COMMONWEALTH OF AUSTRALIA

COMMONWEALTH SCIENTIFIC AND INDUSTRIAL RESEARCH ORGANIZATION

Division of Fisheries and Oceanography

REPORT 42

21st MARINE SCIENCE SCHOOL

MAY 23 - JUNE 2, 1967

Marine Laboratory
Cronulla, Sydney
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INTRODUCTION

The 21st Marine Science School for university students was held at the CSIRO Marine Laboratory, Cronulla, N.S.W., from May 23 to June 2, 1967.

The papers in this report present the results of work done by students under the guidance of research staff from this Laboratory, the Australian Atomic Energy Commission's Lucas Heights Laboratory, and the Universities of New South Wales and Sydney.

The first report of this type was published after the 1966 Marine Science School. However, there is an important difference between that report and this one: the first report was written mainly by the supervising research staff, this report was for the most part prepared by the students. One of the main reasons for extending the School in 1967 was to give the students time to write up their results.

The names of students who attended, their universities, the groups they worked in, and the research staff who guided them are given in the Appendix of this report.

HYDROLOGY

PART 1. THE HYDROLOGICAL REGIME

I. INTRODUCTION

The objects of this programme were:-

To determine the winter chlorinity and temperature distribution along the whole length of Port Hacking and then to contrast this with the chlorinity-temperature pattern in one of the bays, Gunnamatta Bay.

To detect, by oxygen determinations, any stagnation in Gunnamatta Bay.

To estimate, in terms of carbon and phosphorus, the standing stock of organic matter. Dissolved nitrate and orthophosphate were also determined, partly to detect effluent contamination, but also as an indication of potential fertility.

II. METHODS

Chlorinity and temperature were measured in situ with a chlorinity-temperature meter (Hamon 1956). Values were corrected against International Standard Seawater.

Suspended organic matter was collected in a high-speed centrifuge (Davis 1957) transferred with washings to a 15 ml glass centrifuge tube, and spun at 2,000 g for 10 min. The supernatant was discarded and the tube dried at 105°C. Suspended organic carbon content of the dry residue was determined by the chromic acid oxidation method of El Wakeel and Riley (1957).

Suspended phosphorus was determined on a duplicate sample treated as above but subjecting the dry residue to persulphate digestion as in the method of Menzel and Corwin (1965).

Dissolved orthophosphate was estimated by the method of Murphy and Riley (1962). Dissolved nitrate was determined as nitrite after reduction in a cadmium-copper column (Wood, Armstrong, and Richards 1967). Dissolved oxygen was determined by the Winkler method (Jacobsen, Robinson, and Thompson 1950).

III. RESULTS

All data are given in Tables 1 and 2 at the end of this section. Station positions are shown in Figures 1 and 2.

A section of chlorinity distribution along the length of Port Hacking is shown in Figure 3. The same basic pattern was observed this year as in 1966, viz. a balance of saline (chlorinity > 19.65‰)

oceanic water at the mouth against brackish (chlorinity < 19.00 ‰) water in the two miles of narrow river below Audley Weir. The wider area of the estuary contains a fairly uniform water body with a tendency to slightly higher chlorinities in the two deep basins (> 19.50 ‰). The uniform estuarine water body was more saline by about 0.2 ‰ than in 1966, but temperatures for the four water types were all within the range found last year (16.0-16.4°C).

The distribution of chlorinity within Gunnamatta Bay is shown in Figure 4. This is a composite diagram of the situation found repeatedly each day at about high-water. The penetration of saline oceanic water directly into the bay is limited to Station M, but some saline water flows east from the channel along the edge of the sand bank and is reinforced when outside water flows over the bank at high water. Within two hours of the ebb commencing, however, all trace of oceanic water had been swept out of the bay; except on May 31 when strong S.E. winds delayed the ebb flow.

The water within the bay is extremely uniform apart from a few localised drainage effects and the brackish layer along the bottom. This uniformity is thought to be due to nocturnal cooling and mixing. The mixing does not extend to the brackish layer, but the latter consists of a zone, about one metre thick, of suspended black flocculent mud. Presumably the density of this layer is so enhanced by this particulate load, that a positive stability exists. Observation by a diver showed that the low chlorinities encountered at the bottom of the bay were all found within the floc suspension.

Oxygen values were at saturation level at both the top and bottom of the bay, even in the brackish layer, except for a bottom value of 4.8 ml/l at Station B and a surface value of 4.9 ml/l at Station SB. These low values might have been the result of experimental error or of the inclusion of aberrant bacterial contamination as described in Part 2.

Amounts of suspended carbon and phosphorus were higher in Gunnamatta Bay than outside it and tended to higher values in the morning (Table 2). The latter effect could be explained by nocturnal mixing followed by diurnal sedimentation. Carbon to phosphorus ratios were variable, but tended towards 100:1, so that the suspended matter was probably planktonic and alive.

Dissolved inorganic phosphate and nitrate values in the Bay were in general low and similar to those outside. The low nitrogen to phosphorus ratios indicate a relative deficiency of nitrogen.

High oxygen and low nitrate values throughout the Bay argue against any serious pollution, even though the Bay has so little exchange with the outside ocean.

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TABLE 1

TEMPERATURE AND CHLORINITY AT STATIONS ALONG PORT HACKING AND IN GUNNAMATTA BAY

Date	Time	Tide State	Station	Depth (m)	Temperature (°C)	Chlorinity (‰)
25.v.67	1000h	High water 0930h	B	0	16.0	19.50
				1	16.0	19.50
				2	16.0	19.51
				3	16.0	19.53
				4	16.0	19.52
				5	16.0	19.51
				7	15.9	19.51
				9	15.8	19.51
				11	15.7	19.50
				13	15.7	17.98
	1030h		M	0	16.0	19.54
				2	16.0	19.54
				4	15.9	19.53
				6	15.9	19.53
				8	15.8	19.53
				10	15.7	19.53
				12	15.7	16.96
	1100h		C	0	16.0	19.54
				3	16.0	19.38
	1200h		O	0	18.6	19.75
				5	18.0	19.68
26.v.67	1000h	High water 1018h	J	0	16.9	19.59
				2	16.2	19.63
				4	16.1	19.56
				6	16.0	19.56
				8	16.0	19.43
				9	16.0	19.34
	1030h		M	0	16.6	19.59
				2	16.1	19.58
				4	16.1	19.55

Table 1 (Cont.)

Date	Time	Tide State	Station	Depth (m)	Temperature (°C)	Chlorinity (‰)
				6	16.0	19.53
				8	15.9	19.53
				10	15.8	19.53
				12	15.8	19.53
				12.5	15.9	14.43
26.v.67	1100h	High water 1018h	B	0	16.3	19.49
				2	16.2	19.52
				4	16.1	19.53
				6	16.0	19.53
				8	16.0	19.52
				10	16.0	19.53
				12	15.9	19.51
				13	15.8	18.43
	1130h		SB	0	16.0	19.54
				2	16.0	19.53
				4	15.9	19.53
				6	15.8	19.51
				8	15.8	19.51
				10	15.7	19.50
				12	15.8	16.75
	1200h		D1	0	16.3	17.58
				0.5	16.5	17.52
			D2	0	16.0	19.37
				0.5	16.0	19.35
			D3	0	16.6	19.43
				1.0	16.6	19.44
29.v.67	0930h	High water 1300h	1	0	17.4	19.69
				2	17.2	19.70
				4	16.7	19.66
	1000h		2	0	16.5	19.63
				2	16.5	19.65
				4	16.3	7.23

Table 1 (Cont.)

Date	Time	Tide State	Station	Depth (m)	Temperature (°C)	Chlorinity (‰)
	1030h		3	0	16.5	19.64
				2	16.5	19.64
				4	16.5	19.64
				6	16.4	19.63
				8	16.5	19.23
				9	16.5	16.63
	1100h		4	0	16.6	19.63
				2	16.6	19.67
				4	16.6	19.33
				6	16.6	15.43
29.v.67	1600h	High water 1300h	5	0	16.4	19.48
				2	16.4	19.48
				4	16.4	19.48
				6	16.4	19.48
				8	16.4	19.48
				10	16.4	19.48
				12	16.3	19.49
				14	16.3	19.50
				16	16.3	19.51
				18	16.3	19.51
				20	16.3	19.54
				22	16.4	19.57
				23	16.5	19.53
	1230h		6	0	16.4	19.49
				2	16.4	19.50
				4	16.4	19.52
				5.5	16.4	19.56
	1530h		7	0	16.4	19.46
				2	16.4	19.46
				4	16.4	19.46
				6	16.6	19.50
				8	16.6	19.50
				10	16.5	19.50
				12	16.4	19.50
				14	16.4	19.50
				16	16.4	19.50
				18	16.5	19.50
				18.5	16.7	16.40

Table 1 (Cont.)

Date	Time	Tide State	Station	Depth (m)	Temperature (°C)	Chlorinity (‰)
	1500h		8	0	15.8	18.25
				2	15.9	18.98
	1430h		9	0	16.9	18.24
				2	16.6	18.21
				4	16.6	18.21
	1130h		10	0	16.2	19.54
				2	16.2	19.54
				4	16.2	19.54
				6	16.2	19.54
				8	16.2	19.54
				10	16.2	19.54
29.v.67	1145h	High water 1300h	11	0	16.0	19.49
				2	15.9	19.49
				4	15.9	19.51
				6	15.9	19.50
				8	15.8	19.51
				10	15.7	19.50
				12	16.2	19.38
	1210h		12	0	16.3	19.58
				2	16.3	19.58
				4	16.3	19.58
				6	16.3	19.58
				8	16.3	19.55
				10	16.3	19.57
				11	16.4	19.21
30.v.67	1000h	High water 1405h	M	0	15.5	19.54
				2	15.5	19.57
				4	15.5	19.57
				6	15.5	19.57
				8	15.5	19.58
				10	15.5	19.58
				11	15.6	19.57
	1030h		B	0	15.6	19.57
				2	15.6	19.58
				4	15.4	19.58

Table 1 (Cont.)

Date	Time	Tide State	Station	Depth (m)	Temperature (°C)	Chlorinity (‰)
				6	15.4	19.57
				8	15.3	19.58
				10	15.2	19.58
				12	15.2	19.58
	1100h		J	0	15.7	19.56
				2	15.7	19.56
				4	15.6	19.58
				4.5	15.6	19.57
	1130h		F	0	15.6	19.58
				2	15.6	19.58
				4	15.5	19.58
				6	15.6	19.58
				8	15.4	19.61
				8.5	15.4	19.54
30.v.67	1530h	High water 1405h	M	0	16.0	19.50
				2	16.0	19.49
				4	15.9	19.49
				6	15.7	19.49
				8	15.7	19.49
				10	15.6	19.47
				11	15.4	19.48
	1630h		M	0	15.9	19.61
				2	15.9	19.51
				4	15.7	19.48
				6	15.7	19.49
				8	15.4	19.50
				10	15.4	19.50
				11	15.5	17.47
	1645h		J	0	15.8	19.49
				2	15.8	19.53
				4	15.8	19.50
				6	15.8	19.51
				7.5	15.8	19.53

Table 1 (Cont.)

Date	Time	Tide State	Station	Depth (m)	Temperature (°C)	Chlorinity (‰)
	1700h		F	0	15.8	19.53
				2	15.8	19.50
				4	15.8	19.54
				6	15.8	19.54
				8	15.8	19.32
31.v.67	1450h	High water 1500h	F	0	17.8	19.69
				2	15.5	19.59
				4	15.4	19.60
				6	15.3	19.61
				8	15.3	19.64
	1500h		J	0	15.6	19.61
				2	15.6	19.65
				4	15.5	19.63
				6	15.4	19.63
				8	15.3	19.61
	1600h		F	0	15.6	19.66
				2	15.5	19.60
				4	15.4	19.65
				6	15.3	19.65
				8	15.3	19.65
	1610h		J	0	15.6	19.63
				2	15.6	19.65
				4	15.5	19.65
				6	15.5	19.65
				8	15.4	19.65
	1640h		J	0	15.5	19.63
				2	15.5	19.63
				4	15.4	19.64
				6	15.4	19.64
				8	15.4	19.65
	1700h		F	0	15.5	19.65
				2	15.5	19.65
				4	15.4	19.65
				6	15.4	19.65
				8	15.4	19.65

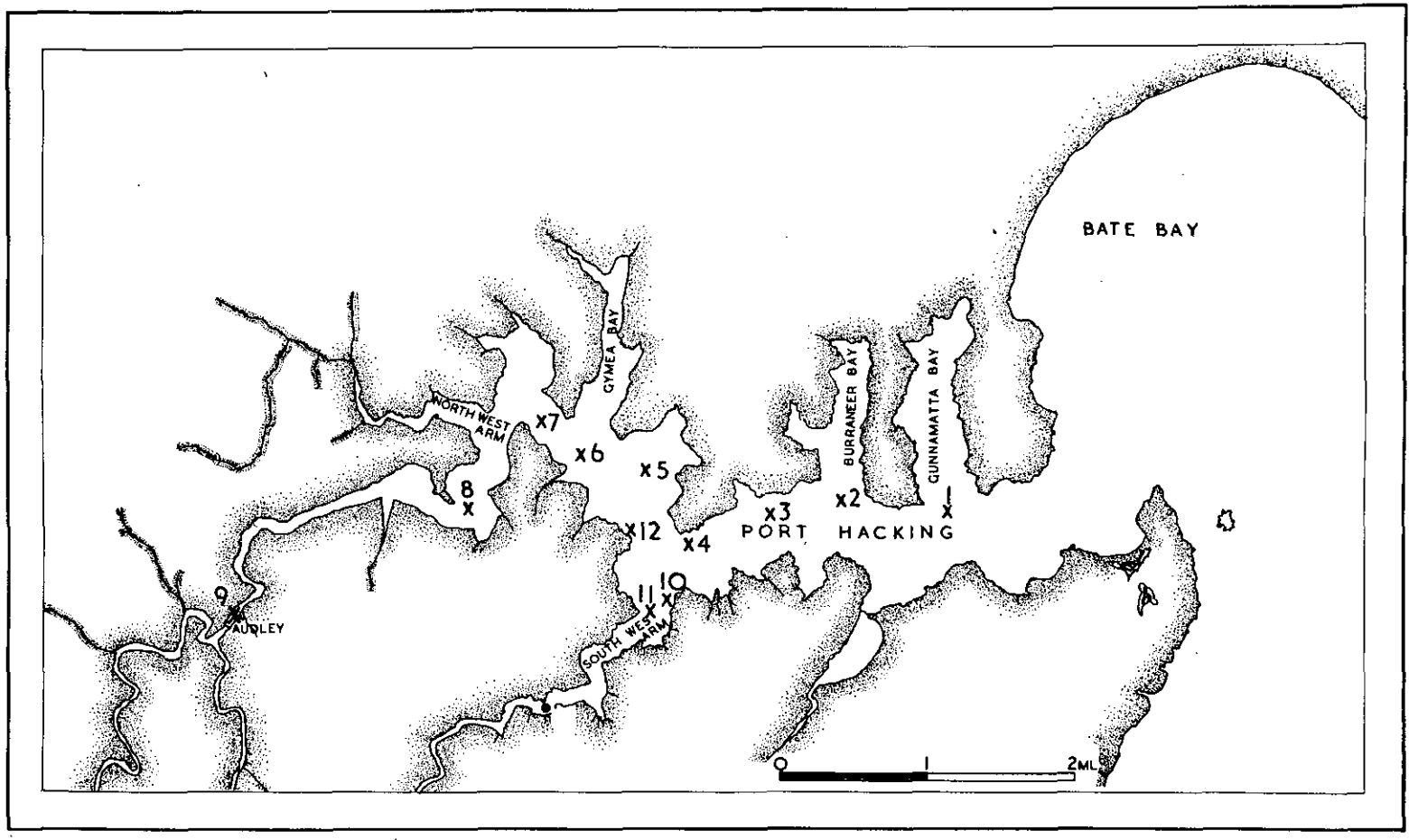


Fig. 1 - Temperature-chlorinity stations along Port Hacking.

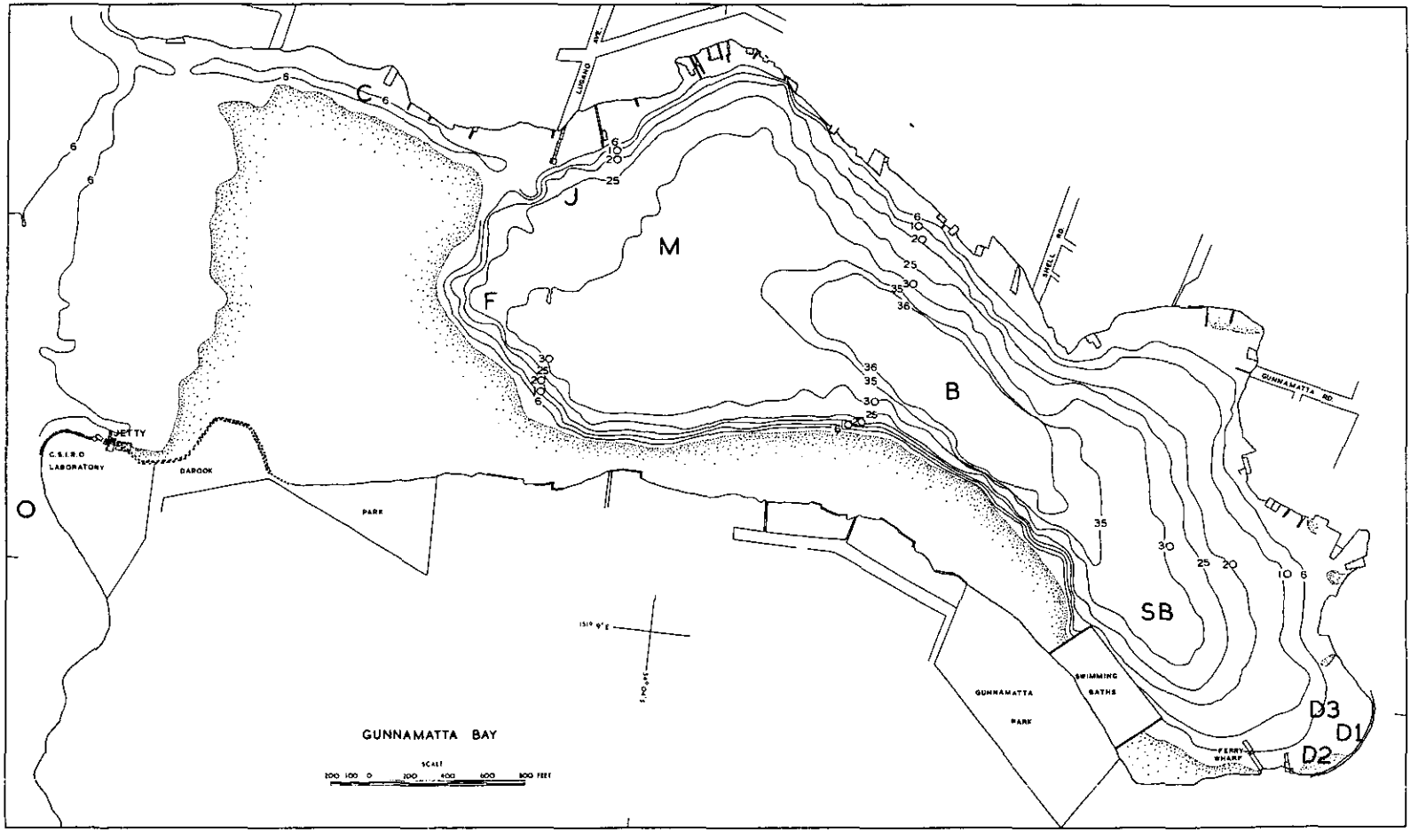


Fig. 2 - Sampling stations in Gunnamatta Bay. Depth contours in feet.

