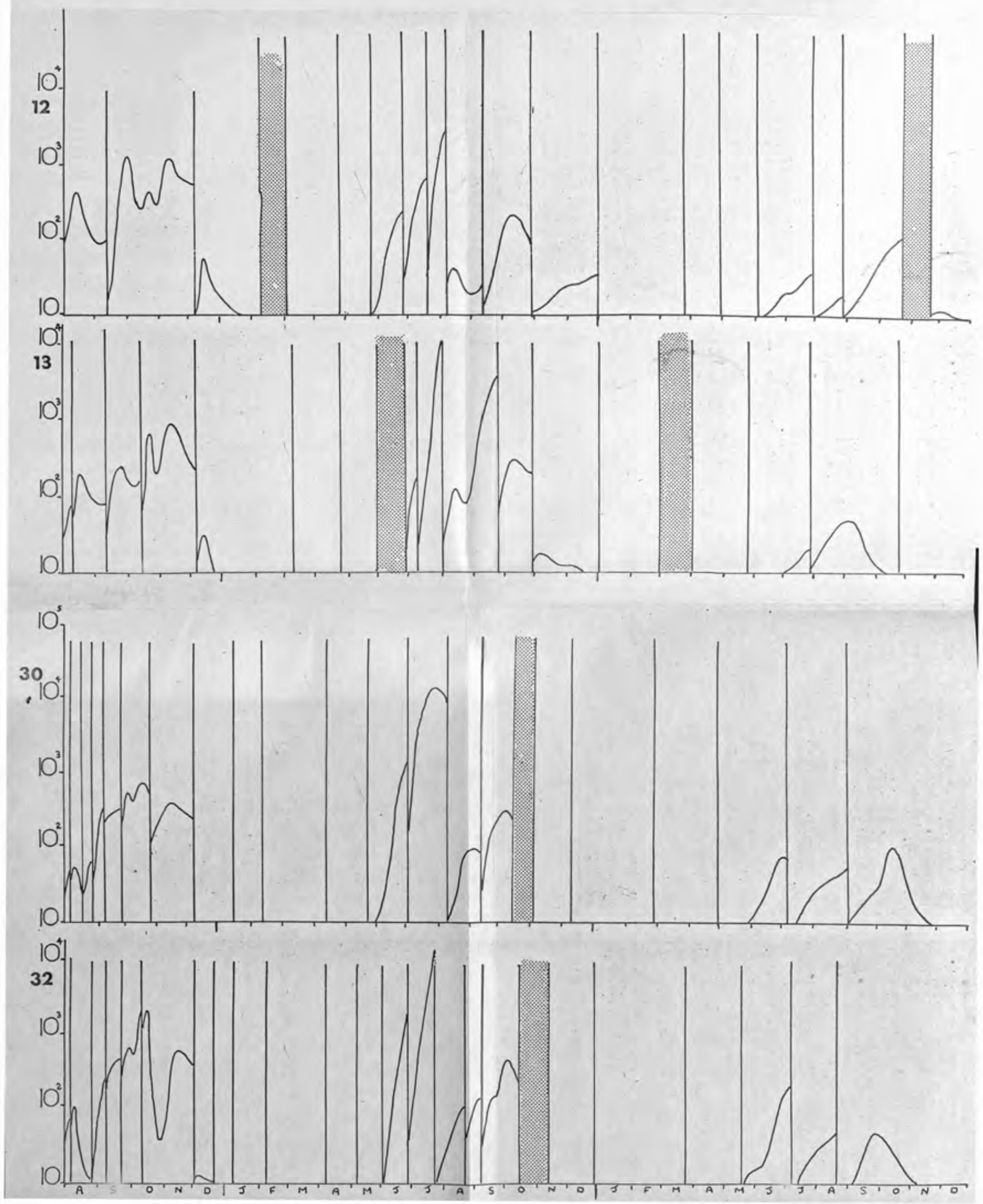

12
13
30
32
10
10²
10³
10⁴
10⁵
A S O N D J F M A M J J A S O N D J F M A M J J A S O N D

beds at times when they were abundant in the reservoir. Many cells were, however, observed to be actively dividing in the filter bed water. Oocystis spp. were most common during the summer and autumn periods (see Figure 21). When large populations of planktonic crustaceans were present Oocystis spp. were observed to decrease because of grazing by these animals. Cells of green algae, probably Oocystis, were observed in the guts of Daphnia pulex and Chironomid larvae (see also page 133 ).

Most of the other species of algae recorded occurred only infrequently in high concentrations in the supernatant water. Many were also present for short periods, e.g. bottom living forms were occasionally swept up into the supernatant water by wind induced turbulence. These, however, soon sedimented out again as soon as conditions became calm. The highest concentrations of algae, other than those normally present in high concentrations or of Chlamydomonas or Oocystis, occurred during the period March 1964 to May 1965. Both before and after this period there were but occasional small growths of algae.

Rhodomonas minuta reached its highest concentrations during the spring months (Figure 22). Larger populations developed in beds 30 and 32 than in beds 12 and 13. It was absent from all four filter beds during November and December 1964 even though relatively large numbers of cells (over 50 per ml.) were present in the inflow water. As the major nutrient concentrations were similar in the filter bed water and in the reservoir water it would seem probable that physical

Figure 21. The periodicity of *Oocystis spp.* in the filter bed supernatant water. The results are expressed as volumes of algae in $\mu^3 \times 10^3$.


12
13
30
32
$10^5$
$10^4$
$10^3$
$10^2$
10
A S O N D J F M A M J J A S O N D J F M A M J J A S O N D

conditions were limiting the growth of *Rhodomonas minuta* in the filter beds. Both light and temperature were also similar in both the reservoir and filter bed waters. Water turbulence was, however, much higher in the filter bed supernatant water. The turbulence was also of a slightly different character, the water being more choppy in the filter beds.

*Cryptomonas spp.* occurred in all four filter beds but at different times of the year (Figure 22). During March 1964 it was present in all four filter beds although in all but bed 13 numbers were low. In bed 12 it did not then reoccur until May 1965. In bed 13, however, a large population developed in late June 1964 but no cells were recorded from then until the end of the period. In June 1964 in bed 32 and in February and May 1965 in beds 30 and 32 small populations of *Cryptomonas spp.* developed. As with *Rhodomonas minuta* in Queen Mary Reservoir, there did not seem to be any correlation between the growth of *Cryptomonas spp.* and either chemical or physical conditions.

*Tribonema bombycinum* occurred in all four filter beds during June and July 1964 but only when it was abundant in the reservoir. This species was effectively removed from the incoming water by the rotary microstrainers (Bellinger, 1968) so that large numbers of cells did not pass onto the filter beds. The cells which were present in the supernatant water did appear to be dividing and the populations increased. Growth could not have been very active, however, as the filter bed populations were never large and only persisted while populations were present in the reservoir water.

Three species of diatoms were commonly present in the filter bed supernatant water. All three were normally abundant in the epipelic or attached populations on the sand surface. Their presence in the supernatant water was probably due to water turbulence which prevented them from sedimenting out. All three species were most abundant at times of fairly high winds. *Cymbella spp.* occurred most frequently throughout the period (Figure 22) although it was absent from all four filter beds during the winter period. Large populations were present in beds 12 and 30 but not in beds 13 and 32, during the late spring, the summer of 1964 and the spring of 1965. *Surirella ovata* was present in all four filter beds in large numbers in the spring of 1964 but much smaller amounts in the spring of 1965. It too was absent throughout the winter period. As with *Surirella ovata*, *Nitzschia linearis* was also more common during the spring of 1964 than that of 1965. Cell concentrations were also much higher in bed 32 than the other filter beds.

When the weather was calm there was very little water movement in the supernatant water. Because of their higher specific gravity, diatoms would tend to sediment out under such calm conditions. An example of such sedimentation can be seen in the third week in March 1964. During this week calm conditions prevailed and there was a general decrease in all species of diatoms in the supernatant water. When filter beds 30 and 32 were cleaned using the "hydra", large numbers of diatoms were resuspended from the sand surface into the supernatant water. If conditions were calm after cleaning these

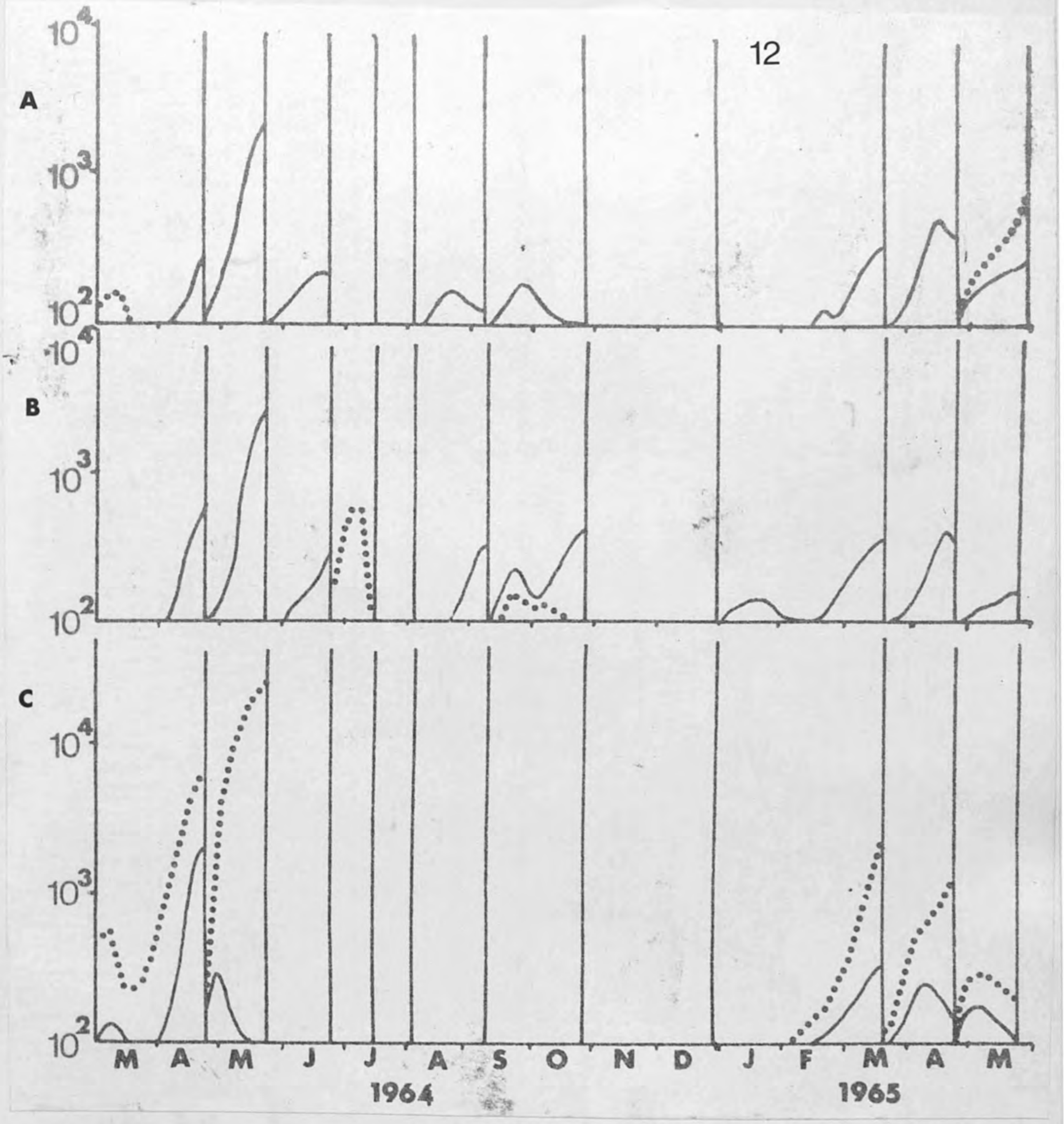



Figure 22. The periodicity of algae in the supernatant water of bed 12. Key; A··· = Cryptomonas spp. —— = Rhodomonas minuta. B··· = Tribonema bombycinum —— = Cymbella spp. C ··· Surirella ovata. —— = Nitzschia linearis.

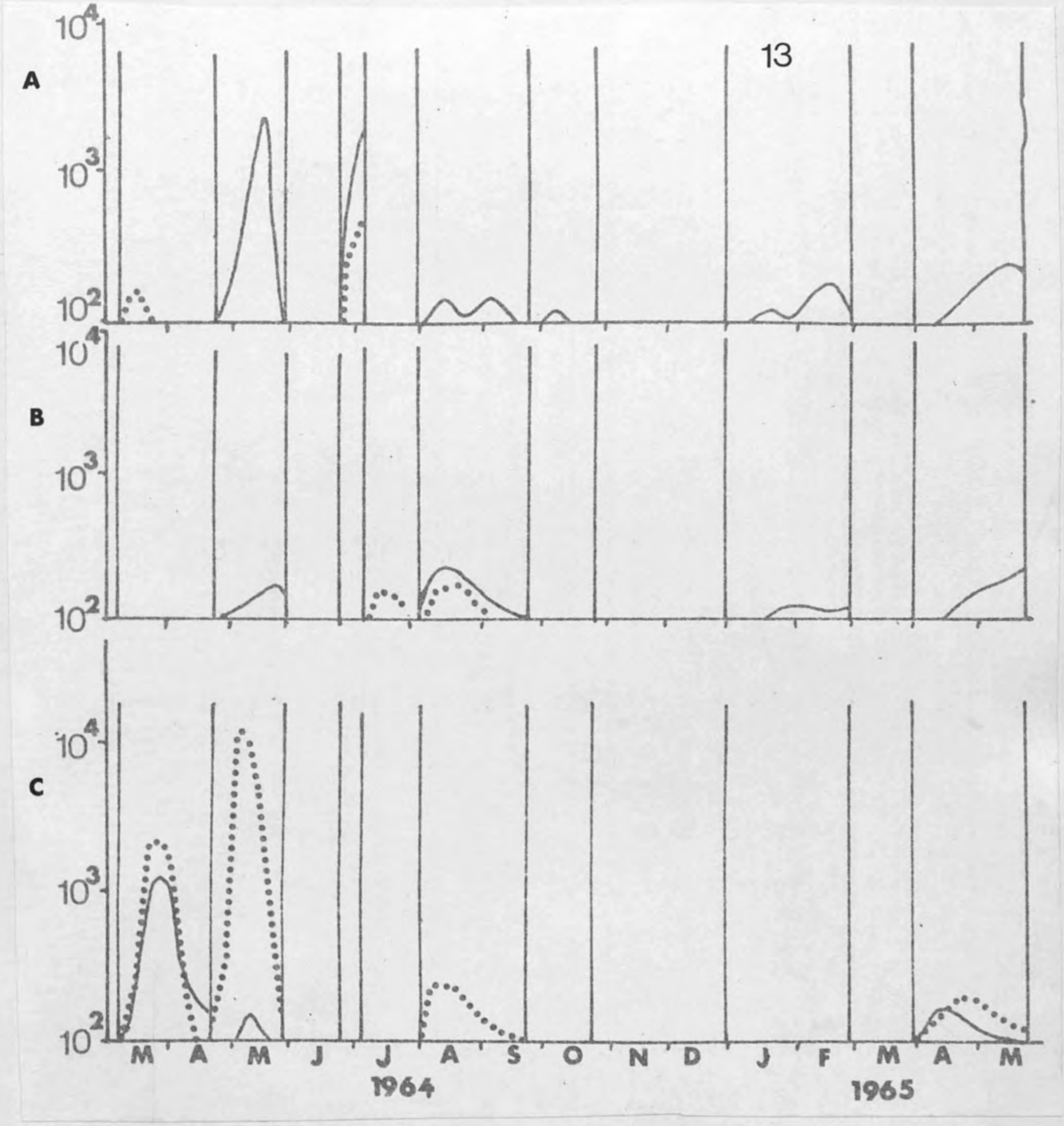



Figure 22. The periodicity of algae in the supernatant water of bed 13.

Key; A ••• = Cryptomonas spp. —— = Rhodomonas minuta. B ••• = Tribonema bombycinum — =Cymbella spp. C ••• =Surirella ovata — = Nitzschia linearis

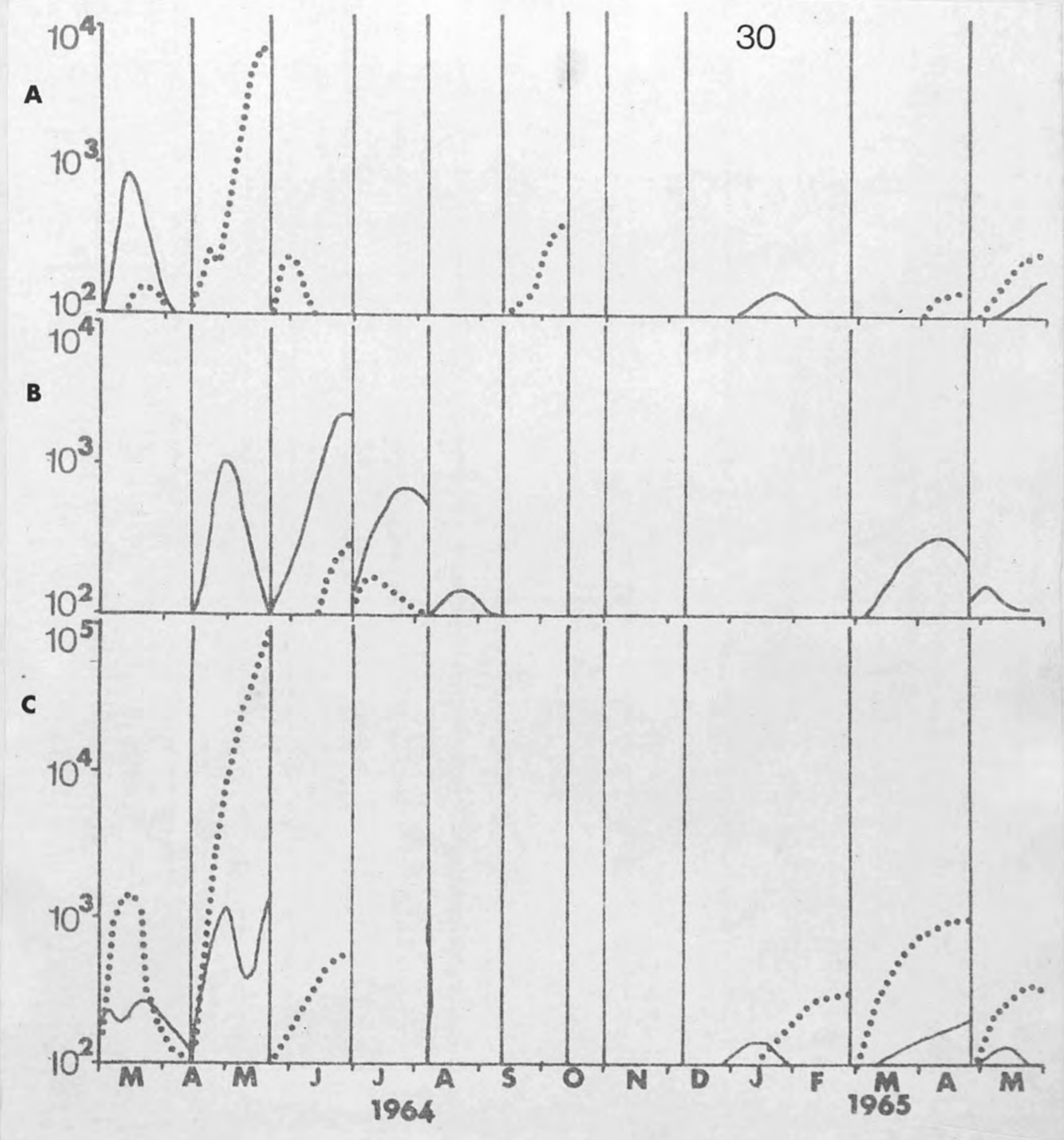



Figure 22. The periodicity of algae in the supernatant water of bed 30.

Key; A = Cryptomonas spp. = Rhodomonas minuta. B =Tribonema bombycinum = Cymbella spp. C = Surirella ovata = Nitzschia lineari s

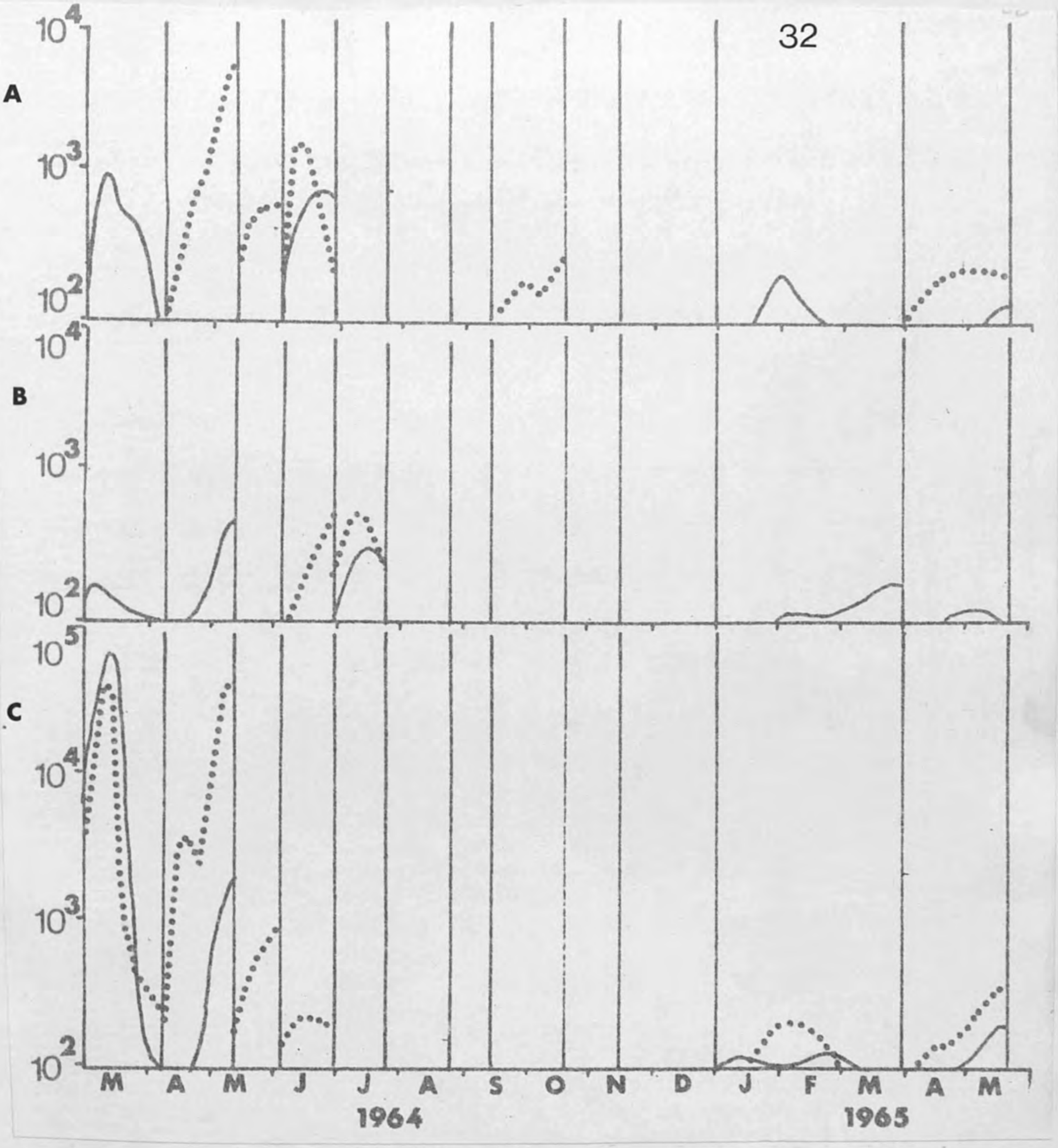



Figure 22. The periodicity of algae in the supernatant water of bed 32. Key; A = Cryptomonas spp. = Rhodomonas minuta. B = Tribonema bombycinum = Cymbella spp. C = Surirella ovata = Nitzschia linearis.

diatoms soon settled out again onto the sand surface. One example of such an occurrence is given in Table 3.

Table 3. The numbers of cells/ml. in the supernatant water of filter bed 32 two days after "Hydra" cleaning.

| Species | Cells per ml. Immediately after cleaning | Cells per ml. 2 days after cleaning |
|---|---|---|
| Melosira varians | 8532 | 2074 |
| Nitzschia palea | 455 | 290 |
| N. linearis | 42000 | 6210 |
| Amphora ovalis | 1860 | 208 |

Each of the filter beds received water from the same source and hence received a similar supply of nutrients. The physical conditions in each filter bed, with the exception of the period of "hydra" cleaning, were also similar. It is difficult, therefore, to explain the floristic difference of the various filter beds. One possible cause could be variations in the numbers of animals present, but these were only observed to differ greatly on one or two occasions and these were often when the algal populations of the filter beds were similar. The main factor of variation between different filter beds was that they were cleaned at different times. This would mean that the seeding populations at the start of each filter bed run would be different. The length of the filter bed run and also the species present determined the depth to which these species penetrated through the sand surface (see Chapter 10). This depth of penetration together with the efficiency of cleaning, would determine how much of a seeding population was carried over from one filter bed run to the next. Chance also plays a part, as, if a few cells are present in the water, any given species may be completely eliminated by the grazing of animals before it has had time to reproduce (see page 108). Because of these variable factors one would expect, therefore, a certain amount of variation in the floras of different filter beds.

On several occasions samples were collected at intervals of one foot (30.5 cm.) through the depth of the filter bed supernatant water. The results of these collections are given in Figures 23 - 25. On many occasions a definite stratification of algae was recorded. These occasions were during the spring, summer or autumn months when calmer conditions prevailed. During the winter months no stratification was observed.

Two main types of stratification were observed. The first was when motile algae were present and these were able to maintain themselves in a position most favourable to growth. Two of the occasions on which this type of stratification occurred are given in Figure 23. On the first Chlamydomonas sp. and on the second Carteria quadrangulata were present in large numbers. On July 12th 1965 Chlamydomonas sp. was present in concentrations exceeding $10^8 \mu^3$/ml. Weather conditions were calm over this period. Cell concentrations were found to be highest at the surface, decreasing towards the bottom. It should be pointed out that on this occasion not all of the cells at the surface were

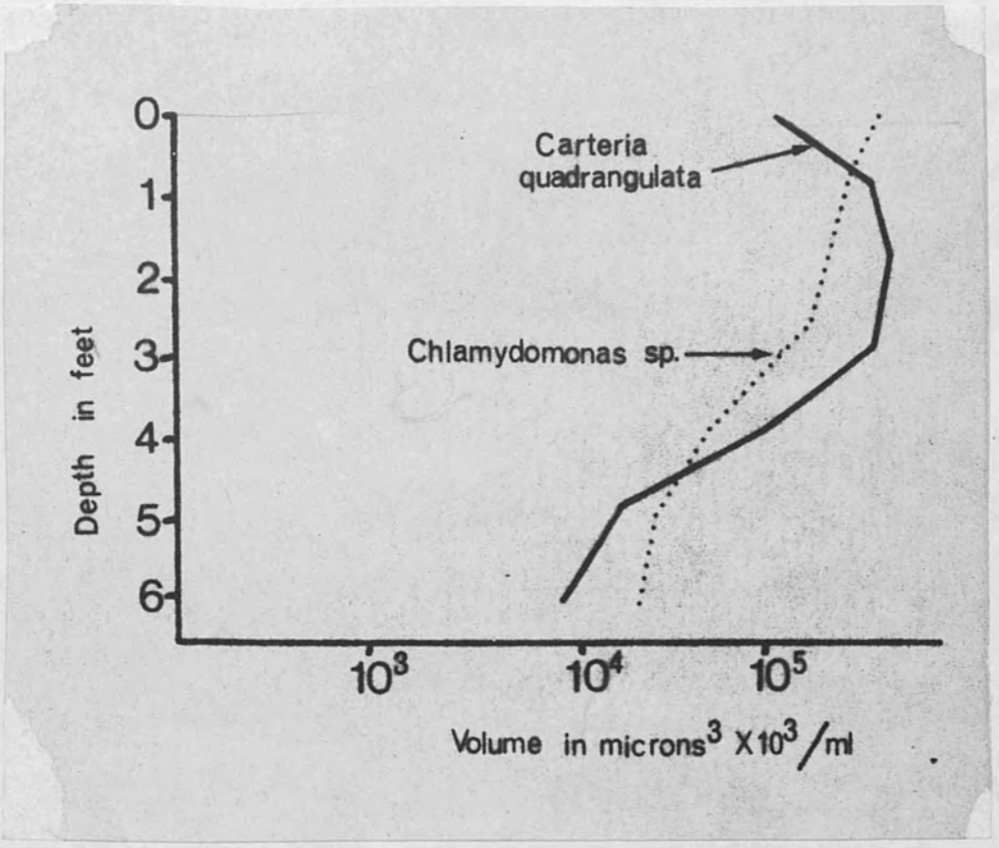


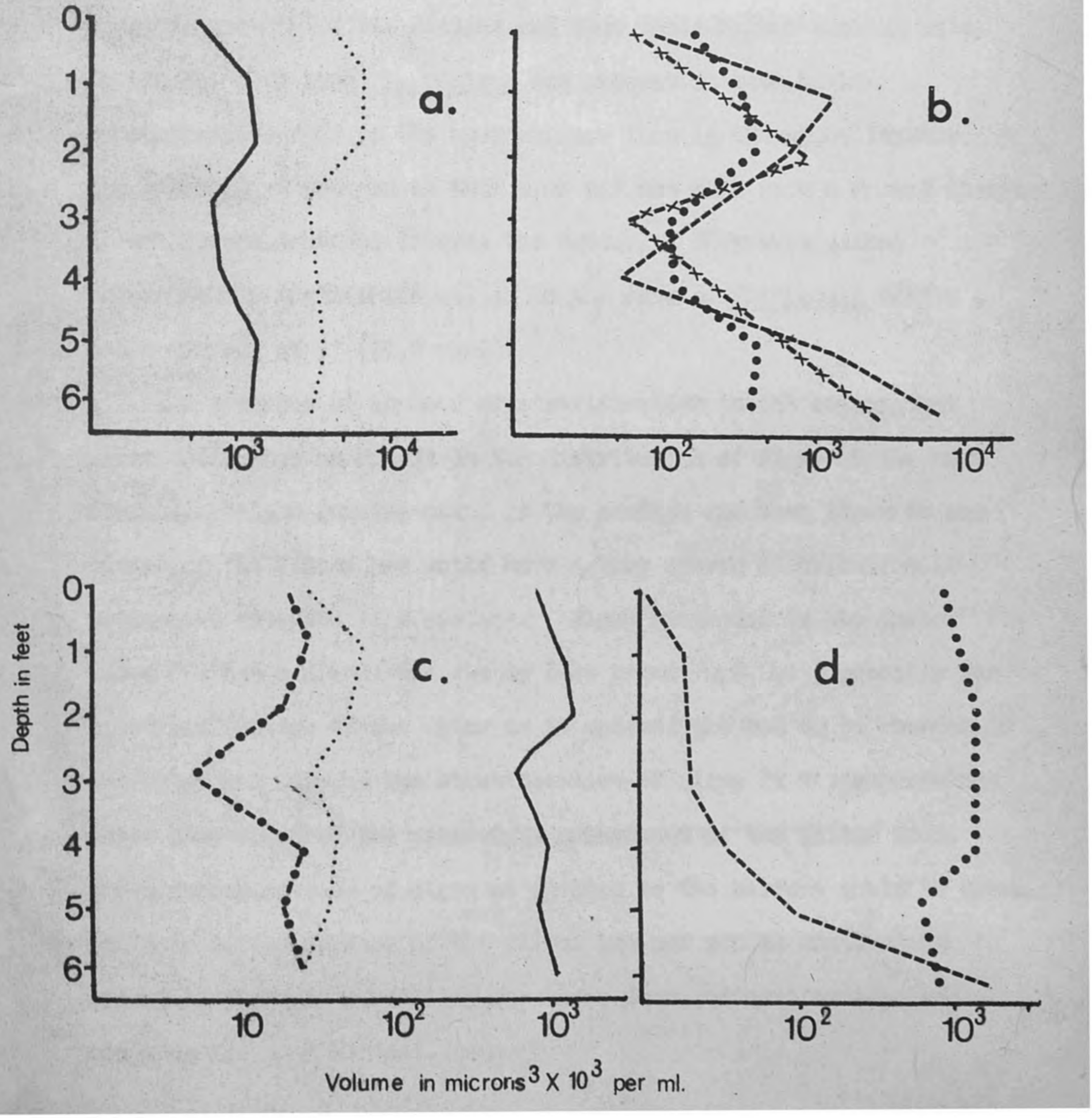

Figure 23. The stratification of Chlamydomonas sp. in July 1965 and of Carteria quadrangulata in September 1965 in the filter bed supernatant water.

motile but some were floating aggregations of cells in the palmelloid stage. On some occasions, when driven by a gentle breeze to one corner of a filter bed, these aggregations of palmelloid Chlamydomonas have been observed to form a scum at the surface more than a centimetre deep. Carteria quadrangulata was present in similar concentrations, exceeding $10^8\mu^3$/ml., in September 1965. Again a distinct stratification occurred but the highest densities occurred between 1-3' (30.5-91.5 cm.) and the lowest at the surface and just above the sand surface. Carteria was never observed to form a palmelloid stage nor to scum at the surface. Motile algae have been reported to avoid the actual water surface possibly due to light inhibition (Goldman et al. 1963). In September and October 1964, Chlamydomonas sp. was present in high concentrations but weather conditions were less favourable and wind induced water turbulence tended to prevent any algal stratification at that time. On October 1st, 1964, a stratified population of Rhodomonas minuta was present (Figure 24). Highest concentrations occurred between o' & 2' (0 - 61.0 cm.) and 4' & 6' (122.0 - 183.0 cm.) There was a distinct drop in concentration around 3' (91.5 cm.) The presence of high numbers near to the surface can be explained by photosensitive movement and the fact that the cells can actively move. Less active and moribund cells would tend to sediment and form high concentrations near to the bottom. The large reduction in cell numbers at 3' (91.5 cm.) might have been merely a reflection of these two processes but as this pattern of stratification also occurred with non-motile as well as motile forms other factors may have been involved.

On September 14th, 1965, a similar pattern of stratification was observed of the diatoms Melosira varians, Stephanodiscus astraea and Nitzschia linearis (see Figure 24). Each of these diatoms exhibited the same sort of depth concentration profile. Minimum concentrations occurred at the sand surface and between 3' & 4' (91.5 - 122 cms.) and maximum concentrations between 1' & 2' (30.5 - 61 cms.) and just above the surface of the sand. Of these diatoms only Nitzschia linearis is capable of active movement and then only of gliding over a surface and not swimming through the water. High numbers near to the sand surface would be expected with non-motile forms due to sedimentation. As there is no possibility of active movement other factors must be involved to account for the high concentrations around 1' - 2'. One possible explanation is the existence of a buoyancy mechanism. Melosira varians has been observed on other occasions to produce gas bubbles during active photosynthesis and so be buoyed up from the sand surface into the supernatant water (see pages 133 ). This mechanism may have been in operation for all three species of diatom enabling them to maintain themselves in the most favourable light climate. An alternative explanation to the buoyancy-sedimentation hypothesis for the paucity of cells at 3' - 4' involves the physical stratification of the water. This could have maintained the inflow water, containing much lower densities of algae, at the 3' - 4' level, as distinct layers.

When the weather conditions were calm after a period of strong

Figure 24 The stratification of algae in the supernatant water of the filter beds, Key; ······ = Chlamydomonas sp. —— = Oocystis spp. • • • • = Stephanodiscus astraea ×-×- = Nitzschia linearis - - - = Melosira varians •-•- = Rhodomonas minuta



winds and water turbulence, as on October 9th 1963, a typical sedimentation profile was observed. Cell concentrations in the surface waters were low but near to the sand they were high (see Figure 25). This type of profile was more marked in the diatoms Melosira varians and Stephanodiscus astraea than in the green algae Coelastrum microporum or Oocystis spp. possibly because of the higher specific gravity of the diatoms and thus their higher sinking rate. On October 14th 1964, M. varians was present in much higher concentrations near to the sand surface than in the upper layers. The other algae present at that time did not show such a marked increase in cell concentrations towards the bottom. They were either homogeneously distributed or, as in the case of Rhodomonas minuta, had a minimum at 3' (91.6 cms.)

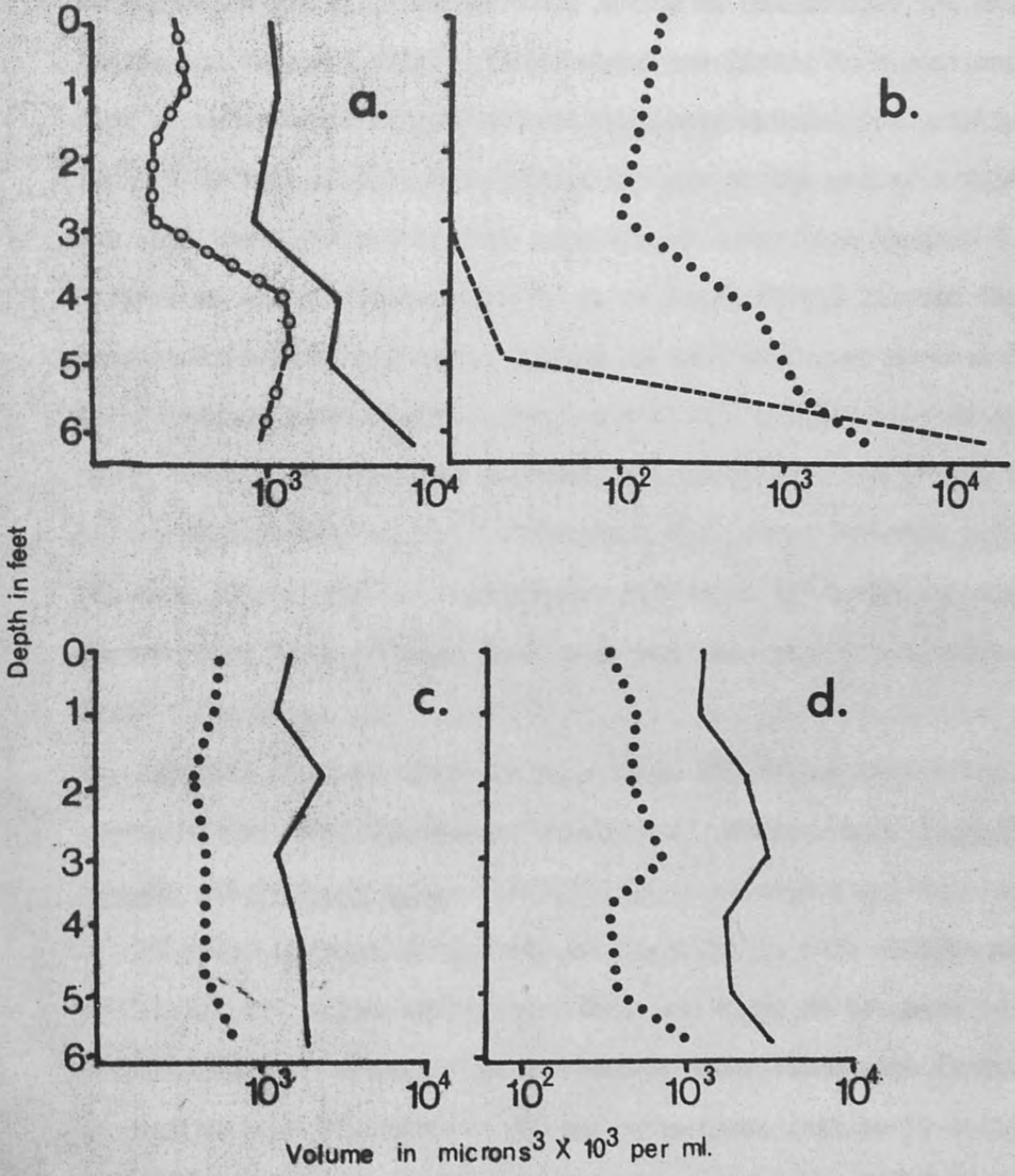
The presence or absence of stratification in the supernatant water could have an effect on the distribution of algae on the sand surface. Algae forming scums at the surface and then blown to one corner of the filter bed would have a very uneven distribution if sedimented onto the sand surface. Algae suspended in the inflow water did not sediment out evenly (see pages 142-148) possibly due to stratification of the water as it entered the bed or to changes in the water velocity. The stratification of algae in the supernatant water also affected the waterworks management of the filter beds. Large concentrations of algae at or near to the surface could be drawn off near to the surface of the filter bed and run to waste along overspill channels preventing the algae from sedimenting onto and clogging the sand surface.

Figure 25. The stratification of algae in the supernatant water.

Key: a. o-o-o Coelastrum microporum, —— Oocystis spp.

b. •••• Stephanodiscus astraea, --- Melosira varians.

c and d. •••• S. astraea, —— Oocystis spp.



B. The unattached bottom living forms.

The unattached bottom living, or epipelic, algal population consists of two main components, (i) non-motile forms resting on the sand surface and (ii) motile forms living on and amongst the sand grains and zoogleal film. These algae are living in a continuous flow of water containing dissolved chemicals and are not usually limited by lack of nutrients except perhaps at the end of a filter bed run when the algal populations become very large (see Chapter V.) Because of the continuous flow of water Brook (1954) likened the filter beds to slow flowing rivers. Light is another factor essential for the growth of these algae. They were always covered with water to a depth of 1.5 - 1.75 m. and received only about 30 - 35% of the incident radiation (see Chapter V.) Throughout the period November to February of each year photosynthetic radiation levels at the sand surface were below 35 cals./cm.$^2$/min. and were possibly limiting for some of the algae.

