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Microheterotrophic communities associated with the degradation of kelp debris

E.A.S. Linley and R.C. Newell

Institute for Marine Environmental Research, Plymouth, Plymouth, U.K.

Abstract

The micro-organisms which colonise kelp debris incubated in seawater show a clear succession. The media are first colonised by bacterial cocci and rods which are subsequently replaced by grazing flagellates and ciliates, as well as amoebae and choanoflagellates in the later stages of the microheterotrophic succession. The biomass of grazing protozoa is generally 10 % of that of the bacteria and estimates of consumption of bacteria by microflagellates of 10 μm^3 body volume suggest that a mean value of 39 x the body weight may be consumed per day. It is suggested that filter- and deposit-feeding organisms utilising degrading plant material are likely to use bacteria rather than protozoa as a principal food resource. Estimates of the rate of turnover of sediments and of the water column near to kelp beds support the belief that for optimal utilisation by consumer organisms detritus is recycled at a rate which is slow enough to promote the development of a bacterial population but which is too fast for protzoa to reduce the bacterial population by grazing.

Introduction

Although there have now been many studies on the role of micro-organisms in the degradation of particulate debris in the sea, especially that derived from sea grasses (FENCHEL 1970; MEYERS and HOPPER 1973; FENCHEL and JØRGENSEN 1976; for review, see NEWELL 1979), the quantitative significance of microbial decomposition is often difficult to assess because some studies have used plate techniques whilst more recent workers have used direct counting methods. Very few have estimated both numbers and cell dimensions to obtain the biomass of micro-organisms associated with the decomposition of plant debris in the marine environment. But, as has been shown in the previous paper by NEWELL and LUCAS (1981), data for the biomass of microheterotrophs associated with the decomposition of known amounts of dissolved and particulate matter is essential for a calculation of the energetics of conversion of debris through the first step of the decomposer food chain into bacteria. It is also important to know what the subsequent energetic losses are, when bacteria themselves become incorporated into higher trophic levels.

Detailed studies of the rate of decomposition of dissolved and particulate components of kelp debris (NEWELL et al. 1980; LUCAS et al. 1981) have therefore been coupled with simultaneous measurements of the numbers and biomass of micro-organisms through at least two steps in the decomposer food chain (LINLEY et al. 1981; STUART et al. 1981). The results show that there is a clear succession in the micro-organisms associated with the decomposition of plant debris, and lead to some general principles

governing the rate of cycling of materials in the sediments and water column which is necessary to optimise utilisation of degraded plant material by deposit – and suspension-feeding consumer organisms.

Material and methods

The material and methods used to quantify the numbers and biomass of micro-organisms colonising dissolved and particulate components of kelp debris have been described in some detail by LINLEY et al. (1981) and STUART et al. (1981). Briefly, known concentrations of dried particulate debris from the kelp *Laminaria pallida*, or dried mucilage from *Ecklonia maxima* and *Laminaria pallida* collected from a kelp bed on the west coast of the Cape Peninsula, South Africa, were incubated in vessels containing freshly-collected 62 μm filtered seawater from the kelp bed and held at the local seawater temperature of 10°C. Control vessels containing sterilised seawater plus kelp debris, and non-sterilised filtered seawater alone were used in all experiments and the media were aerated by a pump which drew 0.2 μm filtered air through the vessels in series.

The number of micro-organisms was assessed by acridine orange direct counting (HOBBIE et al. 1977) whilst scanning electron microscopy based on a combination of methods described by TODD and KERR (1972), PAERL (1975) and BOWDEN (1977) was used to measure cell dimensions and thus to calculate microbial biomass.

Results

A. Microbial communities associated with the degradation of kelp debris

A definite seasonality in the species composition and abundance of morphological types has been described in the microbial community of kelp bed seawater (MAZURE 1978). In the winter, bacterial numbers are low and are dominated by cocci whereas in the summer bacterial numbers are higher and the species composition is different to that in the winter. The types of micro-organisms in the initial inoculum may thus largely determine the nature of the assemblage which develops following the addition of kelp debris under laboratory conditions. Experiments were therefore carried out with seawater collected in the winter (August 1979) and summer (February 1980) from the study site on the west coast of the Cape Peninsula, South Africa.