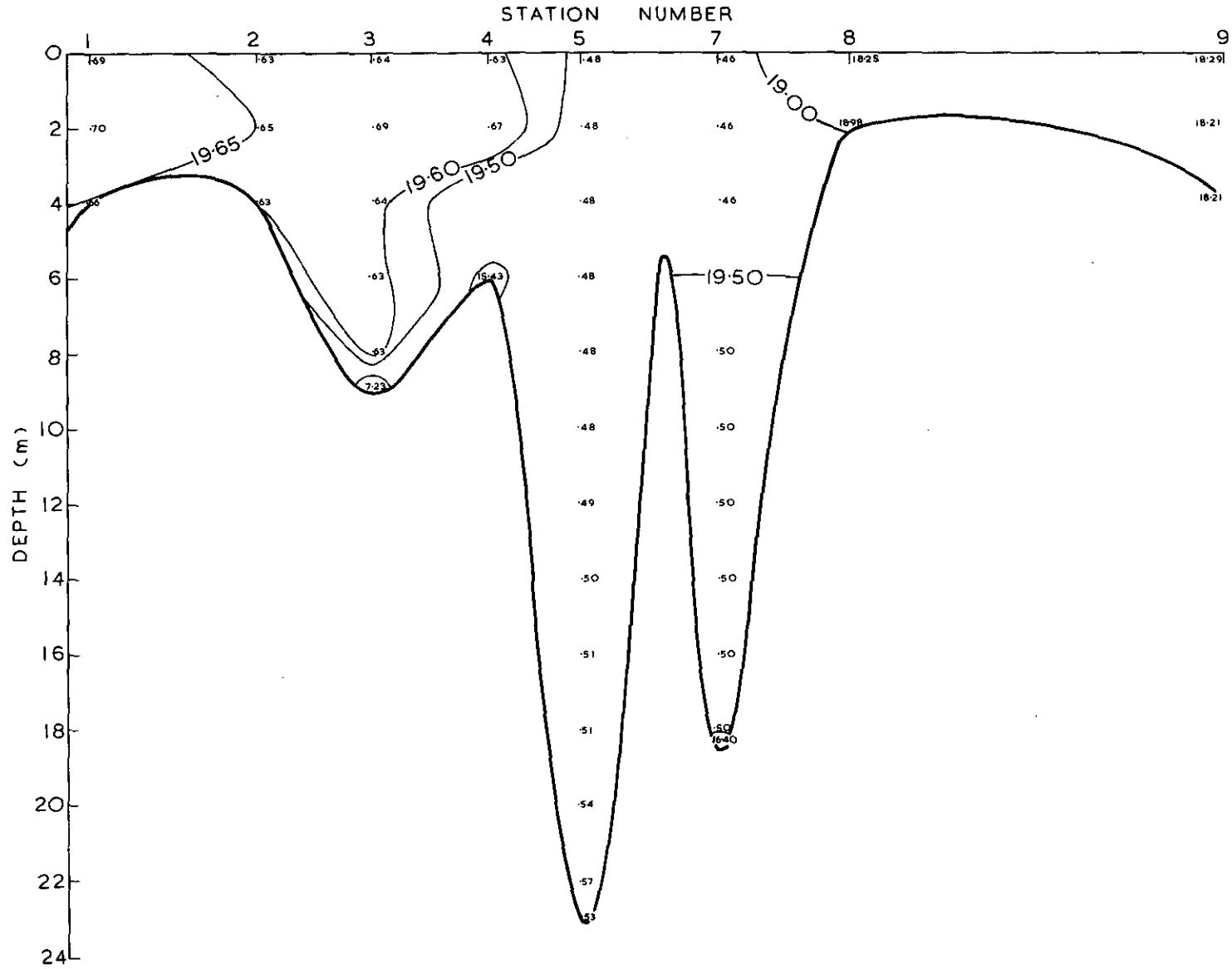


Fig. 3 - Chlorinity (‰) distribution along Port Hacking.

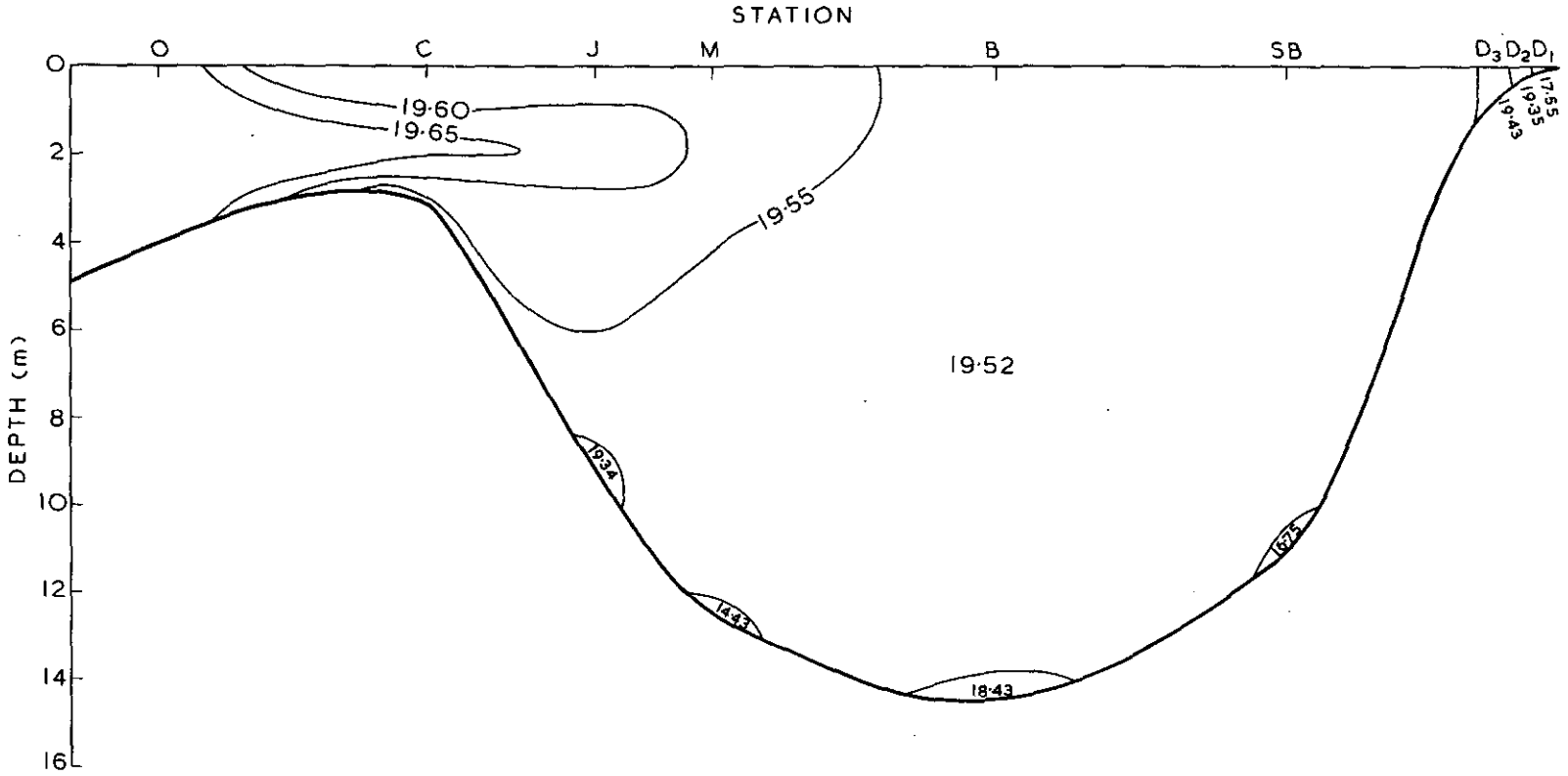


Fig. 4 - Chlorinity (‰) distribution along Gunnamatta Bay.

SUSPENDED CARBON AND PHOSPHORUS, DISSOLVED PHOSPHORUS AND NITRATE, AND DISSOLVED OXYGEN IN GUNNAMATTA BAY

Date & Tide Start	Time	Station	Depth (m)	Suspended Carbon (µg/l)	Suspended Phosphorus (µg/l)	C/P	Dissolved Phosphorus (µg/l)	Dissolved Nitrate (µg/l)	N/P	Dissolved Oxygen (ml/l)
25.v.67 High water 0930h	1000h	B	0	800	4.2	190	18.7	9.0	0.5	5.3
			12							5.3
	1100h	C	0	400	4.6	87	7.5	4.0	0.5	5.3
			3							5.3
	1200h	O	0	200	3.1	64	13.7	6.0	0.4	5.3
			5							5.3
26.v.67 High water 1018h	1000h	J	0				2.0	8.0	4.0	5.4
			9				2.0	7.0	3.5	5.6
	1030h	M	0				< 1.0	2.0		5.5
			12½				2.0	10.0	5.0	5.4
	1100h	B	0	672	6.1	110	< 1.0	2.0		5.4
			13	535	5.4	100	4.0	5.0	1.2	4.9
	1130h	SB	0				< 1.0	3.0		4.9
			11				4.0	5.0	1.2	5.5
29.v.67 High water 1300h	1000h	B	0	389	3.1	125				
			12	746	7.4	100				
30.v.67 High water 1405h	1000h	M	0	337	1.8	187	6.0	5.0	0.8	
			0	224	1.7	132	6.0	5.0	0.8	
	1530h	M	0	162	1.8	90	6.0	5.0	0.8	
			0	327	2.9	112				
	1620h	M	0	162	2.8	58	3.0	5.0	1.6	
			0	146	3.0	49				

HYDROLOGY

PART 2. PARTITION OF ^{32}P BETWEEN DISSOLVED AND PARTICULATE MATTER

I. INTRODUCTION

Analysis of seawater has shown that there are marked seasonal and spatial variations in the concentrations of some of its constituents. The widespread use of radioactive isotopes during the last two decades has provided much valuable information on biological and geological mineral cycles, often showing that the exchange rates of elements between the components of the biosphere and lithosphere are much larger than would have been expected on the basis of net flux measurements. The measurements have shown also that there are wide variations in the turnover rates of elements in systems of apparently similar chemical composition. Pomeroy (1962) presents data on phosphorus turnover in the sea.

The kinetic analysis of element cycles in nature aids interpretation of food chains and gives direct information on the faces of the elements themselves, which is essential to the solution of problems associated with the disposal of radioactive waste.

During the School an attempt was made to examine the partition of ^{32}P between the soluble and particulate phases of the water of Gunnamatta Bay. The experiments were exploratory and aimed at finding whether or not a problem existed. No attempt was made to identify or isolate the components of either phase, which is a vital step in any complete analysis.

II. METHODS

Seawater samples were collected in a polymethacrylate-rubber Van Dorn sampler and stored in polyethylene bottles until required. Stations sampled are indicated in Table 1 and their positions given in Figure 1 of Hydrology Part 1.

A stock solution containing 1 µc of ^{32}P per ml was prepared by adding the carrier-free isotope to seawater from which particulate matter had been removed by membrane filtration. The membranes used were Millipore HA disks of pore size 0.45 µm and diameter 47 mm supported on cellulose pads and held in polymethacrylate holders. A 15 inHg vacuum was used for all filtrations.

The normal pattern of an experiment was to add 3 ml of the stock ^{32}P solution to a soda-glass bottle containing 1000 ml seawater. The bottles were suspended 1 foot below a float in shallow waters around the laboratory jetty, in order to maintain natural illumination and temperature. At intervals, 10 ml aliquots were removed and particulate matter separated by membrane filtration.

The membrane filters were allowed to dry and attached to two inch aluminium dishes with Durofix adhesive. One ml samples of the filtrate were evaporated to dryness on similar dishes using an infrared lamp. After cooling, the samples were protected from atmospheric moisture with a thin film of vinyl lacquer. Radioactivities were measured with a Nuclear Chicago Spectro Shield proportional counter. A minimum of 4000 counts was recorded for each sample which gave acceptable counting statistics. Early experiments showed that self absorption of particles within membrane filters and precipitated salt was negligible so that radioactivities of the two types of sample were comparable.

The results were corrected for isotopic decay and for adsorption of ^{32}P onto membrane filters. The adsorption of ^{32}P onto filters was measured by adding ^{32}P to particle-free seawater and then withdrawing aliquots for filtration at intervals. The radioactivity of the filters obtained in this (Experiment 7B, Fig. 1 (f)) did not change with time and was assumed to be due to adsorption onto membrane filters. The mean of 34 counts per min per ml of solution filtered was subtracted from all other determinations of filterable radioactivity. The results are given as counts per min per ml of solution or per ml of solution filtered.

Soluble and particulate phosphorus and particulate carbon were determined by other members of the hydrology group (Part 1) using methods which have been described already.

In one experiment (6) the loss of ^{32}P from particulate matter was examined. Radioactive particulate matter was isolated by membrane filtration of four 200 ml aliquots of seawater which had been incubated with ^{32}P for 21 hr 45 min. After the initial filtration the filters were left in their separate polymethacrylate holders and particle-free seawater was allowed to pass through. At intervals, the filters were removed, dried, and counted in the normal way.

III. RESULTS

In Figure 1 filterable radioactivity is plotted against time. Supporting information is given in Table 1. There was no detectable adsorption of ^{32}P onto the incubation bottles, the sum of filterable radioactivity and soluble radioactivity remaining constant throughout each experiment. The amount of radioactivity remaining in solution can therefore be calculated by subtraction.

The results of Experiment 6 on loss of ^{32}P from particulate matter, are given in Figure 2 in which residual filterable ^{32}P is plotted against duration of washing. The particulate matter was obtained from the two incubation bottles set up as 5A and 5C. Two aliquots of 200 ml were removed from each bottle. After the initial filtration the membrane filters remained in their holders

during washing (flow rate c. 5 ml/hr). At intervals, filters were removed, dried, and counted in the normal way.

IV. DISCUSSION

Four patterns of behaviour may be distinguished in the fifteen seawater samples which were examined. These were -

(a) Rise in filterable radioactivity during the first five hours of incubation followed by a period of much smaller increase. Samples 1A, 1B, and perhaps 2D.

(b) Partition of ^{32}P between the phases constant throughout the incubation period (3A, 4A, 4B, 5B, 5D).

(c) Steady increase in filterable radioactivity throughout the incubation period (2A, 2B, 2C, 7A).

(d) Exponential rise in filterable radioactivity during incubation period (5A, and perhaps 5C).

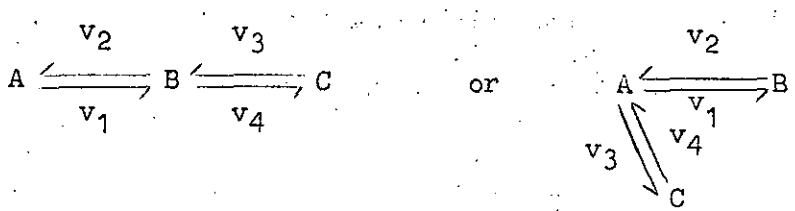
Type a. Rapid rise in the radioactivity of the particulate phase followed by a period with a much smaller rate of increase.

This type of behaviour was observed in 3 samples of surface water, two from the laboratory pier (1A, 1B) and one in a darkened bottle of water from outside the Bay area, 2D.

There are three possible interpretations:-

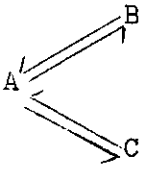
(i) That enclosure causes a change in the seawater leading to a diminution in the rate of movement of ^{32}P into particulate matter. An attempt was made to investigate this hypothesis (Experiment 4) by adding ^{32}P to one part of the seawater sample immediately after collection and to the remainder after it had been in incubation bottles for four hours. No significant result was obtained because the water was of a different type, showing a constant partition of ^{32}P between soluble and particulate matter throughout the experiment.

(ii) That the phosphorus in the water-particulate system can be represented by the equilibrium



where A represents soluble phosphorus, B and C phosphorus in particulate phases, and $v_{1,2,3,4}$ the various partial fluxes. If one compartment attains isotopic equilibrium before the other a graph of activity in the particulate phase against time would be of the two component form that was observed. If the system was in dynamic equilibrium the result should be the same if the experiment is started with either ^{32}P labelled solution or particulate matter. Experiment 6 investigated the validity of this hypothesis, but the water used was of a different type and might not have been in isotopic equilibrium.

At the conclusion of Experiment 2D the ratio of ^{32}P in particulate matter to total ^{32}P was 0.12, the equivalent ratio for phosphorus measured by chemical analysis (Table 1) was 0.19. If the simple model $A \rightleftharpoons B \rightleftharpoons C$ or



is accepted, one of the components of particulate phosphorus has a high turnover rate, equilibrating in under 5 hours and accounts for about 60% of the particulate phosphorus, the other accounts for 30% of the particulate phosphorus and has an insignificant rate of turnover. It is, therefore, possible to accept an even more simplified model of the system,

$$A \underset{v_1}{\overset{v_2}{\rightleftharpoons}} B$$

and to use this to calculate the rate of turnover of soluble phosphorus. At zero time, let the ^{32}P content of the solution, A, be a_o; the ^{32}P content, b, of the particulate matter, B, will be zero. At any later time ^{32}P will exchange between the phases, the rate of change in the particulate phase being written

$$db/dt = k_1(a_o - b) - k_2 b_1 \qquad \ldots\ldots\ldots 1$$

where k_1 and k_2 are rate constants for the two directions of movement.

At equilibrium there is no net movement, let $b = b_e$

$$db/dt = 0$$

TABLE 1

EXPERIMENTAL AND SAMPLING DETAILS AND DATA COLLECTED

Expt.	Date	Station*	Depth (ft)	Time of Sampling	Time of Start of Experiment	Sol. P (µg/l)	Part. P (µg/l)	Carbon (µg/l)	Total Radioactivity (cpm/ml)	Remarks
1A	24.v.67	Jetty	0	1410h	1520h				2331	
1B	"	"	0	1410	1420				2530	Polythene bottle used for incubation
2A	25.v.67	C	0	1000	1420	7.5	4.2	400	1912	A and B from same sample
2B	"	C	0	1000	1425	7.5	4.2	400	1980	Incubation bottle wrapped in aluminium foil
2C	"	0	0	1000	1426	13.1	3.1	200	1992	C and D from same sample
2D	"	0	0	1000	1428	13.1	3.1	200	1997	Incubation bottle wrapped in aluminium foil
3A	26.v.67	B	36	1000	1135	+	5.4		2073	
4A	"	B	36	1000	1440	+	5.4		2116	Water held in incubation bottles for 3 hr prior to addition of ^{32}P
4B	"	B	5	1000	1445	4.0	6.1		2173	
5A	29.v.67	B	36	1000	1415		7.4	750	2159	
5B	"	B	36	1000	1420		7.4	750	2203	Inhibitor methylene blue 5×10^{-4}M
5C	"	B	5	1000	1420		3.1	400	2223	
5D	"	B	5	1000	1428		3.1	400	2224	"
7A	30.v.67	Jetty	0	1400	1445	12.0	20.0		2077	
7B	"	"	0	1400	1445	12.0	20.0		2018	Seawater membrane filtered before start

* See Part 1, Figure 1

\+ Beyond sensitivity limits of the method used

so $k_1(a_o-b_e) = k_2 b_e$

and $k_2 = k_1(a_o-b_e)/b_e$

which may be substituted in equation 1 to give

$$db/dt = k_1(a_o-b) - k_1(b(a_o-b_e)/b_e)$$

which may be rearranged and integrated

$$k_1 = b_e/a_o t.\log_e b_e/b_e-b \qquad \ldots\ldots\ldots 2$$

thus giving a relation which can be used to calculate the partial fluxes, and thus turnover rates. Experiment 1A had a suitable range of values of t (1B and 2D had reached equilibrium before sampling had commenced). Substitution of the data in equation 2 gave values

k_1 = 0.016 per hr (t = 2 hr 24 min), k_1 = 0.022 per hr (t = 6 hr 15 min), and k_1 = 0.009 per hr (t = 25 hr 25 min).

The results mean that during experiment 1A about 15% of the phosphorus in solution was exchanged with the particulate matter each hour.

(iii) A third alternative could be the transformation of ^{32}P into a soluble (filterable) form which differed from the nominal form in its availability to the particulate phase. A situation of this type, for example polyphosphate formation, could only be distinguished by sophisticated chemical analysis.

Type b. Partition of ^{32}P between solution and particulate matter constant throughout the incubation period.

This type of behaviour was observed in the three samples of water taken from the centre of Gunnamatta Bay on May 26. (Samples 3A, 4A, 4B), and in samples taken on May 29 from the same station to which the metabolic inhibitor methylene blue had been added (Samples 5B, 5D).

The average fractions of ^{32}P in the particulate matter of each sample are shown in Table 2.

TABLE 2

Sample	% ^{32}P in particulate matter	% Phosphorus in particulate matter	Mass ratio $\frac{\text{carbon}}{\text{phosphorus}}$ in particulate matter
3A	1.1	100	-
4A	1.2	100	-
4B	1.3	60	-
5B	4.4	-	101
5D	2.4	-	129

The figures indicate a very rapid equilibration of only a small proportion of the total particulate phosphorus and suggest an exchange between ^{32}P and surface-adsorbed phosphorus. There was evidence of this type of exchange in the results of other experiments.

In other water samples which showed more gradual (metabolic?) increases in particulate ^{32}P the graphs of particulate ^{32}P against time did not pass through the origin, but through a point corresponding to 1-2% ^{32}P in the particulate phase at time = 0. The very high values for "absorbed" phosphate obtained in 5B and 5D suggest that there may have been some adsorption onto particulate methylene blue, which is a large organic cation having a tendency to form colloidal particles. The corresponding amounts of particulate ^{32}P during the first 30 minutes in inhibitor free water were 5A, 1.2% (same water sample as 5B), and 5C, 0.8% (same water sample as 5D). An alternative explanation is that methylene blue increased the availability of positive fixed charges in the particulate matter. An interpretation which seems less likely, but was supported by the experimental observation, was that the methylene blue dependent increase in absorbed ^{32}P was in proportion to the amount of particulate matter rather than a simple addition of the fixed charges of the methylene blue particles.