Species of algae normally planktonic in habitat were often found surviving and even reproducing on the sand surface, e.g. Stephanodiscus astraea and Oocystis spp. Although these algae are not true members of the epipelic population, they are included in this section as they are living and competing for nutrients and light in the same environmental situation. In the inflow water there were fluctuations in cell numbers (see Chapter VI) and when these fell to 10 cells/ml. there were no measurable increases in concentration on the sand surface. This indicates that increases on the sand surface of planktonic algae such as Asterionella formosa were largely, if not entirely, due to accumulation rather than growth. This conclusion was confirmed by microscopical observations (when few or no division stages were found). Stephanodiscus astraea was present throughout the entire period in varying concentrations (Figure 26.) Although the maximum concentrations on the sand surface corresponded to periods of maximum growth in the reservoir, the length of the filter bed run also played an important part in its increase. As this alga was present all the time in the inflow water, the longer the filter bed was kept in operation, the longer the period for the accumulation of algae on the sand surface. If all of the algae survived, maximum numbers would be reached at the end of the filter bed run. This was found to be often, though not always, true for S. astraea. In October and November 1963 the living cell concentrations decreased towards the ends of the filter bed runs in beds 12 and 30. This could be correlated with large growths of Melosira varians and Ulothrix tenuissima on the sand surface. (see Figure 26.) Filaments of these algae tended to grow up into the supernatant water causing considerable light cut off to the underlying algae. The reduction in numbers in S. astraea at these times was possibly due to light limitation. Decreases in cell concentrations also occurred towards the end of filter bed runs in May and June 1964 in beds 12, 13 and 30. This was also possibly due to light limitation. In these cases the shading was caused by large populations of Chlamydomonas spp. in the supernatant water (see Figure 20). These populations of Chlamydomonas spp. were capable of causing considerable light attenuation (see page 45 ). Self shading by large populations actually on the sand surface could also be a problem for non-motile forms such as S. astraea as they would tend to be buried under sediment and not be able to move to more favourable positions. Oocystis spp. were recorded on the sand surface only when cells were present in the inflow water (Figure 19). They occurred mainly during the summer and autumn periods and occasionally persisted until the winter. Increases in numbers on the sand surface again seemed mainly to be due to accumulation, few actively dividing cells being observed. Although no quantitative data is available, observations showed that cells of Oocystis were being grazed by animals. Both Daphnia pulex and Daphnia magna were found with cells of Oocystis in their gut. Brook reported several species of algae similar in size to Oocystis present in the guts of insect larvae (Brook, 1952). Grazing may thus have played an important part in reducing the populations of Oocystis as crustaceans, insect larvae and other animals were present on the sand surface.

The true epipelic flora was composed mainly of diatoms with members of the Chlorophyceae becoming co-dominant at times. With the exception of Melosira varians the most abundant species of diatoms were members of the biraphid Pennales. These motile diatoms are able to avoid burial and to maintain themselves in a position favourable to photosynthesis by their movements in the epipelic environment. M. varians and Ulothrix tenuissima, as non motile filamentous forms, overcame the problem of being buried by growing upwards into the supernatant water. The filaments were often made buoyant by the production of gas bubbles. The only other non motile forms abundant on the sand surface were Scenedesmus spp. and the palmelloid stages of Chlamydomonas spp.

Melosira varians has been reported as a summer (Pearsall et al., 1946) and an autumn (Brook, 1954) constituent of the flora of slow sand filter beds. During this present study it was found to be more abundant during the spring, autumn and winter periods and only occasionally so during the summer (Figure 26). In filter beds 12 and 13, M. varians was present in fairly high concentrations throughout most of the period with the exception of the autumn of 1964 and the spring and winter of 1965. The largest concentrations occurred between October 1963 and February 1964. In filter beds 30 and 32 it was present in large concentrations between August 1963 and April 1964 and also July and October 1965. During the intervening period it was either present in low concentrations or not recorded at all. The persistence of high concentrations in beds 30 and 32 between October 1963 and February 1964 corresponded, not only to a period favourable to growth (large concentrations were present in other beds at the same time), but also to the period during which these two beds were cleaned by the "hydra" process (see Chapter II). The beds were not then drained down and skimmed thus any algae remaining in the bed after cleaning could quickly recolonise any bare areas of sand surface.

Ulothrix spp. (mainly a mixture of U. tenuissima and U. tenerrima) were present mainly during the autumn of 1963, the late spring of 1964 and the summer and autumn of 1965. Recorded concentrations on the sand surface were never very high and did not exceed $6 \times 10^5 \mu^3$/cm. On many occasions, however, especially during 1965, mats of these algae were observed floating on the surface of the supernatant water buoyed up by gas bubbles probably largely of oxygen produced during vigorous photosynthesis. On very bright, sunny days very many such mats could be observed in each of the filter beds. This detaching of mats would greatly reduce the sand surface populations, thus the overall populations of these algae on the sand surface was probably much higher than that recorded. The numbers of most filamentous forms tended to be underestimated due to the method of sampling. It was very difficult to obtain a representative sample per unit area of sand surface, especially when the filaments were many centimetres in length, as they were not sucked up by the pump. The occurrence of *Ulothrix spp.* on the sand surface, as was found for other chlorophyceans such as *Spirogyra sp.* by Brook (1954), was mainly during the summer period. The presence of large numbers of *Ulothrix spp.* on the sand surface of bed 12 during the spring of 1964 corresponded with a period of copper sulphate application to that bed. Copper sulphate was applied to give a concentration of 0.5 mg./L. There followed a period of increase of certain algae, especially *Ulothrix*. (For a fuller discussion of this application of copper sulphate, see pages 103 to 105).

Numerous species and varieties of diatoms of the Biraphidineae were present throughout the entire period. The most abundant diatoms present were *Pinnularis microstauron*, *Surirella ovata*, *Cymbella spp.*

Nitzschia palea, N. acicularis and N. linearis. Other species present occurred in smaller numbers, usually in mixed populations with the more common species.

Pinnularia microstauron occurred mainly during the spring period (Figure 26). Although P. microstauron was the most common species of Pinnularia present, P. viridis, P. Debesi, P. divergens and P. microstauron var. Brebissonii also occurred with it in small numbers at various times. Cell concentrations were slightly lower in beds 30 and 32 during the period of "hydra" cleaning (up to January 1964) and in all four beds during the autumn of 1964. The populations produced during the autumn of 1965 were removed by cleaning in beds 12 and 13 and never recovered, whereas those in beds 30 and 32 were able to increase throughout the period and survived in higher concentrations throughout the winter. Cell concentrations generally increased throughout the run of a filter bed, exceptions being the late spring of 1964 in bed 13 and the autumn and winter of 1965 in beds 30 and 32.

Surirella ovata (Figure 26) was most abundant during the spring periods and least abundant during the late autumn and early winter periods. It was generally present in greater concentrations in beds 30 and 32 than in beds 12 and 13. This did not seem to be a function of the "age" of the bed, indeed the highest concentrations recorded were in bed 32 during the spring of 1964 when growth was interrupted by cleaning. These large populations in early 1964 in beds 30 and 32 may have been a direct result of the large residuum of living cells left on the sand surface after the previous cleaning. Throughout the period August 1963 to January 1964 these two beds were cleaned by the "hydra" process. This method of cleaning tended to allow organic material to accumulate in the upper layers of sand. It is possible that Surirella ovata favoured high concentrations of organic matter and was thus able to grow more readily in these two beds.

Nitzschia linearis (Figure 26) was the largest species of Nitzschia found in high concentrations. It occurred mainly during the spring and autumn periods although large growths did occur during the summer of 1965 in beds 13 and 32. Cell concentrations were generally higher in beds 30 and 32 until the spring of 1964. As wich Surirella ovata, this can possibly be correlated with the higher content of organic matter in these beds associated with "hydra" cleaning. After the period of "hydra" cleaning beds 30 and 32 were resanded (see Chapter II). The cell concentrations of N. linearis then fell to levels similar to those in beds 12 and 13. Cell concentrations were low in all of the beds studied during the summer and autumn of 1964 and the autumn of 1965.

Nitzschia acicularis (Figure 26) was not abundant in any of the filter beds between August 1963 and February 1964. There was then a period, until June 1964, of rapid growth and large populations developed. Throughout the rest of the period concentrations remained fairly constant, only increasing slightly in bed 30 in May and June 1965. Cell concentrations were reduced in beds 30 and 32 in 1964 after resanding. This period of resanding corresponded to a period of low cell numbers in the inflow water. The re-establishment of the populations was thus much slower. N. acicularis is also noted for its rapidity of movement (West & Fritsch 1932), a feature of great advantage in competing for a favourable position on the sand surface. This ability to move actively together with the narrowness of the frustules, probably accounts for its ability to penetrate the depths of the sand. Living cells were observed in the filtered water passing out of the filter beds on several occasions indicating that N. acicularis was able to pass through the entire sand column in the dark and still survive.

Nitzschia palea (Figure 26) was the third species of Nitzschia to occur abundantly. It was present mainly during the late spring and early summer periods but high concentrations sometimes persisted into the autumn periods. In all beds cell concentrations decreased during the winter of 1964 to 1965 and 1965 to 1966. The spring growth commenced at about the same time as that of N. acicularis but extended into the summer period. Cell concentrations were never very high, seldom exceeding 2 x 10 µ /cm.$^2$ The widespread occurrence of N. palea in soils (Lund, 1946) and sediments (Round, 1957) is well known especially in areas rich in dissolved substances. It did not appear to be more abundant in beds 30 and 32 between August 1963 and January 1964, however, when concentrations or organic matter were higher.

The species of Cymbella present were mainly C. turgida, C. ventricosa and C. lanceolata. They occurred as mixed populations. These algae did not occur in high numbers in any particular season but at various times of the year (Figure 26). In bed 12 cell concentrations were at their highest during the spring and autumn of 1965. In bed 13 concentrations were relatively low until the late summer and autumn of 1965. In bed 30 maximum concentrations occurred in the spring and summer of 1964 and the spring, summer and autumn of 1965, whilst in bed 32 maximum concentrations occurred in the spring of 1964 and the summer of 1965. Each of the identified species of Cymbella had very short doubling times and were thus capable of producing large populations quickly (see Chapter VIII). This potential never seemed to be realised in the epipelic population. Indeed Cymbella was the dominant species of the attached algal populations (see page 135 ) and it is possible that these should be considered as more or less permanently attached forms rather than epipelic forms.

Chlamydomonas spp. often occurred in large numbers in the supernatant water (see page 74 ), and during these periods cells were also found living on the sand surface (Figure 26). Many of these cells had merely sedimented onto the sand surface and there survived. At times, however, the cells shed their flagella and formed a palmelloid stage, covering parts of the sand surface with a layer of "jelly". It was on these latter occasions that maximum cell concentrations of Chlamydomonas spp. on the sand surface were reached. These palmelloid stages occurred during May and June 1964 and to a lesser extent in June and July 1965. The conditions bringing about these palmelloid stages are not clearly understood (Fritsch, 1935). There may, perhaps, be some correlation between the applications of copper sulphate to Queen Mary Reservoir and subsequent growths of *Chlamydomonas spp.* both in the reservoir (see page 68 ) and in the filter beds. There is a closer correlation, however, between the periods when the supply main from the reservoir was chlorinated for the purpose of mussel control (Greenshields & Ridley, 1957) and the development of the palmelloid stage of *Chlamydomonas spp.* on the filter beds. Periods of chlorination are indicated by solid bands in Figure . It is possible that very small amounts of chlorine were allowed to pass onto the filter beds in the inflow water and this might have been enough to stimulate the development of the palmelloid stage. Cell concentrations were low in all four filter beds during the periods October to December 1964 and 1965. These reductions in concentration were probably not due to low light and temperature levels but to other factors as higher measurable populations were present in January and February of each of these years when light was equally low and temperature levels were lower.

*Scenedesmus spp.* were present throughout most of the period in each of the filter beds studied. Cell concentrations were not usually high. Maximum concentrations occurred just after periods of chlorination (solid black bands, Figure 26), however, indicating possible stimulation of growth or the ability to survive higher than normal concentrations of chlorine. This correlation was more clearly seen in beds 12 and 13 than in beds 30 and 32. Cell concentrations reached $10^8$ $\mu^3$/cm. in bed 12 at one time. Apart from the period of vigorous growth cell concentrations rarely exceeded $10^6 \mu^3$/cm.

Between March 26th and April 24th, 1964, copper sulphate was added to bed 12 at a concentration of 0.5 mg./L for 8 hrs. every day in an attempt to control the algae on the sand surface. The results of these applications are given in Table 4. Bed 12 was already in use when the copper sulphate treatments were started and results for the two weeks previous to March 26th are also included in Table 4.

Concentrations of Chlamydomonas spp. decreased throughout the run of the bed. This decrease had started before the applications of copper sulphate and cannot, therefore, be attributed to it. Ankistrodesmus falcatus, a planktonic alga, did not fluctuate greatly in concentrations. Scenedesmus spp. and Ulothrix spp. were either present in very low numbers or not recorded at all before March 26th. After this date Ulothrix spp. increased rapidly in concentration and Scenedesmus spp., after an initial lag, started to increase by the end of the filter bed run. All of the diatoms but Stephanodiscus astraea had increased in concentration by the end of the filter bed run after the application of copper sulphate on the 27th March.

The largest increases in algal populations, after the start of the copper sulphating, were Pinnularia microstauron, Nitzschia acicularis, N. linearis, and Surirella ovata. Each of these species increased at least forty-fold in concentration in the four weeks following the first application. Another alga which occurred in concentrations higher than usual towards the end of the filter bed run was Lyngbya versicola. This was not recorded on the sand surface before the start of copper sulphating.

**Table 4.** The concentrations of algae, in cubic microns per square centimeter, on the sand surface of bed 12 between March 13th and April 24th 1964.

| Species | March | | | | April | | |
|---|---|---|---|---|---|---|---|
| | 13 | 20 | Copper sulphate application | 27 | 3 | 10 | 24 |
| Chlamydomonas spp. | 474 | 134 | | 172 | 80 | 10 | 16 |
| Ankistrodesmus falcatus | 81 | 236 | | 180 | 142 | 192 | 208 |
| Scenedesmus spp. | 1 | 1 | | 4 | 2 | 1 | 40 |
| Ulothrix spp. | 0 | 0 | | 10 | 105 | 532 | 680 |
| | | | | | | | |
| Melosira varians | 0 | 260 | | 180 | 296 | 624 | 1280 |
| Stephanodiscus astraea | 1365 | 4050 | | 5040 | 9060 | 4660 | 1150 |
| Pinnularia microstauron | 6 | 0 | | 84 | 450 | 390 | 450 |
| Nitzschia acicularis | 8 | 33 | | 360 | 6210 | 12220 | 9200 |
| N. linearis | 45 | 117 | | 1500 | 1680 | 9000 | 14000 |
| N. palea | 0 | 0 | | 7 | 82 | 300 | 1300 |
| Surirella ovata | 117 | 708 | | 764 | 2580 | 28200 | 3200 |
| Cymbella spp. | 0 | 39 | | 48 | 84 | 1038 | 3000 |
| | | | | | | | |
| Lyngbya versicola | 0 | 0 | | 0 | 8 | 200 | 1600 |

The copper sulphate dose of 0.5 mg./L would give a concentration of 0.19 mg./L copper in the supernatant water. In waters such as are found in the Thames Valley, rich in organic materials and of high calcium carbonate hardness, much of the copper sulphate would either be precipitated as basic copper carbonate or bound up into organic complexes leaving much less than 0.19 mg./L copper in the water as a toxic substance. Thus although certain diatoms have been found to have their growth inhibited by 0.2 mg./L copper, and Scenedesmus dimorphus to haveiits growth retarded at the same concentration (Chapter IX), the amount of toxic copper in the filter bed water during this series of applications was probably much less than this. There would also have been a continuous loss to the system of copper dissolved in the water passing out of the filter bed. The overall effect of this series of applications seemed to be to increase the populations of most of the algae present. It is not known whether this was due to stimulation of growth or to other factors such as reduced grazing by killing the animals present.

Persistent vertical migrations have been known in populations of epipelic algae for many years (Bracher, 1919) and have been recently re-investigated (Aleem, 1950; Hopkins, 1963; Palmer & Round, 1965; Round & Palmer, 1966). These investigations were carried out on tidal waters but recently the work has been extended to fresh water habitats (Round & Eaton, 1966; Eaton & Moss, 1966). Such diurnal vertical migrations as were described by these workers were suspected in filter beds but no direct observational data was available. Waterworks engineers have long known that blankets of algae, buoyed up from the sand surface during photosynthesis, left bare patches of sand which increased the volume of water which was able to pass through the filter bed (Pearsall et al. 1946). A large population of epipelic algae present on the sand surface would be expected to offer less resistance to the flow of water than algae that had migrated into the sand interstices and partially blocked them. Similarly, filamentous algae, buoyed up into the supernatant water, would offer less resistance to the flow of water through the filter bed than filaments lying on the sand surface. If the resistance to flow did vary with the position of the algae one might be able to detect this movement by observing changes in head loss (see Chapter II for full explanation of head loss). These changes would only be clear if the flow through the bed were also constant as this can also affect head loss. As the flow rate of water through the filter beds often had to be varied to meet changing consumer demands there were only a limited number of periods for which data are available for head losses when flows were kept constant. Several such periods are shown in Figure 27. Periods of darkness, i.e. the sun below the horizon, are indicated by black bars. From these figures a clear diurnal fluctuation in head loss can be seen with higher head losses at night and lower ones during the day. The algal floras of each of these beds on the occasions recorded in Figure 27 are given in Figures 20 to 26. Many of these species are capable of movement and some have been reported as showing a vertical migration (Round & Eaton, 1966; Eaton & Moss,

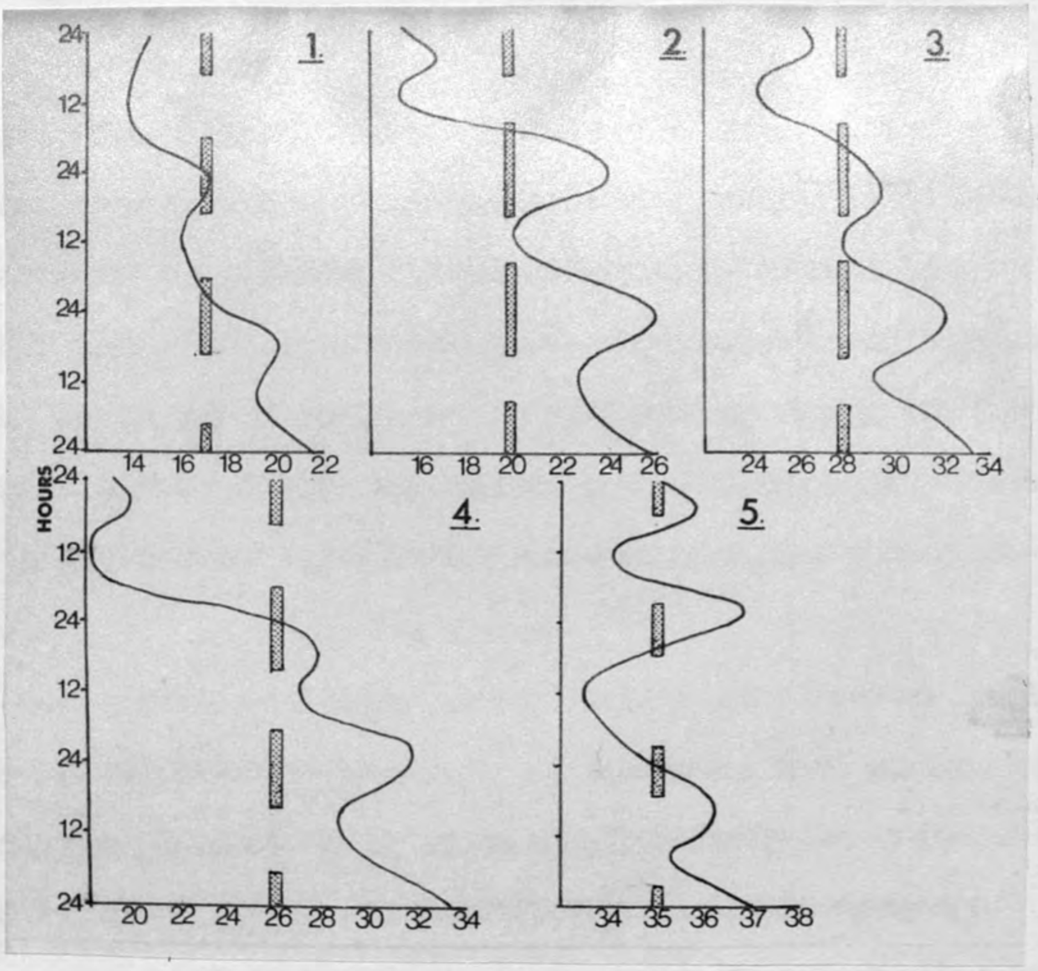
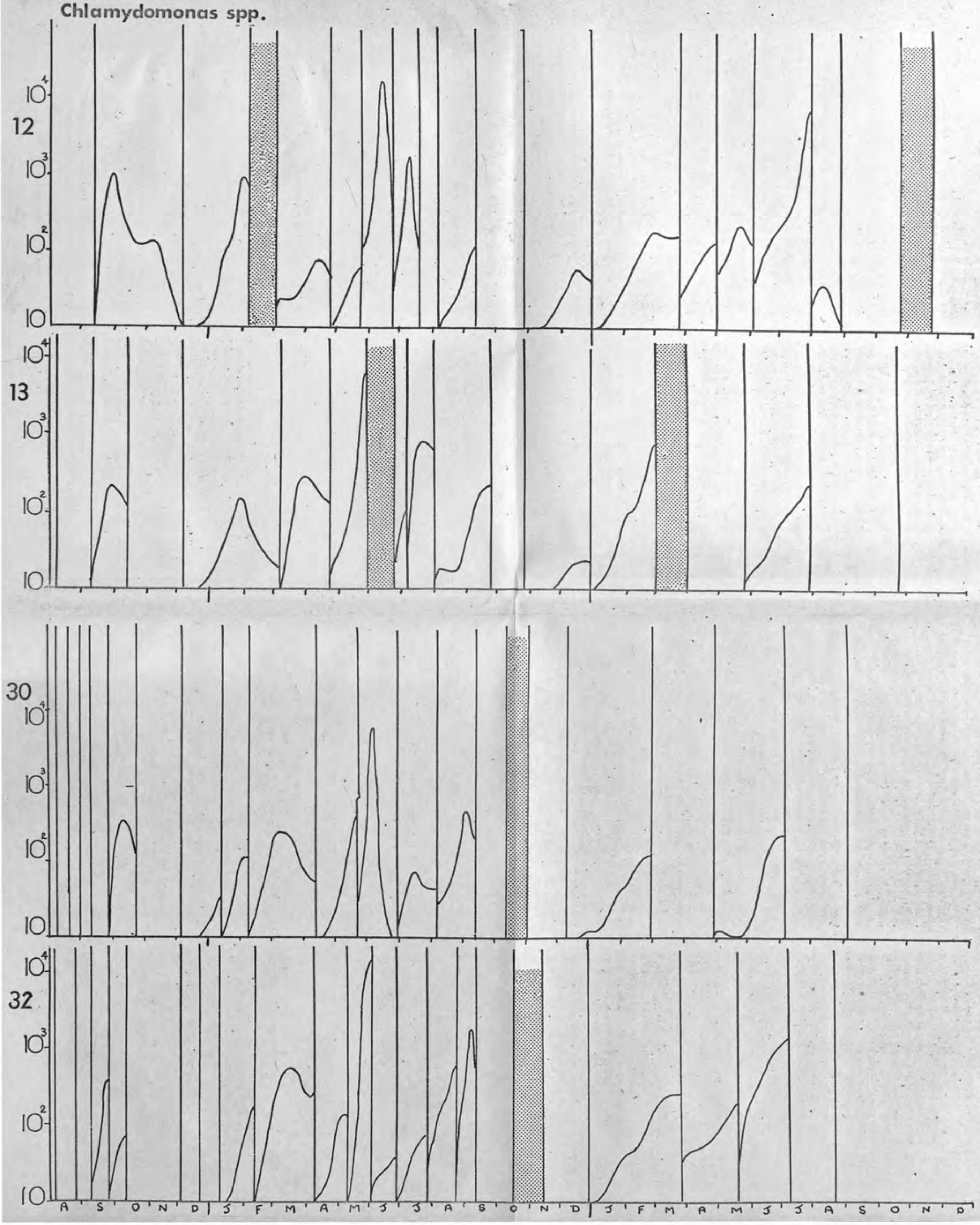


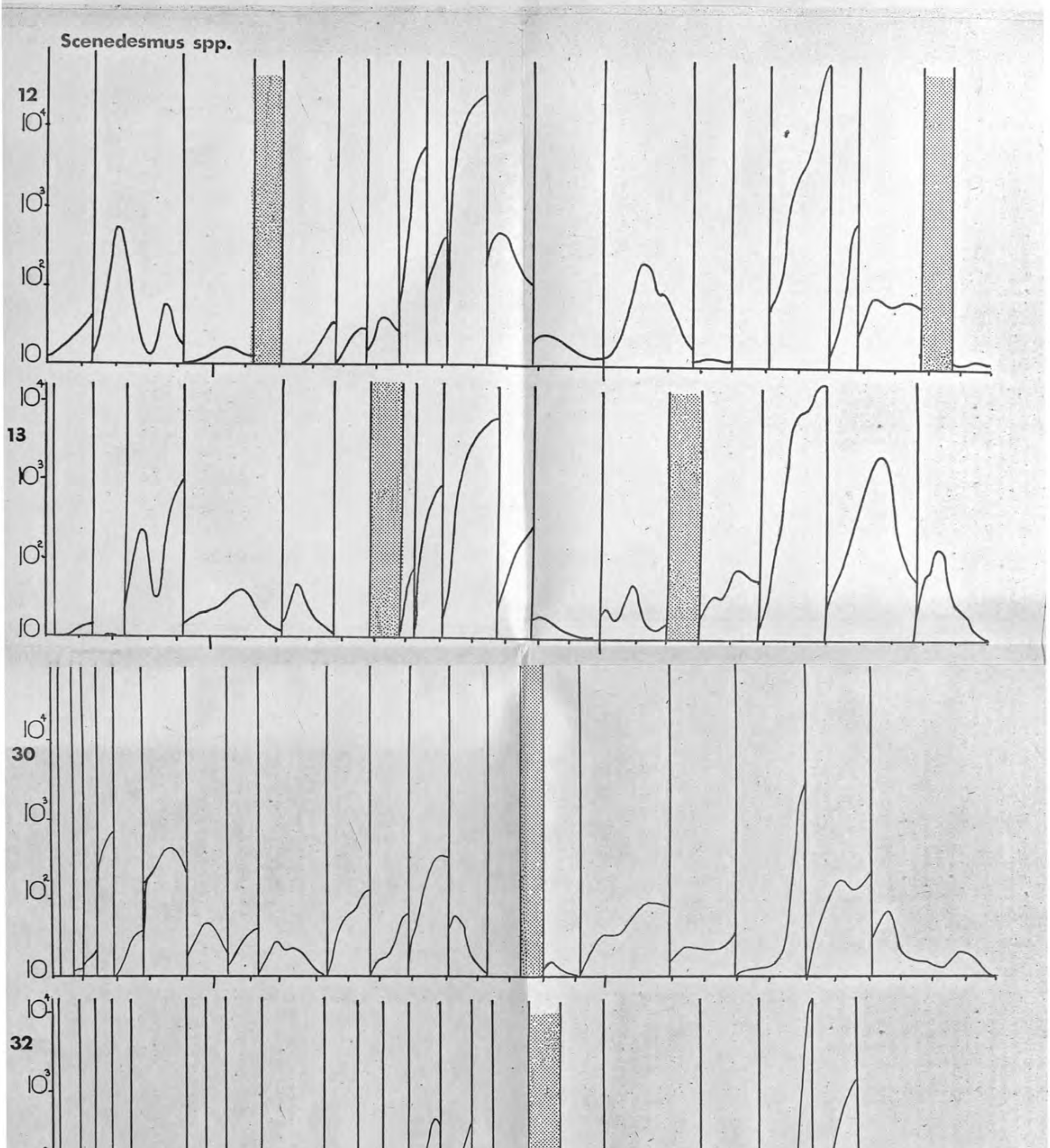

**Figure 27.** **The variations in head loss in inches (horizontal scale) over three day periods. The shaded areas represent periods of darkness. The occasions recorded are as follows ;**
**1. September 10-12th 1964. 2. December 5-7th 1964.**
**3. January 21st-23rd 1965. 4. March 18th-20th 1965.**
**5. June 19th-21st. 1965.**

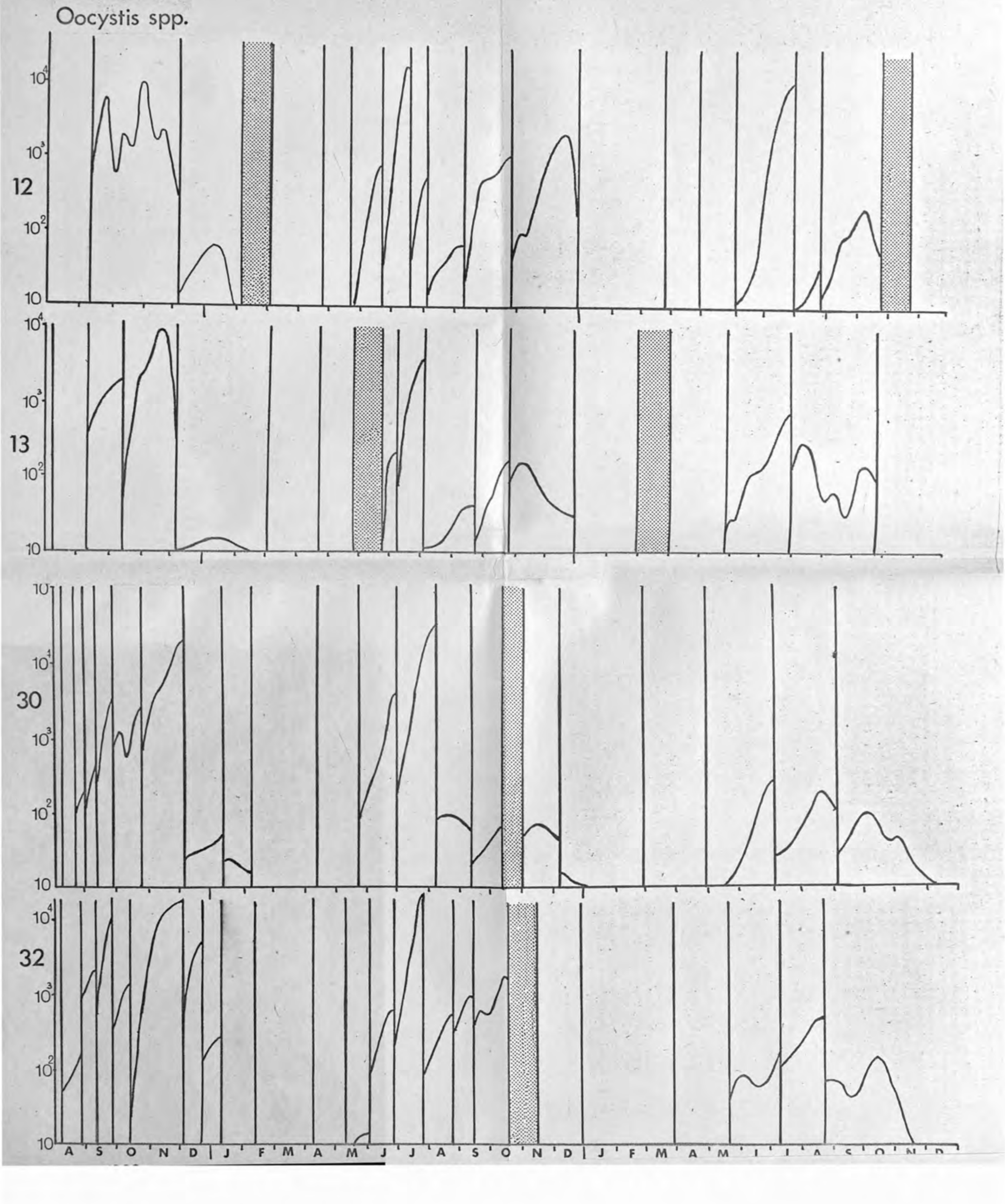
1966). As other physical factors in the filter beds, such as inflow and outflow, were constant during the periods recorded, other factors must have caused the recorded diurnal fluctuations in head loss. It is possible that these other factors were the diurnal upward migrations out of the top layers of sand onto the sand surface during the day and back again at night and also the falling back onto the sand surface at night of filamentous algae normally buoyed up during photosynthetic periods.

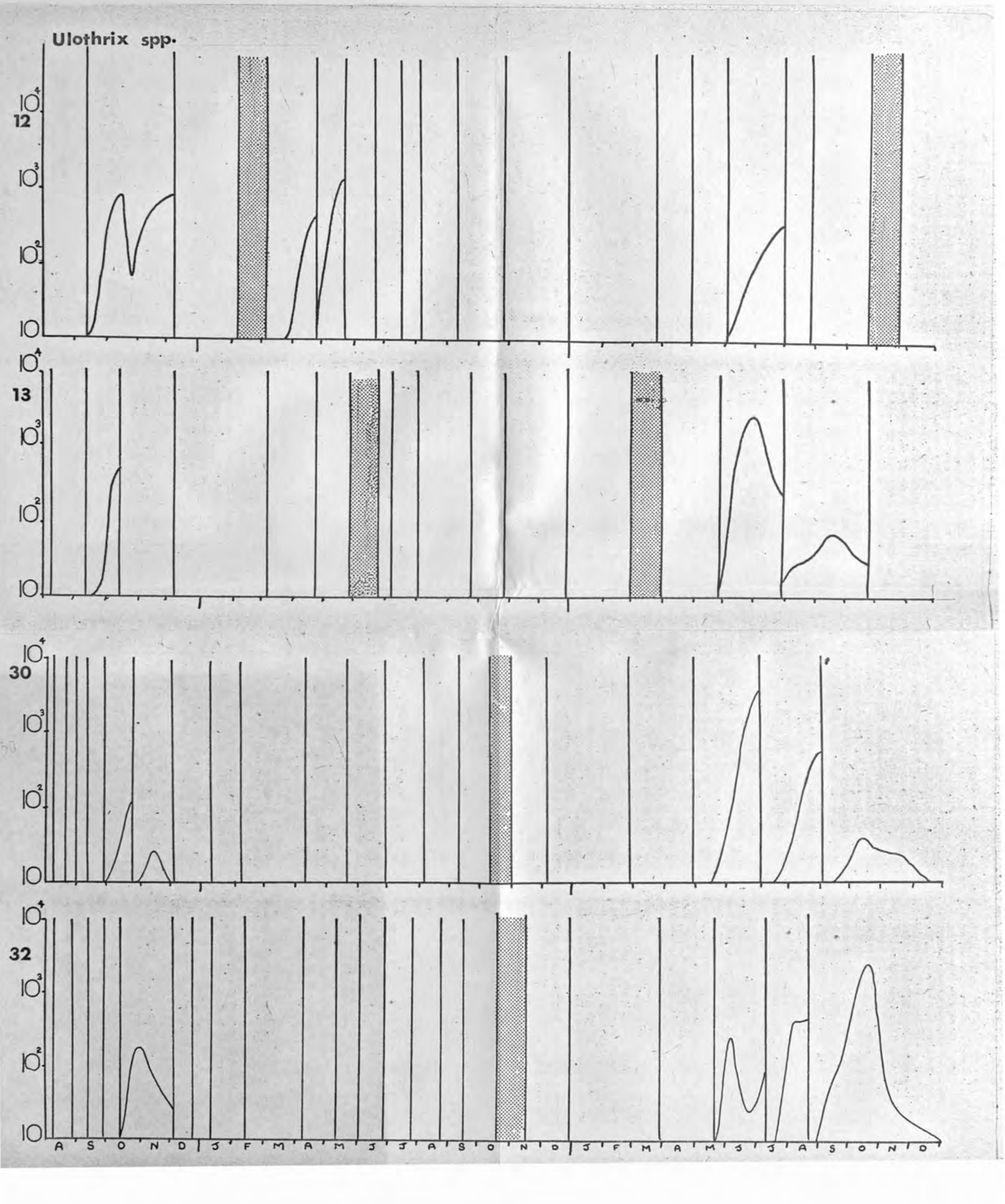
The sand surface of a newly cleaned filter bed offers an ideal situation for colonisation by algae. Although the sand surface is homogeneous the algae are by no means evenly distributed as was suggested by Brook (1954). Fritsch (1931), in referring to the plankton, pointed out that when an algal species is introduced into a body of water, its successful establishment depends not only upon physical and chemical properties in the water but also upon the time of the year. He also pointed out that a slight infection by a few spores or individuals could probably be successful only if the microscopic fauna were at a minimum. The alighting of an alga upon any particular part of the sand surface depends upon such factors as current velocities and sedimentation rates (see page 142) and the establishment of the alga upon the factors outlined by Fritsch. Unlike most lacustrine sediments the chemical conditions dominating the flora on the sand surface are not so much the interface conditions between the sand and water but the chemical conditions in the supernatant water as this is continuously flowing down past the sand surface (see Chapter VIII). The epipelic flora was dominated by diatoms and these were generally more abundant during the spring and autumn than in the winter or summer. The summer flora included members of the Chlorophyceae which at times assumed co-dominance. As there was a constant deposition of organic material on the sand surface there was always a tendency to bury the algae there. To overcome this, and to keep at the surface, requires either active movement and utilisation of energy or, in the case of filamentous forms, being buoyed up into the supernatant water by means of gas bubbles. At times certain populations shaded others on the sand surface, e.g. growths of *Ulothrix spp.* and *Melosira varians*, have been observed to shade the sand surface populations reducing photosynthesis and limiting growth. At other times large growths of *Chlamydomonas spp.* and *Scenedesmus spp.* developed on the sand surface and these may have inhibited the growth of other algae chemically (see Chapter VIII) as well as by shading. Feeding by animals has also been shown to play a part in the regulation of algal population sizes (Brook, 1952 & 1954). As well as the species discussed in detail here many others occurred but only in small numbers or infrequently.

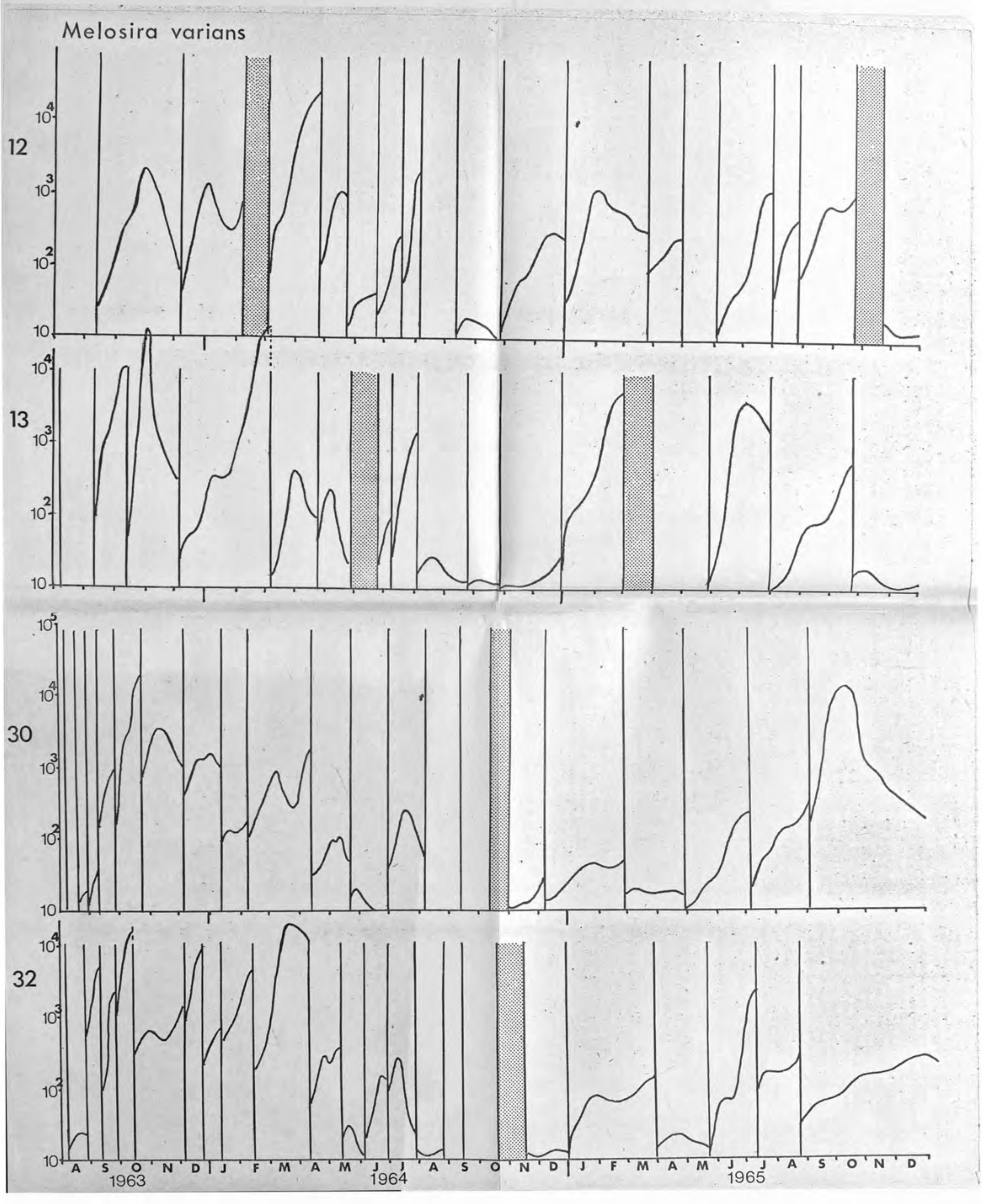
Figure 26. The periodicity of the algae on the sand surface. The results are expressed as volumes of algae in cubic microns x $10^3$ per $mm^2$.