The seawater control samples contained mainly a mixed assemblage of small bacterial rods and cocci whose numbers reached approximately 9×10^6 cells ml^{-1} after 7 days. At the same time, a mixed population of mainly phototrophic flagellates similar to *Micromonas* and *Mallomonas* increased up to approximately 4×10^4 cells ml^{-1} together with a population of diatoms including *Nitzschia* spp, *Thalassiosira*, *Asterionella* and *Skeletonema* (Table 1). The effects of addition of 7.2 g l^{-1} dried mucilage from *Laminaria pallida* to unsterilised seawater are also shown in Table 1, from which it is evident that both the numbers and variety of organisms are affected by the presence of kelp mucilage. Bacterial numbers were almost 100 x higher than in control samples and reached their maximum abundance at day 7 before declining. Further, there is some evidence that cocci reach their maximum abundance and decline before the rods become abundant. Flagellates increased toward the end of the experimental period and were represented by phagotrophic forms similar to *Bodo* and *Rynchomonas* (SIEBURTH 1979). Finally, diatoms failed to appear and occasional ciliates were observed towards the end of the experimental period. In the case of particulate debris incubated in seawater, Cyrtophorine ciliates, choanoflagellates (such as *Stephanoeca* sp.) and amoebae became common after up to 30 days

incubation at 10°C (STUART et al. 1981). The elements of microbial succession which occur in winter samples also occur in the summer incubation experiments but the bacterial population is dominated by large rods which are then replaced by a mixed population of flagellates (mainly organisms similar to *Monas*, *Oikomonas*, *Bodo* and *Rynchomonas)* and ciliates (including types similar to *Uronema*, *Cyclidium* and *Euplotes)* which emerged as major organisms by day 14.

Table 1. *Laminaria pallida*

Day	Seawater Control			Seawater + Mucilage				
	Bacteria	Flagellates	Diatoms	Bacteria			Flagellates	Diatoms
				Rods	Cocci	Total		
	$x10^6$	$x10^4$		$x10^6$	$x10^6$	$x10^6$	$x10^4$	
1	0.64	0.28	—	1.30	0.61	1.91	—	—
2	1.50	0.69	—	2.24	3.09	5.33	—	—
3	2.57	1.75	—	132.5	108.4	240.9	—	—
4	6.53	1.98	—	26.78	508.8	535.6	—	—
5	3.51	2.08	—	322.9	411.0	733.9	—	—
6	1.57	4.10	663	520.1	84.7	604.8	—	—
7	9.13	3.76	897	606.9	107.1	714.0	—	—
8	0.77	2.93	6660	516.6	181.5	698.1	30.3	—
9	0.60	—	7747	17.11	29.14	46.25	144.0	—
10	6.70	—	13730	15.96	33.93	49.89	149.0	—
11	8.76	—	17588	10.72	26.23	36.95	99.5	—

The results suggest, therefore, that the addition of kelp debris results in the inhibition of some components of the mixed assemblage of micro-organisms in natural seawater, and at the same time provides a substrate for a large population of bacterial cocci and rods. These, rather than the organic material from kelp debris, may then provide an energy resource for other heterotrophs including the flagellates and ciliates which appear at a later stage in the incubation experiments (see also HARRISON and MANN 1975a, b; MORITA 1977). A selection of some of the typical components of the microbial community which becomes established on kelp debris is shown in Figure 1.

B. Conversion of organic matter through the decomposer food chain based on kelp

Although the numbers of micro-organisms which develop in the incubation media show a clear succession, with maximal bacterial numbers after 7–9 days followed by an increase in the flagellate and ciliate populations, the data give little indication of the energy transfer through the food chain unless the biomass of micro-organisms is known. These can be calculated from the cell numbers, dimensions and the specific gravity of the component cells (see LINLEY et al. 1981).