The chemical analysis of particulate matter showed that it contained a great deal of phosphorus which did not show any sign of equilibration with ^{32}P (cf 2D). A result of this type could be expected if the central bay area contained recently dead plant and animal matter as suggested by the hydrology, pigment, and zooplankton groups on the basis of C:P ratios, presence of pigment decomposition products, and dead zooplankters, respectively.

Type c. Constant rate of increase in the amount of ^{32}P associated with particulate matter.

The behaviour observed, an influx of ^{32}P into the particulate matter with no hint of equilibration, is typical of many living organisms which did not multiply during the experimental period (cf Type d). The results emphasize the distinction which may be made between organisms which attain flux equilibrium with respect to a particular nutrient and those which do not. Organisms in the latter category (e.g. most planktonic algae with respect to phosphorus, but not potassium, release ions to the environment only upon death or consumption by a predator.

Except for sample 7A for which no information was available the samples which exhibited type c behaviour were all taken from waters in which the pigments group found evidence of active populations of planktonic algae, which may have caused the observed accumulation of ^{32}P in the particulate matter. The results (Table 3) indicated that in 18-19 hr none of the samples approached isotopic equilibrium.

TABLE 3

Experiment	% ^{32}P in particulate form at end of incubation	Duration of incubation (hr)	Total phosphorus concentration µg/l	% total phosphorus in particulate form
2A	6.9	18.75	11.7	36
2B	8.7	18.5	11.7	36
2C	5.6	18.5	16.2	19
7A	4.4	18.75	32.0	63

The variation in the extent of equilibration suggests that the particulate matter was heterogeneous, perhaps consisting of a mixture of living and dead material, a conclusion supported by the non-zero intercepts of plots of particulate ^{32}P against time.

There was no marked difference between the behaviour of the illuminated sample 2A and the darkened sample 2B. In the second sample in series two there was a very marked stimulation of ^{32}P movement into particulate matter in darkness (2D compared with 2C). The finding was surprising and would have to be confirmed before being regarded as significant.

Type d. Exponential rise in ^{32}P associated with particulate matter.

The best example of type d behaviour (5A) was in water collected from the bay station which provided the inactive samples that showed type b behaviour. The form of the curve suggests a bacterial growth curve with, in this instance, a doubling time of slightly less than three hours with over 25% of ^{32}P in particulate form after 21.75 hr. If this interpretation is correct, and it is supported by the complete inhibition of the increase in accumulation capacity by methylene blue, why did it not occur in the earlier inactive samples? Was there a lack of inoculum, or was there a bacteriostatic agent in the water? The surface sample from the bay station (5C and D) showed a similar increase in accumulation capacity but the rate was smaller.

Experiment 6 showed that when particulate matter was washed with ^{32}P and particle-free seawater the efflux of ^{32}P followed first order kinetics, as shown by the linearity of graphs of the log of the amount of ^{32}P remaining in particulate matter against time. The result, that the efflux was proportional only to the amount of ^{32}P present in the particulate matter, infers that the phosphorus in the agent which brought about accumulation in Experiment 5A was also exchanging rapidly with the seawater, but that this exchange was masked by the rapid population growth. If this had not been the case the amount of ^{32}P present in particulate matter during the washing would not have decreased even though the content per cell would have fallen with increasing population size. (An alternative interpretation is that the washing water, taken from the surface at the end of the laboratory jetty contained a toxic principle.)

It is possible that the organisms which caused the putative rapid exchange in type a water were a non-growing population of the type of organisms found in type d water.

V. CONCLUSION

The discussion has been speculative and has emphasized that kinetic analysis of ion movements can only be meaningful if combined with the development of techniques to identify and isolate the compartments. Given these techniques kinetic analysis is a powerful tool in ecological research, without them it is as useful as any tool lacking a large and vital component.

VI. REFERENCE

POMEROY, L.R. (1963).- Experimental studies of the turnover of phosphate in marine environments. In "Radioecology". Ed. by V. Schultz and A.W. Klement jn. pp. 163-166. (Reinhold : New York.)

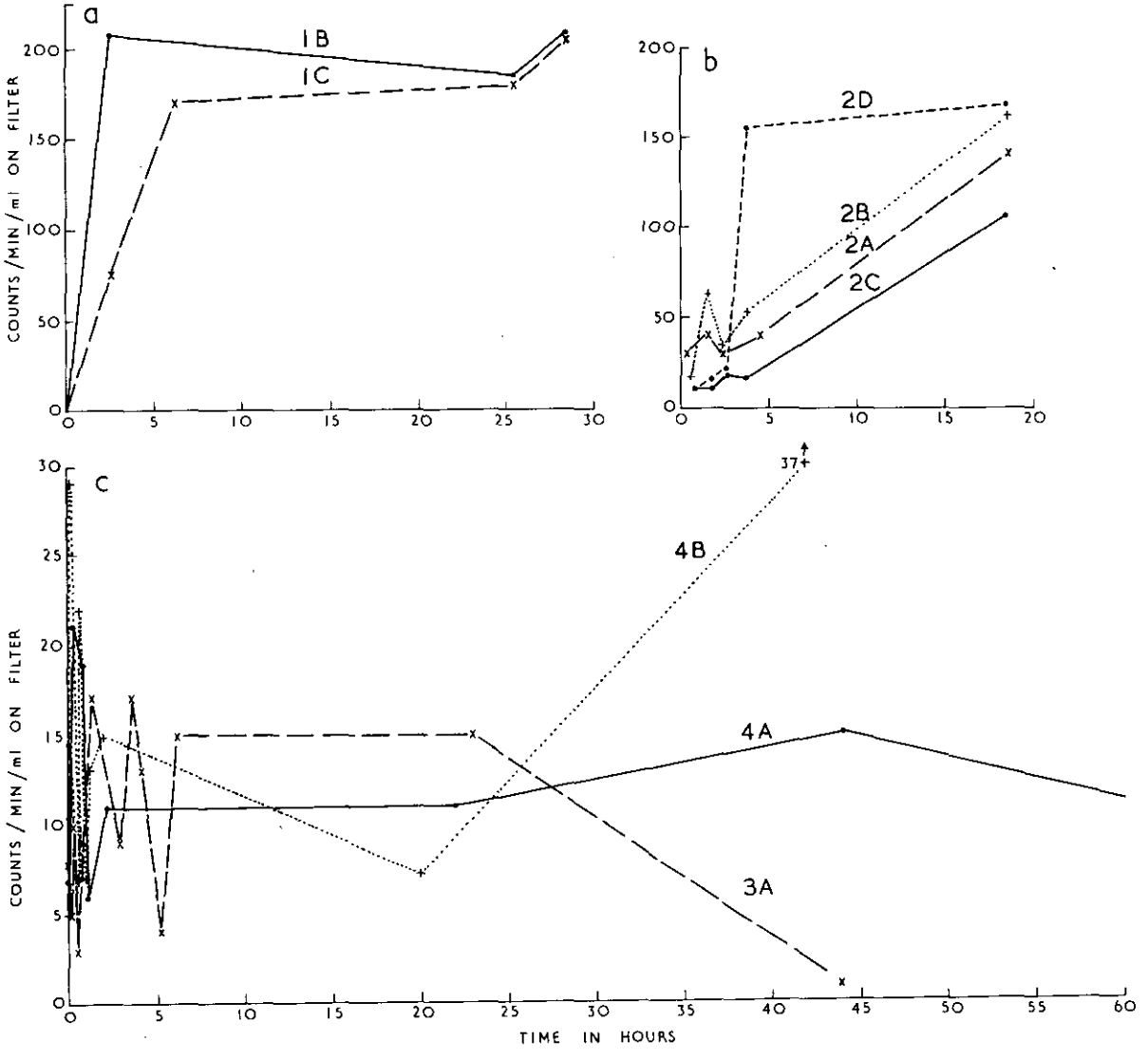


Fig. 1 - Changes in filterable radioactivity with time. Part 1.

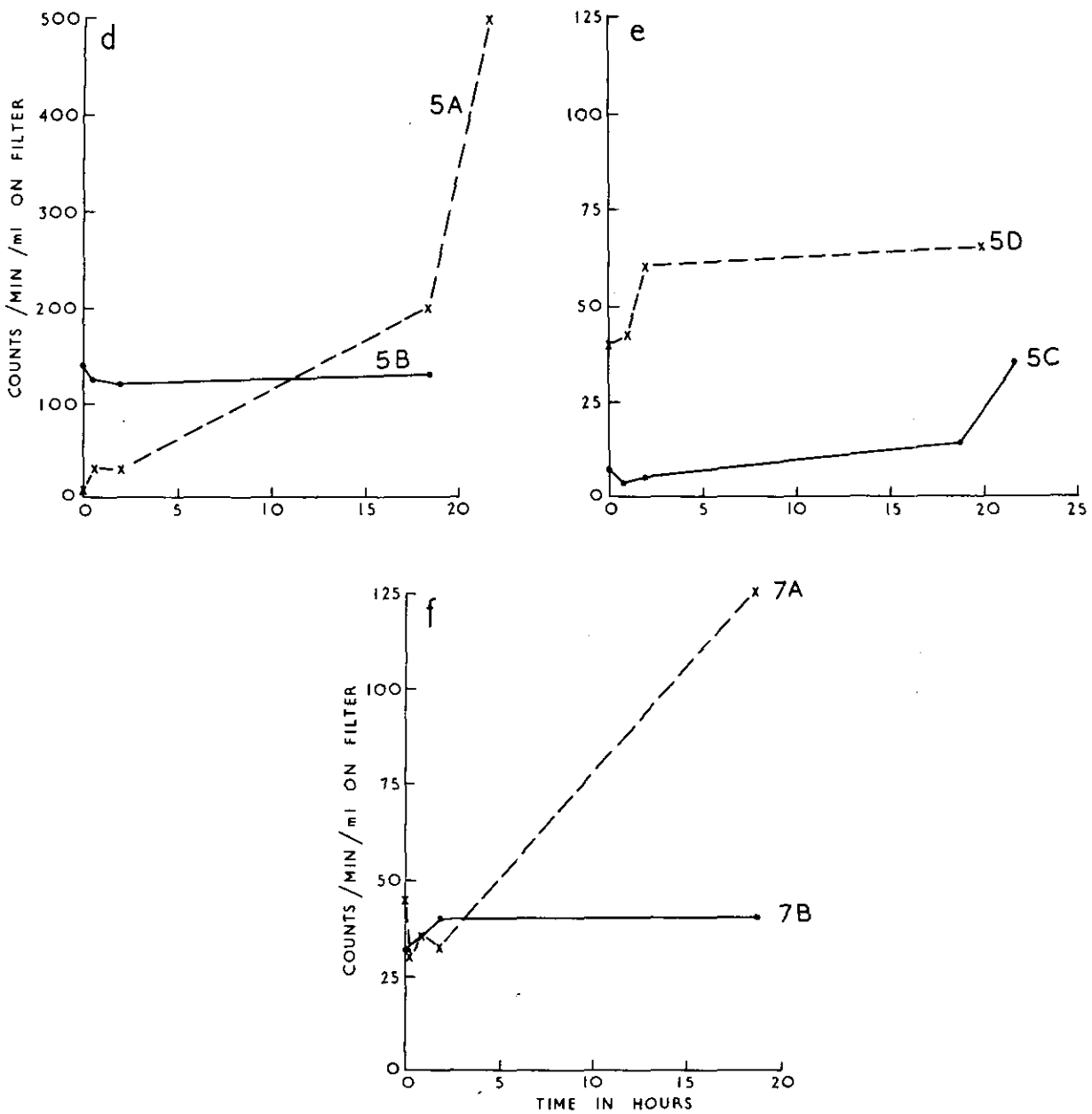


Fig. 1 - Changes in filterable radioactivity with time, Part 2.

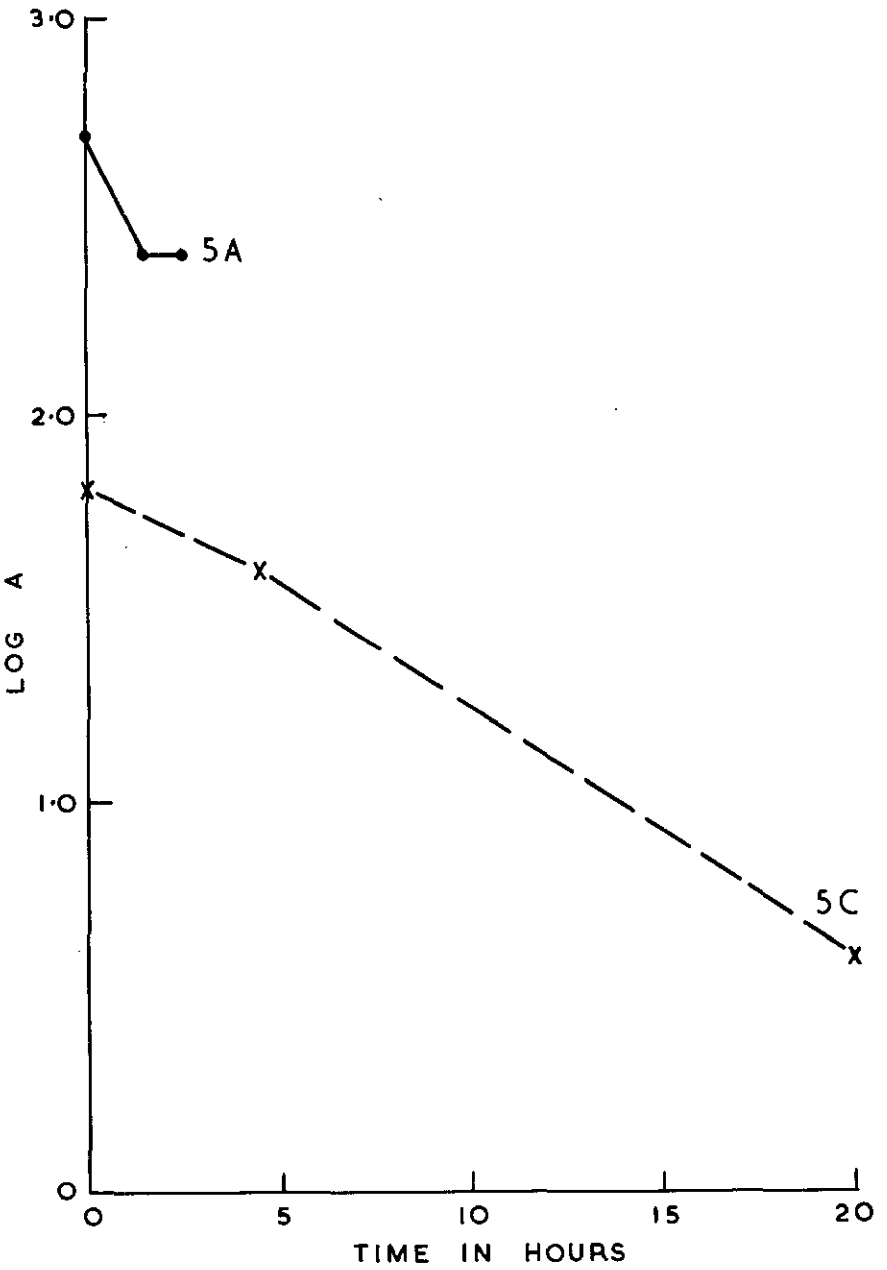


Fig. 2 - Changes in residual filterable radioactivity with duration of washing.

SEDIMENTS

I. INTRODUCTION

The aims of this project were to study -

1. Grain size distribution throughout Gunnamatta Bay for the determination of environmental features of deposition.
2. The acid soluble contents of the sediments, giving a measure of the calcareous benthos and Foraminifera content.
3. The benthic fauna in the sediments and in the weed (both living and dead).

II. METHODS AND MATERIALS

Bottom samples and cores were taken at twelve stations (Fig. 1), fixed by means of prismatic compass and sextant. A Phipps bottom sampler was used for dredging and a 100 lb gravity corer to obtain cores.

Dredge samples were split when wet into approximately 10 g (for acid soluble), 50 g (for grain size analysis), and 500 g (for benthic fauna) subsamples.

The 10 g subsamples were dried, weighed and digested in 10% (v/v) HCl till all reaction had ceased, then filtered through a weighed filter paper, washed and dried, and weighed to obtain the percentage of acid soluble material (mainly calcium carbonate).

The 50 g subsamples were oxidized for 48 to 72 hr with hydrogen peroxide (10 and 50%) to remove the organic material. This was done to prevent clogging of the screens, and to aid dispersion of the clays. The sediment was then washed through a 63 micron screen dividing it into sand and silt-clay fractions.

The silt-clay fraction was analysed wet by pipetting it, after adding a dispersing agent (calcium meta-phosphate), into 32, 16, 8, 4 and 2 micron size fractions and calculating each fraction as a percentage of the total sample.

The sand fraction was dried and weighed, and screened for 20 min in a shaker, using 4 mm to 63 micron screens at $\frac{1}{2}\phi$ intervals ($\phi = -\log_2$ of the diameter in mm). Each size fraction was weighed and calculated as a percentage of the total sample weight.

The results from both the fine and coarse material were plotted on a cumulative percent curve to obtain a median diameter (50 percentile).

The 500 g samples were washed through a 1000 micron screen and a faunal analysis carried out on the coarse fraction (the fine sample was examined by the Foraminifera group). The distribution of species was plotted on a semi-quantitative basis.

Samples of weed were taken with a Danielli rake and rope. The fauna found in the weed was fixed in formalin and preserved in ethylene glycol. A series quantitative formal analysis was then carried out.

The gravity corer was used at Stations 6, 7, 9, 10 and 12. Cores were extruded, split, and logged. The depth of penetration, length of core and compression factors are shown in Table 1.

TABLE 1

SPECIFICATIONS OF CORES TAKEN

Station No.	Penetration	Core Length	Compression Factor
9	6ft	2ft 7in.	0.43
10	6ft	2ft 8in.	0.44
12	8ft	3ft 7in.	0.44
7	7ft 6in.	3ft 6in.	0.47
6	5ft	2ft 4in.	0.47

Eh, pH, and pS^{2-} were recorded along the length of the cores by the physical chemistry group.

Bottom photography was attempted but the camera failed to operate.

III. RESULTS

Figure 2 shows changes in percentage acid soluble material, pS^{2-}, Eh, and grain size through Stations 3, 9, 10 and 11. Figure 3 gives similar information through Stations 1, 2, 4, 11, 12, 7 and 6.

Figures 4 and 5 give the results of sediment size analyses in cumulative frequency curves for each station.

Figures 6 and 7 show the sediment fauna for the station groupings used in Figures 2 and 3 respectively.

Figure 8 shows the structure of cores taken.

IV. DISCUSSION

Generally, coarse grained sediments were found in the channel and finer grained sediments in the bay. The coarse nature of sediments at Station 12 can be explained by the high percentage of carbonate material. From Station 12 through Stations 7 and 6 there was a lowering of acid soluble material which was also reflected in the reduced faunal assemblage and population. The organic content of the sediment appears to increase radially towards the centre of the bay from Station 7 to Station 12.

A study of the five cores showed shelly layers which were traceable throughout the bay and also a distinct change in sedimentation from shelly sand to clay-silt type with few shells. This change can be interpreted in two ways:-

1. Development of the bar which restriced circulation in the bay.
2. Changes in freshwater drainage, possibly as a result of urban development.

Changes in freshwater drainage seem unlikely on the evidence of present drainage patterns.

The finer sediments in the bay showed a bimodal grain size distribution. Two possible reasons for this are:-

1. Two sources of the sediment - from the beaches and fine material from streams.
2. Biological (shell fragments) material, which in general is coarser than detrital grains. The sediment structure at Station 12 suggests a biological factor.

No alternation of the sediment types, suggesting dry and flood periods, can be observed in the cores, and no fine stratification was observed in cores, possibly as a result of biological mixing of fine and coarse material. This is supported by the bimodal character of the grain size distribution curves.

Shelly horizons contained large shells amongst which are pectens in a sandy sediment. This suggests periods of clean sediment with good circulation and might indicate earlier the absence, or less restriction from, the bar at the mouth of the bay.

Two basic faunal groupings were found in the sediments -

1. Large shelly types from outside the bay in the channel Stations 1 to 4. These can be related to the coarse grained sediments.
2. A smaller sized fauna on the north side of the barrier. The sediments in this area are generally fine and contain less sand.

In plotting a traverse across the bay in both a north-south and east-west direction the number of species was found to have a fairly even distribution. This is related to the fact that the centre of the bay is well oxygenated (see Station 12). There was, however, a reduction of species, and more noticeably of numbers, at Stations 6 and 7 at the northern end of the bay.

Weed was found at some stations and its fauna examined. Some of the species were found to be exclusively benthic e.g. Caprella sp, two ostracod species and a copepod species while others such as an ostracod species were found in the zooplankton as well.