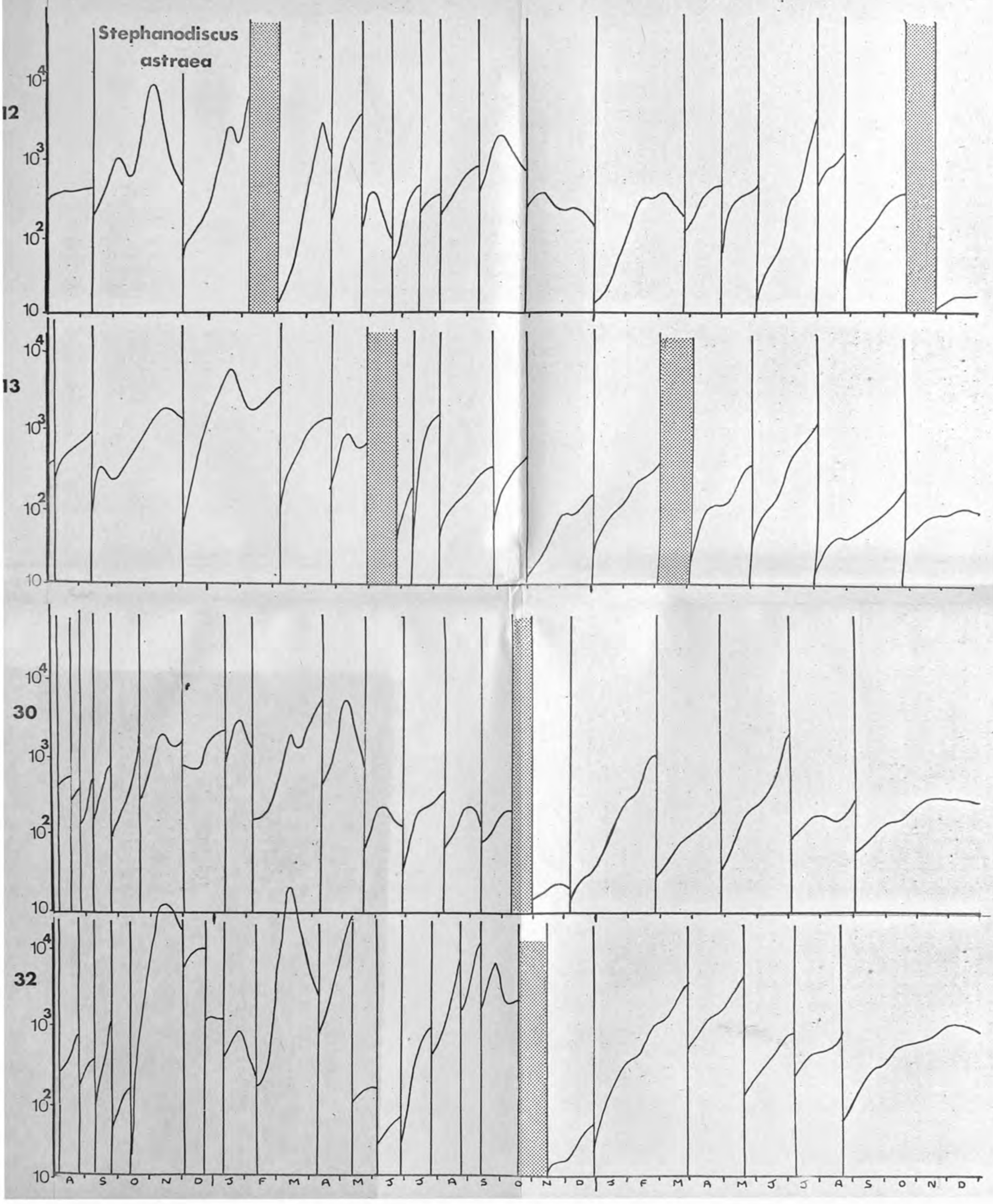

Chlamydomonas spp.
12
13
30
32
10
10²
10³
10⁴
A S O N D J F M A M J J A S O N D J F M A M J J A S O N D

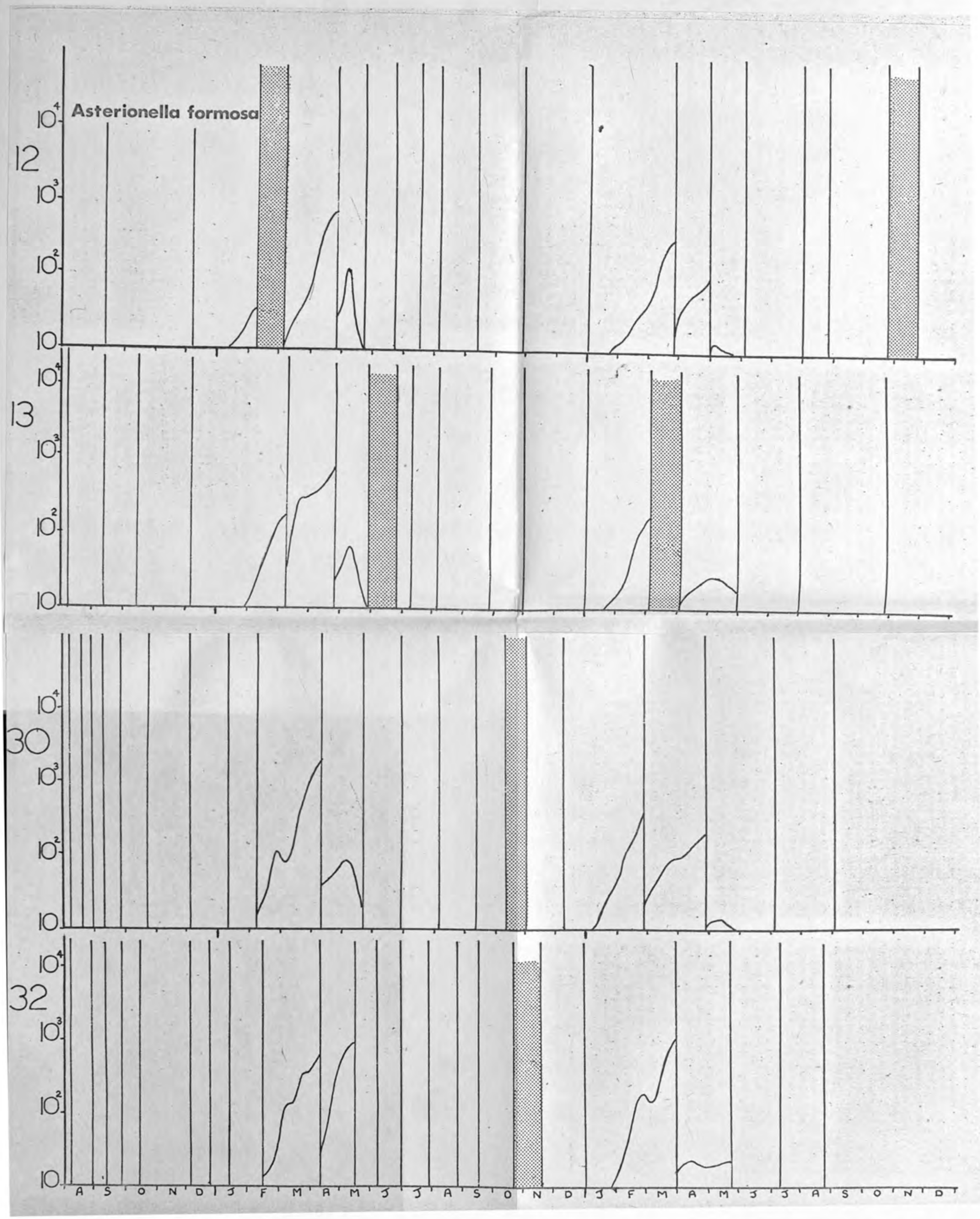

Scenedesmus spp.
12
13
30
32
10
$10^2$
$10^3$
$10^4$
A S O N D J F M A M J J A S O N D J F M A M J J A S O N D

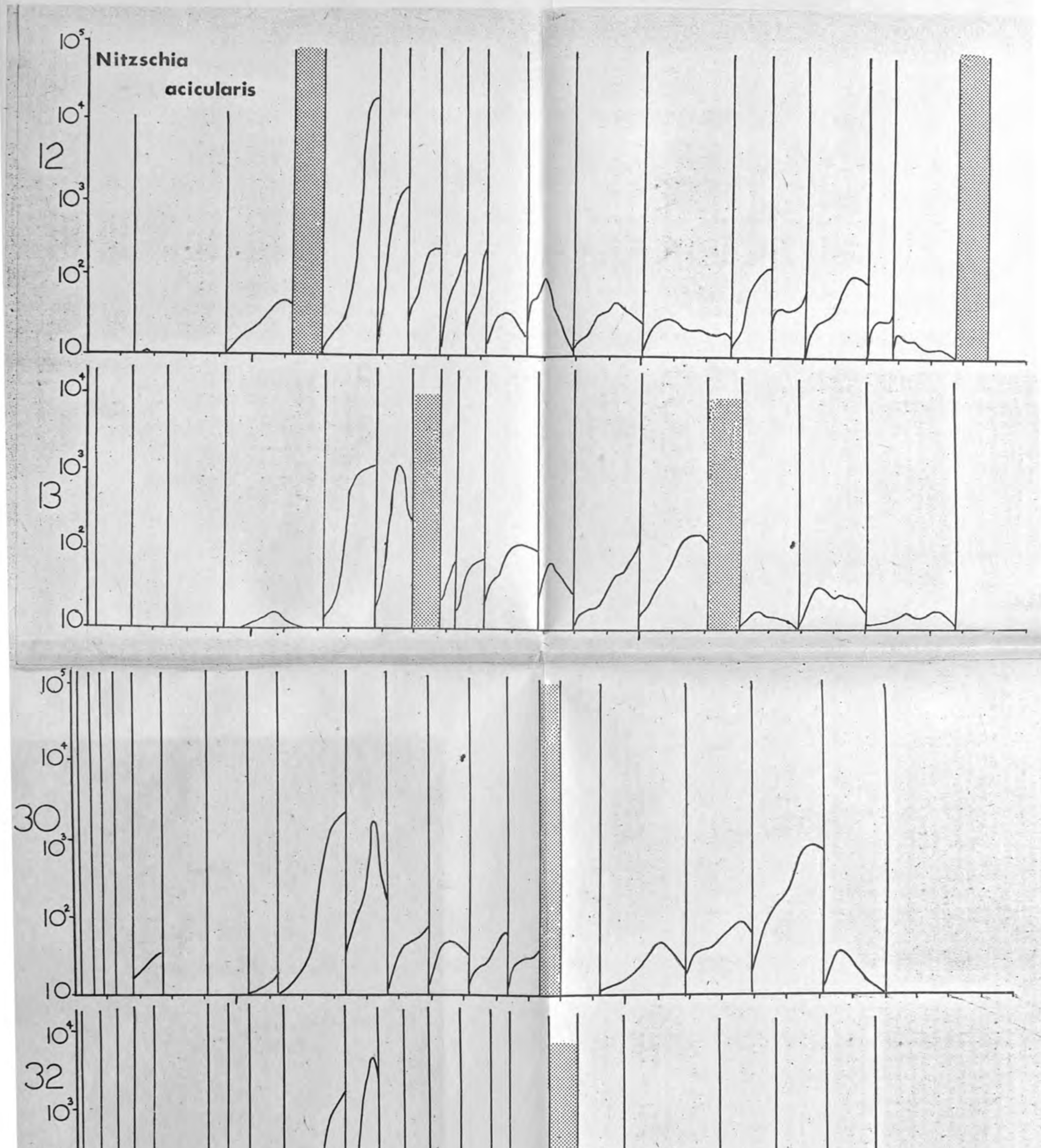

Oocystis spp.
12
13
30
32
10
$10^2$
$10^3$
$10^4$
A S O N D J F M A M J J A S O N D J F M A M J J A S O N D

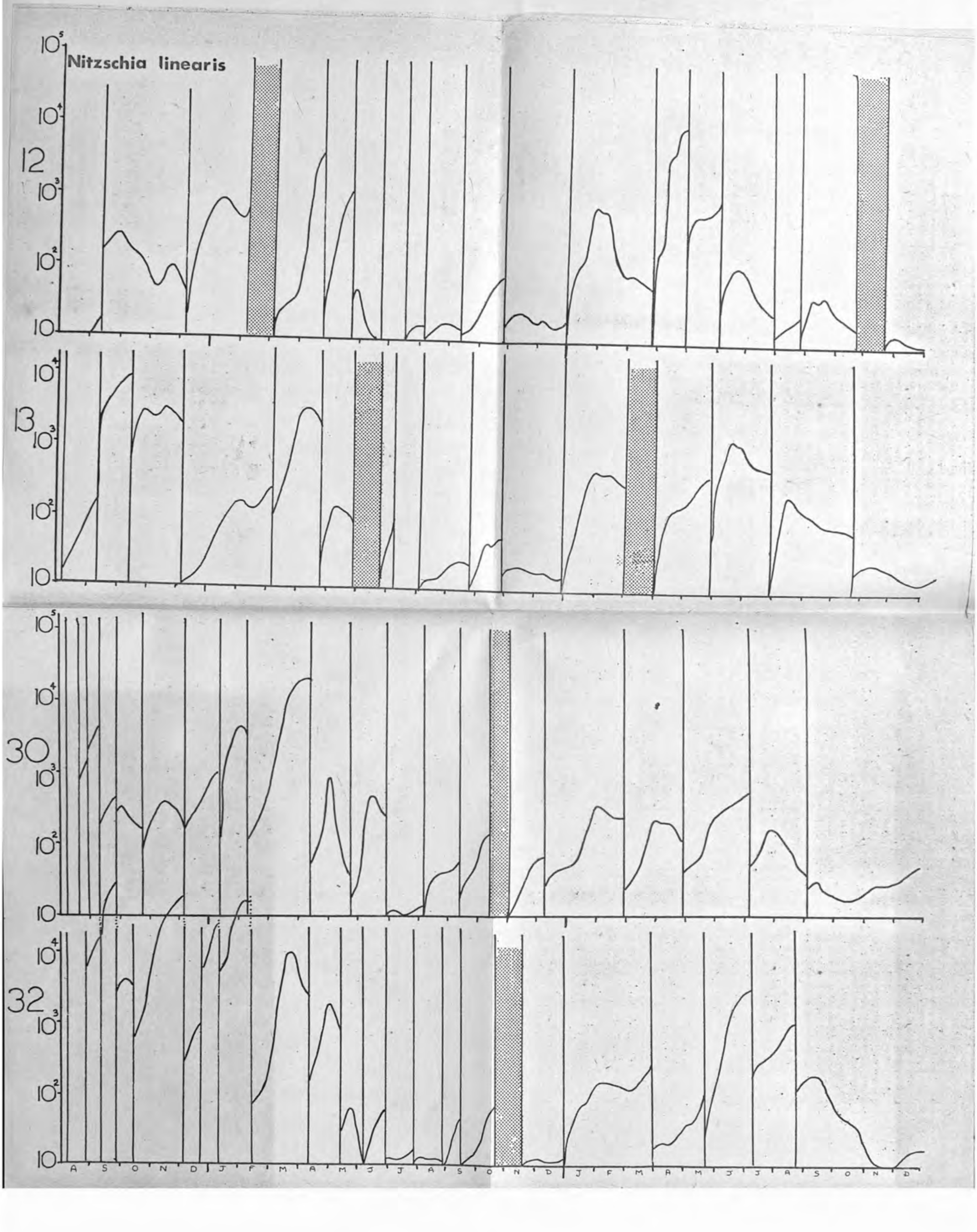

Ulothrix spp.
12
13
30
32
10
$10^2$
$10^3$
$10^4$
A S O N D J F M A M J J A S O N D J F M A M J J A S O N D

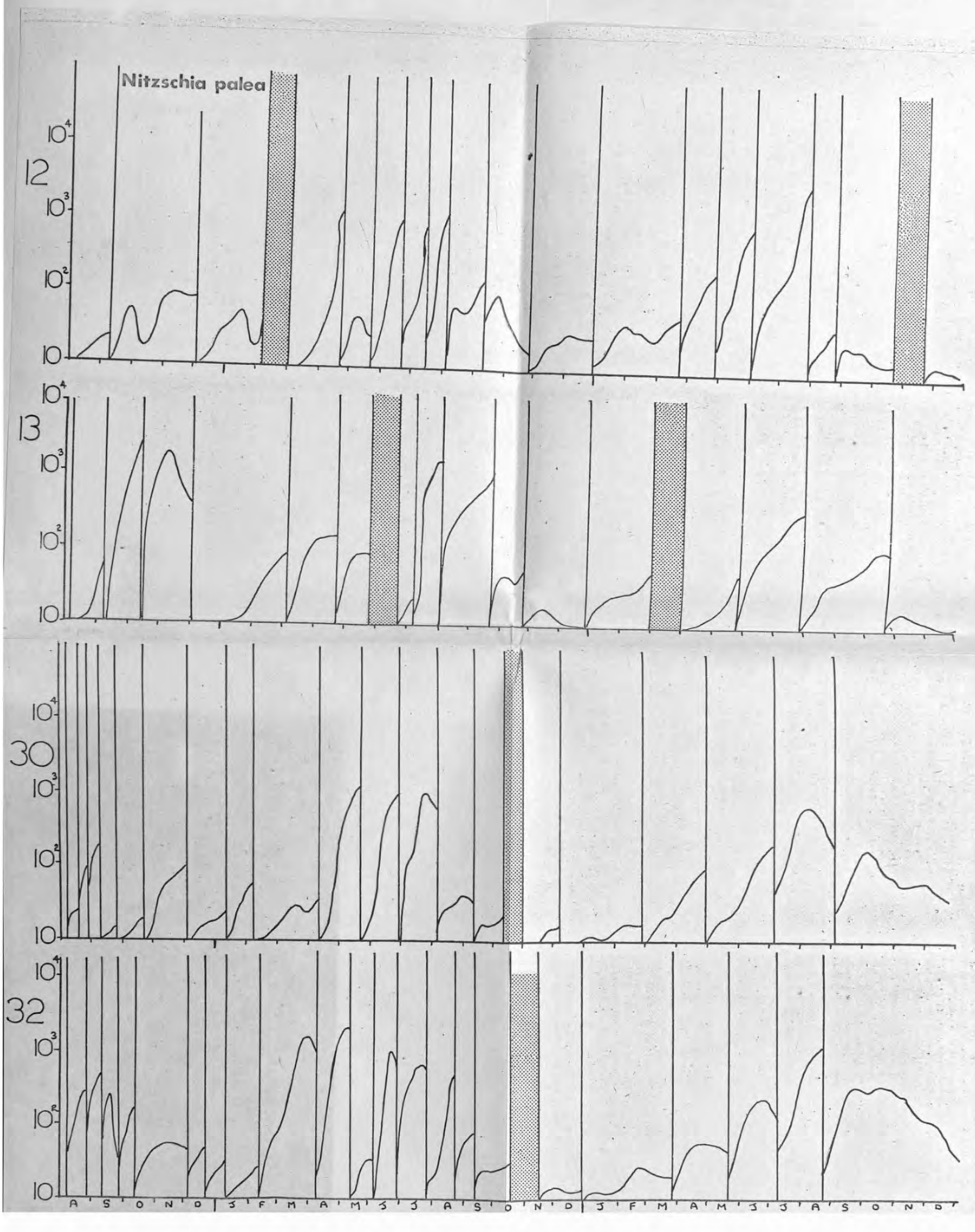

Melosira varians
12
13
30
32
10
10²
10³
10⁴
10⁵
A S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1963
1964
1965

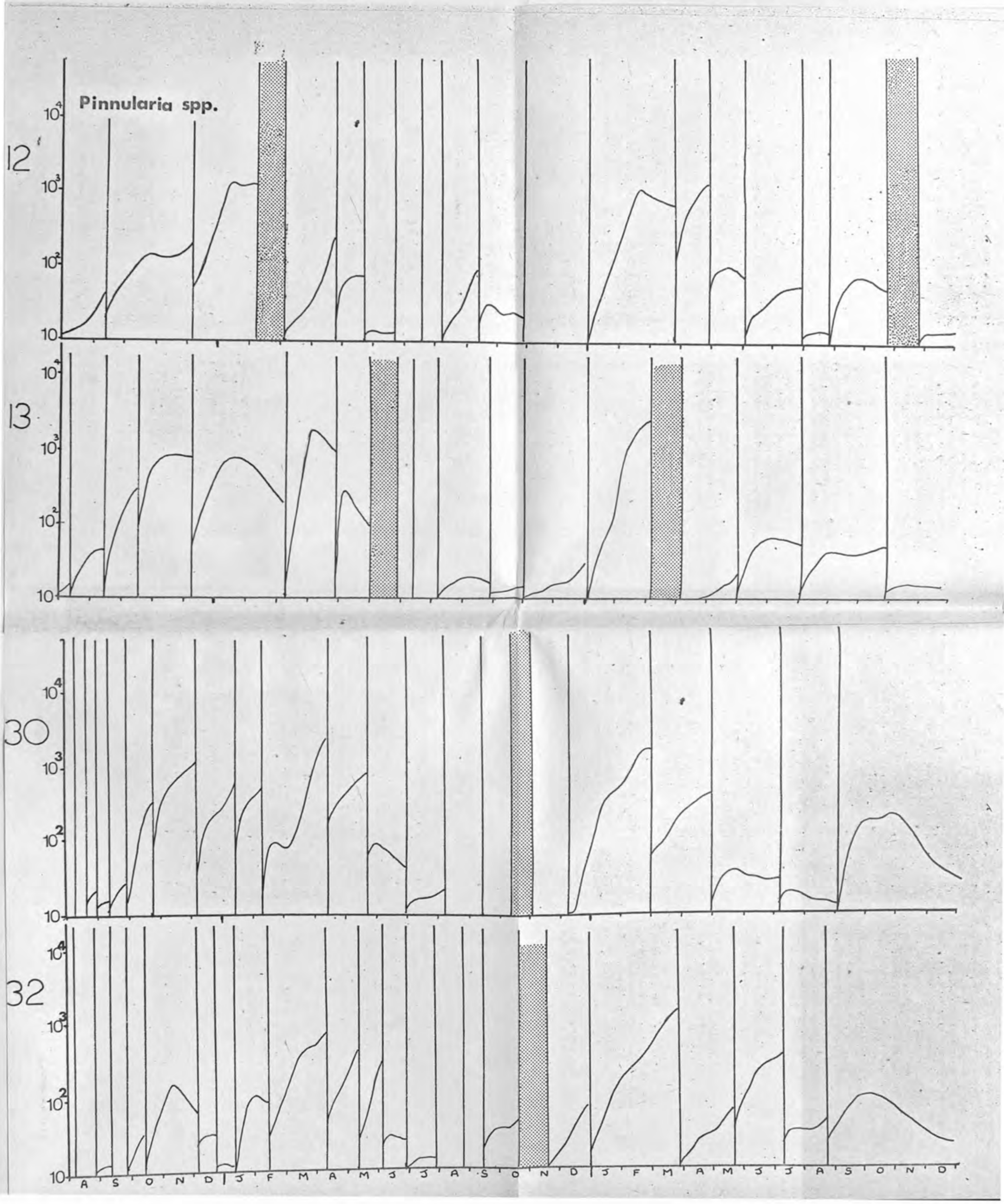

Stephanodiscus astraea
12
13
30
32
10
$10^2$
$10^3$
$10^4$
A S O N D J F M A M J J A S O N D J F M A M J J A S O N D


Asterionella formosa
12
13
30
32
10
10²
10³
10⁴
A S O N D J F M A M J J A S O N D J F M A M J J A S O N D

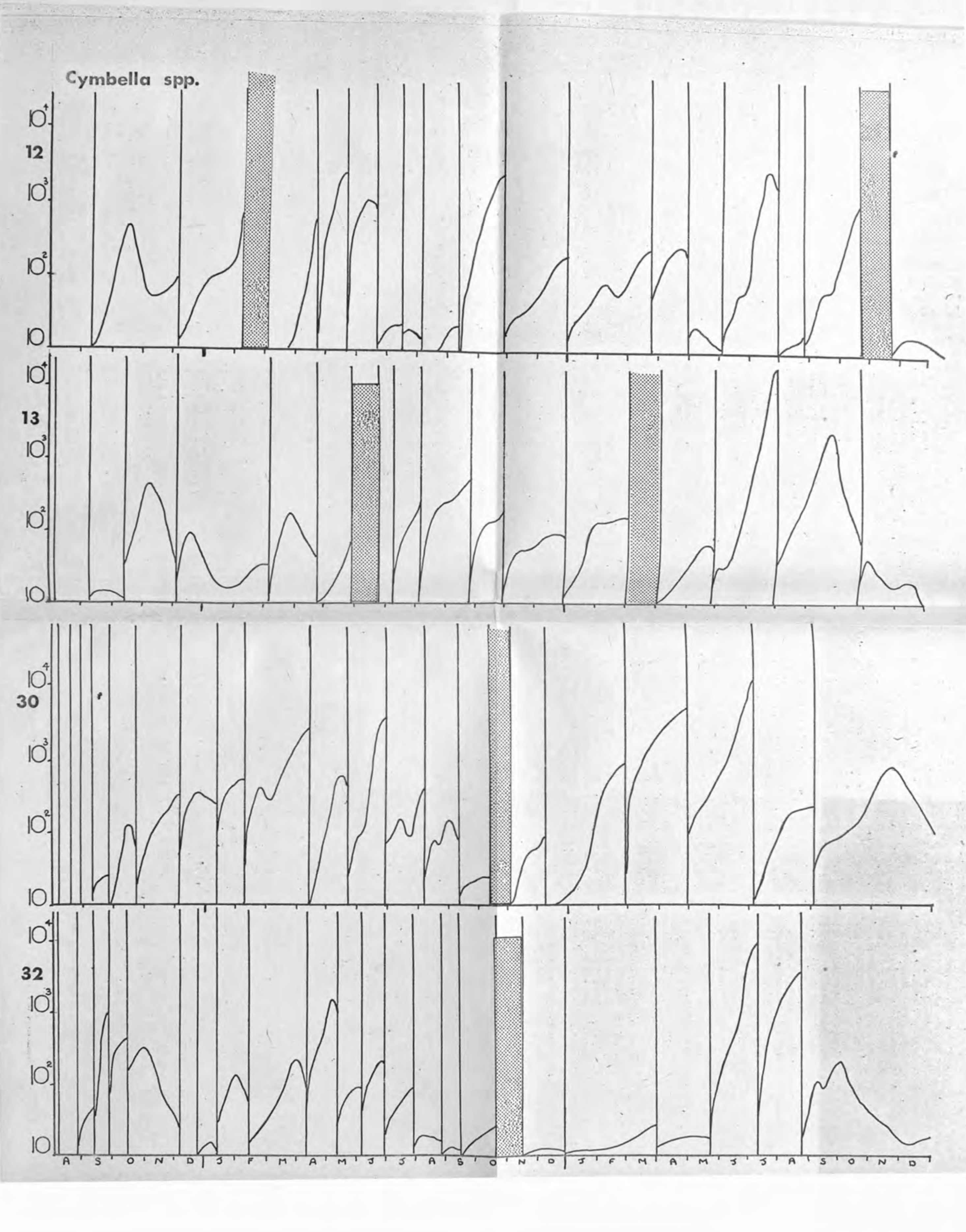

Nitzschia
acicularis
12
13
30
32
10⁵
10⁴
10³
10²
10
A S O N D J F M A M J J A S O N D J F M A M J J A S O N D

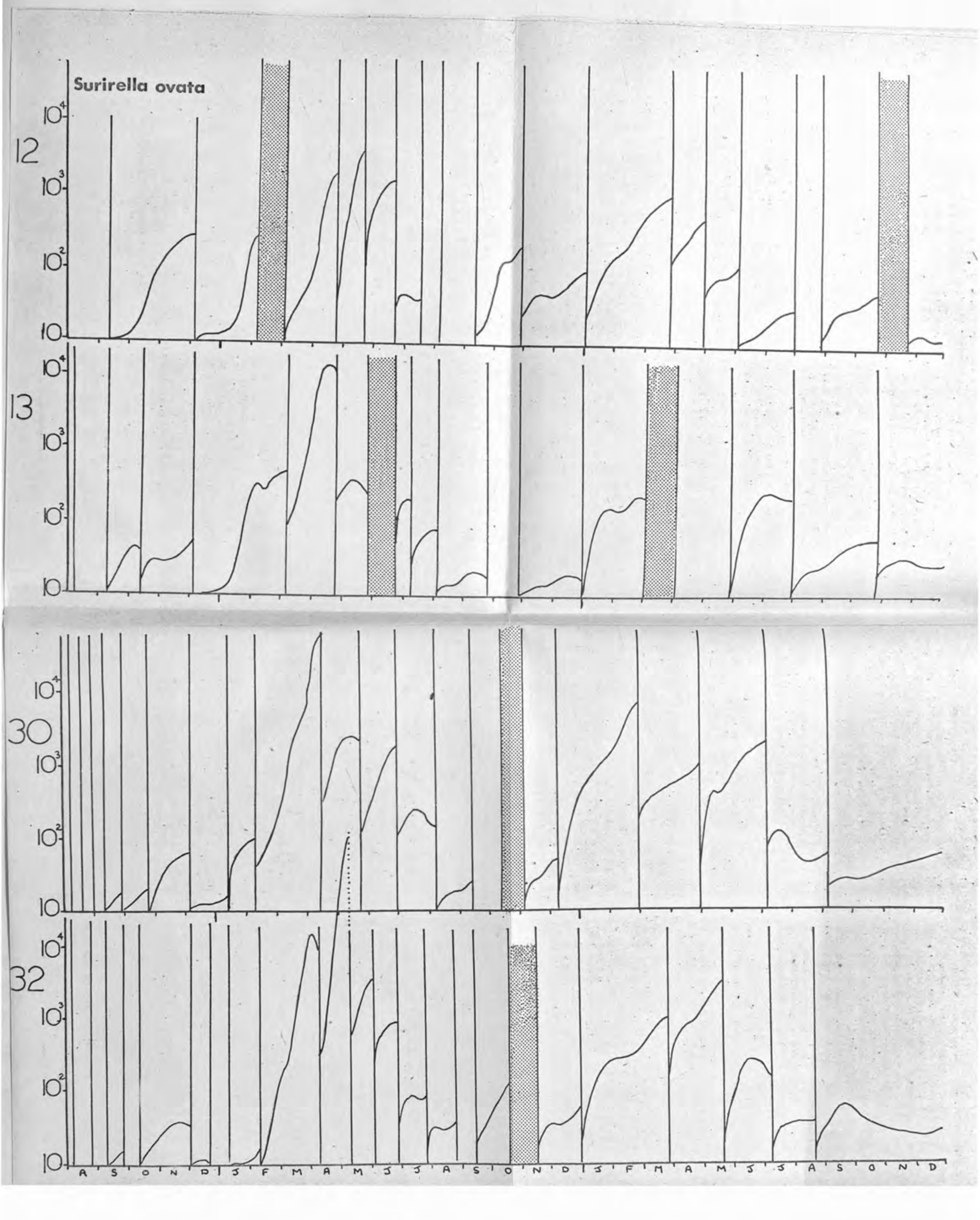

Nitzschia linearis
12
13
30
32
10
10²
10³
10⁴
10⁵
A S O N D J F M A M J J A S O N D J F M A M J J A S O N D


Nitzschia palea
12
13
30
32
10
$10^2$
$10^3$
$10^4$
A S O N D J F M A M J J A S O N D J F M A M J J A S O N D



Pinnularia spp.
12
13
30
32
10
$10^2$
$10^3$
$10^4$
A S O N D J F M A M J J A S O N D J F M A M J J A S O N D


Cymbella spp.
12
13
30
32
10
$10^2$
$10^3$
$10^4$
A S O N D J F M A M J J A S O N D J F M A M J J A S O N D


Surirella ovata
12
13
30
32
10
10²
10³
10⁴
A S O N D J F M A M J J A S O N D J F M A M J J A S O N D

C. The attached sand surface algae.

Several difficulties were experienced in sampling the attached algal flora of the slow sand filter beds. It was hoped to grow these algae on glass slides submerged in the water but this only proved possible if the greatest care was taken over cleaning the glass (see Chapter III). It was also found that the racks of slides could not actually be placed on the sand surface as they were liable to become covered with sand and organic debris. This covering was not due to normal sedimentation but probably to occasional hydraulic back pressures within the filter bed caused by adjustments in the rate of flow of water through the bed. These back pressures resulted in loosely attached sand grains and other particles being washed up into the supernatant water and then possibly settling on the glass slides. As this effect did not extend more than an inch or so above the sand surface it was possible to overcome it by suspending the slides about two inches above the sand surface. Some authors have recommended the use of vertical instead of horizontal slides (e.g. Sladeckova, 1960). According to this author the relative disadvantages of horizontal slides were pointed out as being (i) they collect a great amount of settling seston as well as true attached forms, (ii) there is a marked difference in all populations (both plants and animals) between the upper and lower surfaces of the slide, (iii) development on vertical slides is more even. Finally it was suggested that the vertical position was more convenient in use. Two initial trial exposures were made in 1964 to compare the horizontal and vertical

Table 5. The calculated volume of algae/cm.$^2$ in cubic microns on horizontally exposed compared with vertically exposed slides.

| Slides removed | Sept. 8 | | Oct. 20 | |
|---|---|---|---|---|
| length of exposure in days | 28 | | 28 | |
| Type of exposure | H | V | H | V |
| Tetrastrum staurogeniaeforme | 18 | 4 | 2180 | 1209 |
| Oocystis spp. | 7950 | 6740 | nil | nil |
| Scenedesmus spp. | 9200 | 2770 | nil | nil |
| Stephanodiscus astraea | 640 | 320 | 1700 | 1209 |
| S. hantzschii | 46 | 18 | 850 | 10 |
| Fragilaria crotonensis | 780 | 340 | 364 | 155 |
| Synedra ulna | 1280 | 1300 | 3640 | 1400 |
| Achnanthes sp. | 800 | 325 | 36 | 10 |
| Gomphonema parvulum | nil | nil | 172 | 100 |
| Cymbella spp. | 1008 | 1240 | 110400 | 25200 |
| Amphora ovalis | 40 | 20 | 364 | 432 |
| Nitzschia palea | 620 | 400 | 121 | 144 |
| Lyngbya sp. | 8000 | 6204 | nil | nil |

methods. The results are given in Table 5. From these two sets of exposures it can be seen that there were obviously greater numbers of certain species of algae on the horizontal slides than on the vertical slides. Scenedesmus spp., Stephanodiscus hantzschii, Acnanthes sp. and Cymbella spp. had populations over twice as large on the horizontal slides. Only infrequently were there fewer cells of any particular species on the horizontal slides and on those occasions there were less than 15% fewer cells. There were usually more cells of planktonic species on the horizontal slides than on the vertical slides.

Some of the objections of Sladeckova (1960) to the use of horizontal slides can be overcome. The difficulty over the differences between the upper and the lower surfaces, for example, can be dealt with by counting, for comparative purposes, the upper surface only. It was found that the growth of algae on the sand surface did not seem to correspond to the vertical any more than the horizontal slide, in fact there were probably more horizontal areas on the sand surface than vertical. Because of the generally lower growth found on the vertical slides, horizontal exposure of slides was adopted and the error due to any increased settled seston accepted.

There were three distinct groups of algae colonising the glass slides (a) pennate diatoms, (b) filamentous algae and (c) motile or coccoid unicells or coenobia other than diatoms. The first group, pennate diatoms, included stalked diatoms, i.e. those loosely attached by a mucilaginous pedicel (e.g. Synedra, Gomphonema and Cymbella), diatoms attached by the whole of one surface (e.g. Cocconeis, Acnanthes and Amphora), free living forms (e.g. Pinnularia and Nitzschia) and contaminant diatom species from the phytoplankton such as Stephanodiscus, Asterionella and Fragilaria crotonensis. The filamentous algae included diatoms e.g. Melosira, some Chlorophyceae e.g. Cladophora and Stigeoclonium, and also some Cyanophyceae e.g. Lyngbya and Oscillatoria. Also included were some thalloid algae e.g. Ulvella and Coleochaetae which did not occur in large numbers. The third group consisted mainly of members of the Chlorophyceae e.g. Scenedesmus, Chlamydomonas and Tetrastrum, which, during the summer months, were present in large numbers. Occasionally cells of Merismopedia were present and these were also included in the third group.

The relative abundance of each of these three groups of algae throughout the period and the total volume of algae taken from slides of the same age (four weeks) throughout the period are given in Figures 28 and 29. There was a more or less distinct seasonal variation in the total volume of algae present, with minimum growth during the winter months and late April and maximum growth during March, the summer and the autumn. A similar seasonal variation has been previously recorded by Brook (1954) and Butcher (1946). Superimposed upon this seasonal variation of total algae was a variation within each of the three groups. This variation can be briefly summarised as follows: in January and February pennate diatom and filamentous algae were equally abundant. The pennate forms then

Figure 28. The relative abundance of pennate diatoms (solid line), filamentous algae (fine dotted line) and motile or coccoid unicells or coenobia ( heavy dotted line) on glass slides of the same age throughout the period of study.