Figure 2 shows the biomass of bacteria and of grazing flagellates and ciliates which developed over a period of 28 days in incubation media at 10°C to which 7.2 g l^{-1} dried mucilage from *Laminaria pallida* (= 2.2 g l^{-1} organic matter) had been added. It is obvious that the conversion of organic matter into microbial biomass can be calculated

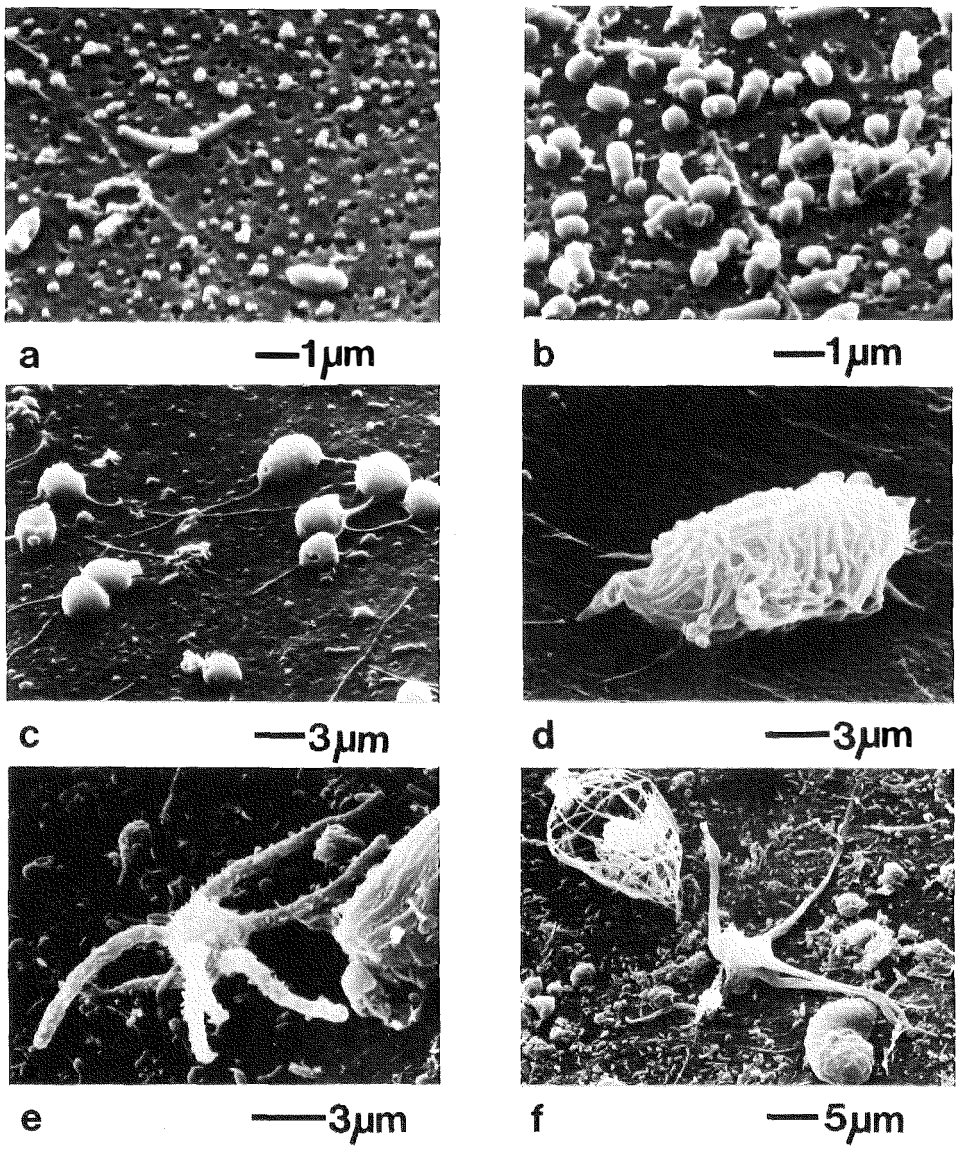

Figure 1

A selection of micro-organisms which develop on kelp debris. a. Population of cocci which dominate the *Laminaria pallida* mucilage media in winter incubations b. Rod and cocci on *Ecklonia maxima* mucilage. c. Phagotrophic flagellates. d. Ciliate with attached cocci. e. An unidentified floating amoeboid form f. A Choanoflagellate (possibly *Stephanoeca* sp.) together with a testacean and an unidentified amoeboid form.

if we know the simultaneous utilisation of organic substrates. As has been shown in the previous paper (NEWELL and LUCAS 1981; also LUCAS et al. 1981, STUART et al. 1981) the annual conversion of organic carbon from kelp via both dissolved and particulate