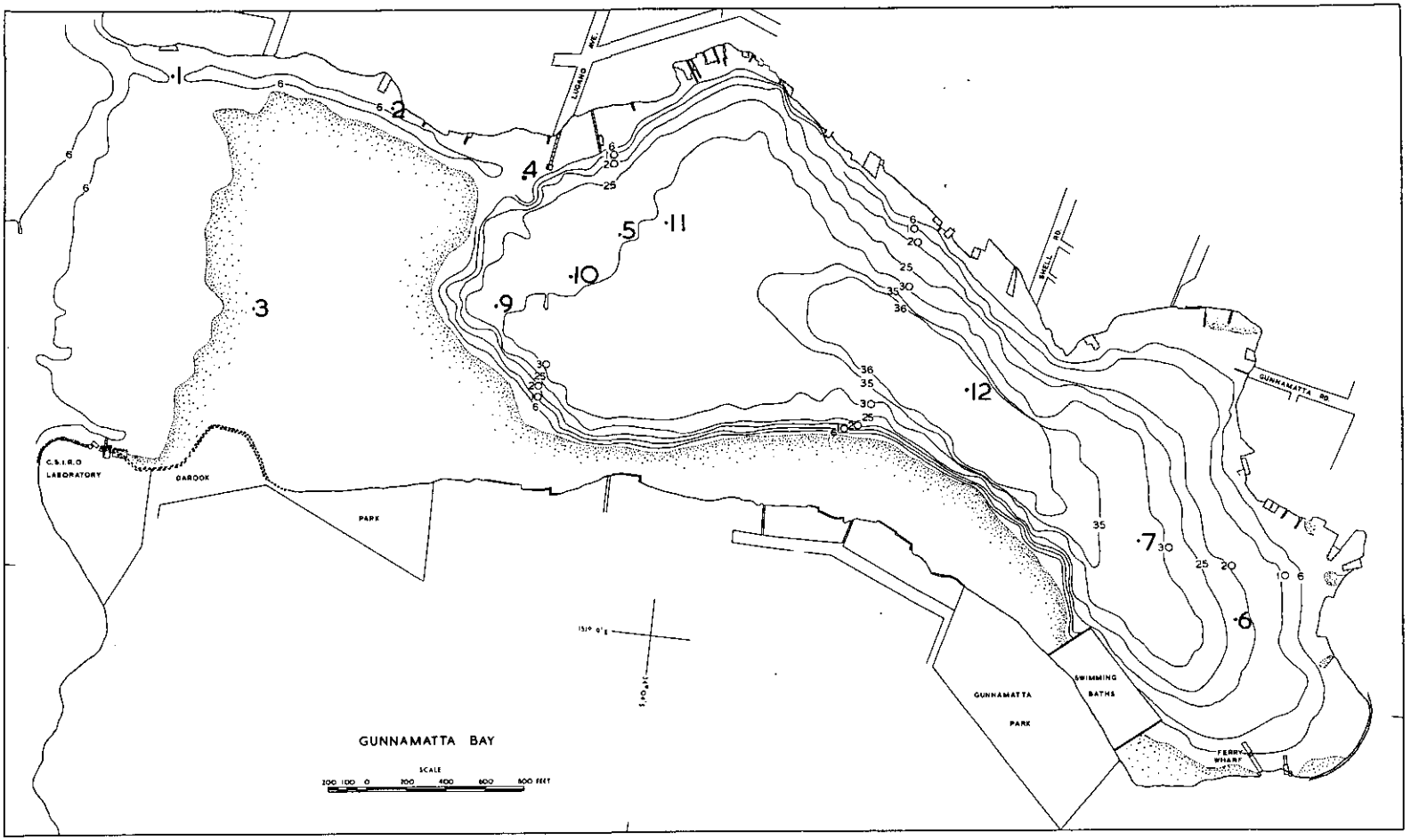


Fig. 1 - Sampling sites in Gunnamatta Bay. Depth contours in feet.

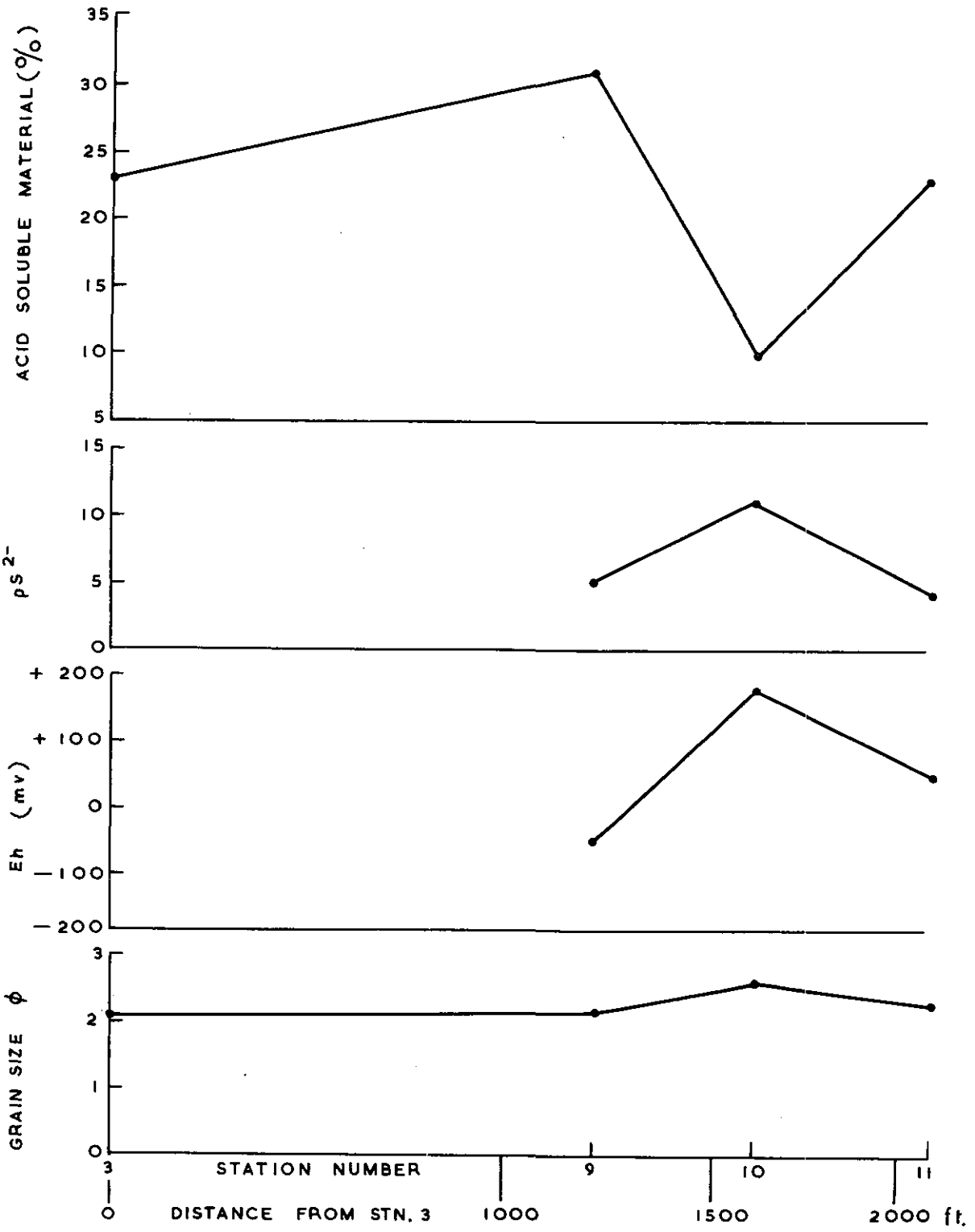


Fig. 2 - Percentage acid soluble material, pS^{2-}, Eh, and grain size at Stations 3, 9, 10, and 11.

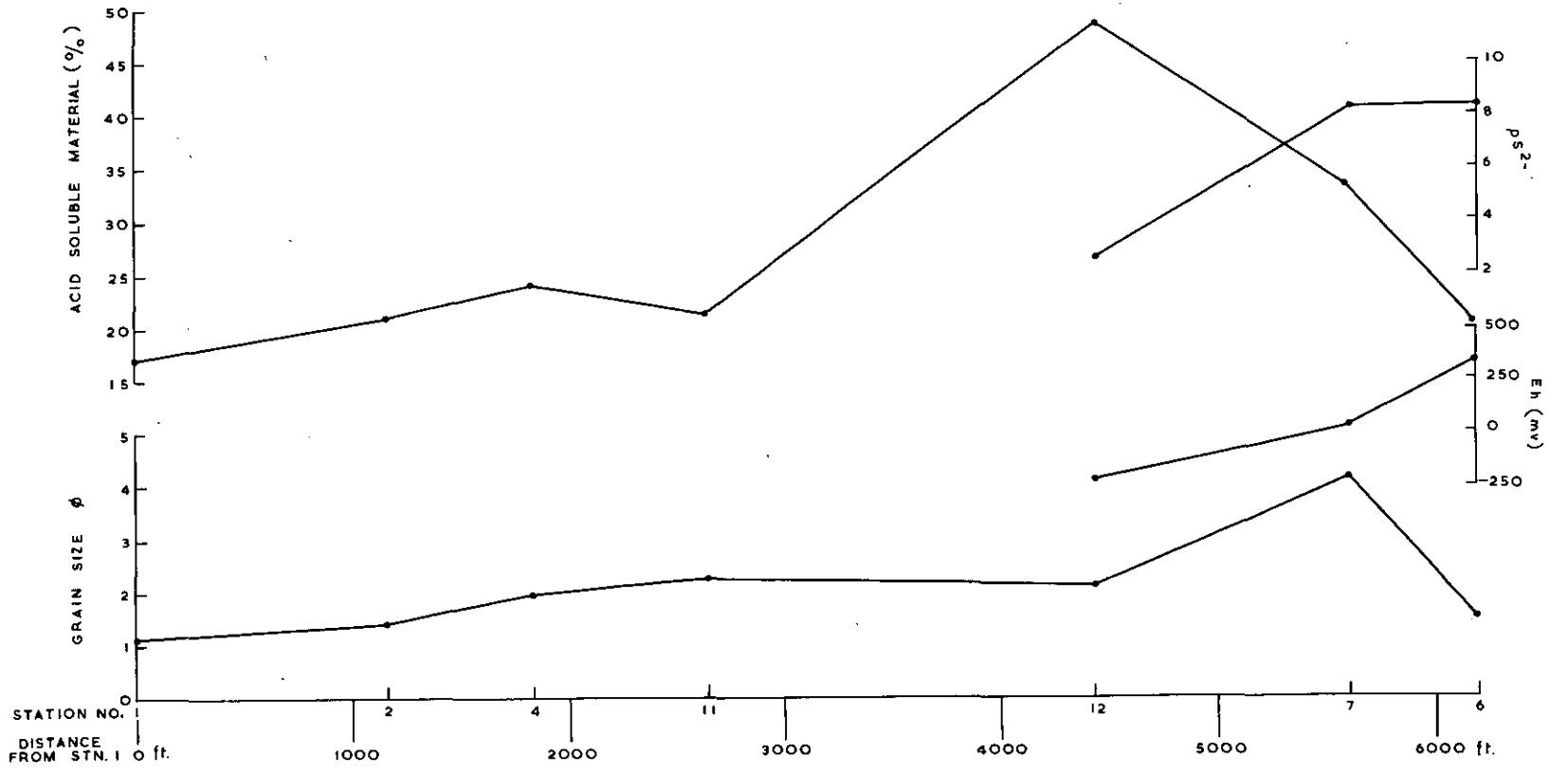


Fig. 3 - Percentage acid soluble material, pS^{2-}, Eh, and grain size at Stations 1, 2, 4, 11, 12, 7, and 6.

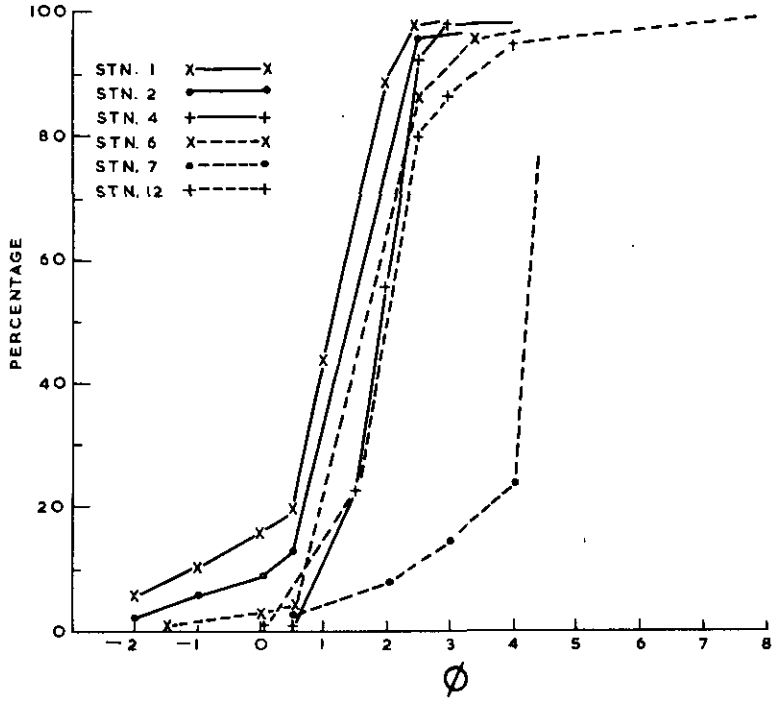


Fig. 4 - Grain size distribution (cumulative frequency) at Stations 1, 2, 4, 6, 7, and 12.

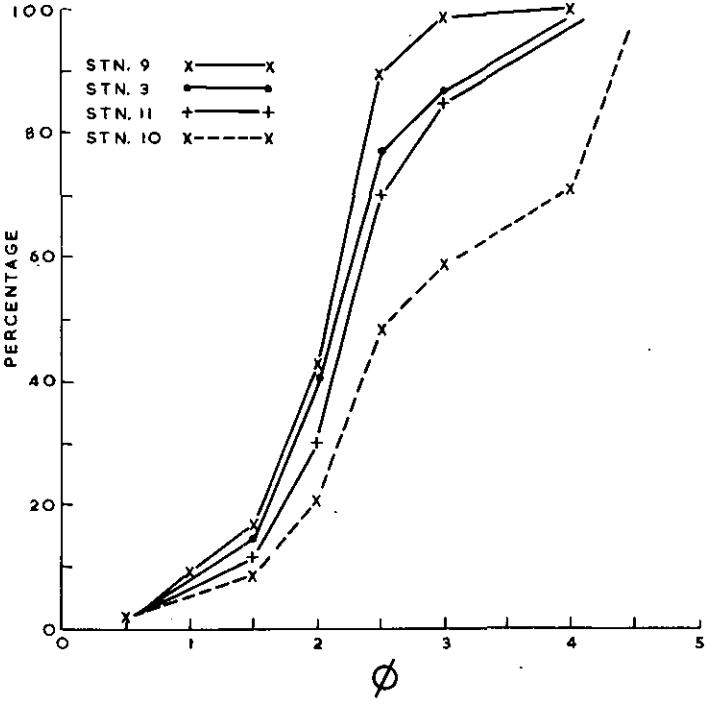


Fig. 5 - Grain size distribution (cumulative frequency) at Stations 9, 3, 11, and 10.

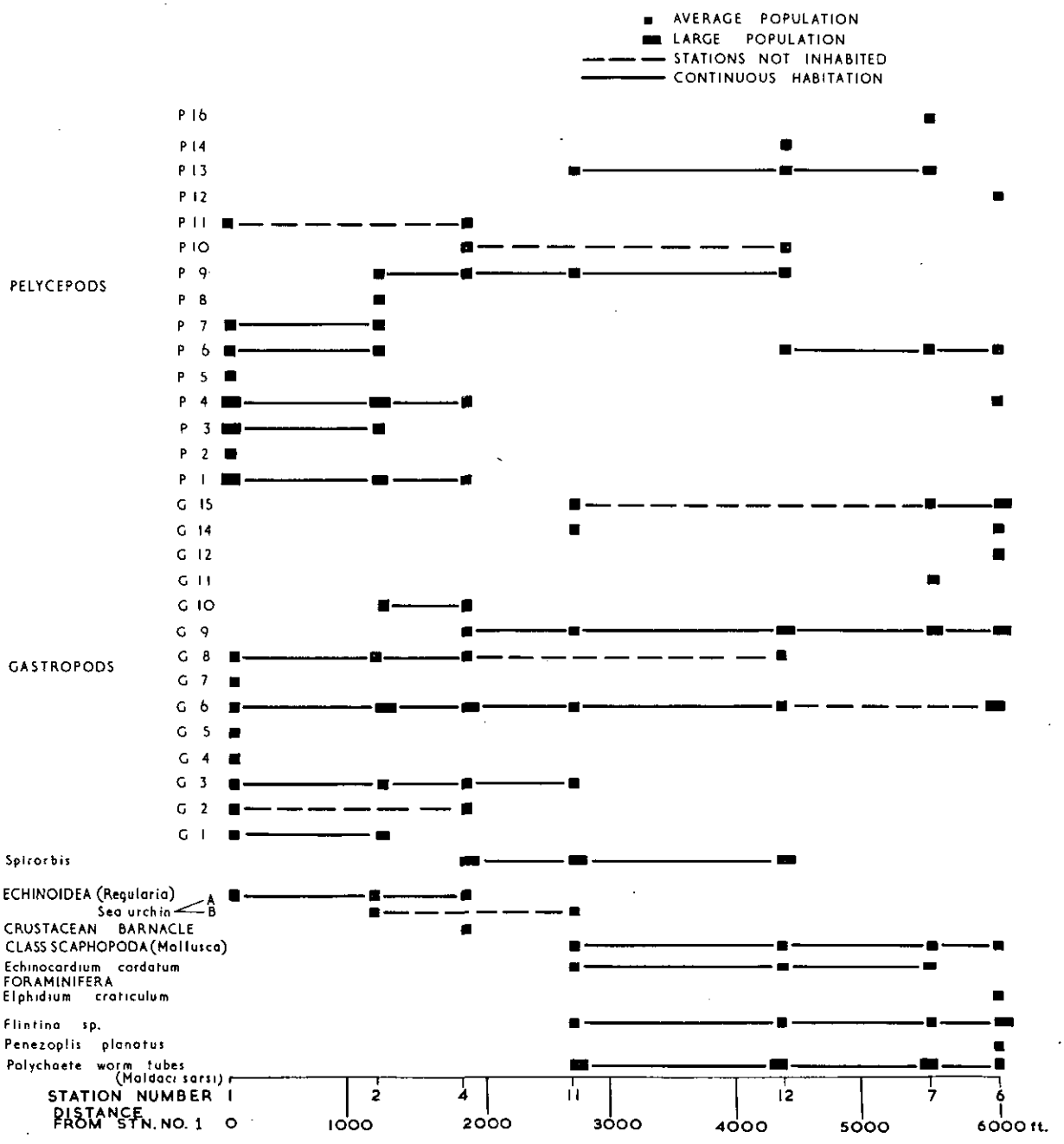


Fig. 6 - Fauna at Stations 1, 2, 4, 11, 12, 7, and 6.
P_{1-16} - pelycepod species G_{1-15} - gastropod species.