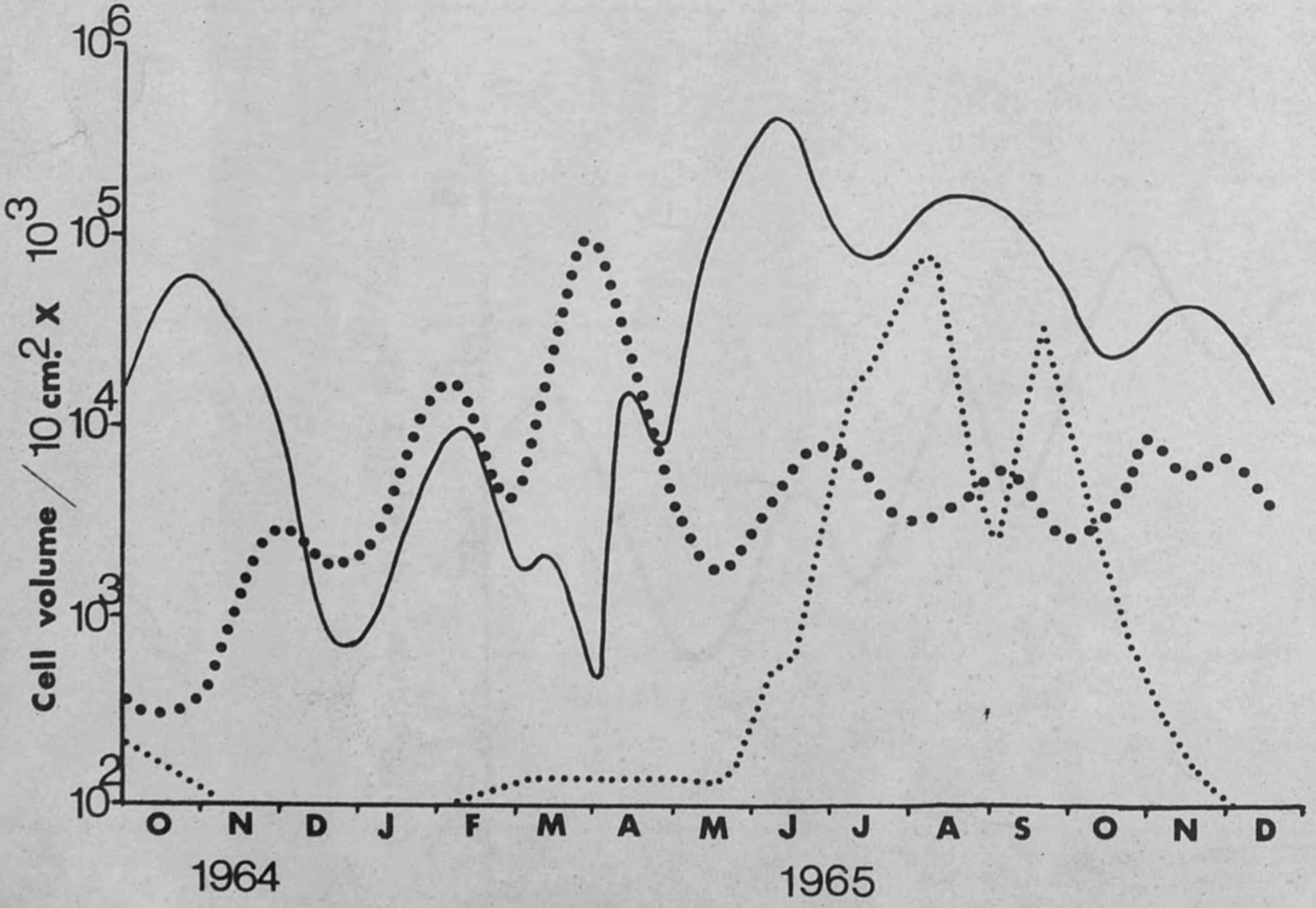
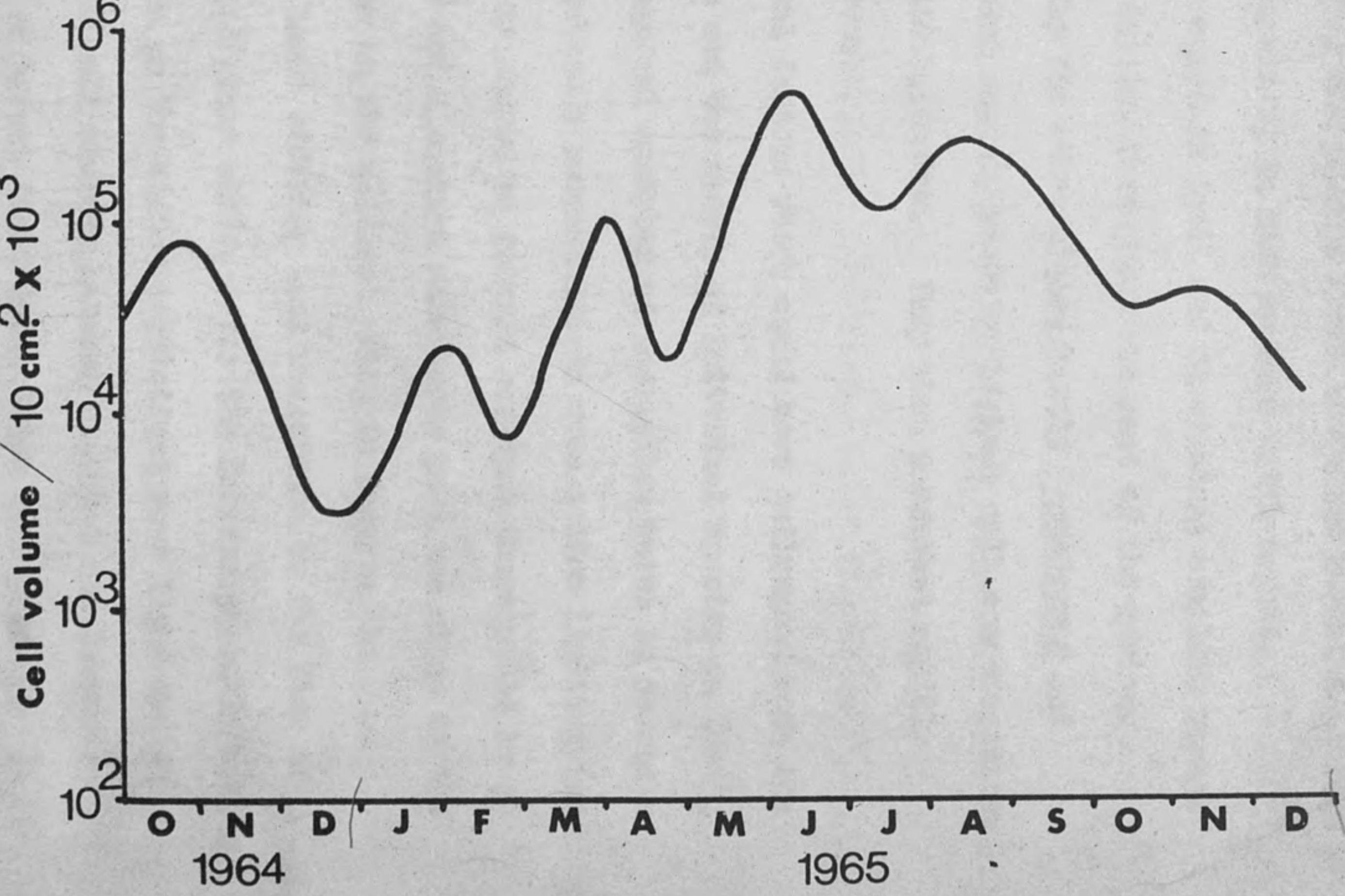


Figure 29. The variation in the total calculated volume of algae taken from glass slides throughout the period

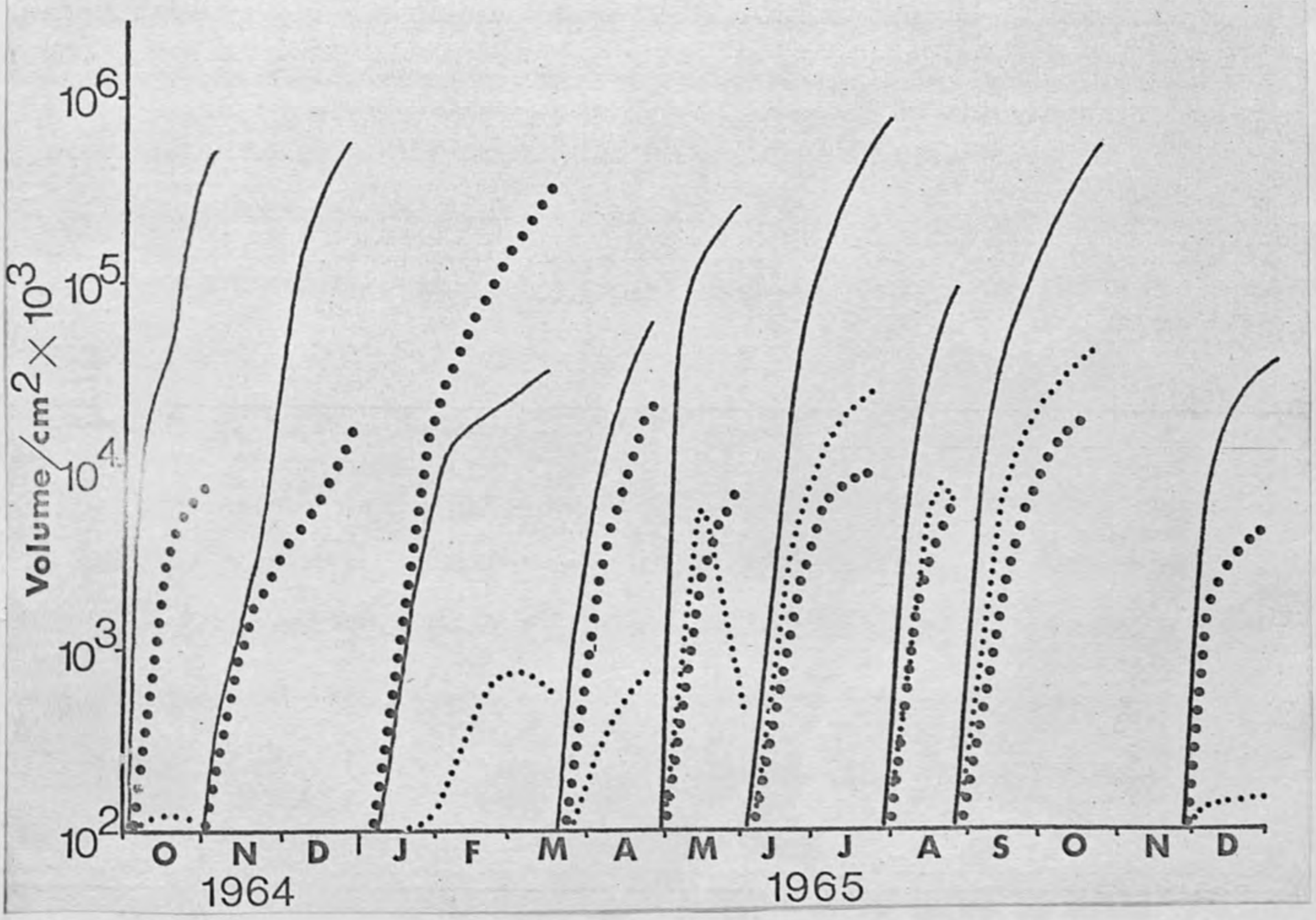


decreased and the filamentous forms increased becoming dominant. From April to early June the pennate forms increased reaching maximum numbers in June and remaining in high numbers until August. Filamentous forms decreased in April and remained at similar, though slightly fluctuating levels, throughout the rest of the period. After the middle of May the other algae, mainly Scenedesmus and Chlamydomonas, increased rapidly reaching highest cell concentrations in early August and mid September. They then decreased rapidly during October and November.

There were several factors which could have influenced both the total growth of algae and the growth of individual species on the slides. From the chemical analyses of the inflow water it seemed probable that major nutrient concentrations were seldom limiting (see Chapter IV) although it should be pointed out that there would be a greater flow of water and dissolved substances past the algae on the sand grains than those on the slides. This is because the sand grains form a porous layer offering less resistance to the flow of water than does a solid glass slide. The two factors probably having the greatest influence on the winter populations were light and temperature. The low cell numbers between November and December occurred at a period of lowest light intensities (see page 40 ). The growth curves were not smooth for either the total algae or for each of the individual groups but showed some irregular fluctuations. These irregular fluctuations were also found by Brook (1954) and Butcher (1946). Butcher, who compared results from static and running water, attributed these irregularities to (i) quickly changing external factors, (ii) the browsing of animals, (iii) the detachment of algal films through the formation of gas bubbles and (iv) the erosive effect of foreign bodies carried by the current. Brook (1954) suggested that only the second and third factors need be considered in relation to the filter bed flora, the others applying to rivers. Filamentous species, especially Melosira varians, Ulothrix tenuissima and Cladophora sp. were observed to become detached from the sand surface and be buoyed up into the supernatant water by gas bubbles, carrying other algae with them. This phenomenon was not, however, directly observed to have occurred on the slides although there was no reason why it should not have done so. The browsing of animals most probably contributed greatly to the fluctuations of the algae on the slides. On occasions large numbers of certain species of algae were removed by animals, mainly Chironomid larvae. It was observed that this removal by animals occurred mainly on the longer exposed slides.

The glass slides provided a clean surface for colonization by algae. The rates of colonization varied throughout the year depending upon the species of algae present and the environmental conditions. Figure 30 gives the volumes of the pennate diatoms, filamentous algae and motile or coccoid unicells or coenobia other than diatoms on exposed slides for various lengths of time throughout the run of the filter bed. The actual rate of colonization is reflected by the slope of the curve, the steeper the slope the faster the rate of

Figure 30. The rates of colonization of glass slides, measured as volumes of cells/cm.$^2$ x 10,$^{-3}$ of the pennate diatoms, filamentous algae and the motile or coccoid unicells or coenobia throughout the period. —— = pennate diatoms, ••• = filamentous algae, ..... = motile or coccoid unicells or coenobia.



colonization.

The rate of colonization of pennate diatoms increased during the spring and reached its maximum in the summer period. This increase in the rate of colonization during the summer was also reported by Brook (1954). After two or three weeks, or when the populations exceeded $10^7 \mu^3/cm^2$, the rate of colonization decreased. Butcher (1946) reported a similar decrease in the rate of colonization after about 20 days in the spring and summer or 30-40 days in the winter. This slowing down occurred as the amount of free space on the slides decreased and the competition for light and dissolved solids increased. The population curve thus tended to level out. The most marked decrease in the rate of colonization occurred in January and February. This was probably due to light limitation as not only were incident radiation levels low, (see page 41), but there was also a large population of filamentous algae present at the same time, competing for nutrients and light, which tended to overshadow the pennate forms.

Except during January and February, filamentous algae either did not colonize the slides or did not increase so rapidly as did the pennate diatoms. The colonization rate of the filamentous forms decreased slightly later than that of the pennate diatoms or the decrease occurred at a lower concentration of cells. This may have been due to a slower growth rate by the filamentous forms or their need for a larger area of clear substratum for the establishment of new filaments. The pennate diatoms, for example, were observed on occasions to form layers on top of each other and to be epiphytic on filamentous forms. Although filaments, once established, could grow upwards away from the slide and into the supernatant water, if they became too long there was a danger that animals or water turbulence would break them off thus reducing the size of the filamentous populations.

The populations of motile or coccoid unicells or coenobia present on the slides (consisting mainly of Scenedesmus spp. and Chlamydomonas sp.) were present only in small numbers throughout the winter and spring. They reached maximum numbers during the summer and early autumn. On three occasions, in early March, late May and mid-August, the populations decreased towards the end of the filter bed run, i.e. with increasing time of exposure. This decrease could be attributed to grazing by animals. Large concentrations of Chironomid larvae and other animals were present, especially in May, on the slides. These animals were observed to have green algae in their guts.

A simple measure of the rate of production of algae on glass slides was suggested by Sladecek and Sladeckova (1963). They divided the amount of standing crop on the slides (expressed as oven-dry weight) by the exposure time in days. This was slightly modified into the following convenient form for the present study:-

$$\text{production rate (uncorrected)} = \frac{\text{volume in } u^3 \times 10^{-3}}{\text{exposure in days}}$$

The term uncorrected production rate was used as no account could be taken of losses from the slides of cells breaking off and being washed away, being buoyed up by gas bubbles and detached, being grazed by animals or lost in any other way. The figure is thus an approximation to the net rate of production. The results for each of the three groups of algae for each of the filter bed runs are given in Table 6.

**Table 6.** The uncorrected Production Rate of the three main groups of algae on glass slides for each of the filter bed runs throughout the period October 1964 to December 1965.

| Period ending | 23.10.64 | 30.12.64 | 11.3.65 | 20.4.65 | 25.5.65 | 17.7.65 | 14.8.65 | 23.10.65 | 18.12.65 |
|---|---|---|---|---|---|---|---|---|---|
| Length of exposure in days | 28 | 63 | 63 | 28 | 28 | 40 | 21 | 63 | 28 |
| Pennate diatoms | 18,000 | 6568 | 690 | 1650 | 4910 | 8290 | 2154 | 6150 | 1066 |
| Filamentous algae | 208 | 375 | 4035 | 555 | 247 | 118 | 344 | 298 | 142 |
| Motile or coccoid unicells or coenobia | 6 | 0 | 8 | 29 | 30 | 3790 | 365 | 289 | 6 |

These results illustrate the seasonal variation in rates of production. The production rates for the filamentous algae and the motile or coccoid unicells or coenobial forms were usually much lower than those for pennate diatoms. The exceptions were the period ending 11.3.65 when the filamentous forms were dominant and the period ending 17.7.65 when the motile or coccoid unicells or coenobial forms became sub-dominant. This index figure can be used to indicate simply the importance, based on production, of any particular group of algae. In the present study pennate diatoms usually had much higher production rates than the other algae present reflecting their dominance throughout most of the period.

Table 7 a, b and c gives the volumes of the major species of algae present on the glass slides of increasing length of exposure during the filter bed run throughout the period of study. Certain species were common and could be regarded as either dominant or co-dominant throughout. Examples of such algae were Cymbella spp. and Melosira varians. Other species became common at various times of the year and, although these altered the species composition of the flora, they tended to be additions to rather than replacements of the basic community. Most of the species of pennate diatoms were present, though at times in small numbers, throughout the entire period. Certain species, however, were absent during some of the months, e.g. Surirella ovata in November and December 1964, Fragilaria capucina in July 1965, Diatoma vulgare in November and December 1964 and Cocconeis placentula during the spring and summer of 1965. Certain other species, which were very common in the epipelic community (see pages 94 to 123 ), were present at irregular intervals on the glass slides. These irregularities were probably a reflection of their preference for the sand surface. The pennate diatoms which occurred in large numbers as additions to the basic community were Synedra ulna in November and December 1964, from April to July 1965 and November to December 1965; Pinnularia spp. in May 1965;

Achnanthes minutissima from April to October 1965; Amphora ovalis in October 1964 and 1965; Fragilaria capucina in December 1964, April to July 1965 and October 1965; Cocconeis placentula in December 1964 and October 1965 and Surirella ovata in April 1965 and July to December 1965. Although these species may, at times, have been co-dominant with Cymbella spp., they never superseded it. Other filamentous forms also occurred throughout the period but these again were merely occasional to the community. The most notable of these additions were Tribonema bombycinum, Cladophora sp. and Stigeoclonium falklandicum during the summer and autumn of 1965. The other algae present, notably Scenedesmus spp. and Chlamydomonas sp., were very common at times becoming co-dominant, e.g. in July 1965, but their presence was probably encouraged by unusual environmental conditions such as the addition of copper sulphate or chlorine to the water (see pages 61 to 72 ). These algae can also be regarded as additions to the basic community.

There also appeared to be little or no succession of algae on the slides with increasing lengths of exposure. Only the concentrations of each species, after their first appearance, increased with time. This situation, of addition and increasing concentration rather than succession of species, conforms to the description of the climax association by Butcher (1946). Only on occasions when conditions were altered artificially, e.g. after the addition of copper sulphate or chlorine to the water, did species other than the climax dominants occur in very large numbers.

Two trial exposures were made, in April and July 1965, of slides at various depths in the supernatant water. The length of exposure was four weeks on each occasion. Little difference in the algal flora of these slides could be found.

<u>Tables 7a,b & c.</u> The volumes of the major species of algae, in $\mu^3 \times 10^3$, present on glass slides of increasing length of exposure during the filter bed run.

1964

| Date | 2.10 | 9.10 | 16.10 | 23.10 | 6.11 | 13.11 | 20.11 | 27.11 | 4.12 | 11.12 | 18.12 | 30.12 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Diatoma vulgare | | | | | | | | | | | | |
| Fragilaria capucina | | p | 100 | 250 | | p | p | 210 | 696 | 1640 | 2800 | 8100 |
| Synedra ulna | 526 | 1440 | 5600 | 4260 | p | 221 | 656 | 1109 | 2997 | 7696 | 20700 | 26750 |
| Cocconeis placentula | | | | | p | 141 | 421 | 734 | 1811 | 3760 | 9100 | 16400 |
| Achnanthes minutissima | p | p | p | 280 | p | p | p | | | p | 111 | 241 |
| Pinnularia spp. | | | | | | | | | | | | |
| Amphora ovalis | p | 265 | 1140 | 8400 | p | p | 183 | 266 | 311 | 371 | 489 | 778 |
| Cymbella spp. | 3110 | 9540 | 18810 | 493800 | 149 | 741 | 2320 | 9764 | 28894 | 49760 | 128000 | 349000 |
| Nitzschia palea | p | p | 121 | 365 | | | | | | p | 121 | 230 |
| N. acicularis | p | p | p | p | p | p | p | p | 160 | 256 | 381 | 771 |
| N. linearis | | | | | p | p | p | p | 108 | 171 | 240 | 130 |
| Surirella ovalis | | | | | | | | | | | | |
| Cladophora sp. | | | | | | | | | | | | |
| Stigeoclonium falklandicum? | | | | | | | | | | | | |
| Melosira varians | p | 990 | 2730 | 7600 | 308 | 326 | 789 | 1676 | 1869 | 3676 | 8270 | 18824 |
| M. granulata | | | | | | | p | 344 | 844 | 1506 | 1414 | 1840 |
| Tribonema bombycinum | | | | | | | | | | | | |
| Chlamydomonas sp. | | | | | | | | | | | | |
| Ankistrodesmus falcatus | | | | | | | | | | | | |
| Scenedesmus spp. | p | p | 100 | 160 | | | | | | | | |
| Tetrastrum staurogeniaeforme | | | | | | | | | | | | |
| Merismopedia glauca | | | | | | | | | | | | |

| | | | 1965 | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| date | 15.1 | 22.1 | 29.1 | 5.2 | 19.2 | 26.2 | 3.3 | 11.3 | 25.3 | 6.4 | 13.4 | 20.4 |
| Diatoma vulgare | | | | | | p | 540 | 2700 | p | 111 | 270 | 576 |
| Fragilaria capucina | p | p | p | p | 237 | 198 | 190 | 290 | | p | 430 | 1492 |
| Synedra ulna | | | | | | | | | p | 318 | 1768 | 5126 |
| Cocconeis placentula | p | p | p | p | p | p | p | p | | | | |
| Achnanthes minutissima | p | p | p | p | p | p | p | 117 | p | 280 | 2000 | 4455 |
| Pinnularia spp. | | | | | | | | | | | | |
| Amphora ovalis | p | p | p | p | | p | | | | | | |
| Cymbella spp. | 129 | 438 | 1630 | 5430 | 8419 | 17410 | 24140 | 30400 | 333 | 980 | 7670 | 14380 |
| Nitzschia palea | p | p | p | p | p | p | p | 320 | | p | 380 | 1090 |
| N. acicularis | | p | p | p | p | p | 108 | 3000 | 110 | 317 | 6780 | 14250 |
| N. linearis | p | p | 184 | 1076 | 1224 | 2720 | 3284 | 5200 | | | 210 | 477 |
| Surirella ovalis | p | p | p | p | p | p | p | 264 | 154 | 648 | 1186 | 3670 |
| Cladophora sp. | | | | | | | | | | | | |
| Stigeoclonium falklandicum? | | | | | | | | | | | | |
| Melosira varians | 134 | 746 | 4500 | 12350 | 25090 | 70800 | 254000 | 264000 | 276 | 1440 | 5840 | 14700 |
| M. granulata | | p | 276 | 470 | 1407 | 270 | 256 | 260 | | | | |
| Tribonema bombycinum | | | | p | p | | | | | | | |
| Chlamydomonas sp. | | | | | | | | | p | p | p | p |
| Ankistrodesmus falcatus | | | | | | p | p | p | p | p | p | p |
| Scenedesmus spp. | | p | 107 | 190 | 260 | 680 | 640 | 535 | | | | |
| Tetrastrum staurogeniaeforme | | | | p | p | p | | | | | | |
| Merismopedia glauca. | | | | | | | | | p | 100 | 180 | 280 |

1965

| Date | 4.5 | 11.5 | 18.5 | 25.5 | 8.6 | 15.6 | 22.6 | 29.6 | 3.7 | 10.7 | 17.7 | 31.7 | 7.8 | 14.8 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Diatoma vulgare | p | 380 | 1580 | 3118 | p | 130 | 224 | 652 | 2159 | 3310 | 4210 | | 200 | 767 |
| Fragilaria capucina | p | 290 | 1384 | 2900 | p | 191 | 470 | 1117 | 3232 | 7198 | 7110 | | | |
| Synedra ulna | 350 | 6780 | 12110 | 17400 | p | 170 | 330 | 760 | 1240 | 6900 | 11640 | 140 | 2760 | 15364 |
| Cocconeis placentula | | | | | | | | | | | | | | |
| Achnanthes minutissima | 580 | 3000 | 7760 | 15780 | p | 200 | 800 | 1500 | 4100 | 8960 | 11000 | 370 | 1870 | 14870 |
| Pinnularia spp. | 260 | 1000 | 3110 | 6200 | p | p | | | | | | | | |
| Amphora ovalis | | | | | | | | | | | | | | |
| Cymbella spp. | 367 | 7767 | 15400 | 67800 | 576 | 1200 | 4200 | 15870 | 76670 | 260000 | 393000 | 710 | 1990 | 4040 |
| Nitzschia palea | 310 | 1990 | 3898 | 6810 | p | 176 | 368 | 967 | 1461 | 5671 | 8760 | 160 | 326 | 1485 |
| N. acicularis | 610 | 1300 | 2810 | 3000 | | | p | p | p | p | p | 266 | 770 | 7315 |
| N. linearis | 228 | 770 | 4770 | 11477 | | p | 241 | 777 | 1187 | 4532 | 17190 | | p | 310 |
| Surirella ovalis | | | | | p | p | 160 | 640 | 1190 | 2610 | 3980 | | p | 270 |
| Cladophora sp. | | | | | | | | | | | | 108 | 580 | 2144 |
| Stigeoclonium falklandicum? | | | p | 240 | | | p | p | 230 | 770 | 1210 | 126 | 860 | 2164 |
| Melosira varians | 156 | 720 | 2000 | 6484 | p | 111 | 200 | 964 | 1670 | 2800 | 3190 | p | p | 774 |
| M. granulata | | | | 240 | | | | | | | | | | |
| Tribonema bombycinum | | p | p | 180 | | p | 380 | 460 | 1110 | 2211 | 4220 | | 260 | 1080 |
| Chlamydomonas sp. | | | | | 490 | 1340 | 6420 | 19660 | 60100 | 101000 | 128000 | 108 | 6016 | 3060 |
| Ankistrodesmus falcatus | | | | | | | | | | | | | | |
| Scenedesmus spp. | 294 | 1880 | 6644 | 800 | 168 | 476 | 1798 | 5760 | 16000 | 58800 | 84000 | 410 | 2470 | 4185 |
| Tetrastrum staurogeniaeforme | p | 100 | 260 | p | | | | | | | | | | |
| Merismopedia glauca | | | | | | | | | | | | | | 280 |

D. Distribution of algae on the sand surface.

During 1965 two surveys of the overall distribution of algae on sand surfaces were carried out, on May 15th on bed 12 and on July 24th on bed 13. Samples of the filter skin of known area (10cms.$^2$) were obtained from more than twenty places in each bed and the numbers and species of algae present determined. The overall distribution of algae on the sand surface of bed 12 on May 15th is given in figure 31 a, b and c. The results are expressed as calculated volumes per cm.$^2$ x $10^{-6}$. Figure 31 a. represents algae normally planktonic in habitat, 31 b. non-filamentous mainly free living pennate diatoms and 31 c. filamentous algae (mainly Ulothrix tenuissima).

The main planktonic constituents were Stephanodiscus astraea, S. hantzschii, Asterionella formosa, Ankistrodesmus falcatus and Rhodomonas minuta, and these increased in number towards the centre of the bed and away from the entrance. The numbers then decreased towards the far end. There was a broad band across the centre of the bed from side to side of high cell concentrations. This could be accounted for by considering the flow of water through the bed. The water passes into the bed through relatively narrow inlet. The velocity of the water is then reduced as the incoming water mixes with the water already there. If the minimum critical velocity for keeping these algae in suspension was reached near the centre of the bed maximum deposition would take place there and then decrease further from the centre.

The non-filamentous free living forms consisted mostly of Nitzschia acicularis, N. linearis, N. palea, species of Cymbella and Surirella and species of Scenedesmus. These algae were not usually present in large numbers in the incoming water and their presence on the sand surface in high concentrations is due to colonization and population growth. Some of these algae (e.g. species of Nitzschia) are motile and can glide over the sand surface and could be expected to concentrate where optimum growth conditions prevailed. The lowest numbers were near to the inlet possibly due to the scouring action of the incoming water on the sand surface. The highest numbers were present in a band stretching from the centre of the west side of the bed towards the far end and across the middle. This band corresponded to areas of low numbers in the planktonic and filamentous forms. This was particularly true in the N.E. corner where there were only small numbers of non-filamentous but very high numbers of filamentous forms. Immediately alongside, to the west, was an area of high numbers of non-filamentous and correspondingly low numbers of filamentous forms. Conditions were not favourable for the non-filamentous forms on the sand surface amongst the filamentous forms probably due to competition for nutrients and also because of shading causing light limitation (see also page 41 ).

Ulothrix tenuissima was the main filamentous form present. Some U. tenerrima also occurred and near to the edges of the bed Melosira varians was present in high numbers. The highest concentrations of filamentous forms occurred in the corners at the north end and in a band across the centre. Again there were low numbers at the entrance to the bed and at the centre of the north end. One reason for larger numbers being present in the corners is because detached fragments of

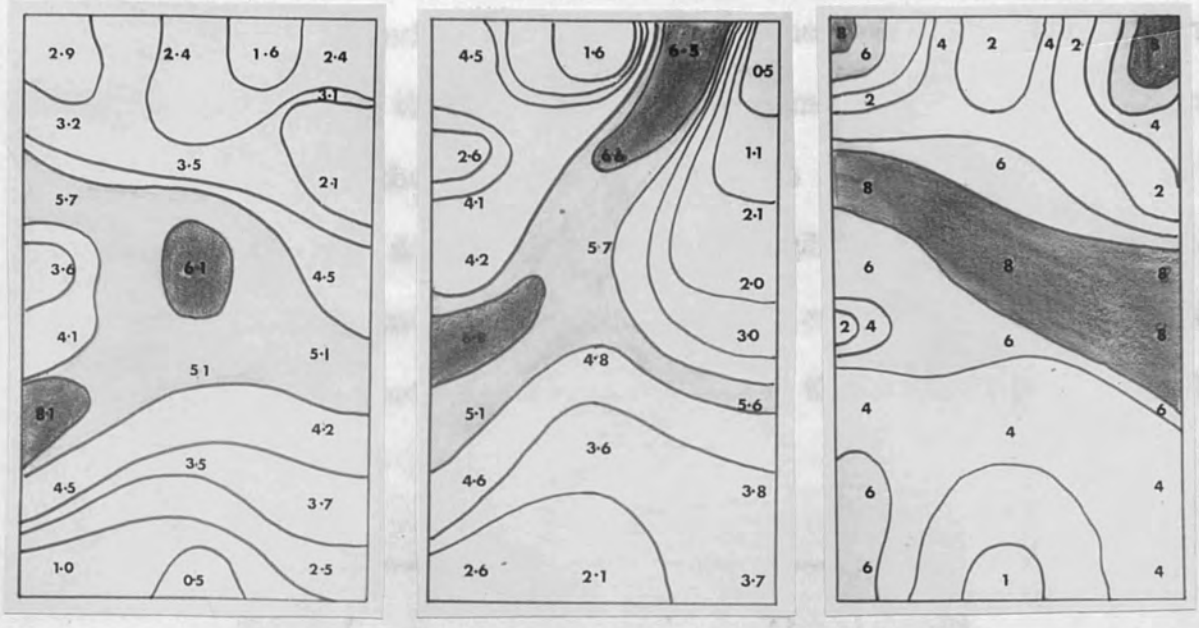



__Figure 31a,b & c.__ The distribution of algae on the sand surface of bed 12 on May 15th 1965. The figures represent the calculated algal volume / $cm^2 x 10^6$. The filter bed inflow is at the bottom of the figure.

(data from the Met. Office Kew Observatory). This had a considerable effect on the currents in the supernatant water. This effect was investigated by following the path of polythene bottles floating just below the surface as the bed was drained down. The general pattern of the currents, on the surface at least, is given in figure 32. The area enclosed by the dotted line was calm and shaded from the wind. Four bottles were launched just west of the entrance. Their passage across the bed is represented by arrows and their final positions by the numbers 1 to 4.

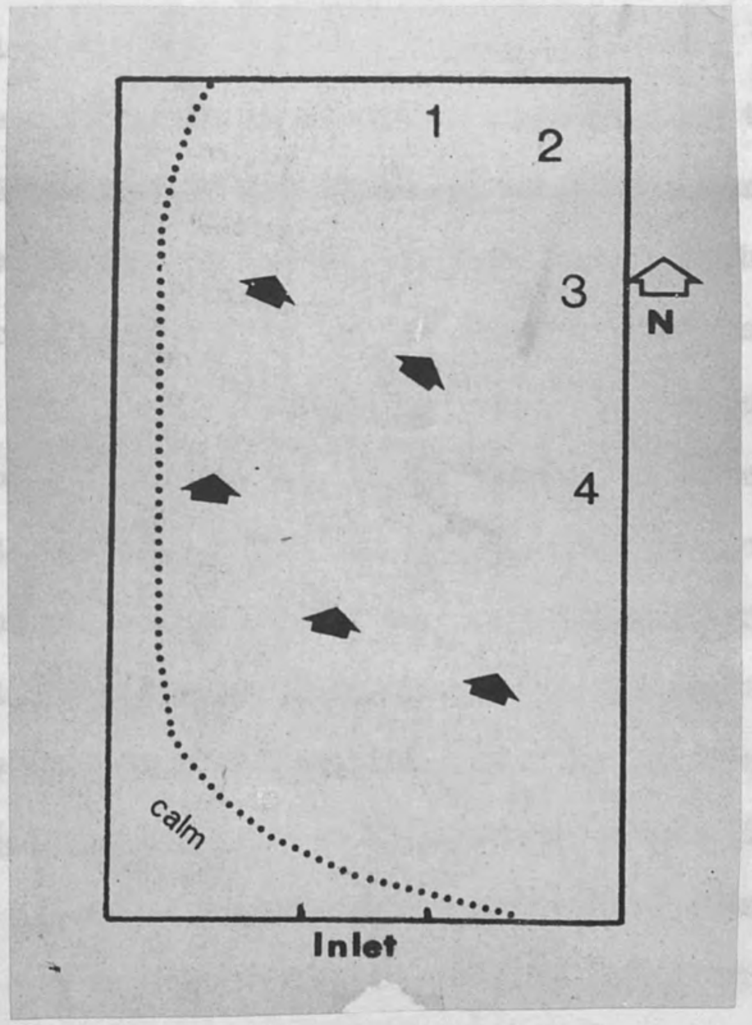



Figure 32. The current directions at the surface of the supernatant water in bed 13 on July 24th, 1965.

The distribution of filamentous algae, mainly Ulothrix tenuissima, was plotted by drawing the observed patches of green filaments on the sand surface. Their distribution is given in Figure 33 c. The shaded areas are patches of filaments and the calculated cell volumes /cm$^2$ are superimposed. Figures 33 a. and b. give the results for planktonic and non-filamentous mainly pennate bottom living forms respectively. The filamentous algae were present in much higher numbers in the sheltered part of the bed. The patches indicated that the wind blowing obliquely across the bed (see Figure 32) may have induced a clockwise motion in the supernatant water. The straggling filaments were deposited in a circular manner around the bed. The wind induced the flow of water towards the N.E. corner with probably a resultant return current towards the S.E. corner. This was supported by the observed surface flow and the directions of the deposited filaments. The planktonic forms were again most concentrated near the middle of the bed but this time displaced to the east. Planktonic forms were less dense at each end, very similar to the pattern observed in bed 12 and might also be explained by sedimentation currents, in this case displaced eastwards by the wind. The non-filamentous free living forms (Figure 33 b.) consisted of the same species as before but Nitzschia acicularis formed a larger percentage (85%) of the population. The highest concentrations occurred near the centre and in the N.E. corner and the lowest concentrations at the entrance and the N. end. The largest numbers were in between the areas of maximum filaments and maximum plankton deposition.

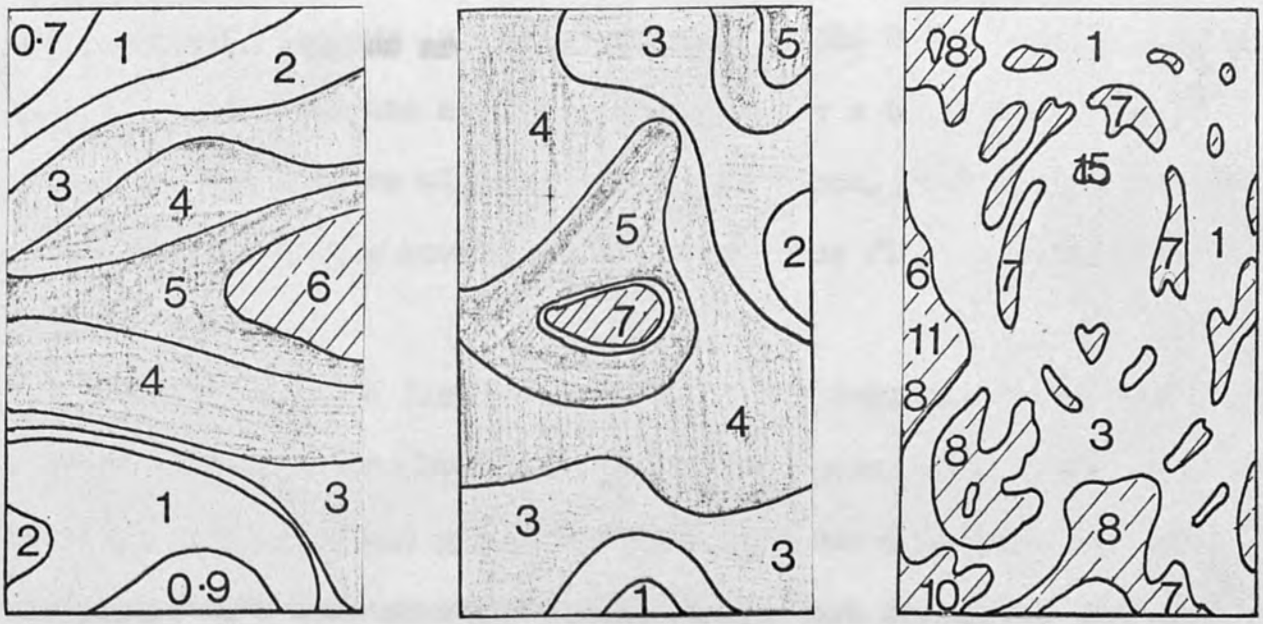



Figure 33 a. b. and c. The distribution of algae on the sand surface of bed 13 on July 24th, 1965. The figures represent the calculated algal volume per $cm^2 \times 10^{-6}$.

These two surveys show that the sand surface of the slow sand filter beds was by no means homogeneous as found by Brook (1954). Populations of algae would develop depending upon the deposition of cells from the incoming water or the relative favourability of the local environment. Deposition of planktonic algae, which did not seem to increase in cell numbers on the sand surface to any great extent, probably depended upon sedimentation currents, whilst filamentous algae growing from the bottom broke away and were redistributed by wind-induced currents. Non-filamentous free living algae tended to accumulate away from the entrance and in areas where numbers of other algae were lower. This uneven distribution emphasises the need for composite sampling of sand surface organisms (see Chapter III) if representative samples are to be obtained.

## VIII. A Polythene Bag Experiment.

During the investigations on slow sand filter beds difficulty was experienced in attempting to relate algal successions with the large measured changes in nutrient levels in the water (see Chapter V). This was mainly because slow sand filters have a continuous flow through them, thus the algae on the sand surface, even though they themselves may not be moving, have a continuous flow of water and nutrients past them.

In an attempt to find out more about the requirements of the algae a suspension of filter bed algae in natural water was enclosed in a polythene bag and their growth followed for over a month. The bag was placed in a slow sand filter bed for 33 days during May and June. Samples of the unattached algae on the sand surface in the polythene bag were collected at varying intervals throughout the experiment.

Practical considerations ruled out the use of a large bag as handling it would have required more than one person. A bag one metre in diameter and two metres in length and of 800 L capacity was used. It was accepted that this size system is open to criticism concerning wall effects, shading etc. and these factors have been taken into account when considering the results.

The construction of the bag and its method of suspension are given in Figure 34.a.

The bag was suspended from the end of a rod stretching out three feet from the bank. The aspect was southerly. The bag was attached to the rod by means of a double funnel. The neck of the bag fitted

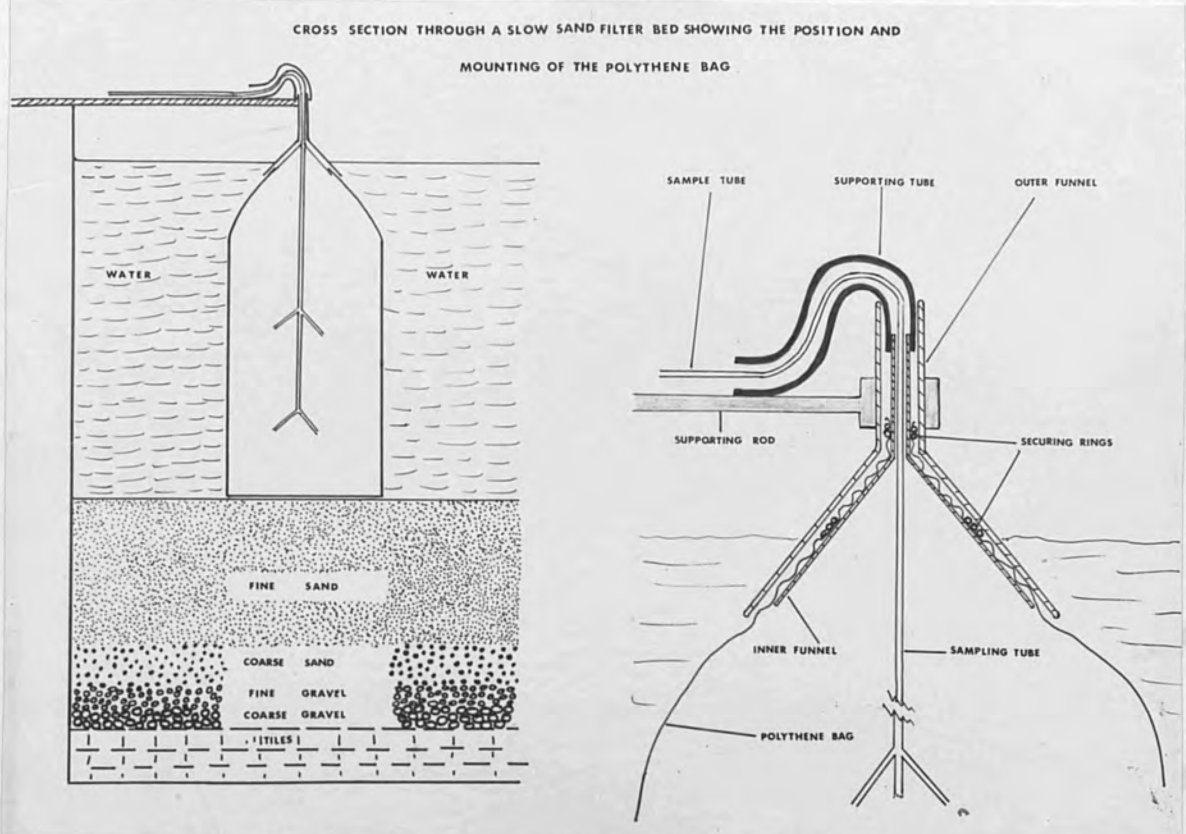



Figure 34.a.

over the smaller inner funnel and was attached to it. This smaller funnel was then firmly pushed inside the larger outer funnel and fixed in place by means of the support tube. The bag was thus fixed in place between the two funnels and was protected from the surface wave action by the outer one which just dipped below the water surface. The double funnel arrangement also served to reduce the exposure of the enclosed water to atmospheric pollution. The sampling and stirring tube was passed through the supporting tube and through the inner funnel into the bag. The bag was suspended so that the bottom was just above the sand surface.

The bag was filled with glass fibre filtered, filter bed supernatant water free from plants and animals. It was then innoculated with oOL of coarse filtered filter bed water free from larger organisms such as crustations and larger rotifers. The innoculum contained various diatoms, Chlorophyceae, Cyanophyceae, euglenoids and cryptomonads. Before each sample was removed the contents were well mixed by bubbling air through the bag. Samples were removed by means of a suction pump and chemically analysed (see Chapter III). Algal numbers were estimated using the membrane filter technique described by McNabb (1960).