pathways into bacterial biomass amounts to at least 14 % rising to as much as 22 % of the total organic carbon production by the kelp bed during the summer months. But the data can also be used to calculate the conversion through the second step in the decomposer food chain from bacteria to protozoa.

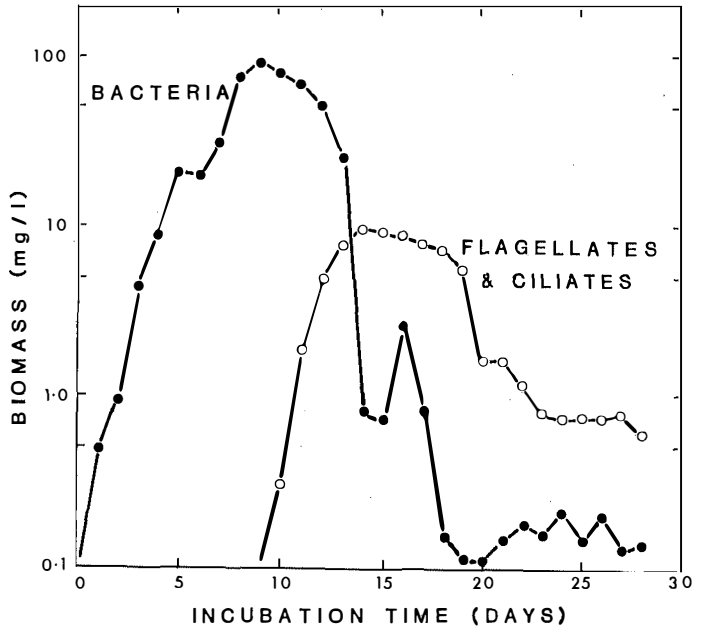

Figure 2

Laminaria pallida. The biomass of micro-organisms (mg l^{-1}) developing over a period of 28 days in incubation media at 10 °C to which 7.2. g l^{-1} of dried mucilage from *Laminaria pallida* (= 2.185 g l^{-1} organic matter) had been added.

It is clear from Figure 2 that there is an approximately 90 % loss in the biomass in the step from bacteria to protozoa, and in fact the biomass of grazing flagellates and ciliates in all our culture experiments was always about 10 % of the bacterial biomass (Figure 3). So from this, and the figure cited above for conversion into bacterial biomass, it can be anticipated that only some 1.4 – 2.2 % of the total kelp production would be converted to protozoa via the bacteria, the remaining 97.8 – 98.6 % being oxidised and returned principally as CO_2 to the environment.