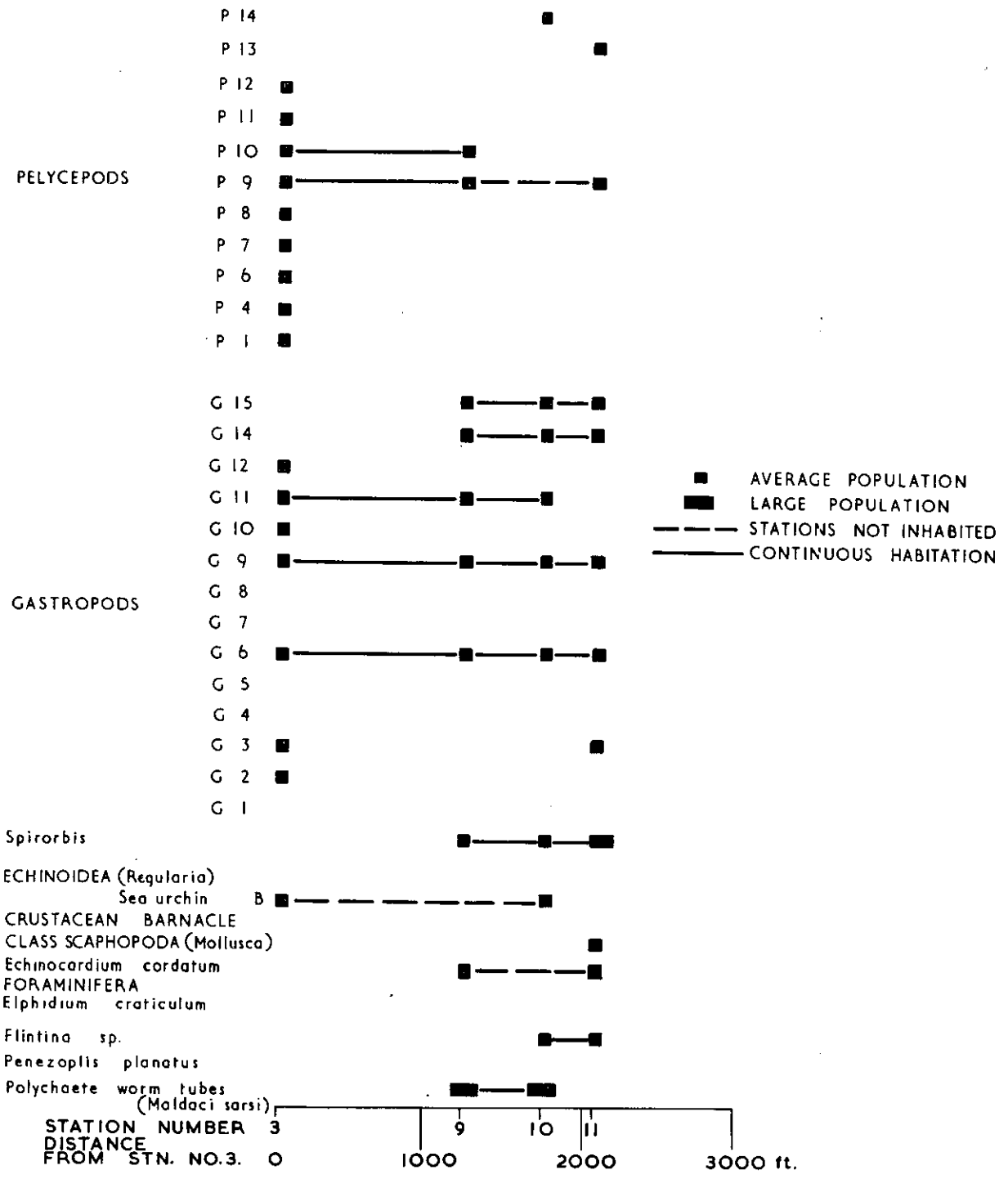


Fig. 7 - Fauna at Stations 3, 9, 10, and 11.
P_{1-14} - pelycepod species G_{1-15} - gastropod species

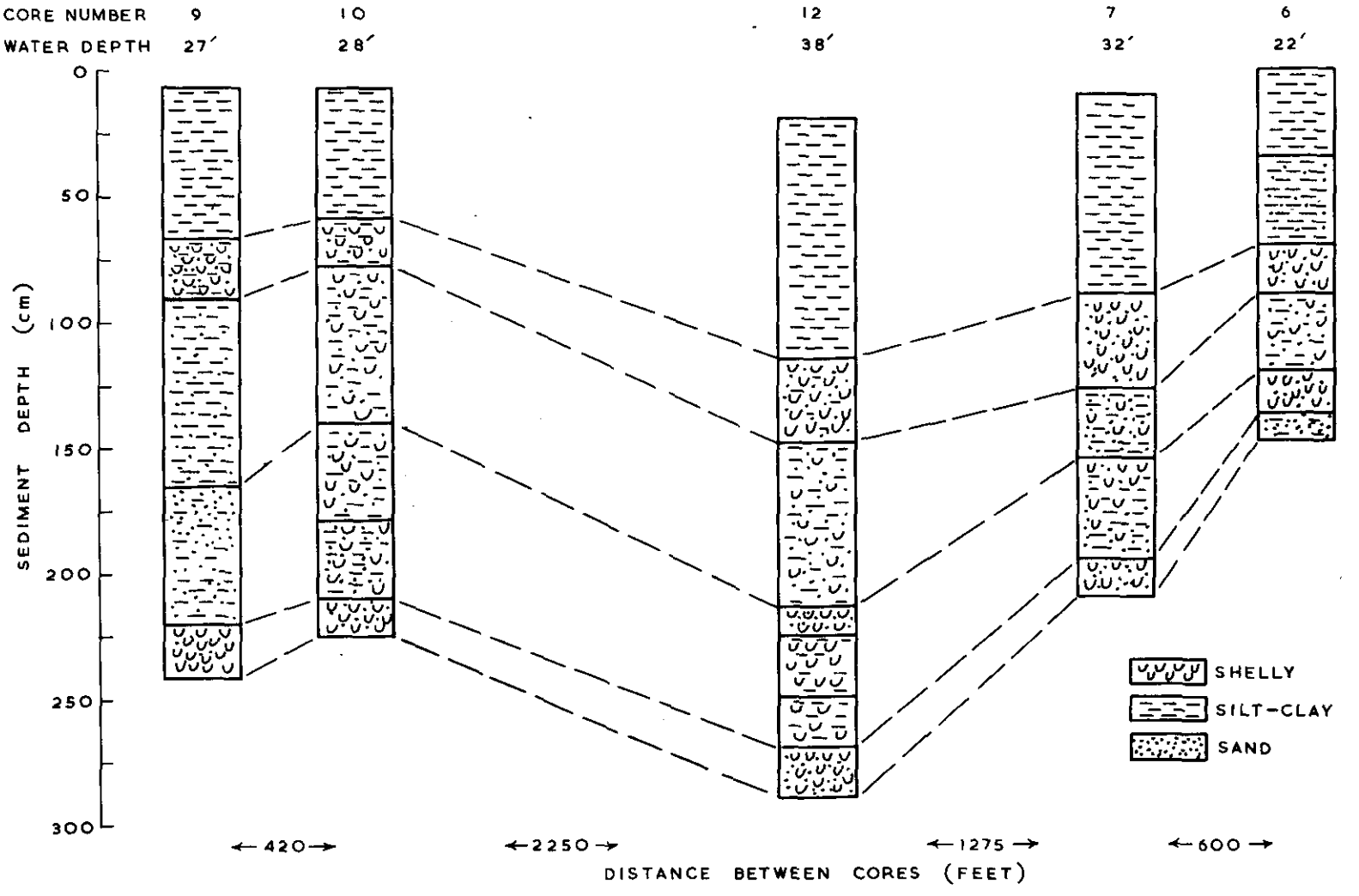


Fig. 8 - Structure of cores taken.

PHYSICAL CHEMISTRY OF SEDIMENTS

I. INTRODUCTION

The application of oxidation-reduction potentials to the study of natural systems, such as a mud-water interface, is complicated by the way in which metal complexes and other ions act together, and by the intervention of bacteria and green plants in the processes. The electron and proton availability measured as Eh and pH, respectively, can characterize such an environment (Baas Becking et al. 1960); and in this context they are used as empirical parameters rather than as electrochemical data capable of thermodynamic interpretation. By measurement of these parameters the overall chemical changes brought about in marine sediments can be followed.

Eh is the electromotive force of an oxidation-reduction system referred to a standard hydrogen half-cell, and is a quantitative expression of its oxidizing and reducing intensity. In general:

$$OX. + ne^- \rightleftharpoons RED \qquad 1$$

In most organic reactions the electron shift is accompanied by a proton shift and the reaction is therefore pH dependent.

$$\text{e.g. } ne^- + mH^+ + OX \rightleftharpoons RED \qquad 2$$

By applying the equilibrium constant to Equation 2, and with simplification of terms, Eh of this reaction can be expressed as:

$$Eh = E_{redox} - \text{const. pH} \qquad 3$$

E_{redox} is a measure of electron availability and the constant depends on the temperature and the nature of the reactions occurring in the system.

To follow specifically the action of sulphate-reducing and other bacteria involved in the formation and utilization of H_2S, it is also necessary to measure the parameter pS^{2-} (i.e. $-\log_{10}$ of the activity of sulphide ions).

In this project, electrode techniques for determining the empirical parameters pH, Eh, and pS^{2-} were applied to the physicochemical measurement of the sedimentary environment in Gunnamatta Bay, Port Hacking. Correlation of these parameters to sediments of different particle size and to the distribution of biological populations was attempted. Eh/Ph changes with depth of sediment were also measured.

The bacterial population of the natural sediments was estimated, and an attempt was made to reproduce this population in laboratory models and to simulate the physico-chemical conditions that occur in the field.

II. METHODS

(a) Field Determinations

Eh was estimated by using a bright platinum electrode, pH by a glass electrode, and pS^{2-} by a sulphide-coated silver electrode. The three electrodes were interchangeable and were coupled to a portable Keithley 601 Electrometer. A standard calomel (Hg/Hg_2Cl_2) half-cell with a remote junction was used as an intermediate reference in all cases. The technique for determining pS^{2-} by using a silver/silver sulphide electrode is that of Berner (1963).

Sediment samples were collected by means of a "torpedo" core sampler and parameters were estimated as soon as possible after each core was collected. No cores were left standing for more than 30 sec.

An attempt was made to measure all three parameters in situ by using specially constructed probes (see Figs. 1, 2, and 3). A different probe design was used for deep water sediments to the one used for exposed sandy flats, the major difference being that the latter probe design was thinner, for easier insertion into the sand. With deep water sediments, the special probe was lowered from a boat until contact with the sediment occurred. Readings were then taken directly. With sandy flats, the probe was pushed into the sediment until the required depth was reached.

Parameters were measured systematically over a range of sediment types in Gunnamatta Bay (Fig. 4). With selected samples, pH/Eh/pS^{2-} changes with depth of sediment core were determined. Measurements were also taken in various submerged plant patches and compared with those from bare patches. In addition, parameters were measured in association with the distribution of benthos on a sand flat.

(b) Estimation of Bacterial Population in Sediments

Two selected sediment samples were centrifuged at 3000 r.p.m. for 15 min and the interstitial water so obtained was used as inoculum on five selective agar media. Incubation was carried out for five days at 23°C under a variety of combinations of light and oxygen access. Plates were then examined for relative abundance of various bacterial types, and an attempt made to determine conditions for optimal growth. Variation in bacterial content between sediment was noted.

The five media used in bacterial culture experiments were:-

1. General - nutrient agar prepared with seawater

2. Selective medium for sulphate-reducing bacteria (Salle 1954)

Sodium lactate	0.5 g
Asparagine	0.1 g
$MgSO_4.7H_2O$	0.1 g
K_2HPO_4	0.05 g
Ferrous ammonium sulphate	Trace
Seawater	1 l.

In 2% agar
pH 7.6

3a. Selective medium for sulphide-oxidizing bacteria (Medium T)

KH_2PO_4	1.0 g
NH_4Cl	1.0 g
$MgCl_2$	0.5 g
NaCl	10.0 g
H_2O	1 l.

Trace elements - add 1.0 ml of the following solution (sterilized separately) to each litre of basic Medium T. $FeCl_3.6H_2O$, 1.5 mg; $CuCl_2.2H_2O$, 1.0 mg; $NiCl_4.6H_2O$, 2.0 mg; Na_2MoO_4, 5.0 mg; $Co(NO_3)_2.6H_2O$, 5.0 mg; $CuSO_4.5H_2O$, 5.0 mg; $MnCl_2.6H_2O$, 5.0 mg; dist. H_2O, 1000 ml.

The following solution was made up more concentrated, sterilized by filtration, and added before use:-

$NaHCO_3$, 2 g/l. of basic medium for all species

pH was adjusted to 7.0-7.2

3b. Similar to 3a except pH 8.0-8.4

4. Medium for iron bacterium, Thiobacillus ferrooxidans (9K Basal Medium).

$(NH_4)_2SO_4$	3.0 g
KCl	0.1 g
K_2HPO_4	0.5 g
$MgSO_4.7H_2O$	0.5 g
$Ca(NO_3)_2$	0.01 g

Seawater to 900 mls

Agar 2%
pH 5.0

A sterile solution (Millipore filtered) of $FeSO_4$ (2% w/v) was added aseptically.

(c) Culture of Sulphur Bacteria

The medium used for culture of sulphur bacteria was that of Postgate (1966).

Two one-litre graduated cylinders were half-filled with a wet slurry of tissue paper and saw-dust to act as a cellulose source. As an organic source one finely diced oyster and some diced Zostera leaves were added. One gram of $CaSO_4$ was added as nutrient for the sulphate-reducing bacteria, as well as some phosphate buffer (0.14 g KH_2PO_4/100 ml seawater). The cylinders were topped up to 850 ml with seawater filtered through Whatman No. 111 paper.

Cylinders were then inoculated with one of the sediment samples used in the bacterial culture experiments. Three grams of wet mud were considered sufficient inoculum. Half of the cylinder surface area was masked with black tape to provide a darkened region. Cylinders were then sealed with "Parafilm" and left standing in the light at 23°C for one week.

The theory tested was that in the cylinder anaerobic SO_4^- reducing bacteria utilize fermentation products and generate H_2S which acts as a substrate for sulphide-oxidising bacteria.

Eh, pH, and pS^{2-} were determined within each cylinder immediately after inoculation and after one week of fermentation.

(d) Laboratory Models

Laboratory models were set up in an attempt to reproduce the important chemical changes attending the bacterial decomposition of organic matter in the marine environment. The technique employed was that of Berner (1963). Two sources of decaying organic matter were used, namely Zostera leaves and oyster.

Nine culture jars were set up in the following manner:-

	Jar No.								
	1	2	3	4	5	6	7	8	9
One diced oyster	+	+	+	+	+	-	-	-	-
Diced Zostera	-	-	-	-	-	+	+	+	+
Sterile sand	-	-	-	-	+	+	+	-	-
Ppt. iron hydroxide trace	+	+	+	-	+	+	+	+	-
$CaCO_3$ 1.5 g	+	+	-	+	+	+	+	-	+
Filtered seawater	+	+	+	+	+	+	+	+	+
Bacteriocide	+	-	-	-	-	+	-	-	-
Contaminated interstitial water (1 ml)	+	+	+	+	+	+	+	+	+
Total Volume (ml)	400	400	400	400	400	200	400	400	400

Jars 1 and 6 had a bacteriocide added and therefore acted as system controls for each organic source. The bacteriocide used was zephiron chloride* at a concentration of 1 p.p. thousand. Sand was added to some jars to increase surface area for bacterial action, and also to simulate natural conditions. $CaCO_3$ was added to simulate animal shells present in the natural environment. Precipitated iron hydroxide was added as an iron source for iron sulphide formation. The effect of the addition of $CaCO_3$ and iron oxide trace could be studied, as both additives were eliminated from some jars.

After inoculation with contaminated interstitial water from sediment samples, Eh, pH, and pS^{2-} were determined periodically for two days. A mercury-filled glass electrode was used to estimate pH, a bright platinium wire electrode for Eh, and a silver/silver sulphide electrode for pS^{2-}. All three electrodes were connected to a Keithley 601BR Electrometer using a remote junction calomel reference half-cell.

III. RESULTS

(a) Field Determinations

All field data for Eh, pH, and pS^{2-} are given in Tables 1, 2, and 3 at the end of this section. Only results from samples collected by coring are presented. The *in situ* probe proved unreliable.

Contours of Eh in Gunnamatta Bay are shown in Figure 5.

Results from samples collected on the beach occupied by the Benthos group were shown in Table 3. The sampling grid used is shown in Figure 6.

The Sediment group obtained cores at selected stations in Gunnamatta Bay. Changes in pH, Eh, and pS^{2-} with depth along the core are shown in Table 2 and Figure 7.

The Eh/pH relationship is shown in Figure 8, Eh/pS^{2-} in Figure 9, and pS^{2-}/pH in Figure 10.

(b) Estimation of Bacterial Population in Sediments

The two sediment samples selected gave similar results. Results are shown in Table 4.

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* Benzalkonium chloride

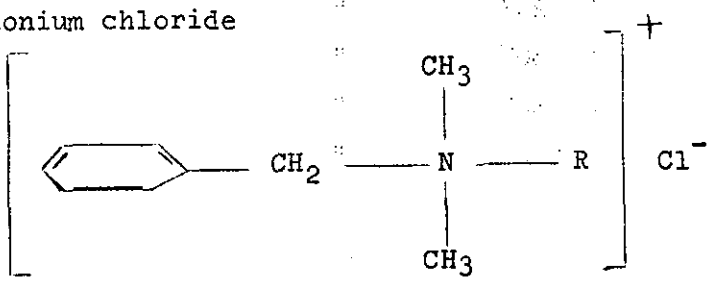

TABLE 4

BACTERIA IN SEDIMENT SAMPLES

Media	Bacterial Group	Optimum Conditions	Relative Abundance*	Appearance
GENERAL	-	Aerobic, light	1	Fungi and bacteria
	-	Aerobic, dark,	3	Fungi and bacteria - agar digested
	-	Anaerobic, light	4	Agar digested
	-	Anaerobic dark	4	Fungi and bacteria - agar digested
	SO_4-reducing, pH 7.6	Anaerobic, dark	3	One type of bacterium only - translucent, white
	Sulphide-oxidizing, pH 7.0	Anaerobic, light or dark	3	Agar digested - dark pigmented bacteria
	Sulphide-oxidizing, pH 8.0	Anaerobic, light or dark	4	Agar digested
	Iron bacteria, pH 5.0	Anaerobic, light or dark	+	One type of bacterium only - poor growth

* + isolated occurrence
1 5% of plate area covered
2 20% "
3 50% "
4 80% "
5 100% "

(c) Culture of Sulphur Bacteria

Changes in the pH, Eh, and pS^{2-} of the culture columns are shown in Table 5. These data are related to those shown in Figure 9.

TABLE 5

CHANGES IN pH, Eh, AND pS^{2-} OF SEDIMENT SAMPLES WITH TIME

	Sediment No. 6			Sediment No. 10		
	pH	Eh	pS^{2-}	pH	Eh	pS^{2-}
25.v.67 Start of Experiment	6.80	+200	16.25	6.80	+200	16.25
1.vi.67 End of Experiment	6.70	-115	4.25*	6.70	-140	4.00*

*H_2S evident - culture blackened

(d) Laboratory Models

Electrode readings of Eh, pH, and pS^{2-} were taken at approx. 4 hr intervals. Results are shown in Table 6.

TABLE 6

Eh, pH, AND pS^{2-} CHANGES IN LABORATORY MODELS OF MARINE BACTERIAL DECOMPOSITION

Jar No.	Time (hr)	Position	Eh	pH	pS^{2-}	Jar No.	Time (hr)	Positio	Eh	pH	pS^{2-}
1	0		357	6.7	22.7	2	0		335	7.0	23
(control)	6		295	6.6	20.8		6		285	6.0	20.8
	10		245	6.5	20.8		10	top	215	5.9	20.8
	20	top	325	6.7	21		"	bottom	-155	6.0	20.8
	"	bottom	255	6.7	20.8		20	top	340	5.8	20
	24	top	325	6.7	20.1		"	bottom	-205	6.0	19.5
	"	bottom	310	6.9	19.9		24	top	295	6.2	19.5
	29	top	345	6.4	19.8		"	bottom	300	6.2	19
	"	bottom	375	6.4	19.8		29	top	330	5.8	19
	32	top	280	6.9	21		"	bottom	330	5.8	19
	"	bottom	255	6.9	21		32	top	20	6.2	19.5
	45		175	6.6	22		"	bottom	-255		
							45	top	145		20.8
							"	bottom	-295	6.4	20.5

TABLE 6 (Cont.)

Jar No.	Time (hr)	Position	Eh	pH	pS^{2-}	Jar No.	Time (hr)	Position	Eh	pH	pS^{2-}
3	0		322	5.8	22.5	4	0		275	6.3	21.5
	6		310	5.4	21		6		300	5.9	21.2
	10	top	45	5.6	20.5		10	top	-85	5.8	19.8
	"	bottom	-255	5.4	20.5		"	bottom	-245	6.0	19.4
	20	top	115	5.6	19.8		20	top	-115	6.4	19
	"	bottom	-205	5.6	20		"	bottom	-205		
	24	top	225	6.1	19		24	top	95	6.1	17.6
	"	bottom	-125	6.0	19		"	bottom	20	6.3	18.6
	29	top	-265	5.7	18.5		29	top	200	6.1	18.9
	"	bottom	-265	5.7	19		"	bottom	125	6.1	19
	32	top	-95	6.2	19.8		32	top	-240	6.3	-
	"	bottom	-315		19		"	bottom	-180		19.5
	45	top	165	6.4			70	top	-215	6.6	17.8
	"	bottom	-235		18.5		"	bottom	-325		
5	0		290	7.2	21	6	0		310	7.0	
	6		325	6.6	21	(con-	6		305	6.9	
	10	top	260	6.6	19.5	trol)	10	top	405	6.9	
	"	bottom	277	6.7	19.5		"	bottom	375	7.1	
	20	top	390	7.0	18.8		20	top	250	7.0	
	"	bottom	420	7.0	18.8		"	bottom	270	7.1	
	24	top	180	7.1	17.5		24	top	305	6.9	
	"	bottom	180	7.1	17.2		"	bottom	290	6.8	
	29	top	268	7.0	15.3		29	top	305	6.7	
	"	bottom	230				"	bottom	275		
	32	top	-65	7.3	19.8		32	top	255	6.8	
	"	bottom	95				"	bottom	225		
	70		-45	7.5	19.8		70		185		
7	0		313	7.1	21.2	8	0		295	6.3	21.2
	6		325	6.4	21		6		290	6.1	21
	10	top	345	6.4	20.1		10	top	385	6.1	20.2
	"	bottom	350	6.4	20.1		"	bottom	370	6.1	20.1
	20	top	225	6.4	19.7		20	top	45	6.1	19.9
	"	bottom	80	6.4	19.7		"	bottom	-35	6.1	19.9
	24	top	285	6.4	18.6		24	top	295	6.2	19.5
	"	bottom	270	6.6	18.6		"	bottom	270	6.0	19.3
	29	top	300	6.7	19.9		29	top	355	6.2	20.2
	"	bottom	305				"	bottom	-245		
	32	top	295	6.8	20.5		32	top	45	6.4	21.1
	"	bottom	280				"	bottom	-155		

TABLE 6 (Cont.)