The experiment was terminated after 33 days as the filter bed was drained for cleaning purposes.

The temperature of the water was 9.5°C at the beginning of the experiment and had increased to 13.0°C by the end. Between days 12 and 18 growths of epiphytic diatoms and filamentous green algae appeared on the outside of the bag. These were eventually removed by brushing but during this period the penetration of light into the bag would have been reduced.

The concentrations of NO3-N, NH4-N and PO4-P passing onto the slow sand filter bed did not vary greatly throughout the period and levels were at all times constantly high (Figure 34b.) Silicon concentrations ranged between 67-100 µg. at./L (4-6mg./L).

In the polythene bag NO3-N, PO4-P and Si concentrations were initially high being similar to the filter bed water from which it was filled. After an initial increase of 95µg. at./L, NO3-N decreased throughout the experiment. After the fifth day the PO4-P concentration also decreased but more rapidly than NO3-N.

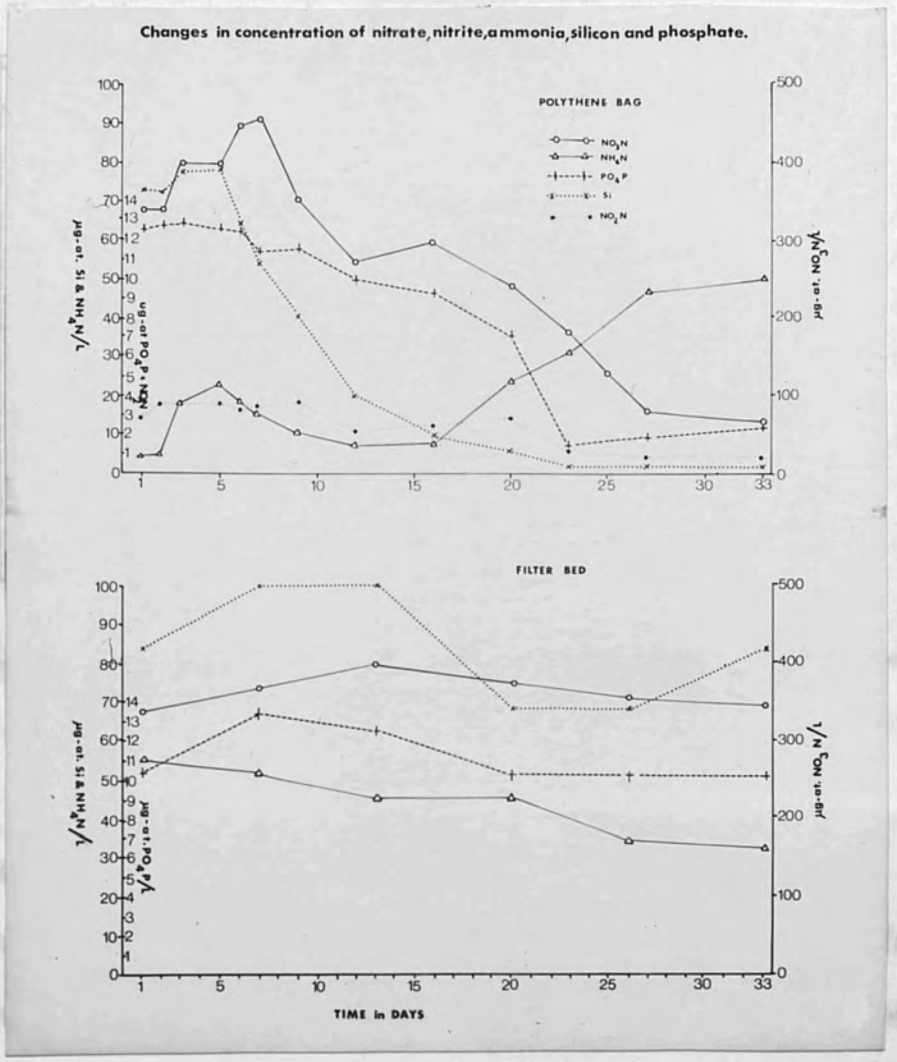



Figure 34.b.

After day 23 PO4-P reached its lowest level although there was a slight increase in concentration from day 25 to the end of the experiment. NH4-N initially increased until day 5 then slowly decreased until day 12. After day 16 it rose steadily in concentration until the end of the experiment. NO2-N remained at a constantly low level throughout.

The initial innocumul of algae into the polythene bag and also those passing onto the filter bed contained 90% by algal volume and

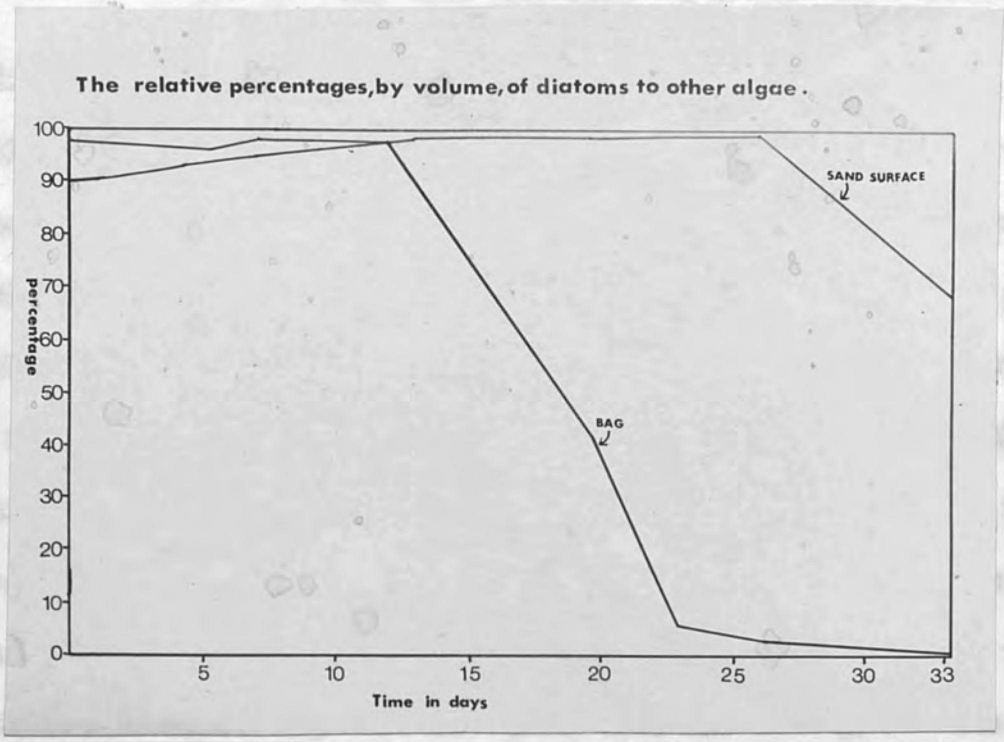



Figure 34 c.

60% by number of diatoms.

Throughout the period of the experiment the sand surface was dominated by diatoms (Figure 34c) although about 30% of the total volume consisted of filamentous green algae and blue green algae by day 33.

The flora of the polythene bag, although initially diatom dominated changed between days 13-23 to complete green algal domination. In the bag the diatom population did not increase after day 20, indeed apart from Surirella ovata the diatom species present had reached their maximum by or before day 12 (Figure 34d). Stephanodiscus astraea did not initially increase and after day 7 it decreased rapidly. This was probably due to unfavourable environmental conditions. A small, still body of water, as existed in the polythene bag, allowed planktonic diatoms to settle out. At the bottom of the bag they formed a self shading sediment probably with local nutrient limitations. Silicon levels were below 9µg. at./L after day 12 and by day 20 they had fallen to 5µg. at./L. in the polythene bag. These low concentrations of silica were probably the cause of the decreases in other diatoms in the polythene bag after day 20. Another factor might have been the reduced light intensity due to the external growths on the bag which occurred between days 12 and 18. The light inside the bag was found to have been reduced from about 60% to 30% of the surface intensity. Ryther (1956) has shown that green algae are better adapted to photosynthesis at lower light intensities than are diatoms. The green algae may thus have increased in the bag as they were better adapted to the reduction in light intensity.

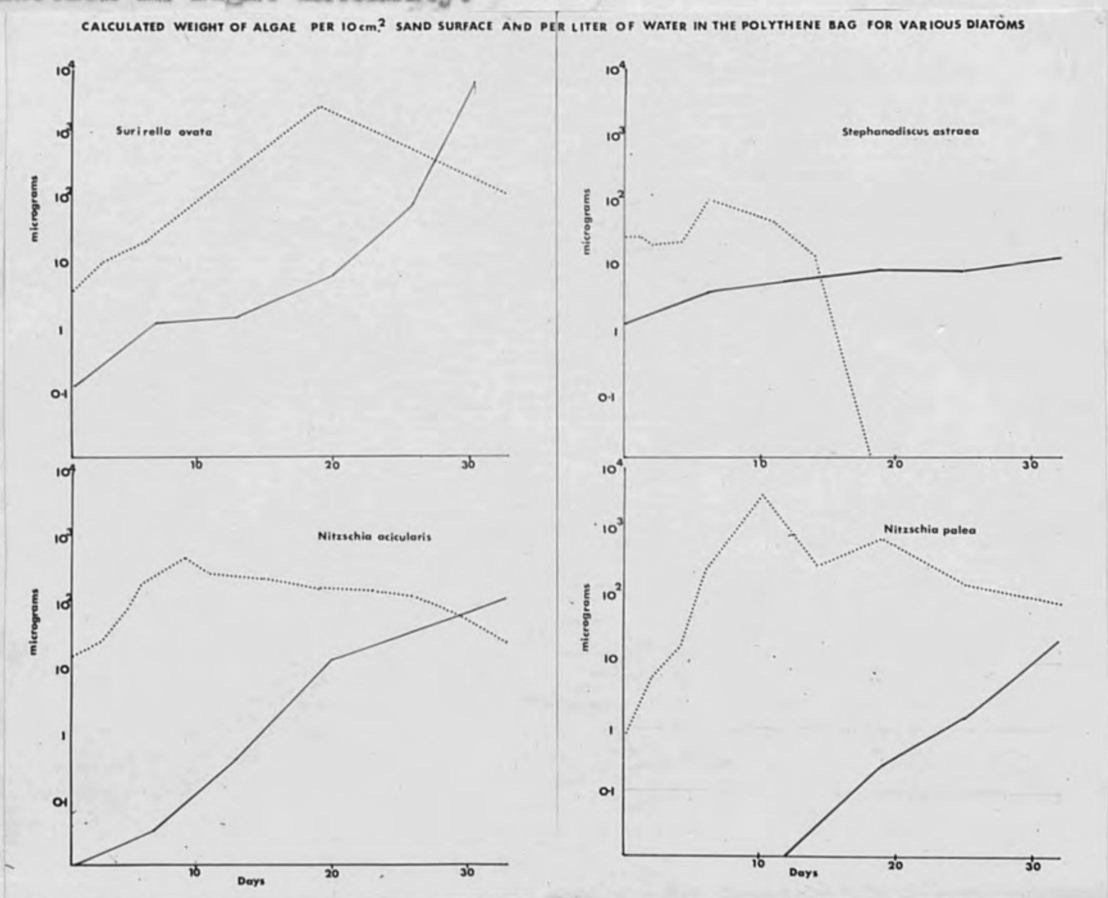



Figure 34d. Solid line represents the algae on the sand surface;broken line represents the algae in the polythene bag.

Diatoms on the sand surface would not have suffered from nutrient limitation during this period. Self shading and subsequent light limitation could have occurred but there was no evidence for this. The numbers of individuals per square centimetre increased until the end of the experiment.

Certain species of algae occurred in large numbers in either the polythene bag or on the sand surface but not both. The growth of

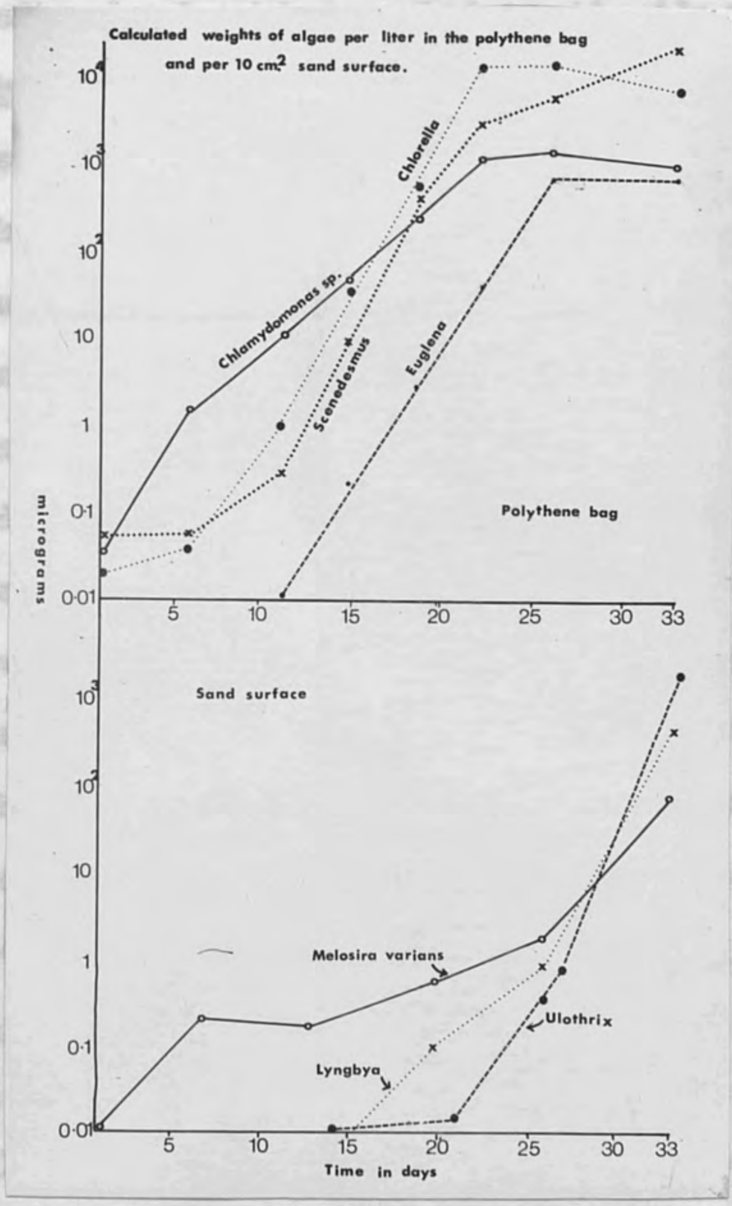



Figure 35.

these is indicated in Figure 35. By day 25, certain filamentous algae established themselves and increased rapidly on the sand surface until day 33. By this time they accounted for 30% by calculated volume of the total sand surface flora. The two major forms present were a species of Lyngbya and Ulothrix tenerrema. They became established only after there had been time for organic matter to build up on the sand surface. In the polythene bag green algae started to increase after day 5 and had reached considerable numbers by day 20. Neither Chlamydomonas sp. nor Chlorella sp. showed marked increases after this day although the calculated weight of the latter was ten times higher than that of the former. Scenedesmus spp. continued to increase up to the end of the experiment.

A species of Euglena increased steadily until day 26 as the organic matter increased. Although no measurements were made, it is considered that dissolved organic matter also increased providing substrates for heterotrophic growth.

Lefevre (1950) proposed that when a particular species of alga multiplies abundantly it might secrete active substances which might prevent the growth of other forms and it is not until the population is reduced or dies off that other resistant species can flourish. Rice (1954) reported natural antagonism between Nitzschia palea and Chlorella sp. and such a system may have operated in the bag although the green algae started to increase even while the numbers of diatoms were still high. Another possible reason for the rapid increase in green algae is that the diatoms might have released accessory growth factors, by excretion or upon dying, which were necessary for the growth of the green algae. Stimulation by organic growth factors of Scenedesmus quadricauda and Chlorella was referred to by Saunders (1957) but without giving clear reference to the original work. The substances (Chlorellin and Scenedesmin) were reported to be self stimulating to the algae secreting them at low concentrations but self inhibiting at high concentrations.

The minimum calculated doubling time of the major species of diatoms in the polythene bag was, with the exception of Nitzschia acicularis, usually less than one half that of the same species on the sand surface. N. acicularis was about the same i.e. 41-46 hrs. N. palea and the species of Cymbella had very short doubling times

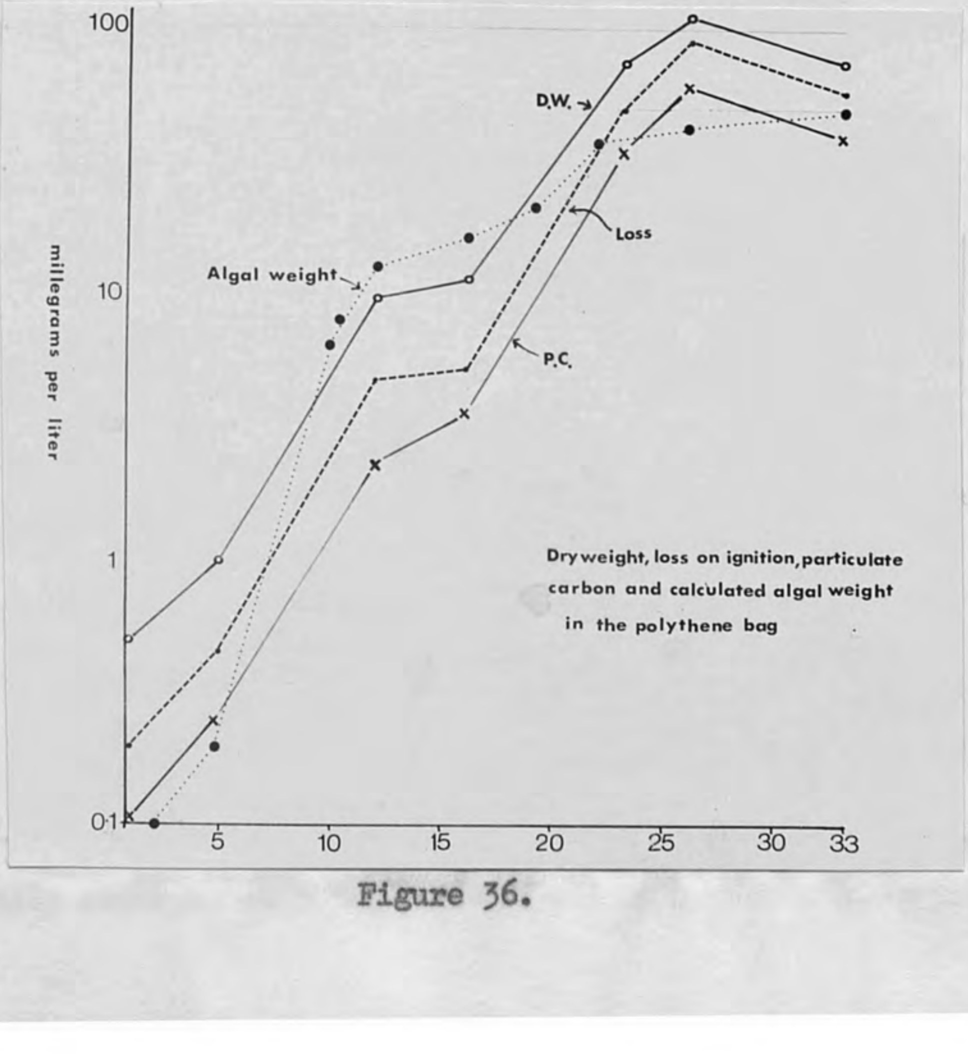



Figure 36.

in the bag of less than 24 hrs. The green algae both on the sand surface and in the polythene bag all had minimum doubling times of less than 24 hrs.

Difficulty was experienced in determining the amount of algal material present on the sand surface and in the polythene bag by means other than counting. This was due to accumulations of algal debris and, on the sand surface, other organic matter. The relation between the weight of algae calculated from their volume, particulate carbon, dry weight and loss on ignition in the polythene bag are given in Figure 36. Although the last three show a close correlation the calculated weight of the algae deviated greatly. The percentage loss on ignition ranged from 48 to 48 during the period of diatom and 70 to 77 during the green algal dominance (Figure 33).

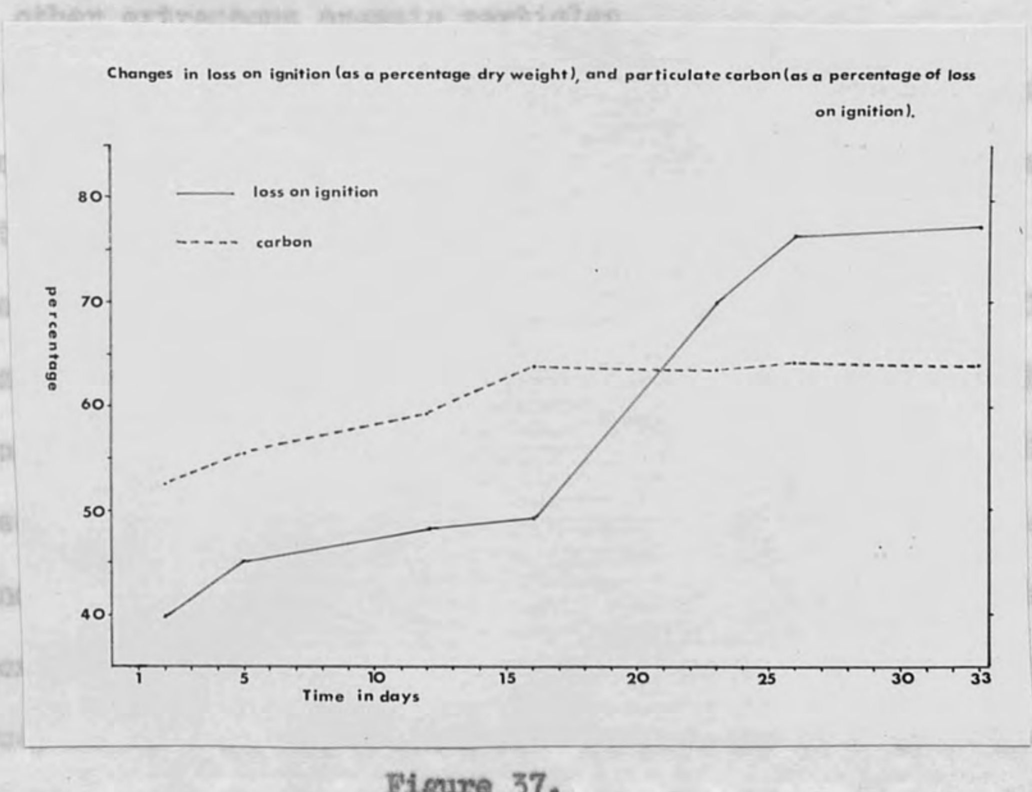



Figure 37.

The percentage carbon of the ash free dry weight was 50 to 59 for the diatom and 60 to 64 for the green algal dominance. These figures are in agreement with those of other workers on diatoms (Lund, 1965) and for green algae (Burlew, 1964).

In the polythene bag, bacterial development was high especially towards the end of the experiment when large amounts of organic matter were present. The bacterial population was determined to be at least 100,000 cells/ml. by day 30. This could contribute up to 10mg./L wet weight. Also large numbers of empty diatom frustules had accumulated by the end of the experiment together with organic debris. As the calculated weight of algae was based only on live cells present considerable discrepancies would be expected. Algal weights calculated from counts would only seem to be valid where there are no accumulations of other extraneous organic particles.

The results from the experiment indicate that, during the period studied, the algae on the sand surface did not suffer from major nutrient limitation, unlike the algae in the polythene bag. This was probably the major reason why the flora of the sand surface remained diatom dominated throughout. This also applied to the sand surface flora throughout most of the period of study (see Chapter VII). Species other than diatoms only started to contribute obviously to the sand surface flora as the amount of biological material present increased. This was also found to be true throughout the rest of the study on the slow sand filter beds when filamentous green algae were usually abundant only towards the end of a filter bed run (see

Chapter VII and Ridley, 1967). If the diatoms were prevented from growing, as in the polythene bag due to nutrient depletion or on the sand surface by copper sulphate applications or infiltration of chlorine, green algae succeeded in dominating the flora.

IX. Culture Experiments

(a) Toxicity of Copper Sulphate.

During March and April 1964, filter bed No. 12 was treated with copper sulphate for 8 hrs. a day to give a final concentration of 0.5 mg./L. The overall effect of this treatment on the algae in the filter bed are discussed elsewhere (see Chapter VII) but during this period some detailed laboratory experiments were carried out to determine the toxicity of various concentrations of copper sulphate to several species of algae.

Several concentrations of copper sulphate were added to laboratory cultures of Asterionella formosa, Fragilaria crotonensis and Scenedesmus dimorphus. The cultures had previously been grown in glass fibre filtered Queen Mary Reservoir water which had been steam sterilised. To this water was added 1% soil extract solution.

Five sets of six 250ml. erlenmeyer flasks, each containing 140 mls. of culture solution (in this case glass fibre filtered filter bed supernatant water which had been steam sterilised and was known to have less than 0.05mg./L copper present) and 10 mls. of algal suspension to give a final concentration of about 1,000 cells/ml., were placed under controlled conditions of light and temperature (two 40 watt fluorescent tubes placed one foot away and a temperature of 18°C). Copper sulphate was then added to give final concentrations of 0.1, 0.2, 0.5 and 1.0 mg./L copper in four of the five sets. The fifth set was used as a control. Growth was measured by filtering the contents of a flask through a glass fibre filter paper (Whatman GF/C) and then determining the loss on ignition. Determinations were made after 2, 7, 15, 24, 29 and 35 days.

The results are given in Figures 38, 39 and 40.

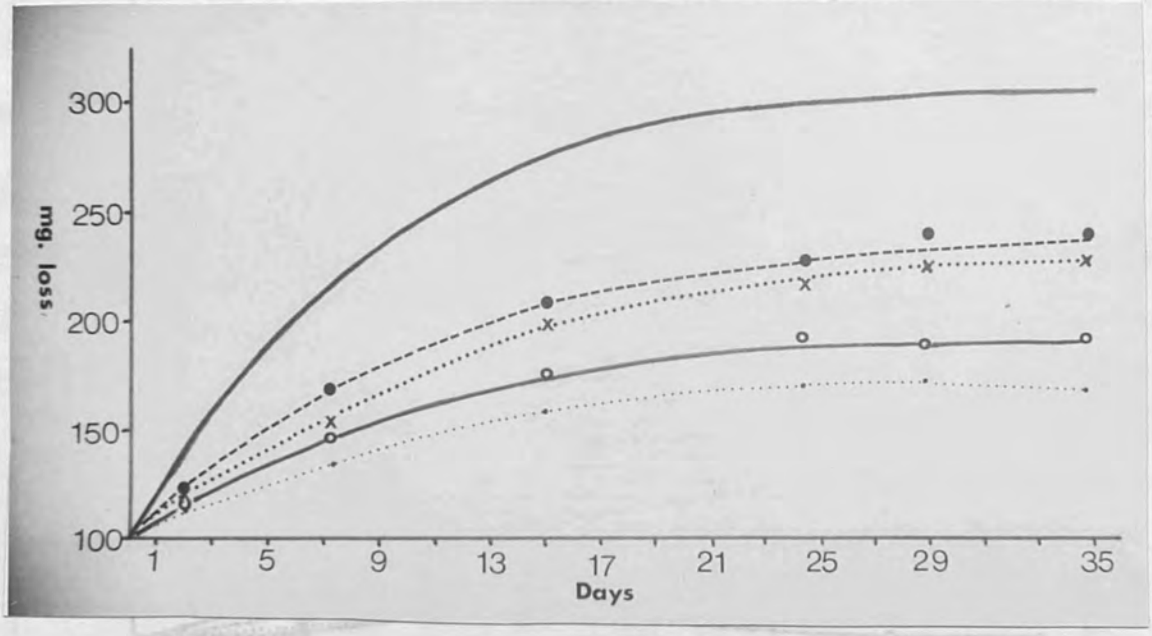



Figure 38. The growth of Scenedesmus dimorphus at different concentrations of copper sulphate. ——— = control, •--• = 0.1mg/L, x·····x = 0.2 mg./L, o—o = 0.5mg./L, •·····• = 1.0 mg./L.

At none of the concentrations used was Scenedesmus dimorphus prevented from growing (Figure 38). The addition of copper sulphate slowed down the rate of growth and also decreased the final yield per culture. The growth curves at 0.1 mg./L and 0.2 mg./L were similar in shape and close to each other as were those at 0.5 mg./L and 1.0 mg./L. The final yields were about two thirds for 0.1 mg./L and 0.2 mg./L and one third for 0.5 mg./L and 1.0 mg./L of that of the control.

Fragilaria crotonensis (Figure 39), unlike Scenedesmus dimorphus, showed little or no growth in the presence of copper above 0.1 mg./L. At 0.2 mg./L there was a small increase but at 0.5 mg./L, after an

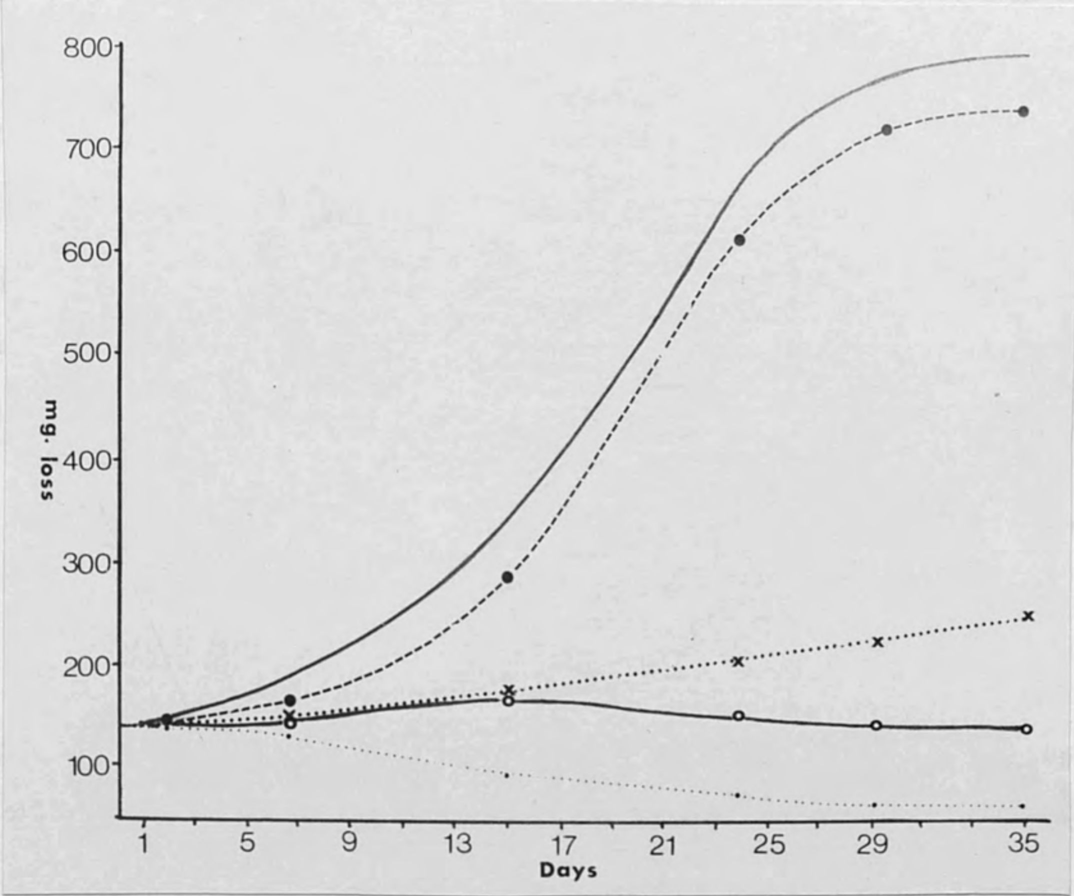



Figure 39. The growth of Fragilaria crotonensis at different concentrations of copper sulphate. —— = control, •---• = 0.1 mg/L, ×·····× = 0.2 mg/L, o—o = mg/L, •·····• = 1.0 mg/L.

initial increase, the final yield was less than the innoculum on day 1. At 1.0 mg./L there was no growth but on the contrary a steady decrease in biomass occurred from day 1 onwards. Growth at 0.1 mg./L was very similar and only slightly less than the control. The initial rate of increase was slightly less as was the total yield.

Asterionella formosa (Figure 40) showed no growth at 0.2 mg./L and decreases at 0.5 mg./L and 1.0 mg./L. After an initial lag phase at 0.1 mg./L the growth increased after day 13 to give a final yield of

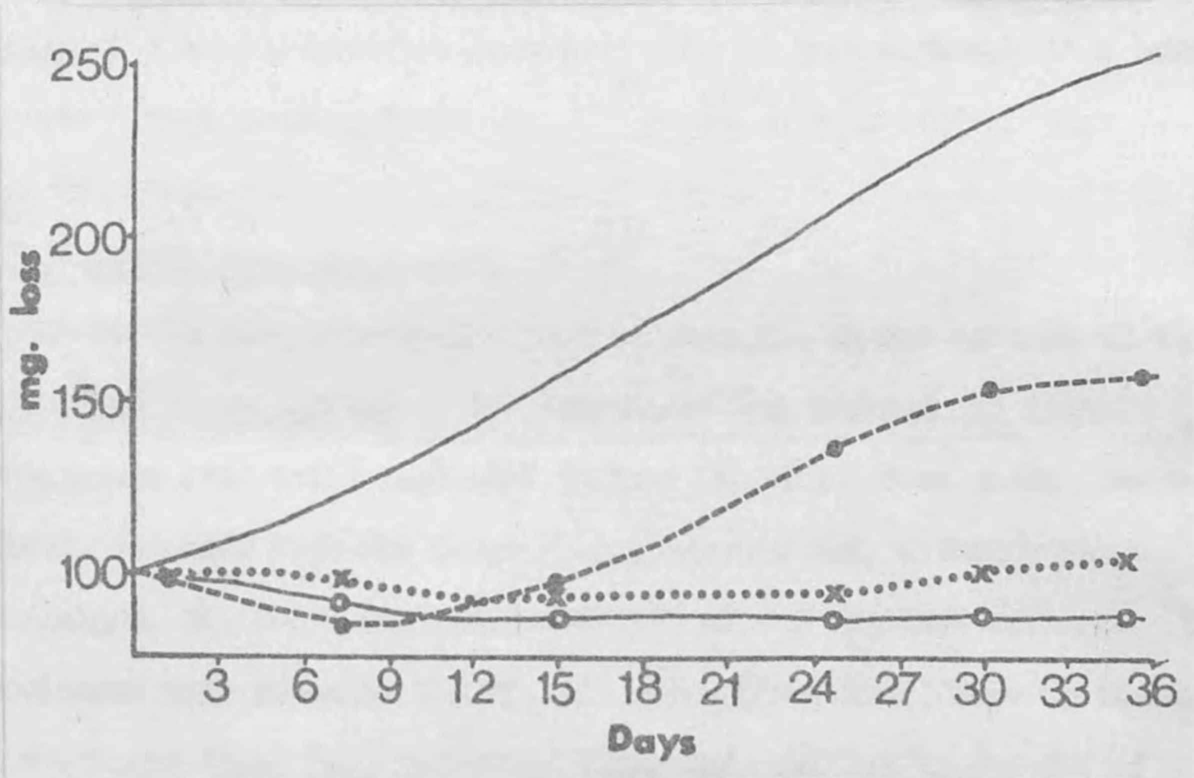



Figure 40. The growth of Asterionella formosa at different concentrations of copper sulphate. = control, = 0.1 mg./L, = 0.2 mg./L, = 0.5 & 1.0 mg./L.

just under half that of the control. The growth rate in culture was much less for Asterionella formosa than for the other two algae. Unlike the previous two algae the growth curve for the control was still showing evidence of growth, albeit slowly, after 35 days. The growth at 0.1 mg./L was also still increasing at 35 days and the final yield could conceivably have been much higher at this concentration.

In this experiment the two diatoms showed little or no growth above 0.1 mg./L copper whilst the green alga was able to increase at all concentrations tested. The final concentration of copper in the water of the filter bed would have been about 0.19 mg./L, enough to virtually prevent the two diatoms from increasing. Scenedesmus dimorphus would, however, have been able to grow although at a lower rate. (see Chapter VII).

(b) Growth Rate Experiments

Growth rate experiments were carried out on one species of alga, Stephanodiscus astraea. The experiment was carried out from September 1964 until September 1965. The algae were grown from net hauls obtained from the Queen Mary Reservoir and, although not unialgal, the hauls contained over 95% of the required species. The cultures were grown in Chu No. 10 medium (Chu, 1942) made up in steam sterilised Queen Mary Reservoir water and modified by the use of the iron source as was recommended by Rodhe (1948) and also the inclusion of 1% soil extract.

The cultures were grown at a temperature within 5°C of the proposed exposure temperature. The algae were exposed in 250 ml. round bottomed glass stoppered flasks which were lowered onto the sand surface of a filter bed on an aluminium framework. The concentration of cells in the bottle at the time of exposure was about 100-200 cells/ml. Three bottles were carried on each framework. These were shaken once every two days to avoid stagnation of the cultures. After seven days the bottles were removed and the samples fixed in Lugol's iodine for inverted microscope counting.

The relative growth rate k' was calculated for each occasion using the formula:

$$k' = \frac{\log N - \log No}{t}$$

where No = original number and N = number after time t in days.

<u>Stephanodiscus astraea</u> showed a distinct seasonal variation. The results are given in Figures 41 and 42; in the former, k' is plotted against photosynthetic radiation (see Chapter V) and in the latter against the water temperature. Each point on the graphs represents the arithmetic mean of the increase in numbers in each of the three light bottles in the seven days. The line in both figures can be divided into three sections; 1. September 1964 to January 1965; 2. January 1965 to April 1965, and 3. April 1965 to September 1965. Figure 41 shows the relationship between photosynthetic potential, as indicated by the amount of photosynthetic radiation available at 1.5 metres depth, and k'. It is doubtful that light intensities were ever so high as to inhibit growth during the period of study.

During September 1964 to January 1965 (line 1) there was a steady decrease in k' and in the photosynthetic radiation. From January 1965 to April 1965 (line 2) although the photosynthetic radiation

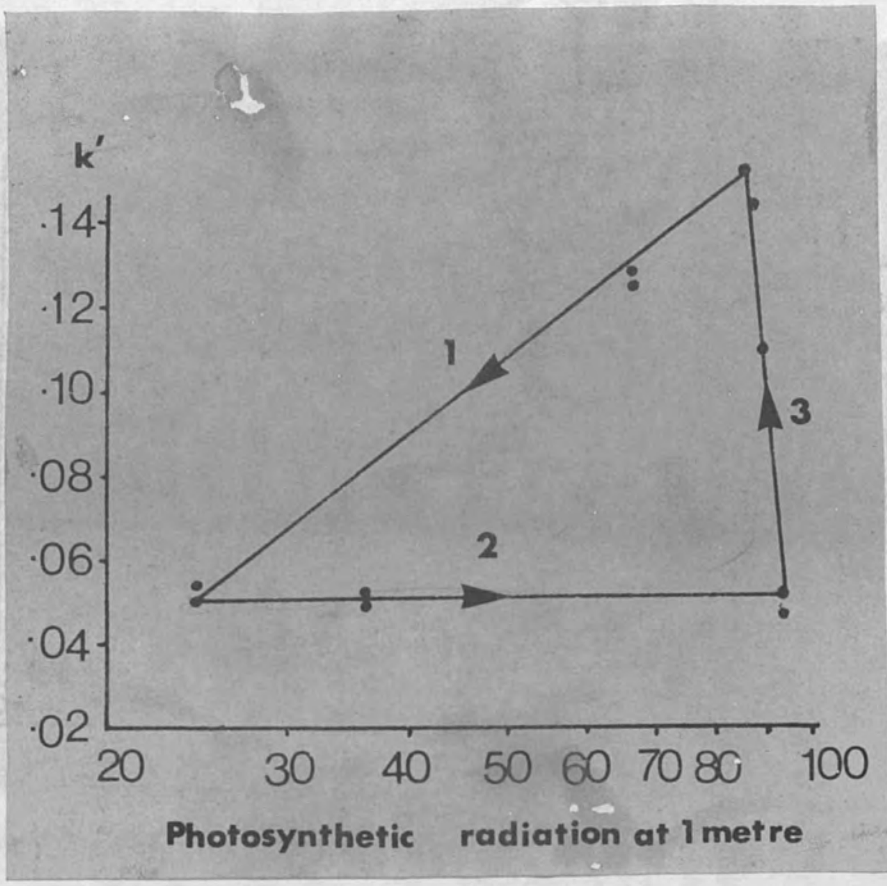



Figure 41. The variation of the relative growth constant, k' with the photosynthetic radiation.

increased considerably, k' remained fairly constant. Between April 1965 and September 1965 (line 3) k' increased rapidly even though there was a slight decrease in photosynthetic radiation. There is a somewhat similar relationship between k' and temperature (Figure 42). As the temperature decreased between September 1964 and January 1965 (line 1) so did k'. It then remained steady until April 1965 (line 2) although the temperature increased. There was then a rapid rise in k' until September 1965 (line 3) associated with a further increase in temperature.