In fact, the rate of consumption of bacteria by grazing protozoa is very high indeed compared with larger consumer organisms. BURKILL (1978) found that the daily consumption of bacteria by the ciliate *Uronema marinum* is approximately 6 x the body weight per day, estimated for a large ciliate of 1000 μm^3. The estimated daily consumption of bacteria by the microflagellates which colonised kelp incubation

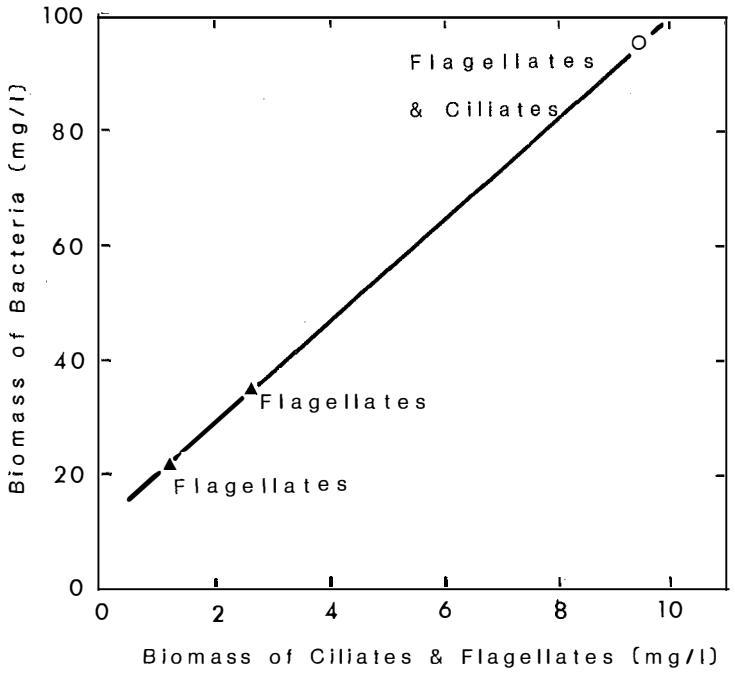

Figure 3

Ecklonia maxima and *Laminaria pallida*. The relationship between maximal biomass attained by bacteria and the biomass of flagellates and ciliates in cultures containing approximately 2 g l^{-1} organic matter from kelp mucilage in seawater at 10°C. Equation of regression $Y = 8.96 X + 10.91$; $r = 0.9998$. (Data from LINLEY et al. 1981.)

media is summarised in Table 2. These organisms had a mean volume of only 10 μm^3 and the results suggest that the mean consumption of bacteria is some 39 x the body weight per day. This is similar to the value of 35 x which would be anticipated on the basis of the well-known allometric relationship between metabolism and body size of $W^{0.75}$. Processing of organic materials through the microheterotrophic food chain may

Table 2. *Ecklonia maxima* The relationship between biomass of flagellates (mg dry mass l^{-1}) and consumption of bacteria (mg day^{-1}) in a culture containing seawater plus 6.4 g l^{-1} dried mucilage from *E. maxima*. (See LINLEY et al. 1981).

Biomass of flagellates (mg l^{-1})	Consumption of bacteria mg day^{-1})
0	0.4
0.215	7.6
0.384	28.0
0.714	28.0
1.143	12.4

also be of considerable significance in the direct consumption of phytoplankton. Recent studies suggest, for example, that total consumption of organic matter from phytoplankton production by marine ciliates may be comparable with that of copepods at some seasons (CAPRIULO and CARPENTER 1980). Turnover of organic matter through the marine microzooplankton thus appears to follow the well-known size-dependence of feeding and metabolism which has been established for higher organisms, and it is clear that much of the organic material from primary production which has been incorporated into bacteria will be oxidised following the establishment of protozoa on decomposing plant detritus.

C. Utilisation of bacteria or protozoa by consumer organisms?

Because of the large loss which occurs at each step in the food chain, it is axiomatic that more energy can be obtained by filter- and deposit-feeding organisms if they utilise the primary decomposers (bacteria) rather than flagellates or even larger ciliates. Within the limits set by their feeding structures, therefore, we would expect to see the large populations of oysters, mussels, sponges, ascidians and other filter-feeders, as well as deposit feeders near to the sites of 'excess' primary production such as macrophyte beds and saltmarshes, utilising bacteria rather than protozoa as a food resource. Utilisation of bacteria by larger consumer organisms allows a conservation of approximately 90% of the energy which would otherwise have been lost in the step from bacteria to protozoa.