Jar No.	Time (hr)	Position	Eh	pH	pS^{2-}
9	0		293	6.8	21.2
	6		355	6.1	21
	10	top	365	5.6	20.1
	"	bottom	225	6.2	20.2
	20	top	170	5.6	20.2
	"	bottom	-85	6.2	17.1
	24	top	265	5.7	19.3
	"	bottom	125	6.4	14.5
	29	top	95	6.2	20.1
	"	bottom	-215		
	32	top	-155	6.4	17.0
	"	bottom	-255		

Only Eh varied markedly with time. Figure 11 shows results for jars containing diced oysters as decaying agent, and Figure 12 results for Zostera jars.

IV. DISCUSSION

(a) Use of In Situ Probes

The use of an in situ probe for measuring physico-chemical parameters is advantageous as it eliminates any oxidising effects on the sample while collecting and transporting it to the boat for measurement. It also enables ease of sampling in sediments which are very fine and therefore difficult to collect by normal core techniques.

Although the probes illustrated in Figures 1 and 2 appear very satisfactory for this purpose, results obtained from them were erratic and non-reproducible. Suggested reasons for failure of the probe system are:-

1. Electrode connections are too fragile and therefore prone to breakage under field conditions.
2. Coaxial connections between electrodes and measuring instrument are too exposed, and therefore easily contaminated with seawater, oil, dirt, etc.

e.g. condition	E_c (mV)
connectors dry	+20
connectors immersed in seawater	-290

3. Probe tip is too blunt and often fails to penetrate sandy sediments.

4. Overall design is too bulky, and offers too great a resistance during free-fall from boat.

When all of the above shortcomings in design were corrected, however, readings obtained were still erratic. This was found to be caused by galvanic action between the metals used in the probe and the surrounding seawater. As the calomel electrode was earthed at the measuring instrument (i.e. connected to seawater), the galvanic potential was effectively introduced as a back-off in the measuring circuit. This effect is illustrated in the following table:

Components Immersed in Seawater	E_c (mV)
Metal guard + electrode (platinium)	-40
Metal guard only (chromium plated brass)	-60
Lead weight only	-60
Whole of probe body	-80
Electrodes only	+20

As this galvanic potential is variable, E_c readings are useless. Future probe design will have to employ an inert casing material (such as perspex or araldite) and contact points on the coaxial connecting cable will have to be effectively covered.

(b) Field Results

Figure 5 illustrated areas of similar Eh within Gunnamatta Bay. The two most prominent features are the large basin of reduced sediment in the middle of the bay, and the effect of oxidised sand at the mouth of the bay. There is a strong confirmation of the reducing sediments in the basin from sulphide determinations, which indicate high sulphide levels (see Table 1).

The data also indicate a marked difference between the north and south slopes of the bay as observed by the Foraminifera group.

The main features of Figures 8 and 10 are the narrow range of pH, and the wide spread of Eh and pS^{2-} respectively. This trend is further shown in Figure 9.

In Figure 9, the negative Eh shows quite a strong correlation with high sulphide levels (see Berner 1963), and this indicates that reactions involving sulphide ions probably control the

system. However, when the Eh is positive, the correlation is more diffuse as one would expect from the instability of sulphide ions under oxidising conditions. This correlation is shown more clearly in Figure 7 (Eh/depth in core).

There were insufficient results to show any clear relationship between animal and plant communities and the parameters measured (Eh, pH, and pS^{2-}).

Measurements made on the beach occupied by the Benthos group indicated a fairly uniform environment down to a depth of 20 cm (Table 3). This is in contrast to the sharp cut-off in the animal population found at about 6 in. depth in the intertidal zone.

(c) Bacteria Plating Experiment

Bacteria grew on all the plates. However, growth was not extensive on the iron medium because of the slow growth rate of the iron bacteria.

Optimum conditions for growth of the sediment bacteria seem to be anaerobic, dark, warm (30°C) conditions. But most of the bacteria were facultative anaerobes and grew almost as well in aerobic conditions and in light.

All the sediment bacteria observed under the microscope were rod-shaped, consistent with the morphology one would expect with marine bacteria. Many of the bacteria were agar digesters, and this is interesting because of the frequency of alginates and other algal breakdown products in seawater. Many of the plates contained fungi as well as bacteria.

There was no observable difference between the frequency of bacteria in the different sediment samples, and it appeared that similar bacteria occurred in both.

(d) Culture Experiment

The results indicate marked successions of oxidising and reducing bacteria (Figs. 11 and 12). The periods of oxidation and reduction were of different lengths, depending on the nature of the organic source.

pS^{2-} and pH changed only slightly and no correlation could be found between them and Eh.

The pH results showed no regular change. Those jars containing $CaCO_3$ or a buffering agent did not differ significantly from jars without $CaCO_3$, i.e. buffering had little effect.

The pS^{2-} values were very low (in the order of 20) and did not change markedly. Therefore, it is assumed that sulphate reducing bacteria were not growing to any significant extent.

As a large range in pS^{2-} was found in field data it must be assumed that we have not reproduced field conditions. Reasons for this could be:-

1. Air being constantly introduced into the jars during electrode readings
2. Shaking of jars, before and after taking the electrode readings
3. Only a small amount of substrate, organic material and inoculum was used.

The above conditions were avoided in the tube experiment (see Table 5). Marked changes in Eh and pS^{2-} were observed; the pH changed only slightly in both cases (6.8→6.7). The Eh changed from an oxidised state to a highly reduced state - this was correlated with a marked increase in pS^{2-}, indicating growth of sulphur reducing bacteria.

The Eh change showed a shift into the regions found in the field for reducing sediments (see Fig. 9).

V. CONCLUSIONS

The composite electrode (Fig. 3) proved to be rapid and convenient in field determinations although the protective bodies (Figs 1 and 2) will have to be redesigned to eliminate the galvanic effect.

The field data indicate the significance of the sulphide ion activity in reduced sediments (Fig. 9) and show that Eh/pS^{2-} diagrams are more useful than Eh/pH diagrams for characterising sediments under these conditions (see Baas Becking et al. 1960).

The laboratory experiments indicate the rapidity with which properly prepared cultures attain the conditions found in reduced basins (Fig. 5) and the importance of an adequate substrate for bacterial growth. The frequent occurrence of fungi and agar-digesting bacteria indicate interesting aspects of the micro-ecology of reduced sediments.

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TABLE 1

Eh, pH, AND pS^{2-} AT STATIONS IN GUNNAMATTA BAY (FIG. 4)

Date	Station	Eh (mV)	pH	pS^{2-}	Comments
29.v.67	1	+145	8.4	11.5	)
	2	+220	-	11.5	)
	3	+135	-	11.3	)
	4	+160	7.2	11.0	)
	5	+110	7.0	11.5	) Water depth of 11 metres
		+285	7.2	12.0	)
	6	+185	8.3	11.5	)
	7	+175	8.1	11.3	)
	8	+135	7.4	10.9	)
30.v.67	J	+405	8.4	7.5	Water sample
		+265	7.7	6.5	Sediment with Zostera growth
		+125	7.8	6.0	Sandy sediment only
	K	+335	7.7	7.3	Sandy bottom
	L	+325	8.2	8.0	Sandy bottom
	M	+265	7.8	7.3	Mangroves + Posidonia
		+345	7.8	8.0	Mangroves
	N	+425	8.2	8.2	Sandy + shells
		+405	7.8	11.0	Posidonia
	O	-245	7.2	3.8	Black mud - depth 11 metre H_2S smell
	P	+185	8.0	7.3	Sand
	Q	+235	7.6	8.0	Thick mud
	R	+275	7.6	7.5	Sandy-clay
	S	+115	7.8	8.0	Silty-sand
	T	+405	7.6	8.2	Yellow sand
	U	+ 80	7.8	8.2	Yellow-brown sand
	V	+395	8.0	8.8	Posidonia, fine silt
	W	+105	7.6	8.2	Black mud at sewer outlet
	X	+275	7.8	8.8	Grey sand
	Y	+ 55	7.4	8.2	Zostera bed
	Z*	+145	7.8	7.5	Silt with shells
	X^1	+ 45	8.0	8.2	Yellow-grey sand
	X^2	+365	8.0	8.2	Silt
	X^3	+160	7.8	8.0	Grey sand
	X^4	+425	7.8	12.0	Yellow sand
	X^5	+335	8.2	8.2	Yellow-grey sand
	X^6	+285	7.6	8.0	Grey sandy silt
	X^7	+285	8.0	8.8	Yellow-grey sand
	X^8	+235	7.0	2.6	Grey silty-mud - H_2S smell
	X^9	+135	7.8	8.0	Grey sand
	X^{10}	+375	7.4	10.2	Yellow-grey sand Mangroves
	X^{11}	+125	7.8	8.2	Edge of Posidonia - black silt
	X^{12}	+265	7.8	9.0	Posidonia
	X^{13}	-195	7.0	4.5	Black silty-mud - H_2S smell
	X^{14}	-215	7.4	3.5	Grey silt - H_2S smell
	X^{15}	-165	7.4	4.0	Black mud - H_2S smell
	X^{16}	+ 75	7.7	8.2	Grey sand

TABLE 1 (cont.)

Date	Station	Eh (mV)	pH	pS^{2-}	Comments
31.v.67	A 1	+455		12.5	Water
	A 1	+435		12.0	Sandy - shallow
	A 2	+445		12.5	Sandy - shallow
	A 3	+410		12.0	Grey sand Zostera
	A 4	+385		12.8	Grey sand
	A 5	+405		12.8	Grey-yellow sand - deep
	A 6	+365		11.3	Fringe of Posidonia - deep
	A 7	+155		12.8	Brown silty sand
	A 8	+160		5.0	Grey silty mud - H_2S smell
	A 9	- 65		5.0	Grey silty sand - H_2S smell
	A10	+335		10.0	Grey-yellow sand - shallow
	A11	+ 80		11.3	Grey-yellow sand Zostera
	A12	+340		11.3	Sandy edge of Zostera
	A13	+380		12.0	Amongst Zostera
	A14	+395		12.0	Grey sand
	A15	+ 60		11.0	Silty sand
	A16	+125		6.5	Grey silty mud - H_2S smell
	A17	+145		10.0	Grey silty mud
	A18	+380		10.0	Grey sandy silt
	A19	+400		12.5	Yellow sand
	A20	+ 70		11.3	Grey sandy silt
	A21	+180		10.3	Grey silty sand
	A22	- 55		5.0	Black grey sand - H_2S smell
	A23	+155		9.0	Grey sand
		+345		11.0	Sea water
	A24	+370		11.8	Zostera bed on sand bank
	A25	+365		10.8	Yellow sand
	A26	+305		6.8	Grey sand
	A27	+235		4.8	Grey sand Zostera
	A28	+ 5		6.8	Very grey sand Zostera
	A29	+ 35		4.8	Grey sand
	A30	+270		10.0	Grey sand
		+355		12.0	Sea water
	A31	+195		11.0	Grey sand Zostera

TABLE 2

Eh, pH, AND pS^{2-} OF DEEP CORE SAMPLES

Core No.	Compression Factor	Corrected Depth (cm)	Eh (mV)	pS^{2-}	pH
6	0.47	0	+ 35	10	7.4
		17	+175	8.5	7.6
		40	+ 95	10.2	7.6
		66	+165	10.5	7.6
		83	+ 75	8.5	7.8
		104	+ 45	10.5	7.8
		130	+175	11.5	7.8
		147	+215	12	8.0
7	0.48	15	+265	10.5	
		33	+125	9.5	
		56	+325	9.0	
		79	- 25	8.2	
		102	- 45	7.25	
		131	- 55	4.5	
		175	- 5	4.5	
		198	+ 25	7.5	
9	0.43	0	+155	11.8	7.6
		21	+105	11	7.6
		42	+105	12.2	7.8
		65	+245	12.2	7.8
		89	+155	11.5	7.8
		114	+245	12.2	7.6
		137	+145	10.2	7.8
		165	+ 45	9.5	7.8
10	0.44	16	+185	11	7.0
		48	+ 5	11.8	7.0
		66	+ 65	9	7.2
		100	+230	9	7.6
		114	+165	9.5	7.6
		127	+165	9.5	7.6
		143	+195	9.5	7.6
		162	+ 5	8.2	7.2
		184	+155	9	7.6

TABLE 2 (Cont.)

Core No.	Compression Factor	Corrected Depth (cm)	Eh (mV)	pS^{2-}	pH
12	0.44	0	+ 65	8.75	
		41	+135	9.8	
		64	+115	8.75	
		84	-155	4.0	
		111	-235	3.75	
		118	-165	3.5	
		141	-145	3.5	
		166	-175	3.0	
		180	-185	3.0	
		196	-185	3.0	
		221	-175	3.0	
		230	-120	3.75	

TABLE 3

Eh AND pS^{2-} OF SAMPLES TAKEN ON BEACH USING HAND-CORER

Core No.	Length (cm)	Sample Position (cm from top)	Eh	pS^{2-}
1	27	3	+235	12.5
		14	+310	12.7
		24	+295	12.3
Seawater			+325	12.0
2	15	3	+300	14.3
		12	+235	13.1
3	13	3	+280	13.8
		11	+300	13.2
4	16	2	+290	13.2
		9	+245	13.0
		14	+265	13.0
5	12	3	+295	13.2
		9	+295	13.1
6	18	3	+325	13.2
		10	+295	13.2
7	17	4	+380	12.1
		14	+290	13.2
8	12	3	+290	15.2
		10	+260	14.5
9	15	2	+295	14.8
		13	+285	14.5
10	16	3	+310	14.6
		13	+295	12.1
11	14	2	+325	14.2
		12	+260	13.7
12	16	2	+305	13.8
		12	+270	13.7

TABLE 3 (Cont.)

Core No.	Length (cm)	Sample Position (cm from top)	Eh	pS^{2-}
13	14	2	+325	12.1
		12	+295	13.7
14	11	2	+325	12.2
		10	+315	13.7
15	18	2	+275	13.2
		4	+275	13.7
		9	+265	13.5
		17	+315	12.1
16	15	2	+295	15.2
		14	+275	14.8
17				
		Electrodes in cover barrel	+275	12.7
	18	2	+325	14.8
		12	+275	14.5
18	16	2	+325	14.5
		11	+265	13.8
19	16	2	+325	14.8
		12	+295	14.5
Electrodes inserted directly into hole in sand			+355	
			+325	
20	16	2	+345	14.2
		14	+325	14.2
21	18	1	+335	12.1
		15	+305	13.8
22	16	12	+310	13.7
		3	+305	12.1
23	14	2	+295	14.2
		12	+285	13.8
24	15	2	+285	14.2
		12	+280	12.1

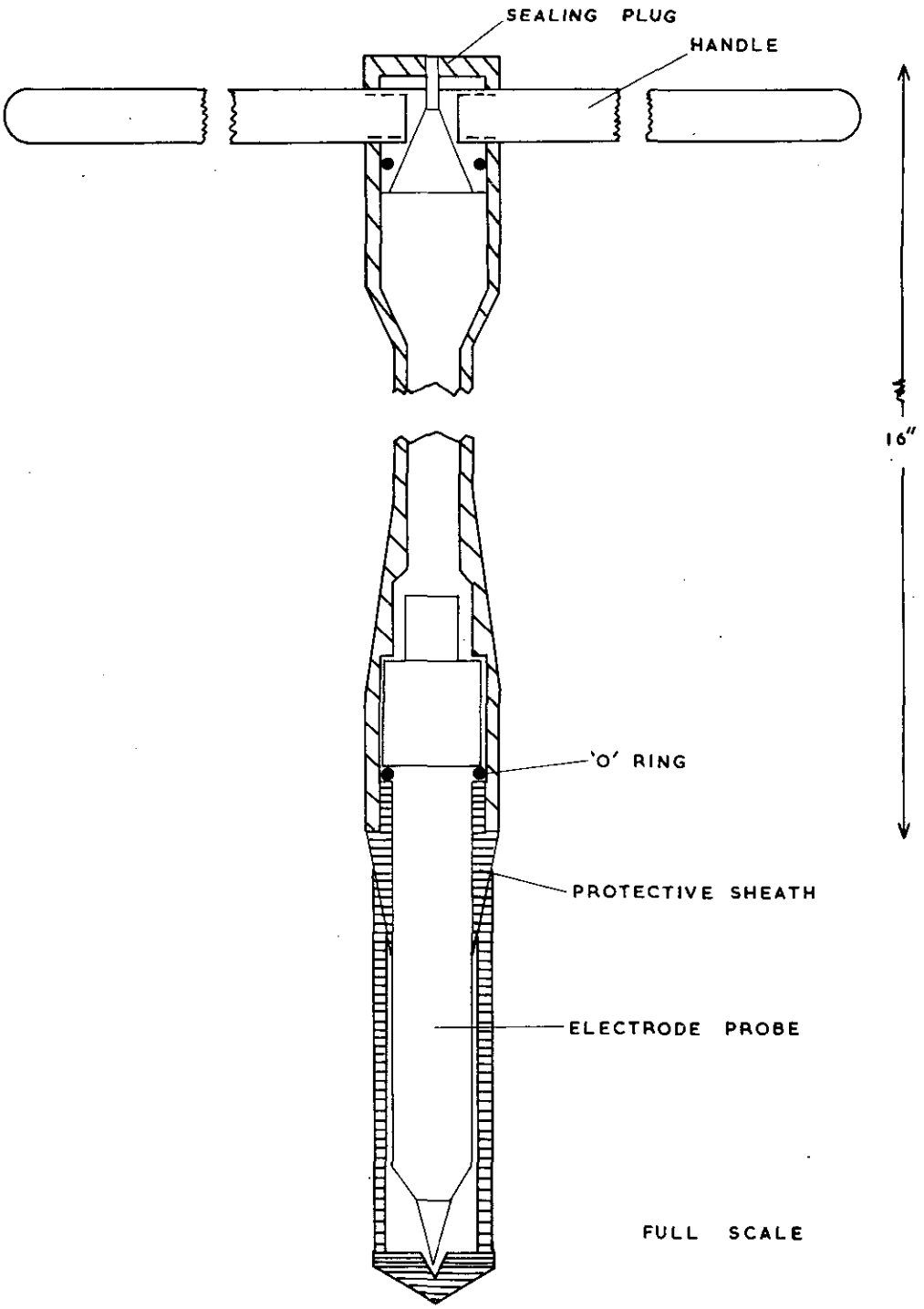

Fig. 1 - Probe casing for use on sandy flats.

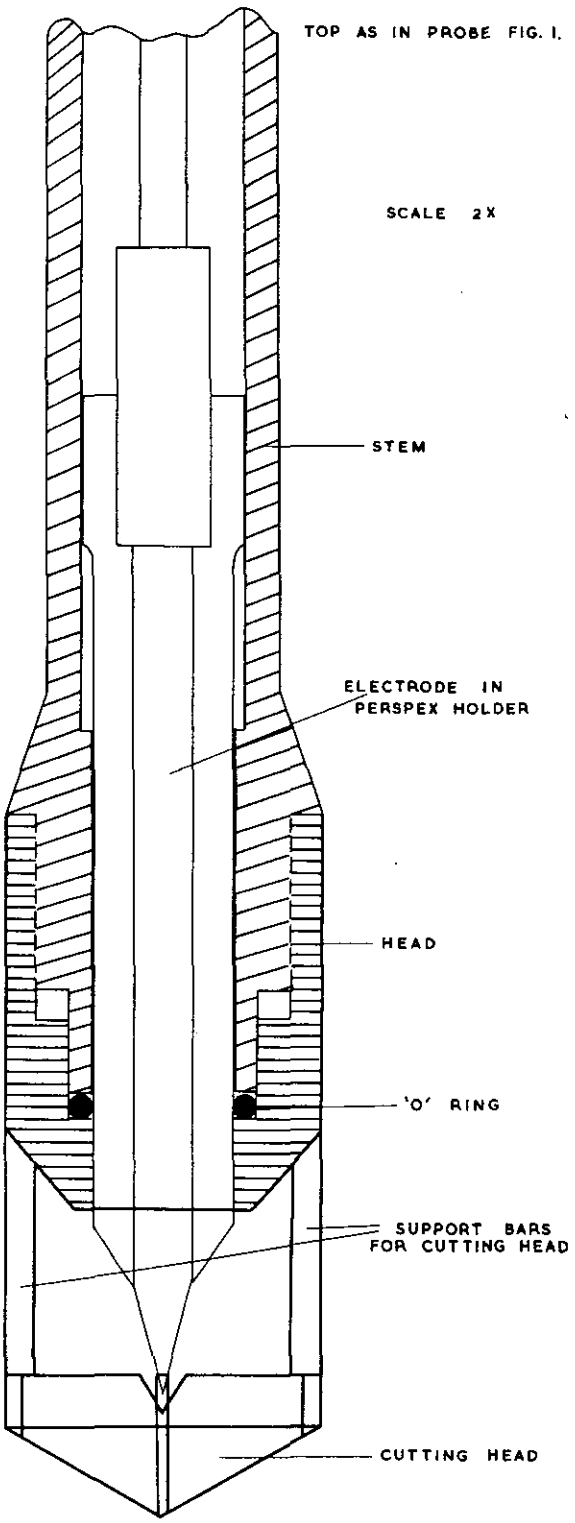

Fig. 2 - Probe casing for deep-water sediments.