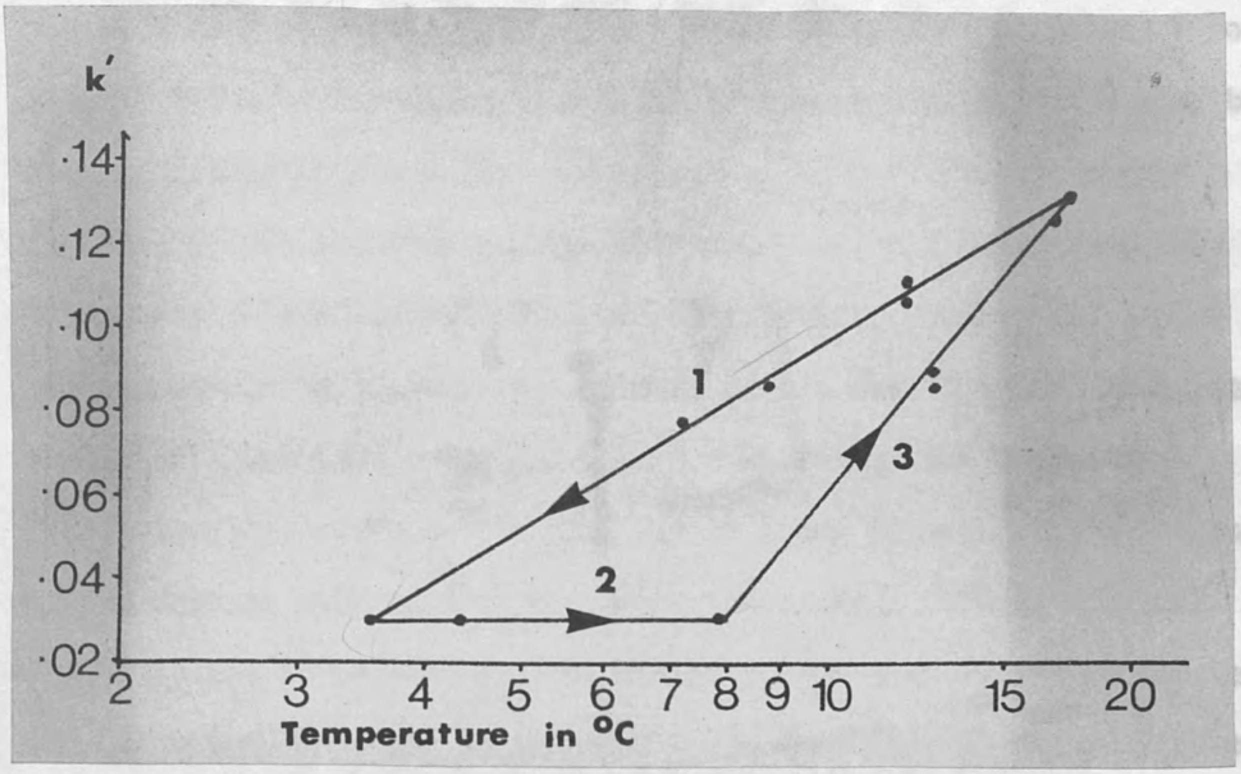



Figure 42. The variation of the relative growth constant, k', with water temperature.

For growth to occur, carbon fixation must take place, and the efficiency of this fixation will determine the rate of growth. Photosynthesis consists of two series of reactions. The first of these is independent of light but dependent on temperature whilst the second is dependent upon light but independent of temperature within the normal ranges encountered in nature. These are the so called photosynthetic "dark and light" reactions.

Figures 41 and 42 show the effect on k' of the light and temperature environments during the experiment. In each figure section 1 indicates the period of decrease for both temperature and light. This decrease would have produced a corresponding decrease in the rate of both the light and the dark reactions. In section 2 both light and temperature increased but k' remained constant. A possible explanation of this was that the amount of enzymes present per cell in January 1965 had been reduced by successive cell divisions, even though the rate of division had been decreasing, so that the amount was inadequate to support an increased growth rate due to limitation of the dark reaction, even though the temperature had increased. Some supporting evidence for this can be found by examining the changes in the average cell volume throughout the period. These increases and decreases correspond approximately to increases and decreases in cell diameters recorded in the open water (see Chapter IV and Figure 6). The average cell volume, calculated from cell diameters, increased from August 1964 reaching a maximum between October 1964 and January 1965. Between January and March 1965 there was a rapid decrease in the average cell volume. Presumably the cells were dividing but not producing enough new materials to meet their metabolic demands and so were utilising stored products. As the volume decreased so the amount of enzyme per cell would also possibly have decreased and it was not until later in the spring that the cells' enzymic activity ceased to limit the light reaction. It was then that the recorded increase in k' in section 3 occurred. Although the spring maximum, in terms of cell numbers, for Stephanodiscus astraea appeared to occur in the late spring, when its growth was not modified by copper sulphate application (as in 1964 and 1965), it continued increasing until June, well into section 3 in Figures 41 and 42.

After the spring maximum the average cell volume steadily increased, together perhaps with an increase in storage products, to reach a maximum volume in the winter. At the start of the spring growth the average cell volume decreased. This cycle corresponded in timing to the observed growth rate cycles (Figures 41 and 42).

X. Penetration of Algae into the sand.

Although the downward vertical flow of water through a slow sand filter bed should remove algae within the top few millimetres of sand, in practice this is not always so. Many diatoms and other algae are known to migrate vertically through mud and sand (Round and Happey, 1965; Round and Palmer, 1966) and this may happen with the sand surface flora (see Chapter VII). Algae are also carried into the sand during the first few days of operation of the slow sand filter bed before the formation of a zoogleal film.

The extent of this penetration can be estimated by taking core samples of the sand but this was not always possible as the filter beds were often drained, cleaned and refilled between sampling times. An attempt was made, using the apparatus described below, to follow the penetration of algae into the sand. The apparatus was designed to simulate as far as possible a column of sand through a slow sand filter bed and was arranged as in Figure 43. A Pyrex glass tube 1½ inches (3.75 cm.) in diameter and 24 inches (60 cm.) in length was used. The bottom was sealed with a rubber bung, the inside surface of which was concave, with a single glass tube running through its centre. The concave inner surface was designed to prevent settlement and trapping of particles on the bung. The outflow was controlled by means of a glass stop cock. About 1 inch depth of glass wool was packed into the bottom of the tube to hold the sand in place. The sides of the tube were then blackened to within 4 inches of the top so that no light was admitted to the lower parts of the tube.

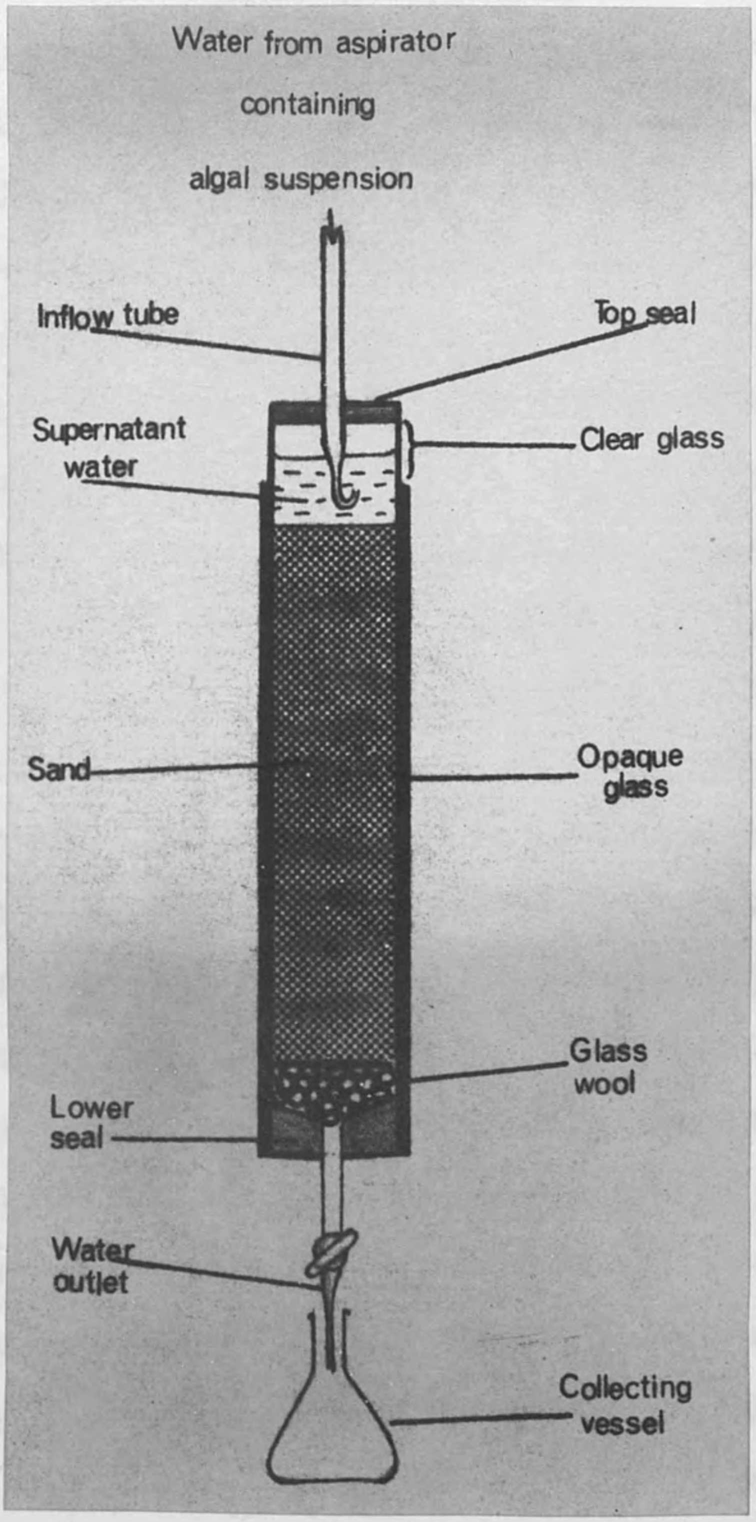



**Figure 43.** The experimental slow sand filter column.

The outlet tube was also blackened. The tube was then clamped into a vertical position, the outlet closed, and half filled with water. Washed and sterilised filter bed sand was then added from the top of the tube till its depth was 1 foot 6 inches (45 cm.). The presence of water in the tube helped to prevent air from being trapped in the sand. The sand was packed down carefully with a rod. Extra sand was added to bring the level to ½" below the top of the blackened section. Glass fibre filtered Queen Mary Reservoir water was then added by upward flow through the sand until there was 2" of supernatant water above the sand. The top of the tube was then sealed with a rubber bung having a single inlet tube passing through it. The end of the tube just dipped beneath the level of the supernatant water. This tube was then connected to an aspirator of water containing an algal suspension. The outlet led into a flask containing Lugol's iodine so that any algae passing through the column were immediately preserved. The apparatus was placed near to a north light which enabled the sand surface to be partially illuminated.

The algal suspension was made up from a net haul obtained from Queen Mary Reservoir resuspended in 40 litres of sterile reservoir water. The algae were kept in suspension by continuously bubbling air through the aspirator. The rate of flow of water through the column was 3" per hour (approx. 82 cc/hr.). The flow was continuous and was kept up for 21 days. The outflow was examined every 2 days and the algae present counted. After 21 days the sand was carefully removed in quarter inch layers and the algae were separated from the sand and counted. The numbers of algae in the aspirator at the beginning and end of the experiment were also determined. The results are given in table 8 and are expressed as cells per ml. of water or per cc. of sand x $10^{-1}$.

The numbers of cells/ml. of Scenedesmus spp. and Ankistrodesmus falcatus in the aspirator water increased throughout the experiment but those of Oocystis solitaria and Pediastrum boryanum decreased. All of the diatoms, with the exception of Melosira varians, had decreased in number by the end of the experiment. This was expected due to the artificial environmental conditions in the aspirator. A "pot" flora had started to develop. Ankistrodesmus falcatus, the smallest alga present, occurred in the largest numbers in the outflow water. Scenedesmus spp. were also frequent but only occasional diatoms occurred. No cells of Oocystis solitaria or Pediastrum boryanum were observed below ¾" and ¼" respectively. Large numbers of Scenedesmus spp. were found throughout the top 2" of sand although their numbers decreased with depth. Three diatoms, M. varians, Rhoicosphenia curvata and Surirella ovata, were not observed below 1", the last two, however, were only present in small numbers so that accurate counting was difficult. The remaining diatoms were present throughout the top 2" either as seemingly healthy or as dead frustules. As many, if not more, live than dead cells of Stephanodiscus astraea and Navicula sp. were present in the lower sand samples. The reverse, however, was true of Nitzschia linearis and Cymbella spp. Some species were present in larger numbers between ¼" and 1¼" depth than through the rest of the column. This could have been due to accumulation specifically at this depth either because of the rate of flow or because the algae preferentially maintained themselves in this position.

Larger algae, such as P. boryanum and M. varians were removed in the uppermost layers. This was because the interstices between the sand grains were too small to allow the passage of these relatively large algae. Live cells of many other species were only present in small numbers below 1". S. astraea and Navicula sp., both of which were below 35u in their maximum dimension, were present throughout the top 2". It is possible that both of these algae penetrated the sand before a biological filtering layer (zoogleal film) had developed on the sand surface. This could also have occurred with the smaller green algae, but if the sub-surface environmental conditions were unfavourable they would have died and decomposed leaving little or no trace (having no siliceous frustules). This may account for the apparent lack of green algae deeper into the sand. Scenedesmus spp. occurred in the largest numbers of all the algae present throughout the depth of the sand column. Although they were not able to photosynthesise at these depths the cells appeared to be healthy. There is evidence that this genus can live heterotrophically (Taylor, 1950; Bristol-Roach, 1926). The dissolved organic matter formed from decomposing cells in the upper layers may have provided sufficient nutrients for Scenedesmus to grow.

Live cells of most of the algae present were recorded below a depth of 1". When a slow sand filter bed is cleaned the usual practice was to skim off the top 1" or less of sand. This practice would have left live cells in the sand to act as a seed when the bed was next filled for use.

It was possible, on two occasions, to compare core samples from a filter bed with the results of the penetration experiment. The distribution of algae in these core samples was similar to that in the experimental sand column. Larger species of algae, such as S. astraea and Nitzschia linearis, were confined to the top 2 mm. Very few diatoms, with the exception of Nitzschia acicularis, were present below $\frac{1}{2}$". In both core samples there were large numbers of N. acicularis (exceeding 500 cells/cc. of sand) between 1" - $1\frac{7}{8}$" depth. About $\frac{3}{4}$ of these cells were living, the rest being empty frustules. N. acicularis is known by waterworks to be able to penetrate slow sand filter beds so its presence below the surface layers of sand is not unexpected. The accumulation at a specific depth, as with certain algae in the sand penetration experiment, was possibly an effect caused by the rate of flow of water through the bed.

Table 8. Cell concentrations in the inflow and outflow water and in the top two inches of sand. The results are expressed as cells per ml. of water or cells per $cm^3$ of sand x $10^{-1}$

| | Aspirator | | Depth of sand layer | | | | | | | | Maximum in |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | Day 1 | Day 21 | 0-1/4 | 1/4-1/2 | 1/2-3/4 | 3/4-1 | 1-1¼ | 1¼-1½ | 1½-1¾ | 1¾-2 | outflow |
| Chlamydomonas sp. | 1 | 8 | | | | | | | | | 1 |
| Ankistrodesmus falcatus | 20 | 40 | | | | | | | | | 5 |
| Oocystis solitaria live | 1264 | 966 | 3100 | 2600 | 65 | | | | | | 1 |
| dead | | | 1500 | 101 | 12 | | | | | | |
| Scenedesmus spp. | 96 | 164 | 1250 | 1040 | 940 | 702 | 687 | 568 | 390 | 130 | 4 |
| Coelastrum microporum | 16 | | | | | | | | | | |
| Actinastrum hantzschii | 4 | | | | | | | | | | |
| Pediastrum boryanum | 64 | 32 | 400 | | | | | | | | |
| Melosira varians live | 2 | 26 | 202 | 64 | | | | | | | |
| dead | | | 78 | 12 | | | | | | | |
| Stephanodiscus astraea live | 368 | 122 | 1350 | 834 | 203 | 200 | 182 | 182 | 108 | 30 | |
| dead | | | 525 | 108 | 65 | 148 | 260 | 180 | 100 | 26 | 1 |
| Navicula sp. live | 1 | | 10 | 108 | 8 | 110 | 135 | 87 | 17 | 2 | 0.1 |
| dead | | | 11 | 12 | 206 | 124 | 102 | 83 | 1 | | |
| Cymbella spp. live | 8 | | 65 | 260 | 113 | 109 | 12 | | | | 0.1 |
| dead | | | 565 | 568 | 1130 | 224 | 284 | 220 | 164 | 12 | |
| Nitzschia linearis live | 16 | | 1501 | 508 | 208 | 65 | 2 | 1 | | | 0.6 |
| dead | | | 108 | 115 | 112 | 117 | 104 | 52 | 2 | | |
| Rhoicosphenia curvata live | 1 | | 6 | 2 | | | | | | | |
| dead | | | 4 | 2 | 3 | 1 | | | | | |
| Surirella ovata live | 1 | | 6 | 3 | 1 | | | | | | |
| dead | | | 5 | 1 | | | | | | | |

XI. Seasonal variations in the cell sizes of Stephanodiscus astraea and Asterionella formosa.

During the period October 1963 to June 1966 the mean cell size of populations of cells referred to Stephanodiscus astraea (Ehr.) Grun. was observed to change. To investigate this cell size change samples of these algae collected from the reservoir and filter beds at various times were cleaned in hot concentrated sulphuric-nitric acid mixture (50% by volume of each). The cells were then washed in distilled water and concentrated by centrifugation. The diameters of at least 50 cells from each sample were measured and the size frequency distribution calculated. The results are given in Figure 44. During the spring diatom growth period of 1966, weekly (or on one occasion fortnightly) samples were taken between February 21st and April 12th. During this period the samples were cleaned as above and measurements made of the diameters of S. astraea and also the length of Asterionella formosa frustules. The size frequency distributions were calculated and the results are given in Figure 45a. and 45 b.

During the spring growth period of 1966, assuming that all the cells of S. astraea were growing actively and dividing equally fast, an overall movement of the population mode from a larger to a smaller diameter would have been expected. Cells would presumably only be able to return to their original size by auxospore formation. Although this mechanism has been referred to (Hustedt, 1930), auxospores were never observed during the present investigation.

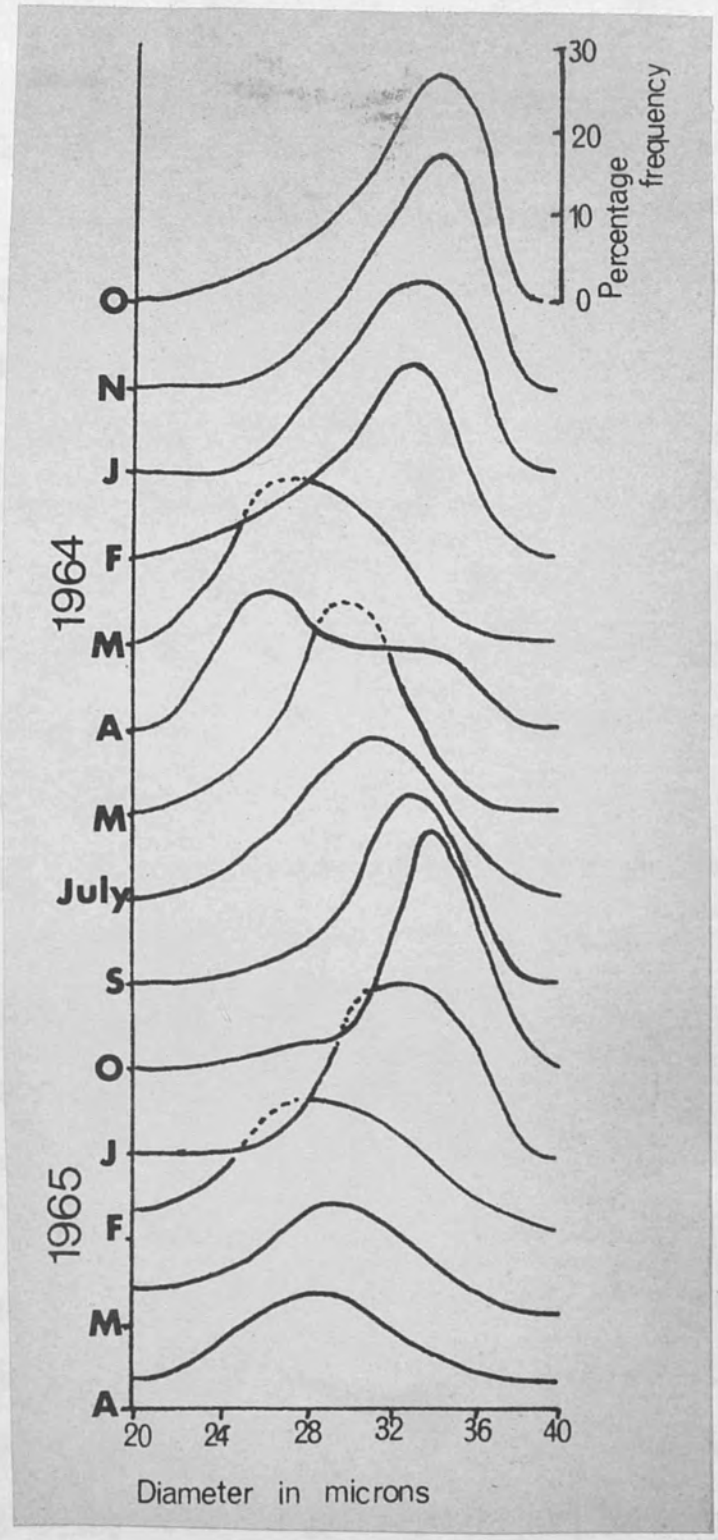



**Figure 44.** The size frequency distribution of the cell diameters of populations of ***Stephanodiscus astraea.***

The winter populations of S. astraea had a diameter mode between 30-35μ (Figure 44). At the time of the onset of the spring growth period this mode shifted to the left (i.e. to the smaller average diameter). When the spring populations had reached, or just passed their spring maximum (see periodicity graph, Chapter VI, Figure 18) the mode was at its minimum level. The mode then shifted to the right (indicating an increase in average diameter). This shift was particularly marked when no large populations occurred in the early summer after the spring maximum. If growth continued throughout the spring period and was not greatly reduced in the summer, as in 1966, then no such rapid shift to the right occurred. No rapid decreases were observed in the average cell diameter during the autumn growths of S. astraea. These were never as large nor as long in duration as the spring populations (see Figure 44).

More detailed measurements at shorter time intervals were made during the spring of 1966 (see Figure 45 a.) On February 21st, the population had a mode between 32-34μ. At this time numbers were still increasing towards their spring maximum. On February 28th, the main mode had decreased in size and a secondary mode between 28-30μ had developed. By March 14th, both the primary and the secondary modes had increased in size and had sharpened, i.e. the population spread was narrower. The mode at 32-34μ had greatly reduced by March 21st, and was replaced by a new main mode between 28-30μ. This new main mode then sharpened, a "shoulder" in the curve being all that remained of the 32-34μ mode. The new mode then decreased and the curve became

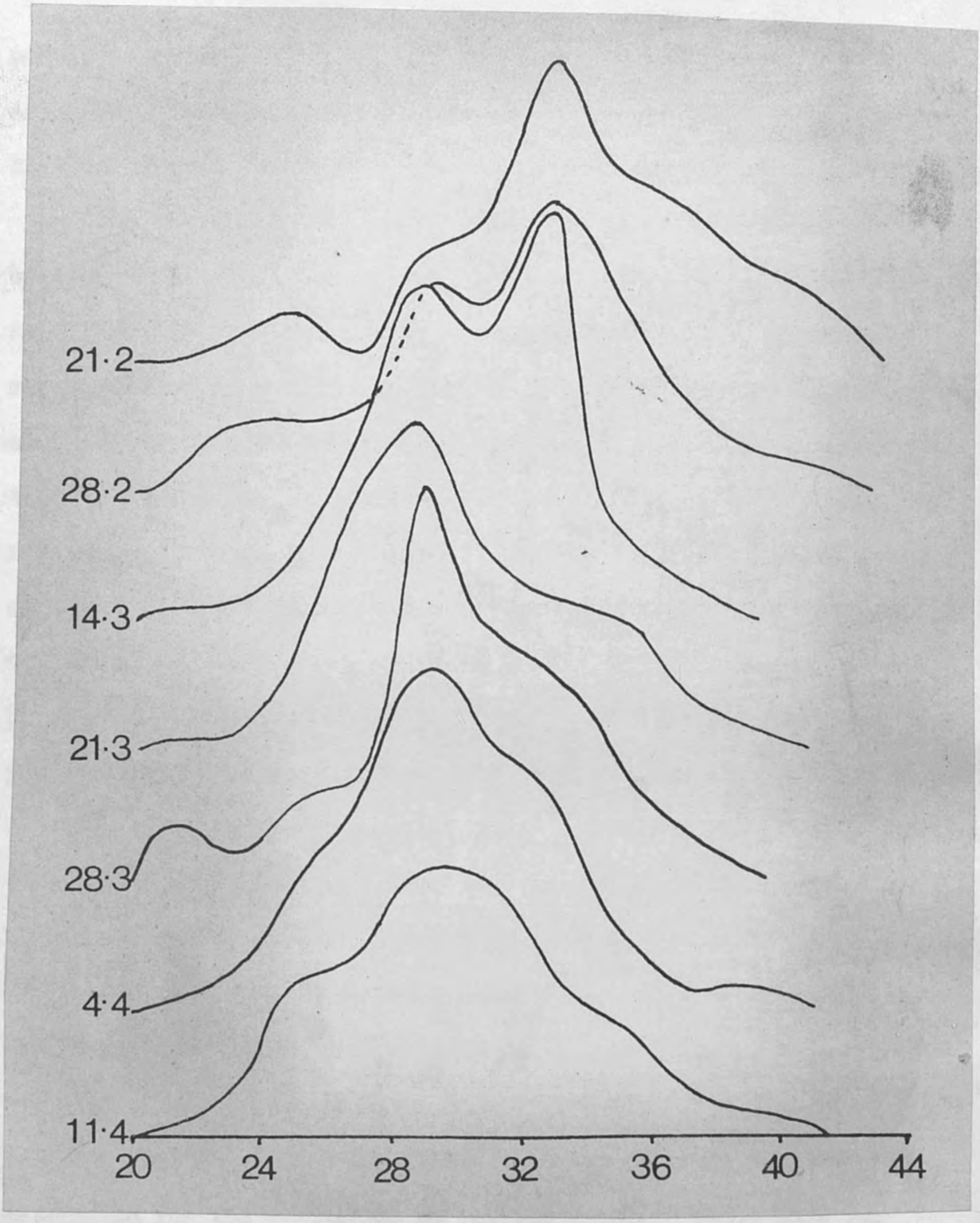



Figure 45a. The size frequency distribution of the cell diameters in microns (horizontal scale) of the spring population of Stephanodiscus astraea in 1966.

more rounded until by April 12th the mode spread between 28-32µ. After April 12th there was an increase in number in cells of larger diameter which caused a shift in the mode to the right (see Figure 45a).

This decrease in the average diameter of the population, followed by size restitution, could possibly be explained by the normal decrease in diameter in the valvar plane during division. The observed size restitution might possibly be explained by auxospore formation, although auxospores were never observed during the period of study. Another possible explanation is that one population may have been replaced by another one adapted to slightly different environmental conditions. This would require a continuous replacement over a period of time in the water being abstracted from the River Thames. S. astraea was not, however, present in the River Thames during this period. Light and electron microscope studies of the sculpturing of the frustules throughout the period did not reveal any such change (see page 184 et seq.) Wesenberg-Lund (1908), as reported in Hutchinson (1967), recorded a similar size change within a population of S. astraea in Lake Rueso in Denmark but as only limited data was available, few conclusions were drawn.

During the spring period of 1966 the length of Asterionella formosa frustules was measured and the size frequency distributions determined for the population at various times. There were two distinct modes present on February 21st (Figure 45 b.) the larger between 57-62µ and the smaller between 77-89µ. By the 14th March both of these modes had shifted slightly to the right, their new

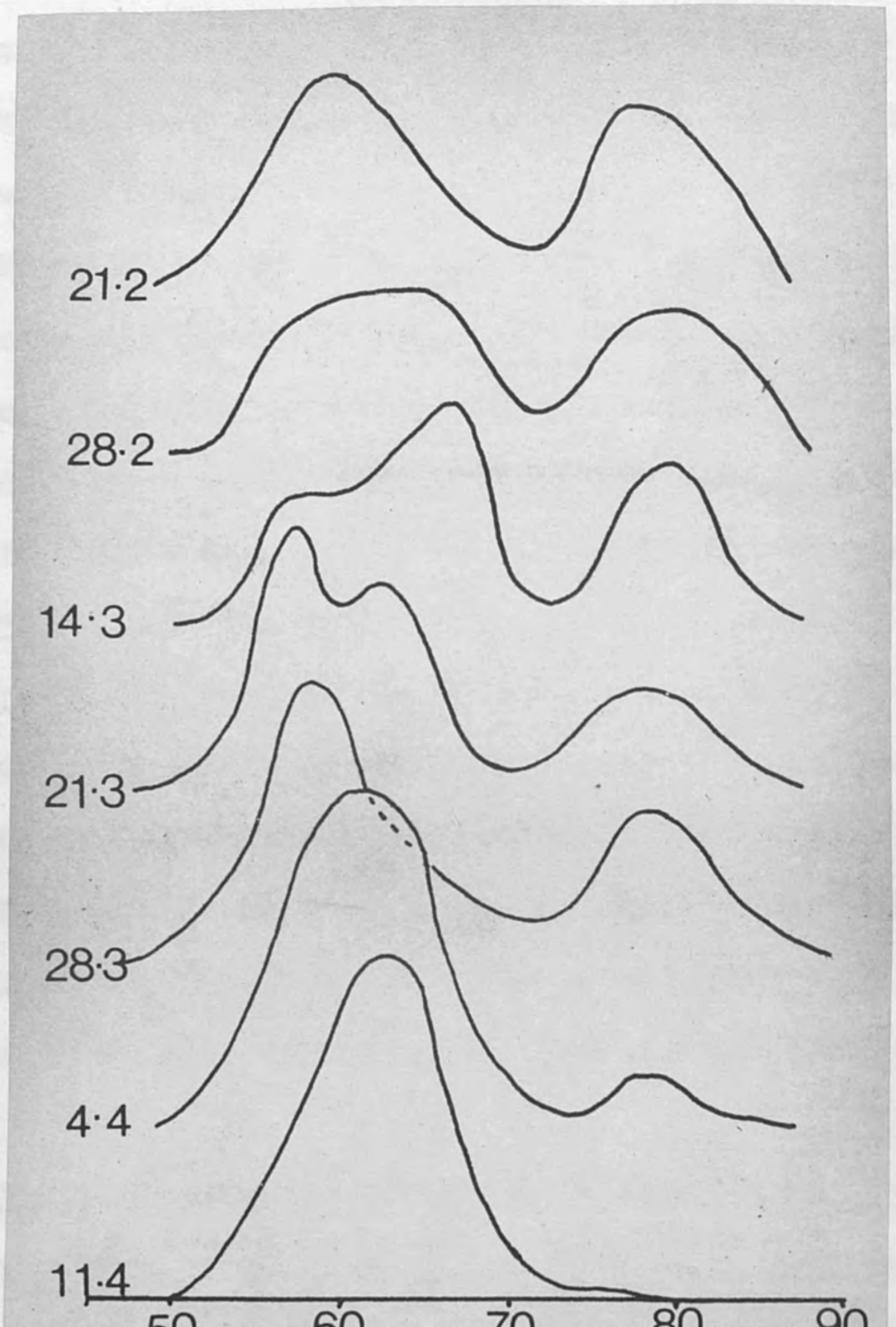



**Figure 45b.** The size frequency distribution of the cell lengths in microns (horizontal scale) of the spring population of Asterionella formosa in 1966.

positions being at 68 and 83µ. After March 14th the 83µ mode shifted to the left and decreased in size, completely disappearing by April 12th. The 68µ mode also moved slightly to the left but this sharpened and increased in height.

Although the period of observation for Asterionella formosa was short (being only through the spring growth) and no information is available for other times of the year, the results suggest that two populations were present. The larger one having a mode between 77-83µ was present in smaller numbers, whereas the shorter one, having a mode between 57-63µ was present in larger numbers. A similar, although more complicated situation, was observed by Lozeron (1903) referred to in Hutchinson (1967), in the Ober-Zurichsee. He found three populations having slightly different cycles in the lake. Wesenberg-Lund (1908), as referred to in Hutchinson (1967), found evidence of size reduction in some Danish lakes. He also observed rapid size restitution over short periods of time, at times as short as three weeks.

Cells from each population sample were observed in detail to ascertain whether or not there was any difference in the species or varieties at various times of the year. The cells of both populations of Asterionella could be referred to Asterionella formosa Hass.

A number of varieties of Stephanodiscus astraea have been described. A summary of these is given in Table 9.

Very few cells were found of diameter above 40µ, the majority being between 20-40µ. This diameter range could include all of the varieties described by Hustedt (1930) and Cleve-Euler (1951). The punctae were generally coarse in most of the specimens regardless of their size. The radial rows were formed from either single rows of pores or in many cases groups of two or three rows tapering to a single row near the centre of the frustule (Figure 46). The numbers of rows at the circumference ranged from 8-10 in 10µ and the numbers of pores 11-18 in 10µ. Most specimens were in the range 9-10 pores/10µ at the circumference and 12-14 pores/10µ along the radius.

If one considers only the cell diameters then two main "varieties" were present, var. "typica" and var. intermedia, although the descriptions of Cleve-Euler indicate that these overlap. Some cells of less than 20µ were present and these could be referred to var. minutula. The majority of cells, which on the basis of diameter could be called var. "typica", had 8-10 pores/10µ at the circumference and between 12 and 16 pores/10µ along a radius. The pores were usually in groups of two at the frustule edge although certain cells (Figure 46) had single rows to the very edge. This latter feature has only been described in var. incertus Cleve-Euler. The cells described as var. intermedia, on the basis of diameter, had 8-9 pores/10µ at the circumference and 14-16 pores/10µ along a radius. The pores were usually in groups of 2-3 at the edge of each ray but some were single and some were in groups of 4 (Figure 46). A group of frustules, representing only a small percentage of the total, had a single row of coarse dots. These had 8-10 pores/10µ at the circumference and 11-13 pores/10µ along a radius. Their diameters ranged from 20µ to 32.5µ. These

Table 9. A summary of the main characteristics of the varieties of Stephanodiscus astraea as described by Hustedt (1930) and Cleve-Euler (1951).

| | Dia. in microns | Rows/10u | Pores/10u | Pores/row at edge | Comments |
|---|---|---|---|---|---|
| **Hustedt** | | | | | |
| v. "typica" | 30-70 | 9 | 12 | 2-4 | |
| v. minutula | 8-30 | - | - | - | weakly silicified |
| v. intermedia | 20-25 | - | - | 3-4 | rows dispersed at edge |
| **Cleve-Euler** | | | | | |
| v. "typica" | 20-70 | 10-15 | - | 2-4 | |
| v. minutula | 8-25 | - | - | 2 | |
| v. intermedia (spinuligerus) | 20-40 | - | - | 3-5 | rows broader towards edge |
| v. incertus | - | - | - | 1 | |
| v. niagarae | up to 70 | - | - | 4-5 | distribution restricted to North America |

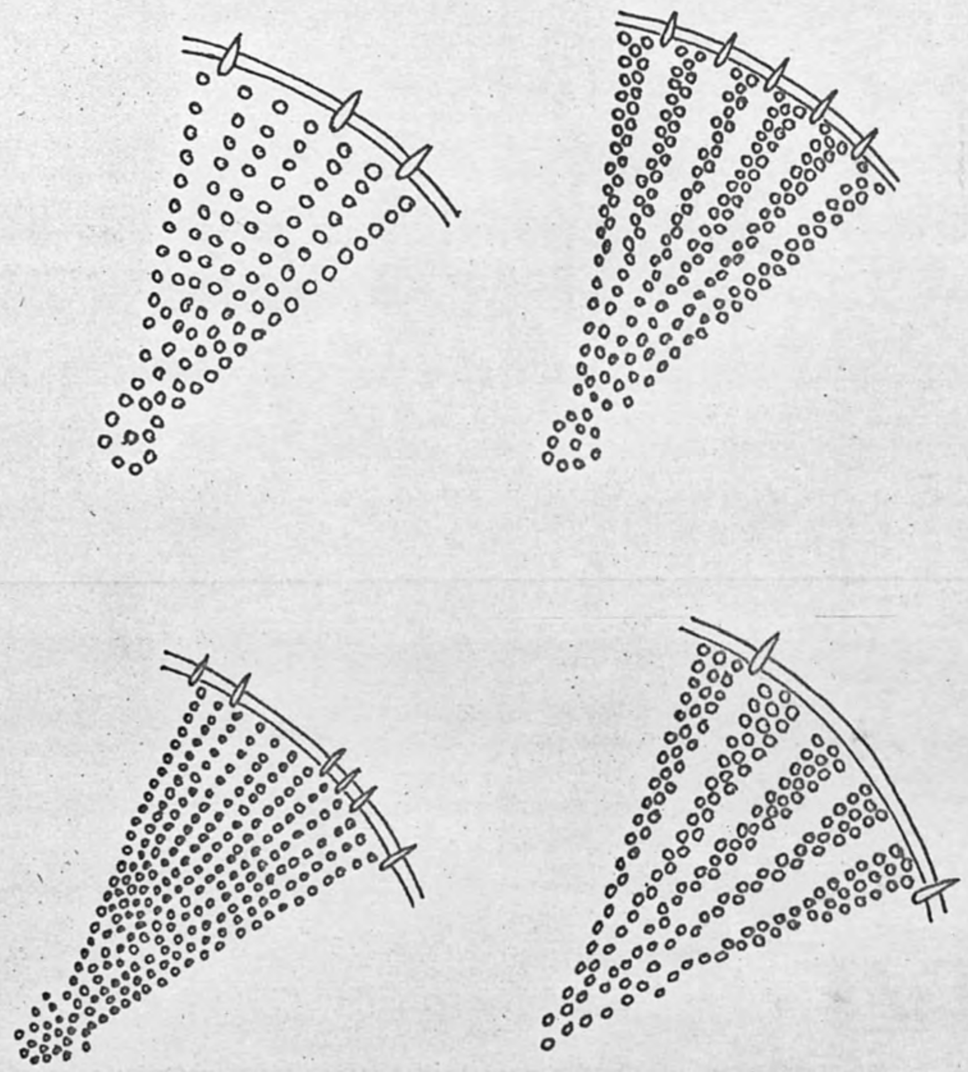
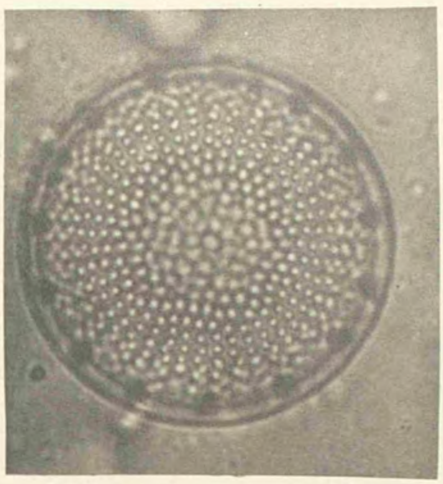


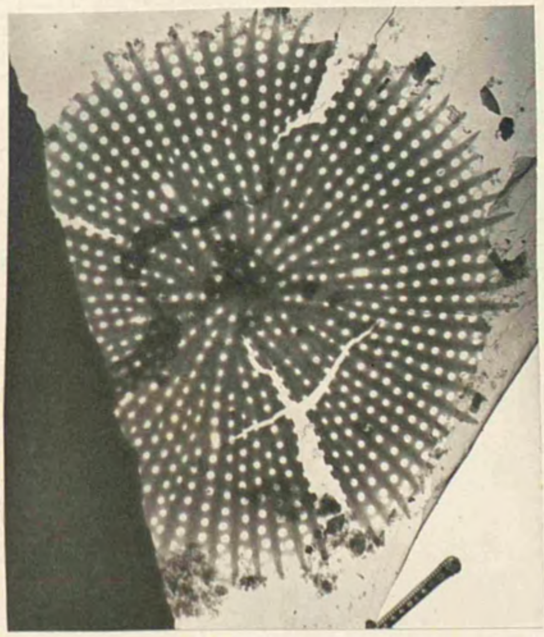
Figure 46. Some of the observed variations in the punctation of frustules of Stephanodiscus astraea.

fit the description of Cleve-Euler's <u>var. incertus</u>. There was, however, no obvious diminishing of the hyaline rays towards the edge of the frustules and although the spines were sometimes smaller this feature was also present in cells of other varieties.

With the observed changes in cell diameter at different times of the year, unless the frustules of <u>S. astraea</u> are definitely large enough, i.e. above 40µ, to be called <u>var. "typica"</u> rather than <u>var. intermedia</u> or definitely small enough i.e. below 20µ, to be called <u>var. minutula</u>, there is extreme difficulty in separating out the different described varieties. The size ranges of all the named varieties overlap between 20 and 40 microns. The punctuation throughout the range of diameters seems to be variable. There was a tendency, though not very marked, for cells in the range of <u>var. intermedia</u> to have broader rays than other varieties (Plate 5). There is also a small proportion of cells fitting the description of <u>var. incertus</u> (Cleve-Euler, 1951) (see Plate 6). As there is considerable difficulty in separating the varieties of <u>S. astraea</u> and as these varieties overlap each other there is a need for more detailed studies. Electron microscopy would be an ideal method of investigating the punctuation but due to variations within a single population, numerous samples collected over a period of time need to be studied.



<u>Plate V.</u> Light microscope photograph of a frustule of <u>Stephanodiscus astraea</u> attributed to var. <u>intermedia</u>.



<u>Plate VI.</u> Electron microscope photograph of a frustule of <u>Stephanodiscus astraea</u> attributed to var. <u>incertus</u>.