Direct experimental evidence for the ingestion and absorption of bacteria by consumer organisms is scarce, although there are numerous reports in the literature of the ability of marine organisms to survive and grow on a diet of bacteria (for reviews, see MORITA 1977; NEWELL 1979). FENCHEL (1972) has, however, shown that bacteria are absorbed during passage of detritus through the gut of the bivalve *Macoma balthica* (Figure 4).

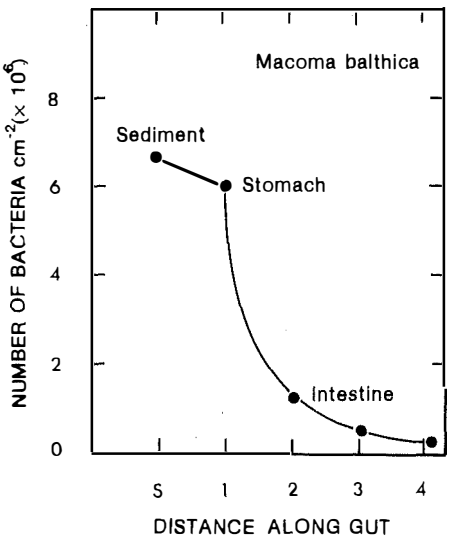

Figure 4

Graph showing the numbers of bacteria cm^{-2} in sediment prior to ingestion (S), in the stomach (1) and in the anterior (2) mid- (3) and hind gut (4) of *Macoma balthica*. (NEWELL 1979; Based on FENCHEL 1972.)

Indirect evidence using carbon and nitrogen analyses of ingested and egested material from several invertebrates suggests that this also occurs in many other deposit-feeders (NEWELL 1965; 1979; LONGBOTTOM 1970; for review, see MORITA 1977). More recently, SOROKIN (1973) has estimated the percentage of bacterial cells assimilated to food consumed (expressed as carbon) for a variety of invertebrates. Values as high as 73–76 % have been obtained in some corals, approximately 82 % in the sponge *Toxadocea violacea*, in the tunicate *Ascidia nigra* and in the holothurian *Ophiodesma spectabilis and 68* % in the bivalve *Crassostrea gigas*, but there is an urgent requirement for further direct studies on the utilisation of micro-heterotrophs as a food resource for consumer organisms.

Discussion

The general proposal that particulate debris may pass through the gut of consumer organisms whilst bacteria are removed (NEWELL 1965; DARNELL 1967a, b) suggests that an optimal turnover time for utilisation of bacteria would be one which was long enough to allow a large population of bacteria to colonise the material, but which was too short to allow protozoa to graze the bacteria. Bacteria reached their maximal abundance in the culture media at 7–10 days at 10 °C whereas protozoa first appeared at 10 days and reached their maximal abundance at 15–17 days (see Figure 2). For optimal utilisation of bacteria therefore, a particle or water column turnover time of less than 10 days at 10 °C would be anticipated, and this may be less than 5 days at 20 °C if bacterial growth follows the usual temperature-dependence for such processes.