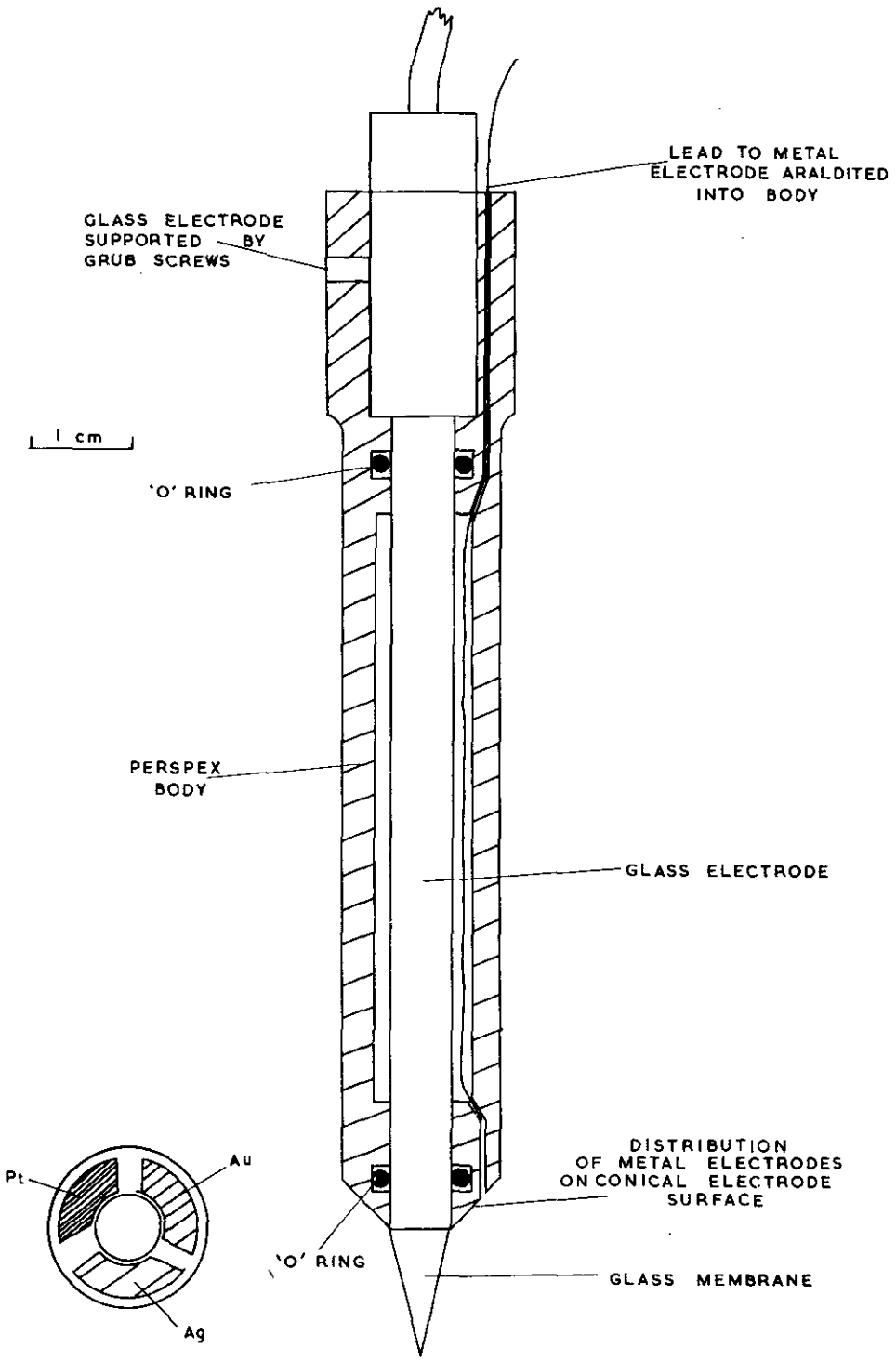

Fig. 3 - Composite electrode for use in either of the casings in Figures 1 and 2.

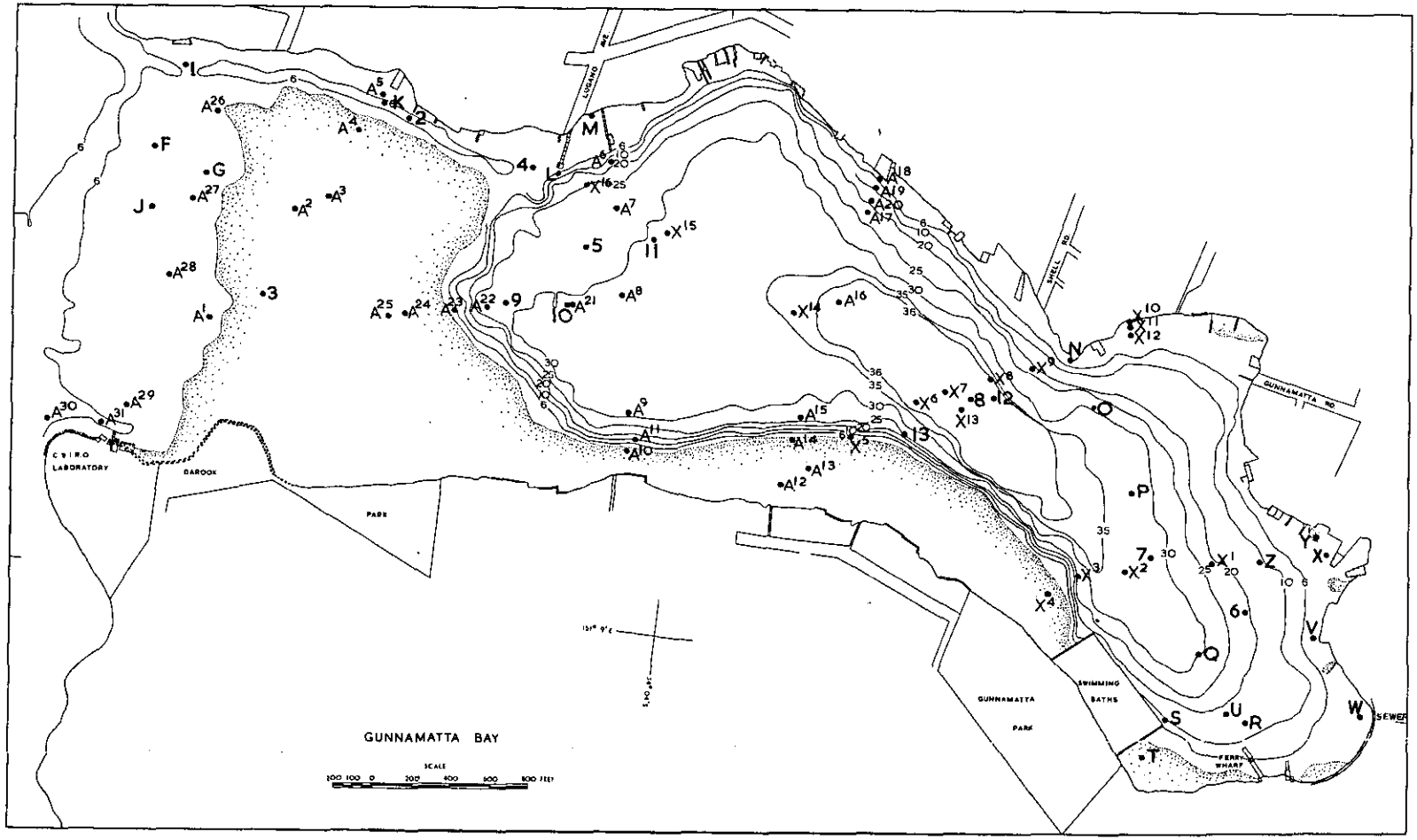

Fig. 4 - Sediment sampling sites with depth contours in feet.

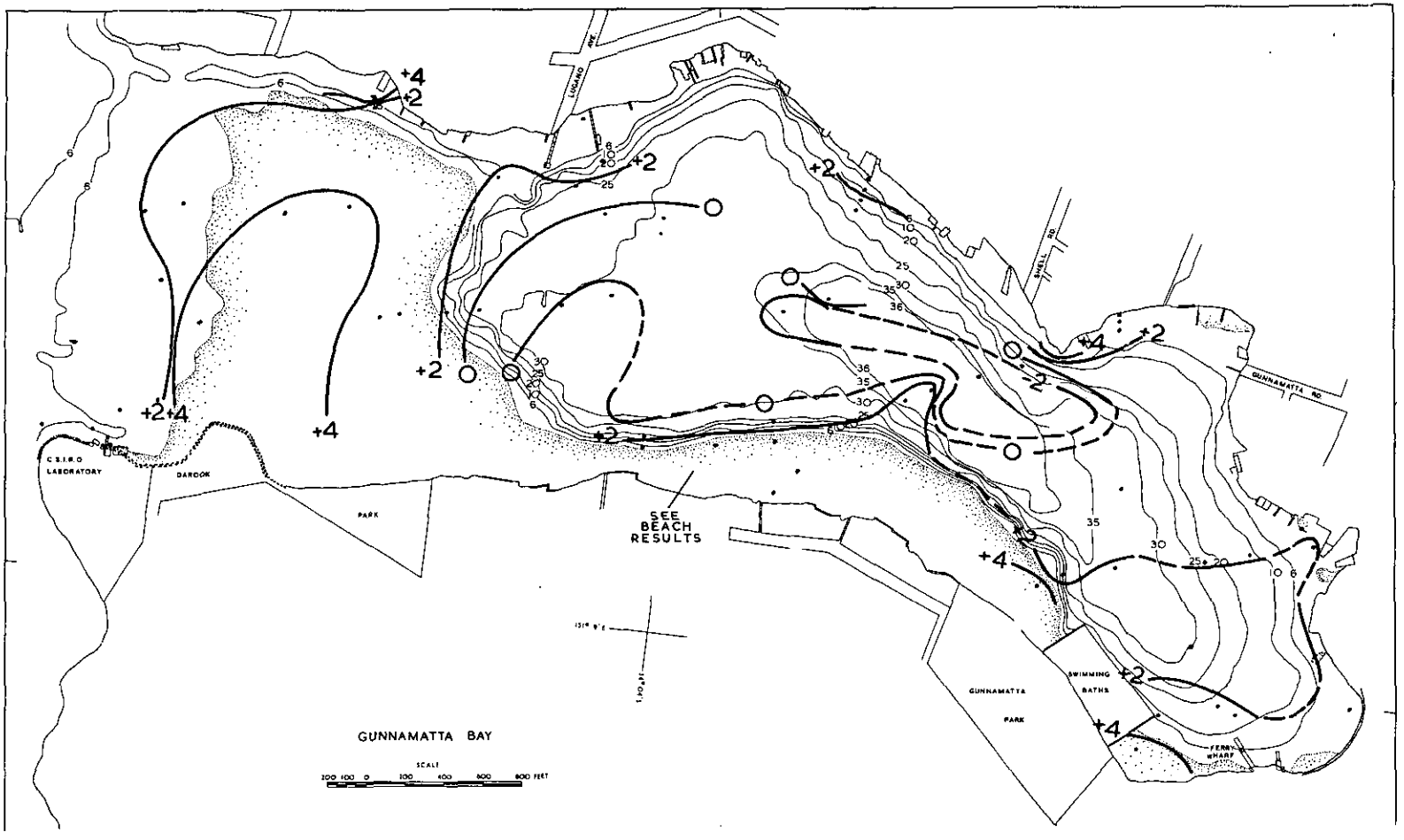

Fig. 5 - Contours (at 200 mV intervals) of Eh in Gunnamatta Bay. Depth contours in feet. -2 = -200 mV and over, 0 = 0 - -200 mV, +2 = 200-400 mV, +4 = 400 mV and over.

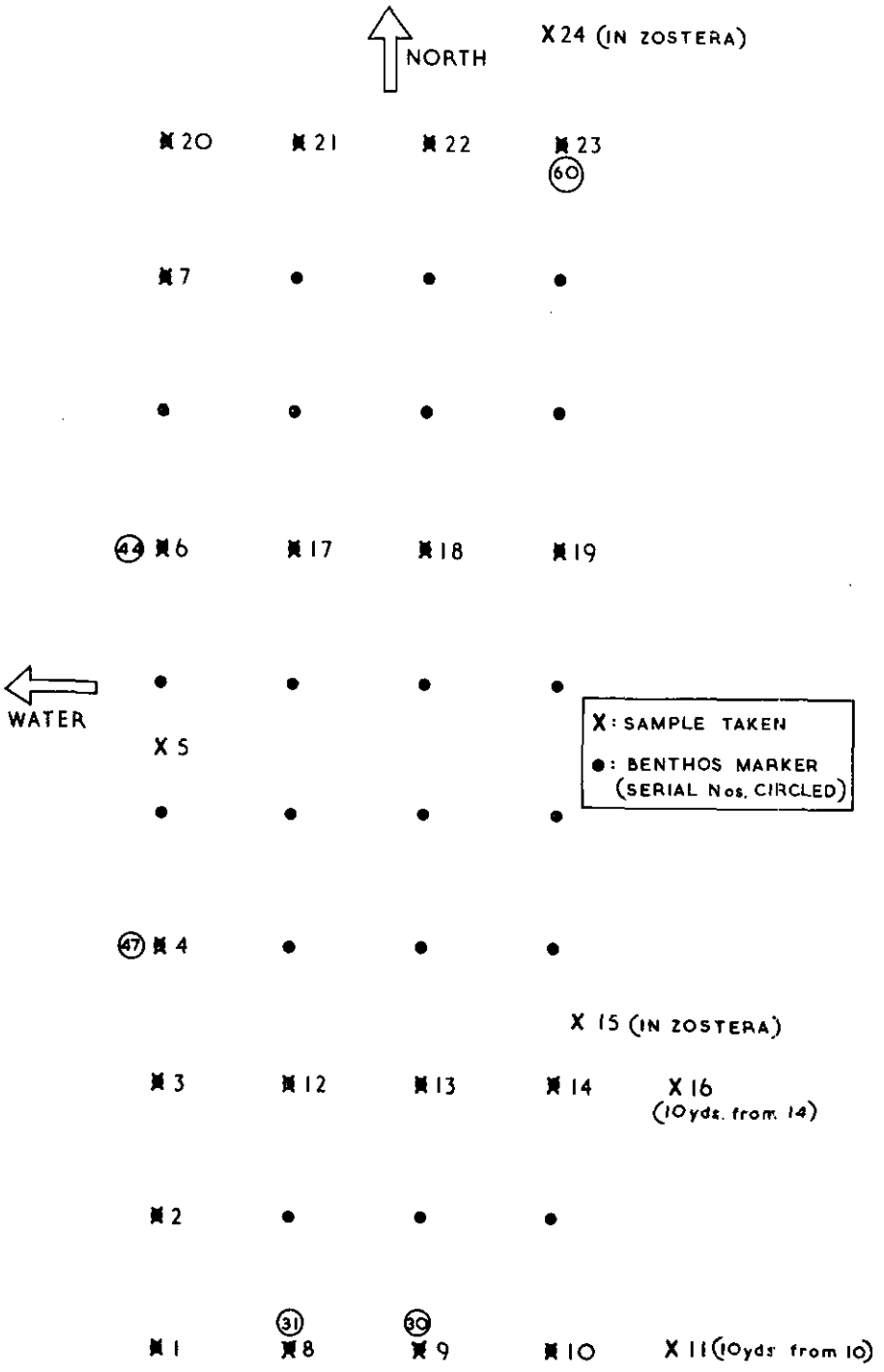

Fig. 6 - Sampling grid on 'Benthos Beach'.

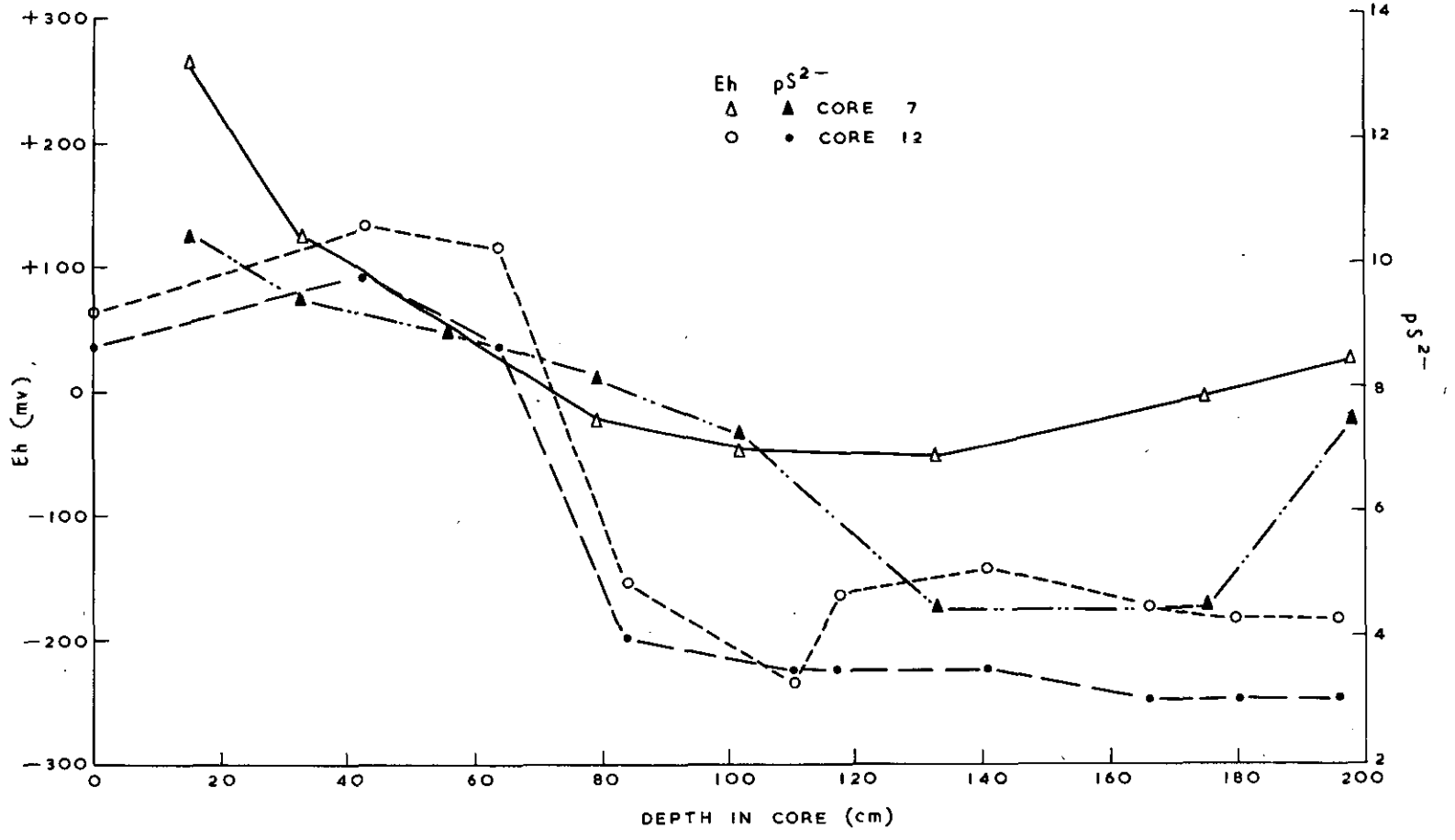

Fig. 7 - Eh and pS^{2-} changes with depth of core.