# PART FOUR.

## XII. Taxonomic Notes.

Of the algae which were identified during this ecological investigation, many occurred in small populations, or only for short periods. Such algae have not been dealt with in detail in the foregoing account but are described as many of them have been recorded only infrequently in this country. A complete list of all species found is given at the end of this chapter.

### CHLOROPHYTA

#### VOLVOCALES

Spermatozopsis exsultans Korshikov. (Figure 48).

This species was recorded on several occasions between the months of June and September in 1964 and 1965. It occurred in small numbers in the supernatant water of all four filter beds studied. The cells were 7-10µ long and 3-4µ wide. Although most of the cells had four equal flagella some were observed with only two. This variation in the number of flagella has also been reported by Korshikov (1913). This species has not been commonly recorded in the British Isles but there are two previous records from the London area. Swale (1964) recorded S. exsultans from the River Lee and Scourfield (1944) recorded it in bomb craters in Epping Forest.

Carteria quadrangulata Pascher (Figure 47).

Cells of this species were ovoid with broadly rounded base and a truncated top. The anterior end four protruding lobes. No papilla was observed. The chromatophore was cup shaped, filling most of the

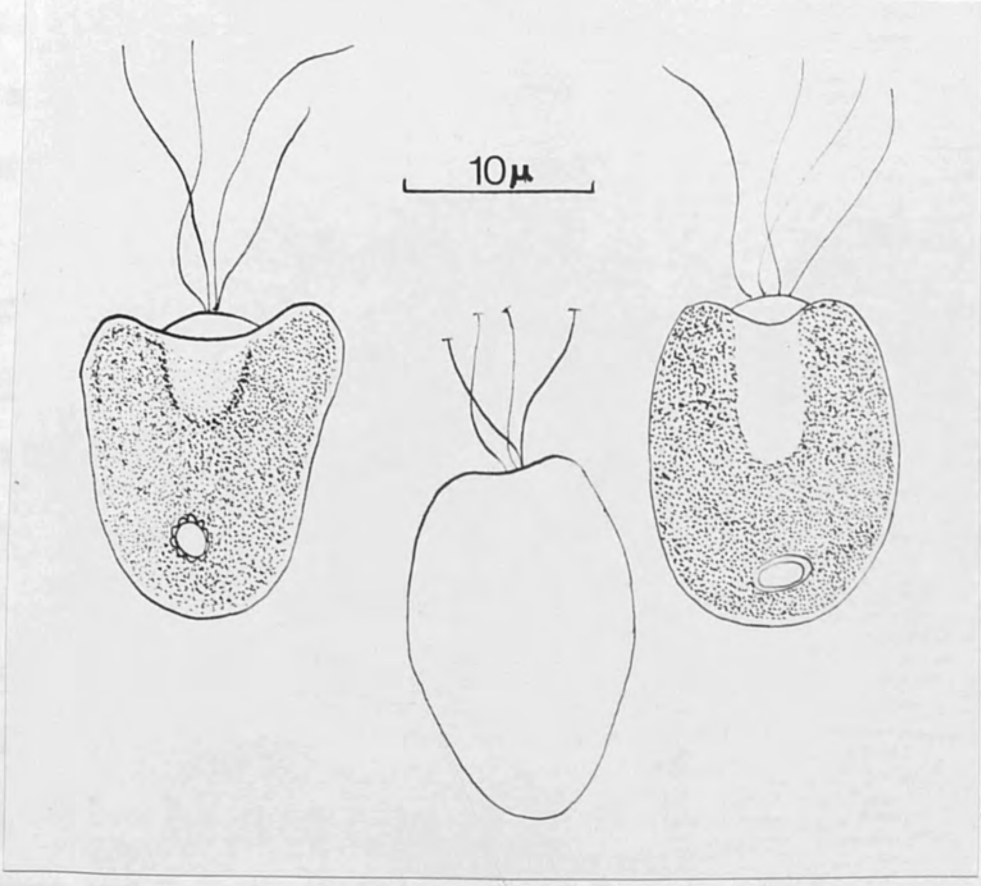



Figure 47.
Carteria quadrangulata Pascher.

cell, with a pyrenoid at its base. This species was recorded in late June and early July of 1964 and 1965 in the supernatant water of each of the filter beds.

CHLOROCOCCALES

Ankistrodesmus pseudomirabilis Korshikov

The cells were 25-50µ long and about 2-3µ wide at the centre. The cells were crescent shaped with attenuated ends. The distance across the ends of the arc varied from 30-40µ. Cells of this species were observed throughout the winter months of 1964 and 1965 in small numbers in the reservoir water.

Tetrastrum Chodat

West and Fritsch (1927) maintain that the division between Tetrastrum and Crucigenia is artificial being based purely on the presence or absence of spines and of occasional syncoenobia. They keep the division only for convenience. The species recorded were identified according to Korshikov (1953). West and Fritsch (1927) stated that species of this genus were uncommon in the British Isles.

Tetrastrum staurogeniaeforme (Schroed) Lemm (Figure 49).

The coenobia were 4 celled with a small space between the cells at the centre of the colony. The cells were triangular in shape with the outer surface rounded. The outside margin bore 3-5 thickened spines. The chromatophore had a single pyrenoid. The cells, without spines, were 5-7.5µ long and the coenobia were 10-20µ across. The spines were 3.5-7µ long. This species occurred commonly on the sand surface especially during the summer months.

Tetrastrum heterocanthum (Nordst.) Chod. (Figure 49).

The coenobia were 4 celled with a fairly large space at the centre. The cells were kidney shaped and bore 2 spines of unequal length. There was a single chromatophore and pyrenoid in each cell. The cells were 6-8.5µ wide, the coenobia 12-18µ across and the spines 3-9µ long. This species was uncommon occurring during the summer months on the sand surface.

Tetrastrum hastiferum (Arnoldi) Korshikov (Figure 49).

The coenobia consisted of 4 cells forming a square with a very small space in the centre. The outside wall of each cell bore a single, long, delicate spine. Each cell had one chromatophore and pyrenoid. The cells were 3-5µ wide and the spines 7.5-12µ long. This species was recorded on July 3 and 10 1965 from filter bed No. 12 sand surface.

Tetrastrum glabrum (Roll) Ahlstr. et Tiff. (Figure 49).

The coenobia consisted of 4 triangular shaped cells with rounded

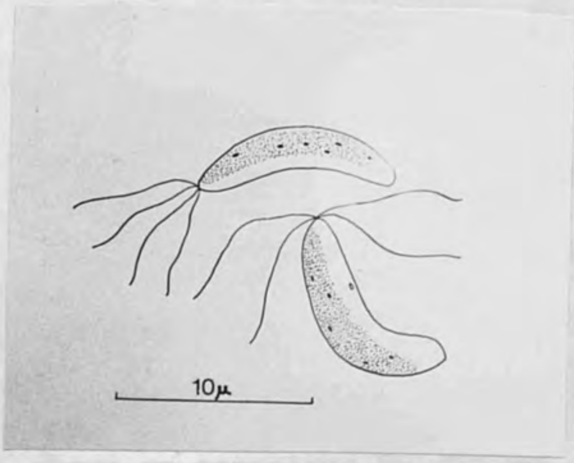



Figure 48. Spermatozopsis exsultans Korshikov

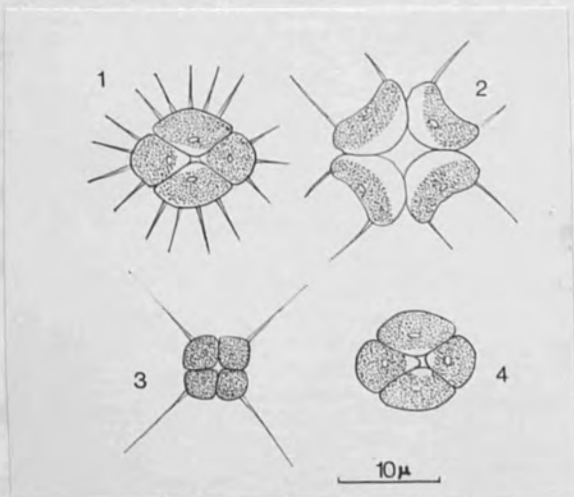



Figure 49. 1.Tetrastrum staurogeniaeforme. 2.T. heterocanthum. 3.T. hastiferum. 4.T. glabrum.

outer surfaces. The coenobia had a mucilaginous envelope. The cells had a single chromatophore with pyrenoid. The cell walls were perfectly smooth having no spines. The cells were 4.5-7µ wide and the coenobia were 10-15µ across. No syncoenobia were observed. This species was found in small numbers on the sand surface during the summers of 1963 and 1964.

SCENEDESMUS Meyen

Certain species of Scenedesmus occurred in large numbers in the slow sand filter beds. Only on rare occasions was one species present at a time, often many species were present with one or perhaps two predominating. For the purpose of the preceding ecological study all of the species were grouped together under the generic name. A more detailed description of the species recorded is now given. The species were identified according to Uherkovich (1966) but reference was also made to Smith (1916), Shen (1956), Korshikov (1953) and Hortobagyi (1959, 1960 a & b). Complete descriptions and lists of synonyms for the following species are given in Uherkovich (1966).

Scenedesmus acutus Meyen

The coenobia consisted mostly of 4 but occasionally 2-8 cells in a linear series. The cells were spindle shaped with a more or less cuspidate end (Figure 50 (1)). Cells 8-12µ long and 2.5-4.5µ wide. This species was very common especially on the sand surface where on occasions it was one of the co-dominant algae.

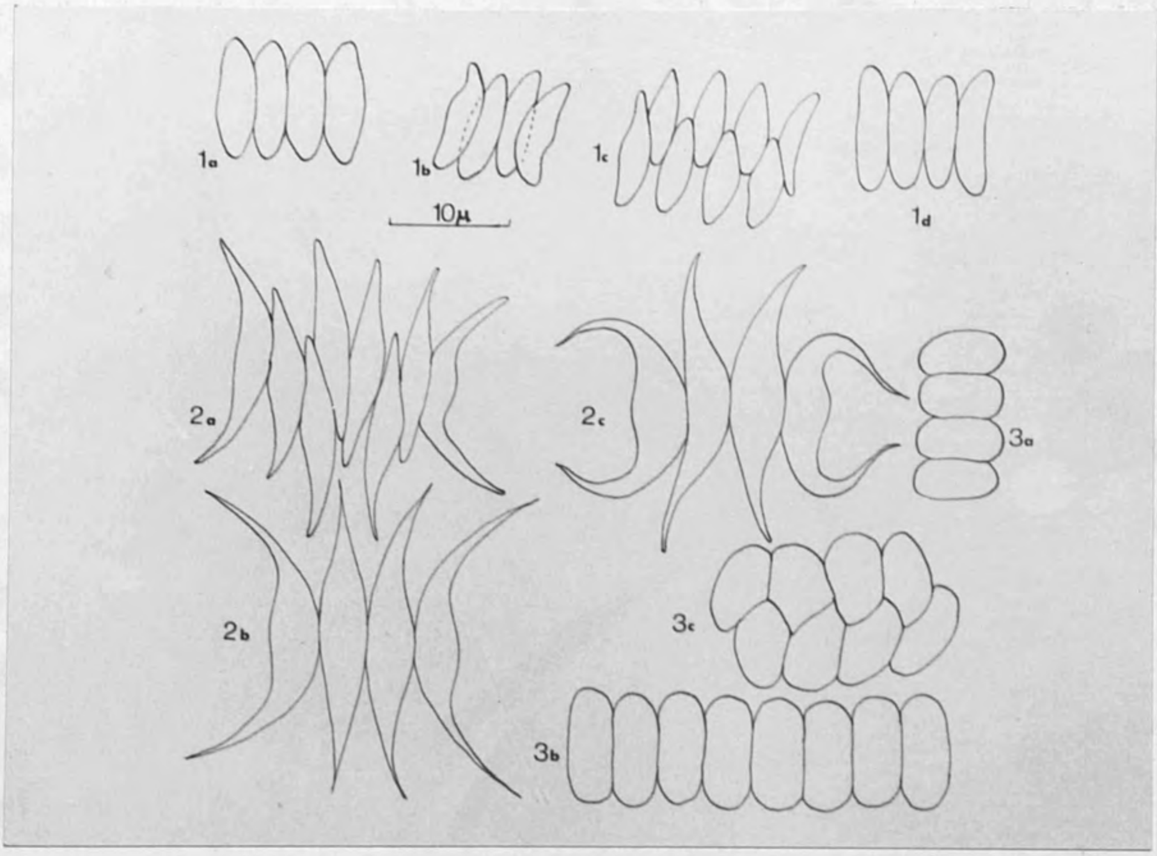



Figure 50. 1a, Scenedesmus acutus. 1b, S. acutus f. tetradesmiformis. 1c, S. acutus f. alternans. 1d, S. acutus f. semiellipticus. 2a, S. acuminatus. 2b, S. acuminatus f. maximus. 2c, S. acuminatus f. tortuosus. 3a and b, S. ecornis. 3c, S. ecornis var. disciformis.

S. acutus f. tetradesmiformis (Woloz) Uherkovich (Figure 50 (1)).

Coenobia similar to the basic form but the cells are curved along their longitudinal axis and nearly tetradesmoid in form. The cells were 8-10µ long and 2.5-4µ wide. Occasional coenobia were found in the reservoir water and filter bed supernatant water during the late summer of 1965.

S. acutus f. alternans Hortob. (Figure 50 (1)).

Coenobia of 8 and occasionally 4 alternating cells, 10-15µ long, 3.5-4.5µ wide. This species occurred frequently throughout the period though not in large numbers.

S. acutus f. semiellipticus Uherkovich (Figure 50 (1)).

Coenobia of 4 cells in a linear series. The outer cells were curved and spindle shaped, the inner ones straight and cylindrical. The ends of all of the cells were rounded. The cells were 10-15µ long and 3-4.5µ wide. This species was only recorded on two occasions, both during September 1965, in the supernatant water of filter bed No. 12.

Scenedesmus acuminatus (Lagerh.) Chod. (Figure 50 (2)).

The coenobia usually consisted of 4 cells in a linear series. The cells were spindle shaped with attenuate ends. The inner cells were more or less straight but the outer cells were curved. The cells were 14-20µ long and 2.5-5µ wide. This was a common species in the reservoir and filter beds throughout the period.

S. acuminatus f. maximus Uherkovich (Figure 50 (2)).

The coenobia were of 4 or 8 cells similar in shape to the main species but much larger. The end cells of the coenobia were 28-36µ long and 4-5µ wide. This form could be distinguished from the main species as it was distinctly larger. It occurred occasionally during the summer periods of each of the years studied both in the supernatant water and on the sand surface of the filter beds.

S. acuminatus f. tortuosus. (Skuja) Uherkovich (Figure 50 (2)).

The coenobia consisted of 4 strongly curved or spirally twisted cells of similar size to that of the main species. It occurred in 3 samples collected in July 1964 from the sand surface of filter bed number 32.

Scenedesmus ecornis (Ralfs) Chod. (Figure 50 (3)).

The coenobia consisted of 2-4-8-16 and on rare occasions 32 cells in a flat plate. The cells were oval 7-15µ long and 3-5µ wide. This was a very common species especially on the sand surface during the summer months when it was at times one of the co-dominant species.

S. ecornis var. disciformis. Chod. (Figure 50 (3)).

The coenobia were 8 celled. The cells were ovoid and formed an alternating plate like series. They were 7-10µ long and 3-5µ wide. This species was common on the sand surface, often forming agregations around particles of organic detritus.

Scenedesmus ovalternus. Chod. (Figure 51 (1)).

The coenobia were of 8 cells which were in contact for but a small part of the length of their lateral walls and formed a drawn out regularly alternating series. The cells were 5-8µ long and 3-4µ wide. This species was not common and occurred only occasionally during the

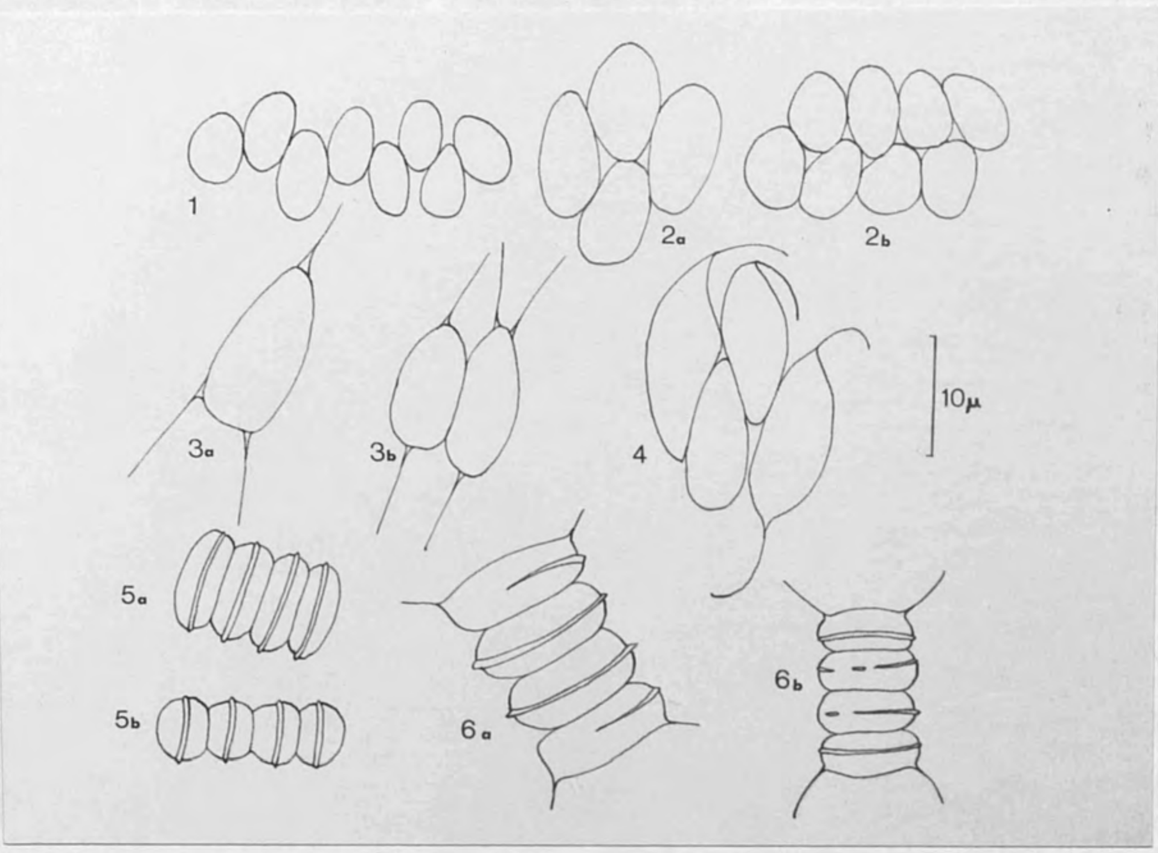



__Figure 51.__ 1, __Scenedesmus ovalternus.__ 2a and b, __S. arcuatus.__ 3a and b, __S. pecsensis.__ 4, __S. naegeli.__ 5a and b, __S. acutiformis.__ 6a and b, __S. armatus.__

summer of 1964 on the sand surface of the filter beds.

Scenedesmus arcuatus Lemm. (Figure 51 (2)).

The coenobia consisted of 4-8-16 cells which were oval or slightly curved in shape. The cells were arranged in two rows with spaces between the inner cells of the coenobium. The cells were 7-12µ long and 3-6µ wide. This species was common on the sand surface especially during the summer months.

Scenedesmus naegli Breb. (Figure 51 (4)).

The coenobia consisted of 2 or 4 cells which formed an alternating series. Each cell was pear shaped with the ends rounded or with small papillae or with a single curved spine 5-8µ long. Several specimens were recorded on the sand surface of bed 12 during September 1964. It was not recorded on any other occasions.

Scenedesmus ? pecsensis Uherkovich. (Figure 51 (3).)

The cells observed were either solitary or in pairs. They were broadly oval in shape sometimes being slightly narrow towards one end. Adjacent cells of coenobia were only in contact for about ½-⅔ their length. Each cell had, at its pole, 1 or 2 strong spines. There was no consistency between different cells in the number of spines per cell. The cells were 12-18µ long and 5-7µ wide. This species occurred on one occasion during September 1965 from the sand surface of filter bed 12. It was in a mixed population with S. acutus and S. quadricauda. Swale (1967) found cells similar to S. pecsensis in a clone culture isolated as Chodatella quadriseta. There were also 4 cell coenobia in the culture which resembled S. quadricauda. It is possible that the cells recorded from the filter beds and tentatively identified as S. pecsensis represented a case of pleomorphism being merely independent stages of S. quadricauda.

Scenedesmus acutiformis Schroeder. (Figure 51 (5)).

The coenobia consisted of 2-4 broad oval to spindle shaped cells in a single linear row. Each cell had a straight rib down either side. The cells were 9-14µ long and 3-4.5µ wide. Coenobia of this species were recorded occasionally throughout the summer months in each of the filter beds.

Scenedesmus armatus Chod. (Figure 51 (6)).

The coenobia were of 2-4 cylindrical to spindle shaped cells, 8-14µ long and 3-6µ wide, in a linear series. The end cells of the coenobia bore a spine $\frac{2}{3}$ the length of the cell at each pole. There was a rib extending down the side of each cell. These ribs were not always complete. Coenobia of this species were found in cultured material collected from the supernatant water of filter bed number 12 on September 13th, 1963.

Scenedesmus quadricauda (Turp.) Breb. (Figure 52 (1)).

The coenobia 2-4-8 cells in a linear row. The cells were oval to cylindrical with rounded ends. The outer cells of the coenobium had one curved spine at each pole. The cells were 8-20µ long and 2.5-7µ wide. Both the size of the cells and the length of their spines was variable. This was a common species occurring frequently throughout the period in all of the locations studied.

Although there are several varieties of S. quadricauda described,

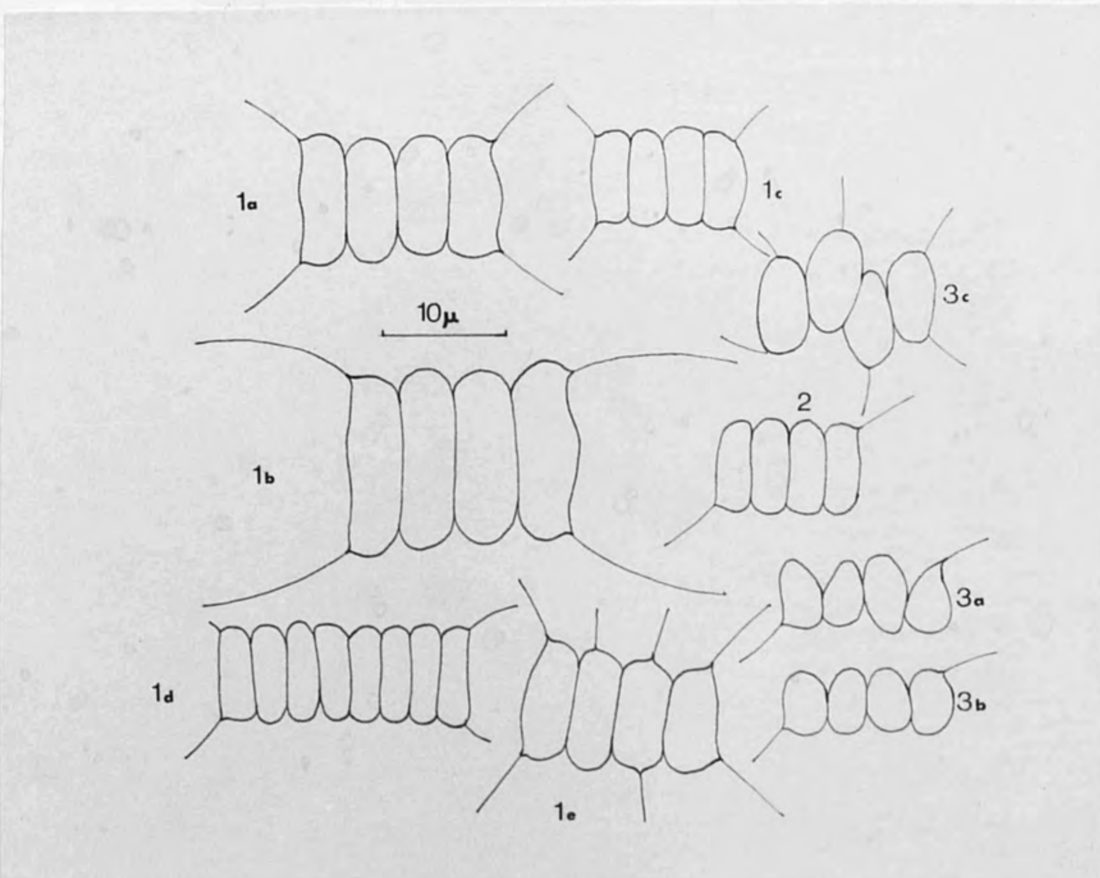



Figure 52. 1a, Scenedesmus quadricauda. 1b, S. quadricauda var. longispina. 1c, S. quadricauda var. quadrispina. 1d, S. quadricauda var. westii f. heterospinosus. 1e, S. quadricauda var. biornatus. 2, S. bicaudatus. 3a and b, S. intermedius var. bicaudatus. 3c, S. intermedius var. balatonicus.

many of these are thought to be local races which have developed due to varying physiological conditions and the inherent variability of the cells (Korshikov 1953). Some of the more distinctive varieties are described below.

S. quadricauda var. maximus W. et G.S. West.

The coenobia were 4 celled. The cells were larger than in the main species being 20-28µ long and 8-11µ wide with spines almost as long as the cell. This variety occasionally occurred in the reservoir water mostly during the summer months.

S. quadricauda var. longispina (Chod) G.M. Smith (Figure 52 (1).

The coenobia consisted of 4 cells which were slightly smaller than those of the main species and were at least 2½ times as long as they were wide. The spines were much longer than in the main species. Coenobia occurred occasionally on the sand surface.

S. quadricauda var. quadrispina (Chod) G.M. Smith (Figure 52 (1)).

The coenobia were mostly 4 but occasionally 2 celled. These cells were oval and smaller than the main species being only 7-8µ long and 2.5-3.5µ wide. The spines on the outer cells were much finer and shorter (¼-⅓ the length of the cell) than in the main species. This variety occurred occasionally in the filter beds during the spring of 1965.

S. quadricauda var. westii f. heterospinosus (Hortob.) Uherkovich Figure 52 (1)).

Only 8 celled coenobia were recorded, the cells being 8-10µ long and 3-4µ wide. One pair of diagonally opposite corners on the end cells bore long spines 4.5-8μ long whilst the other pair bore short spines 1.5-3μ long. This species was recorded on one occasion, October 1st, 1964, from the sand surface of filter bed No. 12.

<u>S. quadricauda var. biornatus</u> Kiss (Figure 52 (1)).

The coenobia were all of 4 longish eliptical cells 9-11μ long and 3.5-5μ wide in a linear series. The outside cells had a long spine ($\frac{2}{3}$ the length of the cell) at each of their poles. The inner cells either had a shorter finer spine at their poles or had rounded smooth ends. This variety was recorded on two occasions, October 1st and 8th, 1964, from the sand surface of filter bed No. 12.

<u>Scenedesmus bicaudatus</u> (Hansg.) Chod. (Figure 52 (2)).

The coenobia consisted of 2-4 oval cells 7-9μ long and 2-3μ wide. The poles of the cells were regularly rounded. The outside cells had, on one pair of diagonally opposite poles, one curved spine $\frac{2}{3}$-$\frac{3}{4}$ the length of the cell. The other pole, together with the poles of the inner cells, was smooth. This species was not common occurring occasionally in the reservoir water during the spring and summer months.

<u>S. bicaudatus var. brevicaudatus</u> Hortob.

4 and occasionally 2 celled coenobia occurred. The cells were 7-8μ long and 3.5-4μ wide. This variety was similar to the main species but the spines were much shorter and finter being less than the length of the cell. Occasional coenobia were recorded in the reservoir and filter bed supernatant water during the summer months.

<u>Scenedesmus intermedius var. bicaudatus</u> Hortob. (Figure 52 (3)).

The coenobia consisted of 4 loosely attached oval to eliptical cells. These cells were attached for less than $\frac{1}{2}$ the length of their adjacent walls. The celles were 5-6µ long and 2.5-3.5µ wide. The diagonally opposite pair of poles of the end cells had one spine $\frac{1}{2}$-1 times the length of the cell. The other pair of poles, together with the poles of the inner cells, were smooth. Coenobia of this species were recorded on July 3rd 1964 on the sand surface of filter bed 12.

<u>S. intermedius var. balatonicus</u> Hortob. (Figure 52 (3)).

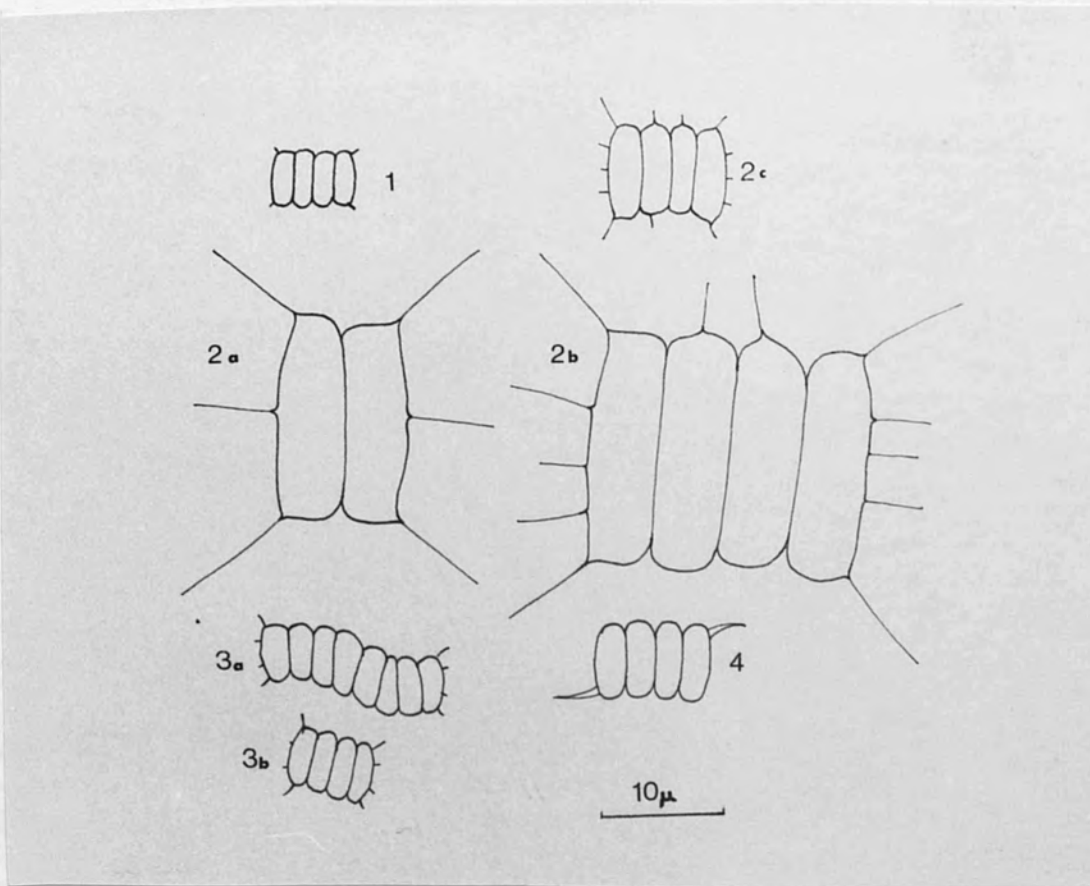
The coenobia were of 4 alternating oval cells 7-8µ long and 3-4µ wide. Each pole of the outside cells had one spine about $\frac{1}{3}$-$\frac{1}{2}$ the length of the cell. One pair of diagonally opposite poles of the two inside cells also bore a spine. Coenobia of this variety were recorded in May 1965 from the sand surface of filter bed 30.

<u>Scenedesmus microspina</u> Chod. (Figure 53 (1)).

The coenobia were composed of 4-8 cylindrical shaped cells, 4.5-6µ long and 1.5-2.5µ wide, in a linear series. The poles of the outside cells each had one fine spine about $\frac{1}{4}$-$\frac{1}{3}$ the length of the cell. Coenobia of this species were recorded from a mixed culture of filter bed algae collected from filter bed number 12 on June 18th, 1965.

<u>Scenedesmus spinosus</u> Chod. (Figure 53 (2)).

The coenobia were of 2-4 cells in a linear series. The cells were 17-20µ long and 4-6µ wide and were cylindrical in shape. Each pole of the outside cells bore a spine $\frac{1}{3}$-$\frac{3}{4}$ the length of the cell. The outside cells also bore 1-3 spines near the centre of the outside edge and at right angles to the cell. These latter spines were $\frac{1}{4}$-$\frac{1}{2}$




<u>Figure 53</u>. 1, <u>Senedesmus microspina</u>. 2a, <u>S. spinosus</u>. 2b, <u>S. spinosus</u> var. <u>bicaudatus</u>. 2c, <u>S. spinosus</u> var. <u>brevicauda</u>. 3a and b, <u>S. spicatus</u>. 4, <u>S. longispina</u> var. <u>asymmetricus</u> f. <u>crassicaudatus</u>.

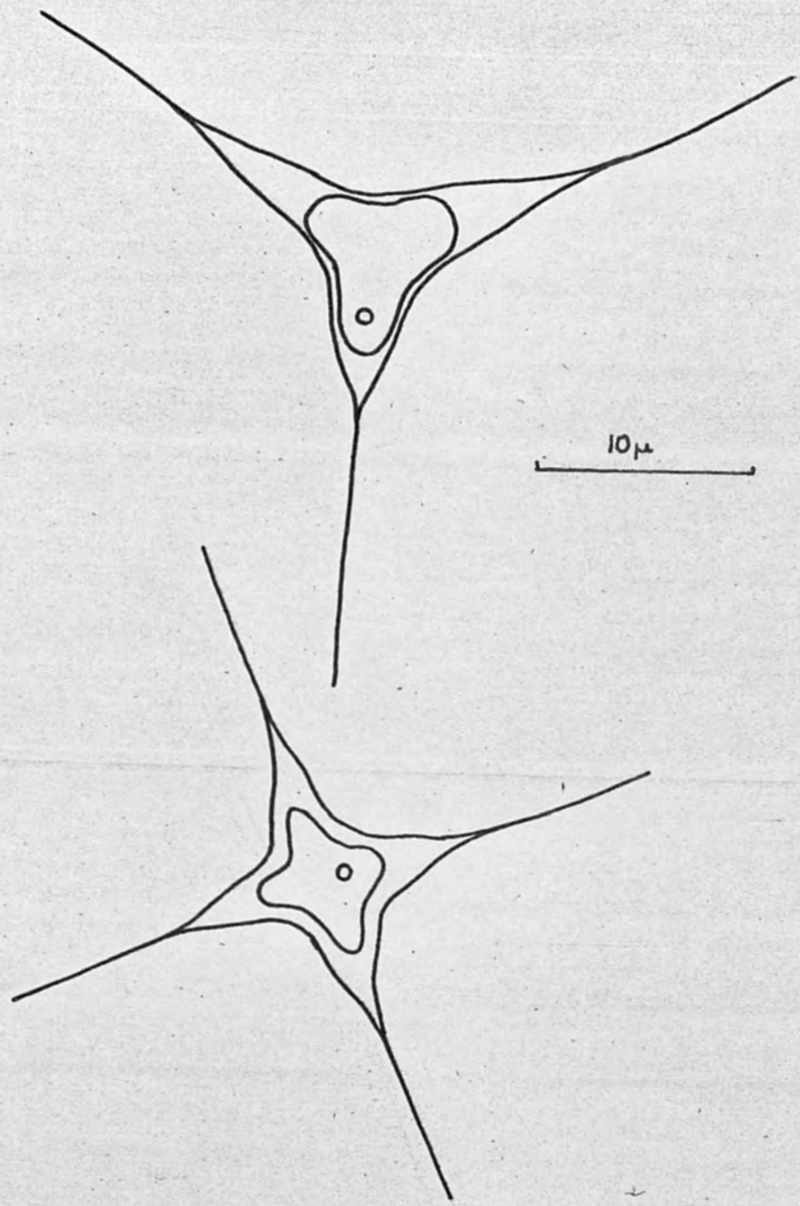
the length of the cell. The inner cells were either free of spines or bore one short spine at their poles. This species was recorded from the same culture as S. microspina. Although the morphology of the cells recorded was the same as those described by Uherkovich (1966) they were of much larger size, 17-20μ long and 4-6μ wide as compared with 5.5-12μ long and 2-4.5μ wide. It is possible that the increased size was due to exceptionally favourable growth conditions in the culture.

Scenedesmus spicatus W. et G.S. West. (Figure 53 (3)).

The coenobia consisted of 2-4-8 oval to eliptical cells 5-8μ long and 2.5-5μ wide in a linear series. The outside cells bore a fine, short ($^1/_6$ - $\frac{1}{3}$ the length of the cell) spine at each pole and two spines on the lateral wall of similar length. The inner cells were without spines. Coenobia of this species were recorded on May 29th, 1964, from the sand surface of filter bed number 32.

Treubaria setigera Bernard (Figure 54).

The cells were 3 and sometimes 4 angled. The angles were rounded and extending from each was a long delicately tapering spine thickened at the base. The cells were pyramidal or cruciform in shape with the surfaces between the angles concave. There appeared to be one or more chromatophores and at least one pyrenoid in each cell. The cells were 6-8μ wide and the spines 17-20μ long. This species was observed during the late summer periods in the reservoir water.




<u>Figure 54.</u> Treubaria setigera.

XANTHOPHYTA

Tribonema Debres & Solier.

This alga often occurred in large numbers in the inflow water to the slow sand filter beds. A large percentage of these algae was removed by the rotary microstrainers and those which were not did not seem to play an important part in the ecology of the filter beds. As the populations often consisted of a mixture of species they were, for convenience, grouped under the specific name of T. bombycinum (Ag.) Debr. & Sol. This species and its varieties has, more recently, been sub-divided into several distinct species, (Huber-Pestalozzi, 1962). Two of these species were commonly present; T. viride Pasch. cells 10-15µ br. and about three times as long as broad and T. vulgare Pasch. cells 6-8µ br. and about 3-4 times as long as broad. The former species was the more common.

BACILLARIOPHYTA

Cyclotella pseudostelligera Hustedt (Figure 56).

Cells were 6-10µ in diameter and 3-7.5µ deep. Spines were present but, owing to their small size, could only be observed with difficulty and their arrangement could not be determined. The markings on the valve were variable. The striae around the margin were between 1 and 3µ long. The central area was approximately circular and bore an inner ring of 5-8 striae (Figure 56). Cells of this species were found on the sand surface of each of the filter beds during April 1965, but not in large numbers. Owing to the small size

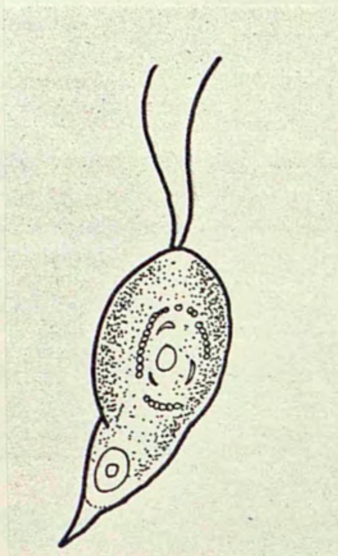


Figure 55.
Rhodomonas minuta

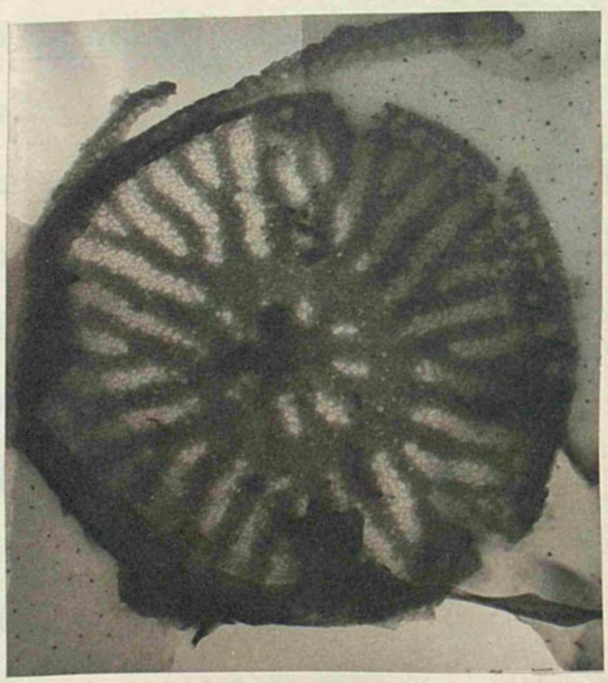


Figure 56.
Electron micrograph of Cyclotella pseudostelligera. Cells 6 microns in diameter.

of the cells and the fact that the cell contents were often indistinct after preservation, C. pseudostelligera may have occurred more often than they were observed during normal routine counting.

CRYPTOPHYTA

Rhodomonas minuta Skuja (Figure 55).

The cells were pyriform and were distinctly curved in the side view (Figure 55). They were 8-15µ long and 4-7µ wide. There was one greenish brown parietal chromatophore which ended either at or just above a shining globule, possibly volutin (Skuja 1948), at the base. There was a prominent pyrenoid sheathed with starch. This starch sheath appeared to be bipartite (see Lund, 1962). Most of the cells observed had a curved acute base. The range in size of the cells recorded covers both the type and the variety nannoplanctica (size 8-9µ long and 5-6µ wide). All sizes of cell occurred commonly in mixed populations and often in large numbers (see Chapter VI) in the reservoir throughout the period of study. As stated by Lund (1962), it may be open to doubt whether R. minuta should be separated into two varieties.