Interestingly, MYERS (1977) in a study which was unrelated to the utilisation of bacteria, has found that the surface of marine sediments in July at Charlestown Pond, Rhode Island, was extensively reworked by deposit-feeding animals and that the turnover time at 0–1 cm depth was 0.7–4.0 days whereas at 1–2 cm depth it was 2.4–11.8 days (Figure 5). In much the same way, data for the biomass and filtration rate of the principal components of the kelp bed communities can be used to estimate the 'turnover time' of the water column near to the sites of primary production by kelp. Dr. C.L. GRIFFITHS at the University of Cape Town (personal communication) has estimated that the population of the mussel *Aulacomya ater*, which is the dominant filter-feeder in the kelp bed at Oudekraal, on the west coast of the Cape Peninsula, may reach as much as 1098 individuals m^{-2} and the population as a whole could filter up to 1300 l h^{-1} m^{-2}. The mussels alone could thus filter a 10 m water column in 7.5 h although a rather longer turnover time is likely to be characteristic of the kelp bed as a whole. It is thus clear, that both the rate of reworking within the top 2 cm of sediments by deposit-feeding animals and of the water column by suspension-feeding animals near to the site of production from salt marshes and kelp beds is strikingly close to what we would predict for a bacteria-driven system (see also NEWELL 1981) although it is evident that other sources of organic material, including phytoplankton, may form a component of their diet.

Since the average production of a kelp bed is approximately 1172 g carbon m^{-2} y^{-1} (see NEWELL et al. 1980; FIELD et al 1980) and an annual conversion efficiency of up to 14 % from carbon into bacterial biomass, rising to as much as 22 % during the summer months may be achieved, the dry bacterial biomass supported by a kelp bed of 700 hectares will be approximately 115 x 10^4 kg y^{-1} (see also NEWELL and LUCAS 1981). It appears from the data of SOROKIN (1973) that this material may be absorbed with a high efficiency of approximately 80 % by principal consumer organisms. Transformation of kelp production via bacteria into consumer organisms could thus yield as much

as 92 x 10^4 kg dry mass y^{-1} available for secondary production by the dense communities of filter- and deposit-feeding organisms which characterise the kelp bed community.

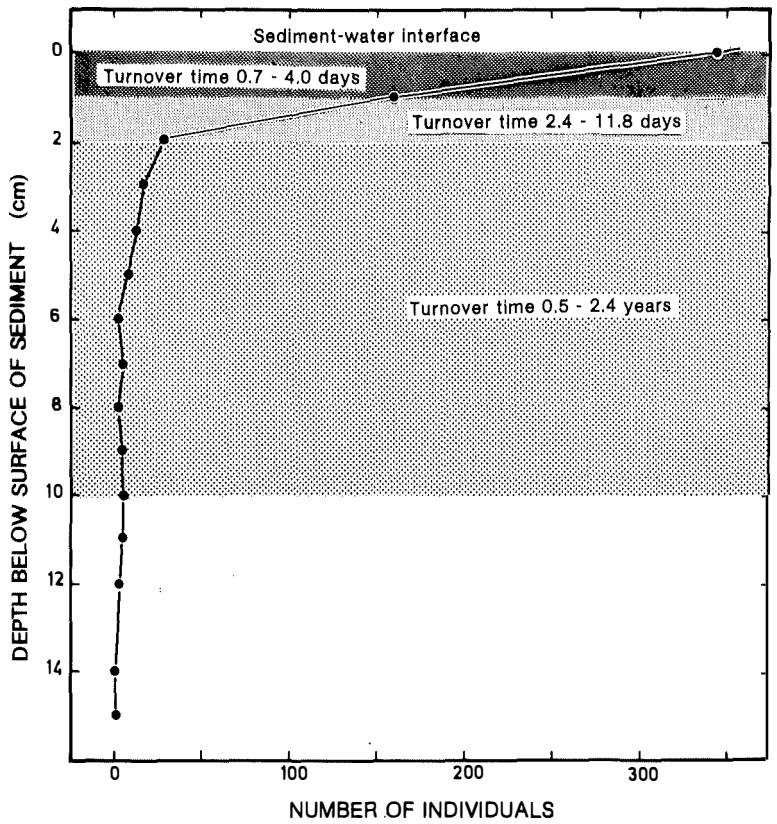

Figure 5

Graph showing the total numbers of individuals recorded in a series of 25 cores, each 15 cm long by 5 cm in diameter, taken in July 1970 at Charlestown Pond, Rhode Island. Sediment turnover times are also shown. (NEWELL 1979; Data from MYERS 1977.)

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