List of Algal Species.

A complete list of species recorded from all habitats studied has been prepared and is given below. The frequency of occurrence of each species is indicated by one of the following symbols:-

a = abundant, c = common, o = occasional, r = rare.

CHLOROPHYTA

Volvocales

| | |
|---|---|
| Spermatozopsis exsultans Korshikov | o |
| Carteria quadrangulata Pascher | c |
| Chlamydomonas spp. | a |
| Chlorogonium elongatum (Dangeard) France | r |
| Gonium pectorale Mull. | o |
| Pandorina morum (Mull.) Bory | c |
| Eudorina elegans Ehr. | c |
| Volvox aureus Ehr. | c |

Chlorococcales

| | |
|---|---|
| Pediastrum boryanum (Turp.) Menegh | c |
| P. clathratum Lemm. | c |
| P. duplex Meyen | c |
| P. tetras (Ehr.) Ralfs | o |
| Chlorella sp. | o |
| Micractinium pusillum Fres. | c |
| Oocystis elliptica West | a |
| O. lacustris | o |
| O. crassa Wittr. | c |
| O. solitaria Wittr. | c |
| Chodatella subsala Lemm. | o |
| Tetraedron minimum (A. Br.) Hansg. | c |
| T. caudatum (Corda) Hansg. | o |

| | |
|---|---|
| T. regulare Kitz. | c |
| Actinastrum hantzschii Lagerh. | o |
| Dictyosphaerium pulchellum Wood | o |
| Crucigenia tetrapedia (Kirch.) W. et E.S. West | o |
| Ankistrodesmus falcatus (Corda) Ralfs | a |
| A. pseudomirabilis Korshikov | o |
| Tetrastrum staurogeniaeforme (Schroed.) Lemm. | c |
| T. heterocanthum (Nordst.) Chod. | o |
| T. hastiferum (Arnoldi) Korshikov | r |
| T. glabrum (Roll) Ahlstr. et Tiff. | o |
| Scenedesmus acutus Meyen | a |
| S. acutus f. tetradesmiformis (Woloz) Uherk. | r |
| S. acutus f. alternans Hortob. | o |
| S. acutus f. semiellipticus Uherk. | r |
| S. acuminatus (Lagerh.) Chod. | c |
| S. acuminatus f. maximus Uherk. | o |
| S. acuminatus f. tortuosus (Skuja) Uherk. | r |
| S. ecornis (Ralfs) Chod. | a |
| S. ecornis var. disciformis Chod. | c |
| S. ovalalternans Chod. | o |
| S. arcuatus Lemm. | c |
| S. naegli Breb | r |
| S. ? pecsensis Uherk. | r |
| S. acutiformis Schroeder | o |
| S. armatus Chod. | r |

| | |
|---|---|
| S. quadricauda (Turp.) Breb. | c |
| S. quadricauda var. maxima W. et E.S. West | o |
| S. quadricauda var. longispina (Chod.) E.M. Smith | o |
| S. quadricauda var. quadrispina (Chod.) E.M. Smith | o |
| S. quadricauda var. westuf. heterospinosus (Hortob) | r |
| S. quadricauda var. biornatus Kiss | r |
| S. bicaudatus (Hansg.) Chod. | o |
| S. bicaudatus var. brevicaudatus Hortob | o |
| S. intermedius var. bicaudatus | r |
| S. intermedius var. balatonicus Hortob | r |
| S. microspina Chod. | r |
| S. spinosus Chod. | r |
| S. spicatus W. et E.S. West | r |
| Coelastrum microporum Nag. | c |
| Ulotrichales | |
| Ulothrix zonata (Web. et Mohr) Kutz. | o |
| U. tenuissima Kitz. | a |
| U. tenerrima | a |
| Geminella sp. | r |
| Cladophora sp. | a |
| Chaetophorales | |
| Chaetophora sp. | r |
| Stigeoclonium tenue Kutz. | o |

| | |
|---|---|
| S. ? falklandicum Butcher | c |
| Coleochaete sp. | r |
| Ulvella sp. | c |
| **Conjugales** | |
| Spirogyra varians (Hass.) Kutz. | r |
| Closterium moniliferum Ehr. | o |
| C. gracile Breb. | o |
| Cosmarium sp. | o |
| Arthrodesmus incus (Breb.) Hass. | r |
| Staurastrum sp. | o |
| <u>BACILLARIOPHYTA</u> | |
| **Centrales** | |
| Melosira varians C.A. Ag. | a |
| M. granulata (Ehr.) Ralfs | o |
| M. granulata v. angustissima | o |
| Cyclotella meneghiniana Kutz | o |
| C. compta (Ehr.) Kutz. | o |
| C. pseudostelligera Hustedt | o |
| Stephanodiscus astraea (Ehr.) Grun | c |
| S. astraea v. intermedia Fricke | a |
| S. astraea v. minutula (Kutz.) Grunow | c |
| S. astraea ? v. incertus Cleve. | o |
| S. hantzschii Grun. | a |

Pennales

| | |
|---|---|
| Tabellaria fenestrata (Lyngb.) Kutz | o |
| Meridion circulare (Grev.) Ag. | o |
| Diatoma vulgare Ag. | c |
| D. elongatum Ag. | o |
| Fragilaria crotonensis Kitton | a |
| F. capucina Desmazieres | c |
| Asterionalle formosa Hass | a |
| Synedra ulna (Nitzsch) Ehr. | c |
| S. acus Kutz. | c |
| Achnanthes minutissima Kutz. | a |
| A. affinis Grun. | o |
| Achnanthes sp. | r |
| Cocconeis placentula Ehr. | c |
| Rhoicosphenia curvata Grun. | c |
| Navicula cryptocephala Kutz. | o |
| N. viridis Kutz. | o |
| N. radiosa Kutz. | r |
| N. anglica Ralfs | r |
| N. hasta Pantocsek | r |
| Pinnularia microstauron Ehr. Cl. | c |
| P. microstauron v. Brebisonii (Kutz.) Hustedt | o |
| P. viridis (Nitzsch.) Ehr. | o |
| P. debesi Hustedt | o |
| P. divergens W. Sm. | o |

| | |
|---|---|
| Stauroneis sp. | o |
| Gyrosigma sp. | r |
| Gomphonema parvulum Ehr. | o |
| Amphora ovalis Kutz. | c |
| Cymbella turgida (Greg.) Cl. | c |
| C. ventricosa Kutz. | a |
| C. lanceolata Ehr. | a |
| C. prostrata Berk. | o |
| C. cistula Hemp | o |
| C. subaequalis Grun. et Van Heurck. | o |
| C. caespitosa Kutz. | c |
| Hantzschia amphioxys (Ehr.) Grun. | o |
| Nitzschia tryblionella Hantz | o |
| N. dissipata (Kutz.) Grun. | o |
| N. palea (Kutz.) W. Sm. | c |
| N. sigmoidea (Ehr.) W. Sm. | o |
| N. acicularis W. Sm. | a |
| N. linearis W. Sm. | a |
| N. baccata | o |
| N. dubia W. Sm. | r |
| N. recta Hantzsch. | o |
| Cymatopleura solea (Breb.) W. Sm. | c |
| C. elliptica (Breb.) W. Sm. | o |
| Surirella turgida W. Sm. | o |
| S. ovata Kutz. | a |

| | |
|---|---|
| S. robusta Ehr. | o |
| S. ovalis Breb. | o |
| Campylodiscus noricul v. hibernica (Ehr.) Grun. | r |
| CHRYSOPHYTA | |
| Synura ulvella Ehr. | o |
| Dinobryon divergens Imhob. | r |
| Mallomonas sp. | r |
| XANTHOPHYTA | |
| Tribonema viride Pasch. | c |
| T. vulgare Pasch. | o |
| EUGLENOPHYTA | |
| Euglena viridis Ehr. | o |
| Euglena sp. | o |
| Phacus sp. | o |
| CRYPTOPHYTA | |
| Cryptomonas ovata Ehr. | c |
| C. curvata Ehr. | o |
| Rhodomonas minuta Skuja | a |
| PYRROPHYTA | |
| Gymnodinium sp. | r |

| | |
|---|---|
| Peridinium sp. | r |
| Ceratium hirundinella O.F.M. | o |
| CYANOPHYTA | |
| Merismopedia glauca (Ehr.) Nag. | o |
| M. elegans A. Br. | o |
| Microcystis aeruginosa Kutz. | o |
| M. flos-aquae (Wittr.) Kirch. | o |
| Oscillatoria limosa Ag. | o |
| O. tenuis Ag. | o |
| O. rubescens D.C. | o |
| Spirulina major Kutz. | r |
| Lyngbya sp. | o |
| L. versicola | c |
| Anabaena flos-aquae (Lyngb.) Breb. | o |
| A. circinalis (Kutz.) Hansg. | o |
| Aphanizomenon flos-aquae (L.) Ralfs | o |
| Phormidium sp. | o |

## XIII. Summary and Conclusions.

1. During the period August 1963 to June 1966 an investigation of the algal flora of slow sand filter beds was carried out. This investigation included more than two years field observations (August, 1963 to December, 1965) of the algal flora in the filter beds.

2. A. Three groups of algae were studied: (i) Planktonic, (ii) Epipelic and (iii) Attached bottom living.

B. Several aspects of the ecology of the algae were investigated. The major chemical inorganic nutrients were measured and meteorological data was obtained from Kew Observatory. All of these factors were analysed with respect to the environmental conditions in the filter beds.

3. The algal flora of a filter bed can be divided into two categories: (i) Those which are present in large numbers simply because of accumulation. These species, planktonic in origin, were derived from large populations in the supply reservoir. They passed onto the filter beds in large numbers and accumulated but did not increase in numbers to any extent by active division. Algae in this category include <u>Stephanodiscus hantzschii</u>, <u>S. astraea</u>, <u>Asterionella formosa</u> and <u>Fragilaria crotonensis</u>.

(ii) The second category includes those species which entered the filter beds, often in small numbers, and then produced large populations by active division. Algae in this category include <u>Chamydomonas sp.</u>, <u>Scenedesmus spp.</u>, <u>Melosira varians</u>, <u>Nitzschia linearis</u> and <u>Cymbella spp.</u>

4. Certain broad conclusions can be drawn concerning the periodicity of the algae in the filter beds.

(i) Certain species, which increased by accumulation, had an annual periodicity which was a close reflection of the reservoir populations, e.g. Stephanodiscus astraea and Asterionella formosa.

(ii) The species which increased greatly by active division in the filter bed supernatant water often had their growth initiated by unusual conditions such as the treatment of the water with chemicals, e.g. chlorine and copper sulphate.

(iii) The epipelic populations were dominated by diatoms throughout most of the year. On occasions when chemical treatments were applied to the water, other algae became co-dominant, e.g. Scenedesmus spp.

(iv) The attached algal flora was dominated by pennate diatoms for most of the year. During the spring filamentous became dominant. Species such as Scenedesmus spp. became co-dominant after periods of chemical treatment. The highest rates of colonization of the glass slides were usually shown by the pennate diatoms. In January and February, however, filamentous algae showed a higher rate of colonization.

5. The distribution of the algae both in the supernatant water and on the sand surface was also investigated. During calm periods the algae in the supernatant water were found to form stratified populations often with minimum concentrations between 3'-4' (91.5-122cms.) possibly because the inflow water did not mix immediately with the water already there. Motile forms maintained themselves near the surface in a position most favourable to photosynthesis. When present in large numbers, however, they often shaded the sand surface populations and

may have caused light limitation. The sand surface populations were distributed according to their origin. Planktonic species which had settled out occurred in maximum numbers near to the centre of the bed. The distribution of the filamentous species was variable but often depended upon wind induced current directions. The unattached bottom living diatoms often avoided areas densely populated by filamentous forms, possibly to avoid shading and thus light limitation and also because of nutrient competition.

6. During the period of study certain experiments were carried out to clarify some of the ecological data. They were as follows:-

(i) The flora of an 800 L in-situ enclosed polythene bag, filled with filtered filter bed supernatant water and seeded with filter bed algae, was compared with that of the sand surface of a filter bed at the same time. The flora of the sand surface, which was probably not subject to nutrient limitation, was diatom dominated, whereas in the polythene bag Chlorophyceae assumed dominance after silicon became limiting. It was possible that there was some interaction between certain species causing inhibition. It is believed that this occurred between Nitzschia palea and Chlorella sp. In addition, it is possible that self-stimulation and inhibition occurred, e.g. in Scenedesmus (Saunders, 1957).

(ii) a. The toxicity of copper sulphate to laboratory cultures of Asterionella formosa, Fragilaria crotonensis and Scenedesmus dimorphus was tested. The growth of the two diatoms was severely impaired at concentrations above 0.1mg/L copper whereas that of Scenedesmus dimorphus was only slightly reduced. Copper sulphate treatment of the reservoir and filter bed water gave final concentrations of 0.19mg/L copper, enough to virtually prevent these two diatoms from increasing.

(ii) b. Growth rate experiments were carried out on cultured populations of *Stephanodiscus astraea* exposed in glass bottles in filter beds throughout one year. A three phase cycle was observed dependent upon light and temperature. The timing of the growth rate cycles corresponded to observed increases and decreases in the average cell diameters of the natural populations.

(iii) An experiment to determine the penetration of the algae into the sand of a slow sand filter bed was carried out. Live cells of most of the species of algae recorded in the inflow water were found below a depth of 7" (2.5cms.). At this depth many cells would be left behind after a filter bed was cleaned to act as an innoculum when the bed was filled for use.

7. Observations were made on the cell sizes of *Stephanodiscus astraea* and *Asterionella formosa*. An annual increase and decrease in size of frustules of these species was observed. The variation in the ornamentation and size of the frustules of *S. astraea* was also investigated. Considerable variation within each population was discovered, covering all of the named varieties of *Stephanodiscus* with the exception of *S. astraea v. niagarae*. It is possible that some of the differences in ornamentation ascribed to different varieties is just variation within one population.

Appendix 1.

The Construction and operation of the submersible light meters.

The rate of photosynthesis and hence the potential growth rate of algae is dependent upon the amount and quality of the available light. Data for solar radiation incident upon the water surface was obtained from the Meteorological Office Kew Observatory (see Chapter III). To measure the percentage of this incident radiation reaching the sand surface of the filter beds a submersible light meter was constructed.

The design of Atkins et al. (1938) was used for the construction of the submersible light meters, but with brass and not gun metal as the housing material. A matched pair of Sangamo Weston model 127 electroselenium cells were used as light sensors. The current produced was measured on a 500 microampere galvonometer of low effective internal resistance. The cells and the meter were wired into an electrical circuit as shown in Figure 57. The circuit was arranged so that the cells were back to back (i.e. the +ve of one was connected to the -ve of the other) so that, with both of them in circuit, the difference between the surface and submerged cells could be read directly on the galvonometer.

For the purpose of this study it was sufficient to be able to measure the underwater illumination as a percentage of the surface illumination at the same spectral band. It was thus only necessary to standardise the two cells relative to each other and not to calibrate them absolutely. The cells were standardised by placing them side by side on the shore, filling the filter wells with water, placing diffuser glasses in each holder and then balancing the two cells by means of the potentiometer to give a zero reading on the galvonometer. Care was taken to ensure that both cells were receiving the same amounts of light during the procedure. The cells were standardised before taking measurements at each of the wavebands studied.

Measurements were made, after standardisation, by placing one cell on the side and lowering the other cell into the filter bed to the required depth. The cell at the surface was then switched on to give the total incident radiation reading. Both cells were then switched on to give the difference between the surface and the submerged illumination. The percentage of the surface illumination at any given depth could thus be determined. Readings were taken at several wavebands by using coloured glass filters of known absorption (see Chapter III). Care was taken, when using filters, to have the filter well of both cells full of water before inserting the filter glass so as not to trap any air bubbles between the filters and the glass of the cell holder. The diffuser glass was always placed on top of the filters. These precautions minimised light losses through the layers of glass and also by reflection at the glass surface. Care was also taken to ensure that both the shore and the submerged cells remained horizontal whilst readings were being taken.

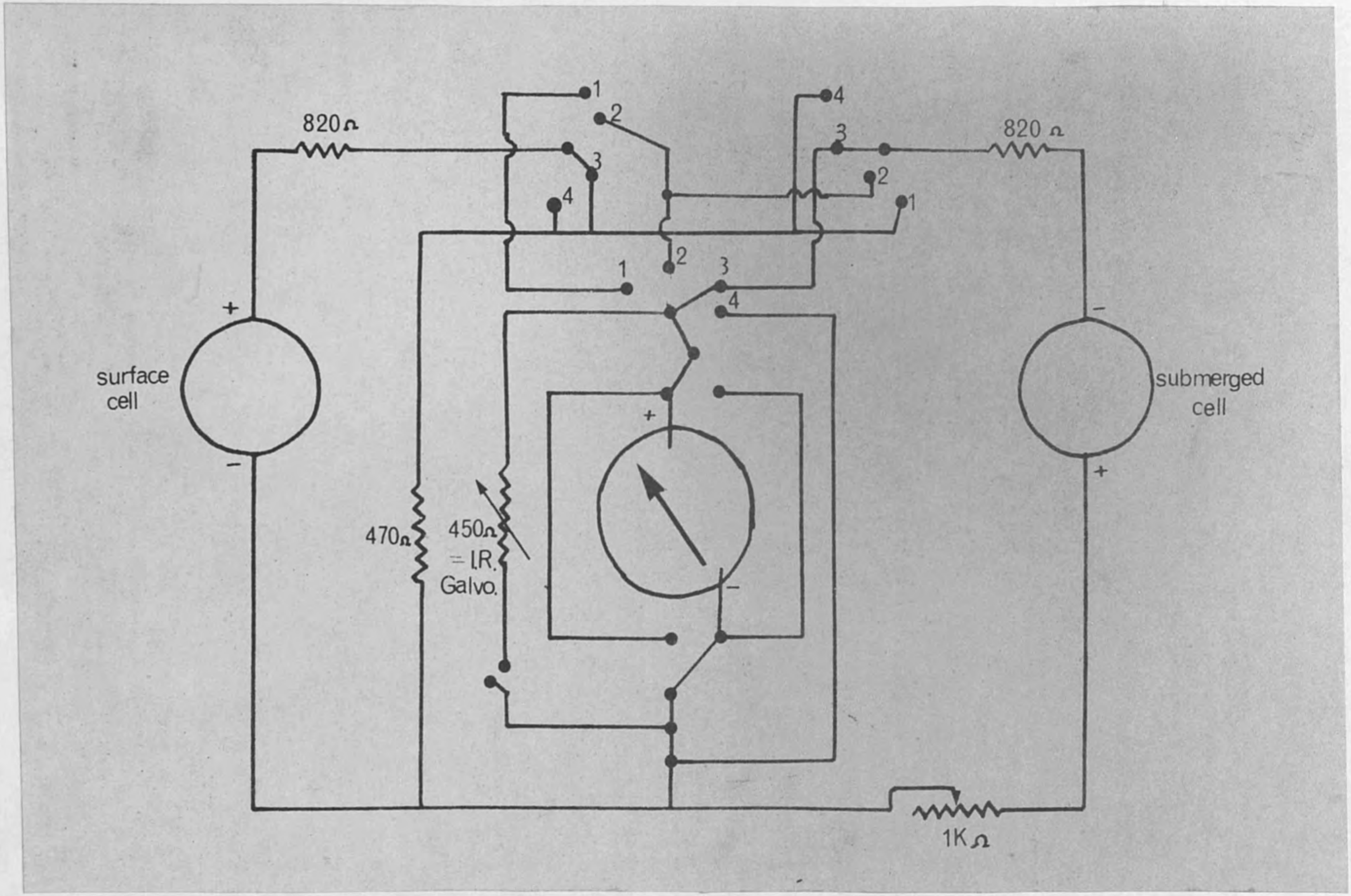



__Figure 57__. The circuit diagram for the wiring of the submersible light meters.

Appendix 2.

Statistical analyses.

In planning the sampling programme for the algae in the filter beds a preliminary series of collections were made and the results tested statistically. Several arbitrary sampling stations (between 6-10) were decided upon and all samples from the supernatant water and the unattached algae of the sand surface were subsequently taken from, or near to, these stations. The traditional belief of Metropolitan Water Board Biologists has been that the algal populations on the sand surface of a filter bed are homogeneous. This belief was also stated by Brook (1954). As the earliest samples collected, however, suggested that this thesis is not always true for all of the species, present tests were made, species by species, and by ecological groups, as to its general validity. For the purpose of this study representative samples of the filter bed algae were required so the randomness of distribution of the algae was tested.

For the first series of samples, collected on July 28th, 1963, the filter bed was divided into six plots, each plot consisting of two samples. The variance within each plot and between separate plots of the various algae present was then determined using Snedecor's F test (Snedecor, 1940). The results are given in table 11. Included in the table are the degrees of freedom and the variance ratio, F, and conclusions at a significance level $p = 0.05$. From this analysis it is clear that the planktonic species were homogeneously distributed both in the supernatant water and on the sand surface. The epipelic algae

Table 11. The variance ratio, F, of individual species within samples and between groups of samples collected on July 28th 1963.

| Species | Sample | n1 | n2 | F | Conclusion (at p= 0.05) |
|---|---|---|---|---|---|
| Oocystis spp. | Supernatant water | 6 | 5 | 2.18 | Homogeneous |
| .. | Sand surface | 6 | 5 | 3.14 | .. |
| Rhodomonas minuta | Supernatant water | 6 | 5 | 3.81 | .. |
| .. | Sand surface | 6 | 5 | 3.42 | .. |
| Stephanodiscus astraea | Supernatant water | 6 | 5 | 2.16 | .. |
| .. | Sand surface | 6 | 5 | 2.73 | .. |
| Melosira varians | .. | 6 | 5 | 11.63 | Several populations |
| Nitzschia palea | .. | 6 | 5 | 2.22 | Homogeneous |
| N. linearis | .. | 6 | 5 | 2.72 | .. |
| Cymbella spp. | .. | 6 | 5 | 3.66 | .. |
| Lyngbya sp. | .. | 6 | 5 | 30.60 | Several populations |

were also homogeneously distributed but the filamentous algae, such as Melosira and Lyngbya, were present as several distinct populations.

A second series of samples from the sand surface was collected on 17th September, 1963. The filter bed was again sub-divided but this time into five plots, each plot consisting of two samples. With this series of samples one half of each sample was counted separately and the other half from each of the ten samples shaken together in a 5L flask. Ten aliquots were then removed for counting. The variation of these aliquots from the original individual samples was then determined. The variance ratio, F, was again determined and the results are given in table 12, together with conclusions at a significance level of $p = 0.05$. The planktonic algae on the sand surface were homogeneously distributed but both the epipelic and the filamentous algae came from several distinct populations. A possible explanation for the epipelic population being homogeneous in the first collection and not in the second is that the first series were collected from a filter bed which had been in operation for a much longer time and the populations were much larger. Instead of forming discreet patches the populations had grown to such an extent that they overlapped each other. The result was one large population formed by the smaller ones coalescing. The aliquots taken after shaking together one set of halves of the samples were not significantly different from the individual sample means for each of the population types.

To obtain a representative sample of the algae on the sand surface, therefore, one could not just take one sample from one station. To

<u>Table 12.</u> The variance ratio, F, of each ecological group within and between samples collected on September 17th 1963 and between aliquots from a bulked sample and the original samples.

| Ecological group | Sample | n1 | n2 | F | Conclusion |
|---|---|---|---|---|---|
| Planktonic | sand surface | 5 | 4 | 1.67 | Homogeneous |
| Epipelic | .. | 4 | 5 | 8.14 | Several populations |
| Filamentous | .. | 4 | 5 | 5.14 | .. |
| Planktonic | Aliquots | 14 | 1 | 34.87 | Homogeneous |
| Epipelic | .. | 14 | 1 | 4.51 | .. |
| Filamentous | .. | 14 | 1 | 51.39 | .. |

obtain such a representative sample collections were made from at least ten stations around the filter bed. These samples were then shaken together in a 5L. flask and an aliquot removed as the representative sample for counting.

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**Slow sand filter bed algae and their production in an *in situ* polythene bag**

An 800 l. enclosed polythene bag was suspended in a slow sand filter bed and filled with glass-fibre filtered filter bed water free from plants and animals. It was then inoculated with a natural population of filter bed algae. Their growth was followed and compared with that of the algae on the sand surface of the filter bed. Regular analyses were made for particulate carbon, dry weight, loss-on-ignition, nitrogen, phosphorus and silicon.

The flora of the sand surface, which was probably not subject to limitation by nutrients, was predominantly diatom dominated, whereas in the polythene bag Chlorophyceae assumed dominance after silicon became limiting. The possibility of stimulation and inhibition by growth factors was discussed and a comparison was made between production as measured by particulate carbon, loss-on-ignition, dry weight and weight calculated from volume.

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# THE REMOVAL OF ALGAE BY MICROSTRAINING

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*(Metropolitan Water Board)**

## INTRODUCTION

Rotary microstrainers have been in use as first stage filters at the Ashford Common Works of the Metropolitan Water Board since 1958. The efficiency of rotary microstrainers and rapid gravity sand filters as alternative methods of first stage filtration has been discussed in terms of filterability indices (Mackenzie[1] and Windle Taylor[2]). The relative merits of these two systems of removing suspended matter and the subsequent effects on slow sand filters have been reported by Ridley[3]. In this paper an attempt has been made to determine the numbers and size range of algae which are either retained by or pass through the mark 1 mesh of the rotary microstrainers.

## EXPERIMENTAL

The source of supply to Ashford Common Works is normally Queen Mary reservoir (Windle Taylor[4-8]). The inflowing water and the effluent from the rotary microstrainers were examined at weekly intervals during 1964 and 1965. The numbers of algae were determined according to Lund[9] and Lund et al[10]. Fourteen genera of algae, varying considerably in shape and size, occurred as major constituents of the phytoplankton during the period of study. They included unicellular algae, colonial forms, filaments and loosely attached chains. Expressing the results as numbers of algal cells per ml is somewhat misleading because of this

**Table 1**

**Size Range and Volume of Single Cells of the Algae Recorded**

| Species | Dimensions in microns Maximum | Dimensions in microns Minimum | Average volume in cubic microns |
|---|---|---|---|
| *Melosira varians* | 35 | 8 | 2 000 |
| *Asterionella formosa* | 130 | 2 | 756 |
| *Fragilaria crotonensis* | 150 | 2 | 640 |
| *Tribonema bombycinum* | 45 | 6 | 1 080 |
| *Chlamydomonas spp* | 30 | 6 | 1 760 |
| *Scenedesmus spp* | 30 | 3 | 1 000 |
| *Cryptomonas spp* | 60 | 5 | 2 000 |
| *Rhodomonas minuta* | 15 | 4 | 392 |
| *Oocystis spp* | 30 | 5 | 3 050 |
| *Ankistrodesmus spp* | 150 | 2 | 314 |
| *Stephanodiscus astraea* | 70 | 8 | 13 700 |
| *S. hantzchii* | 20 | 5 | 795 |
| *Synedra spp* | 350 | 5 | 4 220 |
| *Nitzschia spp* | 500 | 3 | 3 450 |
| Microstrainer mesh | 45 | 38 | — |

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variation in size and form; for example, when trying to compare a large alga (e.g. *Cladophora*) with a small one (e.g. *Ankistrodesmus*). To overcome this difficulty results are here expressed as the total volume of each algal species present per ml of water (for detailed discussion see Bellinger[11]). The average volume and the maximum and minimum dimensions of the major species of algae present are given in Table 1.

The maximum and minimum dimensions of the micromesh screen are 45 and 38 microns respectively. Table 1 shows that any of the algae present could pass through the micromesh screen provided they were presented to an orifice at an angle which ensured alignment with their smallest dimension. Some algae were present as filaments or colonies and these were more likely to be retained by the strainers. When, however, the colonies or filaments were broken up, either by water turbulence or by algicidal treatment, individual cells or small aggregations were able to pass through the screens.

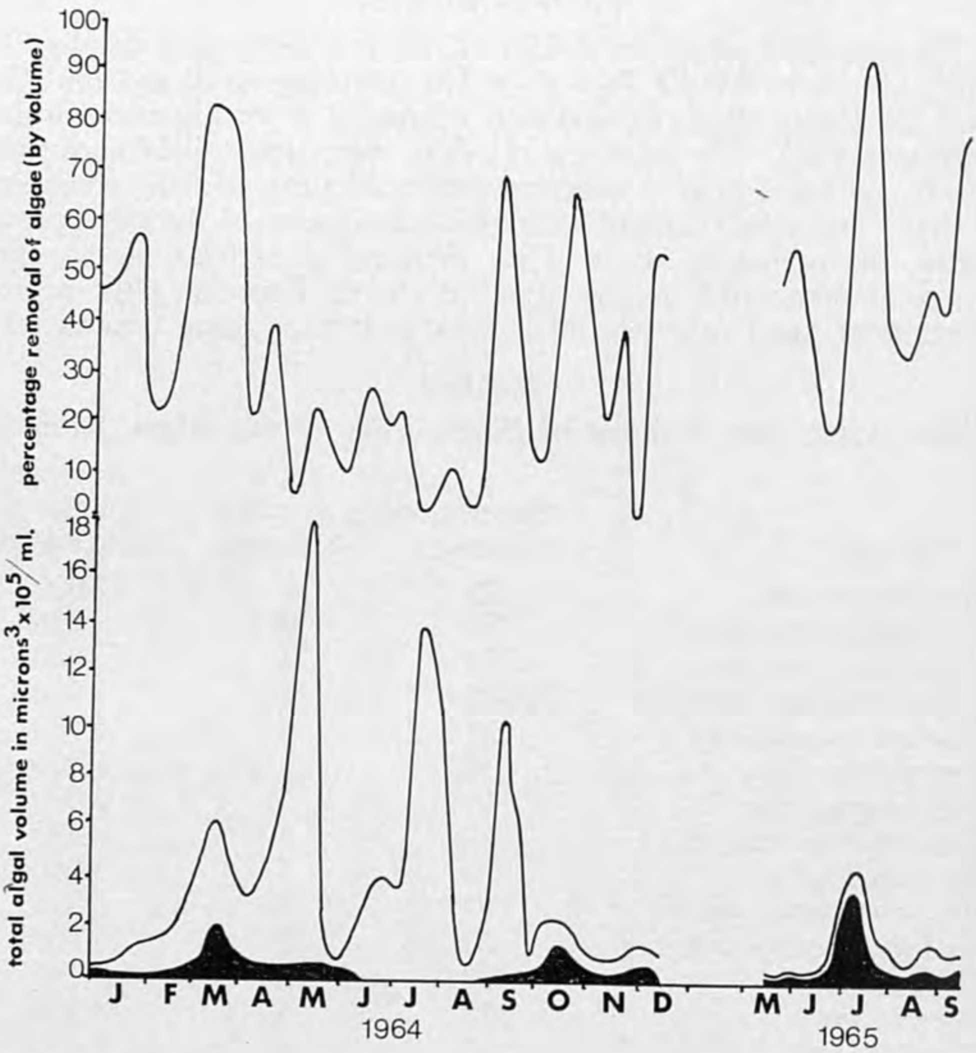


**Fig. 1.** Total volume of algae and percentage retention by microstrainers *(Note: Solid black portion is volume of A. formosa, M. varians, F. crotonensis and T. bombycinum)*

Fig. 1 gives the results of periodic counts expressed as volumes of algal material in the water before and after microstraining. Fluctuations in biological quality can be seen, but substantial reductions in numbers of algae in the incoming water were often due to algicidal control measures in the reservoir supplying the microstrainers. The removal of algae ranged from below 10% to above 90%. As the process of filtration by microstrainers is entirely physical, the removal of algae is discussed according to their physical shape and size.

FILAMENTOUS COLONIAL ALGAE

The algae in this group included the diatoms *Melosira varians* and *Fragilaria crotonensis* and the yellow green alga *Tribonema bombycinum*. Of these, *M. varians* was the least abundant throughout the period, occurring in but small concentrations during the spring and summer of 1964. In early December, 1964, the highest numbers were recorded and over 90% removal by the microstrainers was effected. This high removal occurred on other occasions when high numbers were present. *T. bombycinum* occurred for a short time during the autumn of 1964 and for a longer period during the summer and autumn of 1965. High percentage removals were obtained on all of these occasions. *F. crotonensis* occurred during the autumns of 1964 and 1965. On all occasions removals were high, usually above 80%.

NON-FILAMENTOUS COLONIAL ALGAE

*Asterionella formosa*, a diatom, and *Scenedesmus spp*, chlorophyceans, were the main species recorded in this group. *A. formosa* forms stellate colonies of up to 32 cells; it occurred mainly during the spring of 1964, and on every occasion percentage removals were high. *Scenedesmus spp* form 2–4–8 celled colonies which may tend to clump together during periods of rapid growth. These algae were present throughout most of the period, maximum numbers of cells being recorded during the summer and autumn months, but the percentage removal was almost always extremely low.

UNICELLULAR ALGAE

This group included members of the Chlorophyceae, Cryptophyceae and Bacillariophyceae. *Chlamydomonas spp*, motile chlorophyceans, occurred in greatest numbers during the late winter and early spring of 1964 and also in the autumn of 1964. The percentage removed by the microstrainers was high when the growths were at their maxima. *Cryptomonas spp* and *Rhodomonas minuta*, both cryptophyceans, are also motile forms. *Cryptomonas spp* were recorded on a number of occasions but were not efficiently removed by the microstrainers. *Rhodomonas minuta*, one of the smallest algae recorded, occurred throughout most of the period. The densest populations were present during both the spring and autumn of 1964 and also the autumn of 1965. On only two occasions did removal exceed 50%.

Non-motile chlorophyceans present included *Oocystis spp* and *Ankistrodesmus spp*. The former occurred mainly during the summer and autumn of 1964 and 1965. Removal exceeded 50% on only one occasion, even though the number of cells present in the inflow water often exceeded 250/ml. *Ankistrodesmus spp* included the smallest algae recorded and were present throughout the entire period, the highest concentrations occurring during the spring of 1964. High percentage removals were recorded on only two occasions.

*Stephanodiscus hantzschii* and *S. astraea* were the discoid, and *Nitzschia spp* and *Synedra spp* the pennate, diatoms present. Of these *S. hantzschii* occasionally occurred in loose chains of 6–8 cells. When it was present during the late spring of 1964 the percentage removed by the microstrainers was low but in the summer and autumn of 1965 percentage removals were fairly high on two occasions. *S. astraea* was present throughout the entire period with maximum numbers occurring in the late spring of 1964. High percentage removals were obtained frequently during the spring of 1964 but only once during September 1964, when numbers in the inflow water reached 1 200 cells per ml. *Synedra spp* occurred only intermittently throughout the period and cell concentrations were always low. *Nitzschia spp* were nearly always present and reached maximum numbers during the summer and autumn of 1964. Neither of these pennate diatoms was efficiently removed by the rotary microstrainers.

## DISCUSSION

The main purpose of rotary microstrainers is to remove the larger organic and inorganic particles from the inflow water, and thus to reduce the amount of suspended material passing to the slow sand filters. Improved secondary filtration economy is not necessarily always attained, despite high percentage removal of algae by the microstrainers. This applies to rapid sand filtration as well as to microstraining (Ridley[7]). Certain types of algae, small enough to pass through the micromesh screen, will effectively clog the surface of slow sand filters at a rate depending upon the numbers in the microstrainer effluent. Other algae, however, whilst passing through the rotary microstrainers in only small numbers, rapidly multiply in the supernatant water and on the sand surface of the secondary filter basin and clog the sand interstices. During these studies both of the above types of algae were encountered. Typical of the first type was *S. hantzschii* and of the second *Chlamydomonas spp* and *Scenedesmus spp*.

Every alga recorded could have passed through the microstrainer mesh if present as single cells. As they do not always do so other factors must be involved.

Algae present as colonies may have been too large to pass through the micromesh screen and this would account for the high removal of *A. formosa, F. crotonensis* and *T. bombycinum*. When these species were retained they tended to form a mesh-like layer on the inside surface of the micromesh gauze. This would have acted as an important additional filtering mechanism, although only for short periods as the gauze was backwashed every few minutes as the drum rotated. This additional layer would have a smaller effective orifice size and would thus retain smaller particles. Some evidence of this effect is shown in Table 2 where occasions on which 50% or more removal occurred are recorded. On these occasions either *A. formosa, F. crotonensis* and *T. bombycinum* or the filamentous diatom *Melosira varians* was present. When these larger algae were absent, the percentage of algae retained by the microstrainers was usually reduced. Removal of the algae by the microstrainers was not solely a function of the numbers present but also of the size or shape of particular genera or species.

**Table 2**
**Occurrence and Removal of Algae**

*1964*

| | January 31st | February 7th | February 21st | February 28th | March 6th | March 13th | March 20th | April 3rd | April 17th | April 24th | May 1st | May 8th | July 17th | August 21st | September 4th | September 18th | September 25th | October 2nd | October 16th | October 30th | November 6th | November 13th | December 4th |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| *Chlamydomonas spp* | R | R | R | + | R | R | + | R | R | + | + | | | | | R | | | | | + | R | |
| *Oocystis spp* | | | | | | | | | | | | | + | + | + | + | + | + | + | + | | | |
| *Ankistrodesmus spp* | + | + | + | R | + | R | + | + | + | + | + | + | + | + | + | + | + | + | + | + | | | + |
| *Scenedesmus spp* | | | | | | | R | | | + | | | R | R | | R | | | | | | | |
| *Stephanodiscus astraea* | + | + | + | R | R | R | + | + | + | + | + | R | + | + | R | + | + | + | + | + | + | + | + |
| *S. hantzschii* | | | | | | | | | | | + | + | + | + | + | + | | | | | | | |
| *Synedra spp* | | | + | | | + | + | | + | + | + | + | | | | | | + | | | | | |
| *Nitzschia spp* | + | + | + | + | + | + | + | + | + | R | + | + | + | + | + | + | + | + | + | + | + | + | + |
| *Rhodomonas minuta* | + | + | + | R | R | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| *Cryptomonas spp* | + | + | + | + | R | R | + | + | + | + | + | + | | | | | | | + | R | R | + | + |
| *Melosira varians* | | | | | | | | | | | | | | | | | | | | | | | R |
| *Asterionella formosa* | | R | R | R | R | R | R | R | R | R | R | R | | | | | | | | | | | |
| *Fragilaria crotonensis* | | | | | | | | | | | | | | | | | | R | R | R | | | |
| *Tribonema bombycinum* | | | | | | | | | | | | | | | | R | R | | | | | | |

*1965*

| | May 18th | July 3rd | July 10th | August 2nd | August 9th | August 17th | August 24th | September 1st |
|---|---|---|---|---|---|---|---|---|
| *Chlamydomonas spp* | | + | + | | | | | |
| *Oocystis spp* | | + | + | R | + | + | + | + |
| *Ankistrodesmus spp* | | + | + | + | + | | + | + |
| *Scenedesmus spp* | | | | | | | | |
| *Stephanodiscus astraea* | + | + | + | + | + | + | + | + |
| *S. hantzschii* | | R | + | | | + | R | |
| *Synedra spp* | | | | | | + | + | + |
| *Nitzschia spp* | | | + | | + | | + | + |
| *Rhodomonas minuta* | + | + | + | + | + | + | + | + |
| *Cryptomonas spp* | | | | | | | | |
| *Melosira varians* | | R | | | | | | |
| *Asterionella formosa* | | | | | | | | |
| *Fragilaria crotonensis* | | | | R | R | R | R | R |
| *Tribonema bombycinum* | | R | R | R | R | R | R | R |

*Key:* + = *Not less than* 100 *cells of algae present per ml.*
R = 50% *or more removed by microstraining when present at a concentration of at least* 100 *cells per ml.*

Some exceptions did occur as, for example, when high percentage removals of some algae were obtained even when the larger algae were absent. On September 4th, 1964, *S. astraea* contributed more than 90% of the total algal population present, a cell count of 1 280 per ml being recorded in the inflow water. These cells were fairly large, with an average diameter of 36 microns, and as they were present in such large numbers they could themselves have formed a secondary filtering layer within the microstrainer and thus accounted for the high percentage removal.

*Scenedesmus spp* were also retained in large numbers on some occasions. Some species of *Scenedesmus* form colonies which tend to group together in clumps, for example *S. bijuga*. When this species formed a high percentage of the population, removal by the microstrainers was high, probably because the clumps were too large to pass through the micromesh apertures. On one occasion over 50% of the cells of *Chlamydomonas spp* and *Cryptomonas spp* were retained by the microstrainers. At this time none of the algae capable of forming an additional filtering layer was present in large numbers. As both of the above species are unicellular forms which do not tend to form aggregations, the removal of large numbers by microstraining cannot be explained.

Large volumes of cells do not necessarily cause an increase in filtering efficiency (see Fig. 1). In the spring and summer of 1964 large numbers of unicellular chlorophyceans and cryptophyceans were present in the incoming water but the percentage removal was low. The microstrainers worked most efficiently in the spring and autumn, when a variable mixture of species was present, and in the summer if larger filamentous and colonial forms were present.

Rotary microstrainers, using meshes of various sizes, are in use all over the world (Lynch et al[12], Evans[13]) and can be an extremely efficient method of increasing the volume of water obtained during the run of a slow sand filter. There are of course limitations, for example when the source contains large amounts of very small particles, but this applies to many methods of first stage filtration, including rapid gravity sand filters, unless coagulation processes are included. Despite these limitations the rotary microstrainer has often proved to be a valuable contribution to waterworks economy.

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Addendum.

To the captions of figures 20,21,22 and 26 add; Vertical lines indicate the times of filter bed cleaning and the vertical stippling indicates the times of filter bed resanding.

To the captions of figures 45a and 45b add; The dates of the sample collections are given on the left hand side.

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