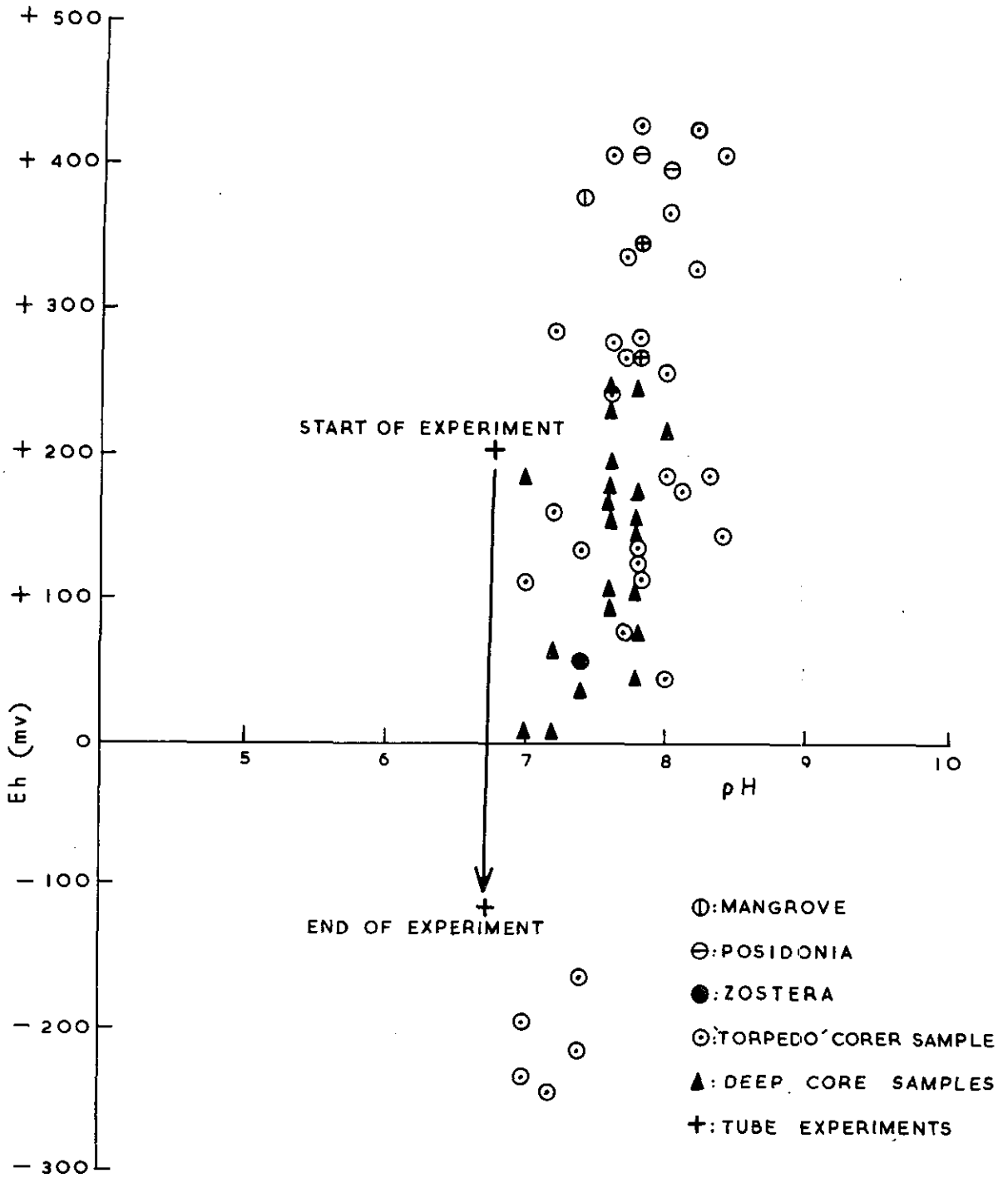

Fig. 8 - Sediment Eh/pH relations in Gunnamatta Bay.

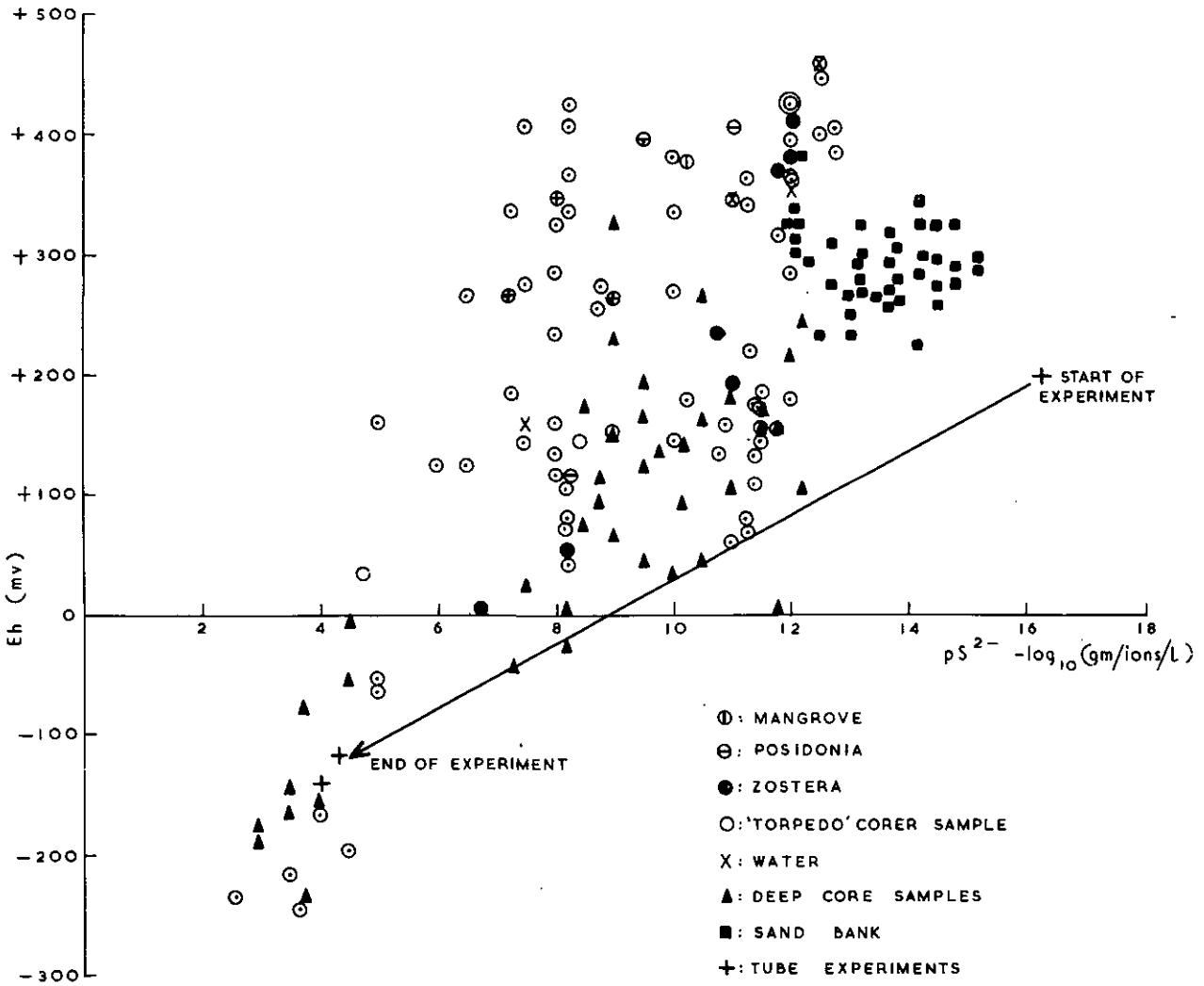

Fig. 9 - Sediment Eh/pS^{2-} relations in Gunnamatta Bay.

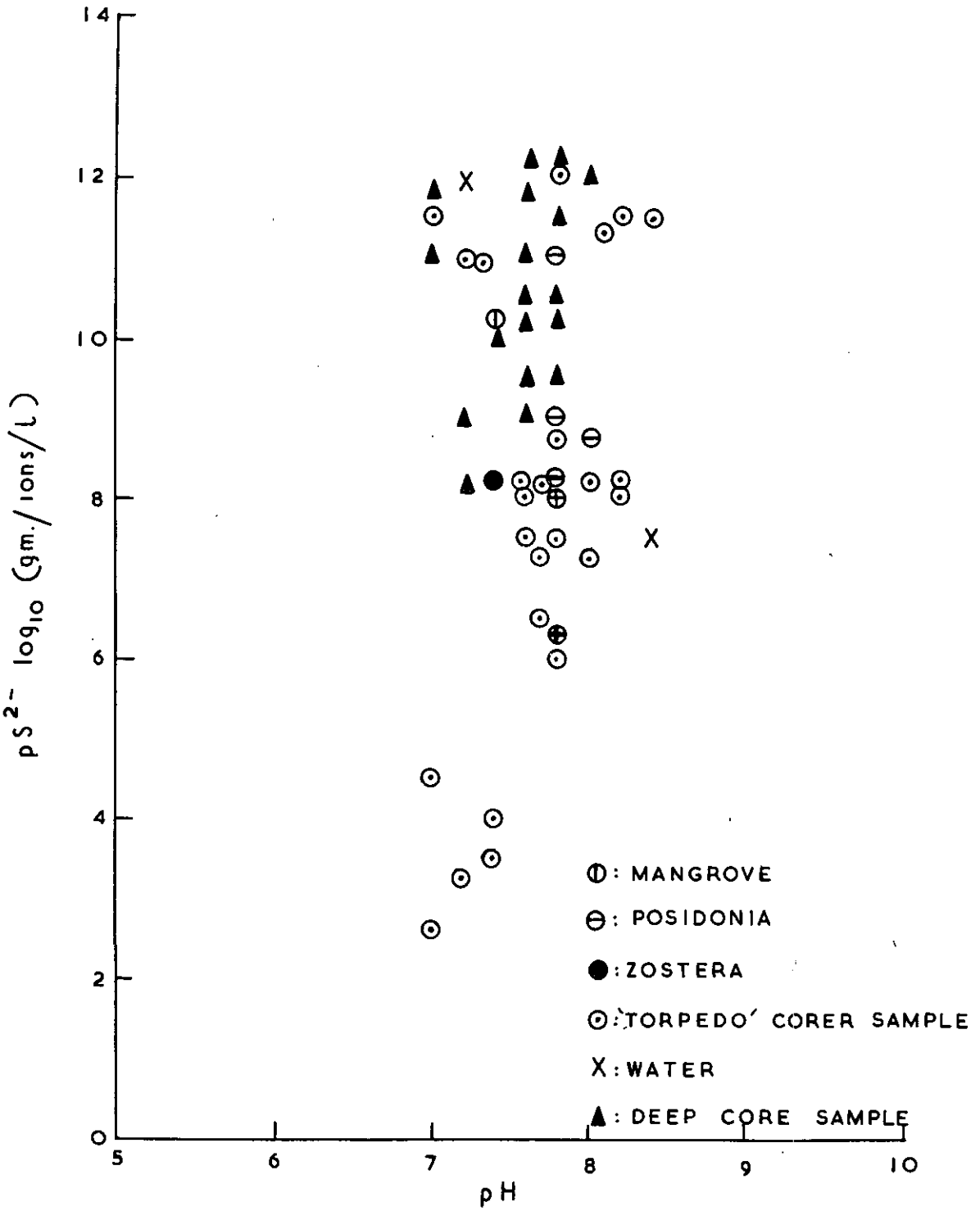

Fig. 10 - Sediment pS^{2-}/pH relations in Gunnamatta Bay.

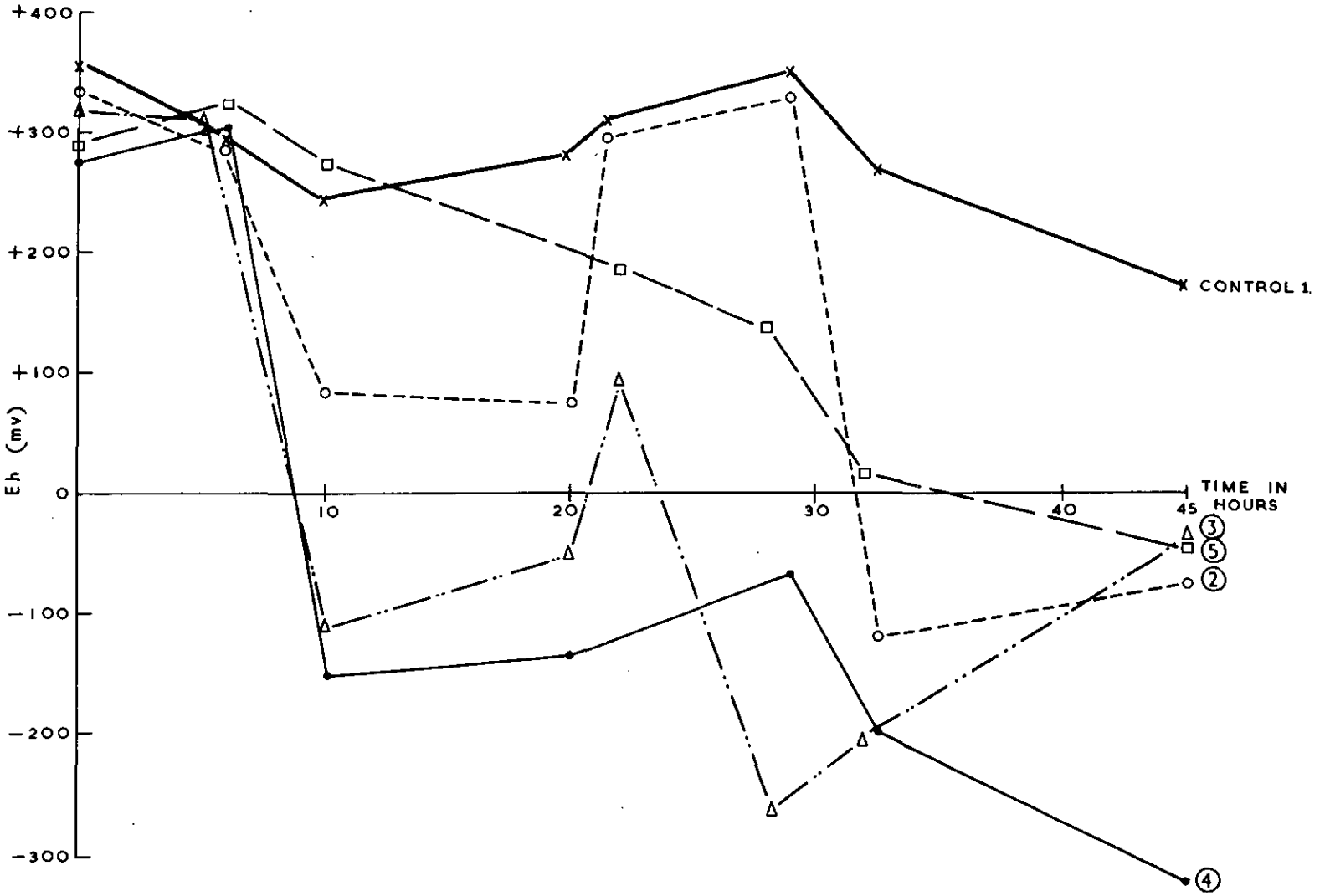

Fig. 11 - Changes in Eh with time in culture experiments with diced oyster as organic source.

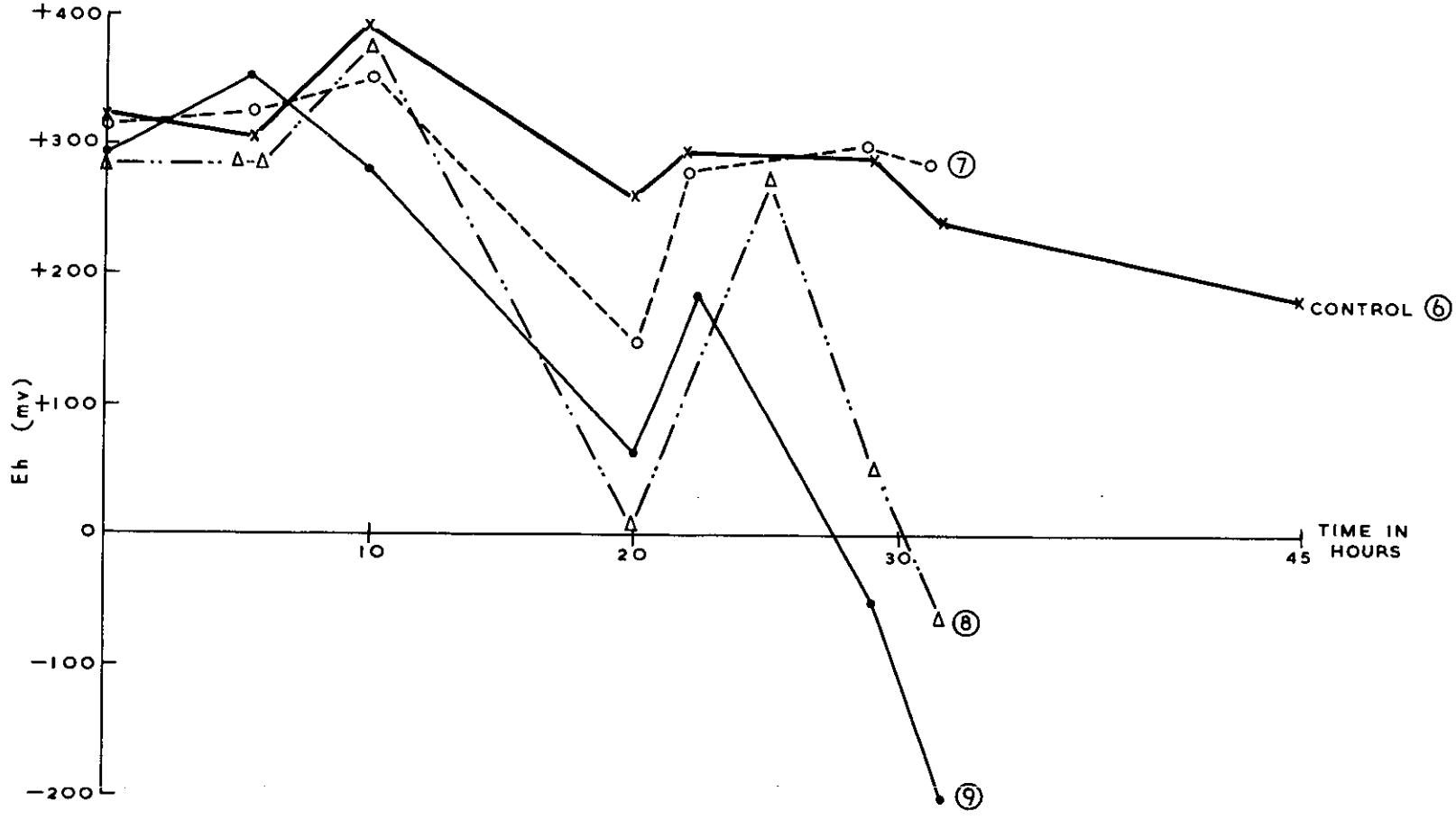

Fig. 12 - Changes in Eh with time in culture experiments with *Zostera* as organic source.

BENTHOS

I. INTRODUCTION

A general survey of Gunnamatta Bay to obtain a preliminary plan of the distribution of animals, plants, and substrates in the Bay was carried out. This survey was made in two parts by the entire group:-

(i) A general survey of the beach indicated by "a" in Figure 1. Data were computer analysed in an attempt to show patterns of distribution and relationships between the fauna on the sandy shore.

(ii) Observations were made of the floor of the Bay. Subsequently, dredges were made from the R.V. Saga in regions of differing substrate in order to correlate the organisms with their most favourable substrate.

Following this preliminary survey individual projects were carried out as indicated below. These were correlated with the sandy shore observations as far as possible.

(iii) Clam Distribution. The spatial and size distribution of Amesodesma angusta and A. cuneata were investigated in the upper littoral zone of the sandy beach near Darook Park (Fig. 1b).

(iv) Distribution of Conuber melastoma and Astropecten polycanthus on the Sandy Shore. The gastropod, Conuber melastoma, and asteroid, Astropecten polycanthus, are the two large predators of the sand flats. The project was initiated primarily to determine the distribution of each species, and secondarily to correlate this with their prey distribution.

(v) Rocky Shore. This project compared the distribution of organisms on an exposed and a sheltered part of the rocky shore of Gunnamatta Bay (Fig. 1d and e). The rocky shore as a whole was then briefly compared with the sandy shore.

(vi) Faunal Associations Inhabiting the Galeolaria and Trichomya Zones of the Rocky Shore in Gunnamatta Bay. This study was directed towards making a comparison of the animal community living amongst the living tube masses of the serpulid polychaete, Galeolaria caespitosa, with that living amongst the byssal hairs of the mussel, Trichomya hirsuta.

Galeolaria and Trichomya, by virtue of the fact that they grow in dense masses, provide a habitat for numerous smaller animals and plants. Whereas Galeolaria and Trichomya are primarily zoned

according to their respective desiccation tolerances, the associated communities are secondarily zoned in that they depend on the particular kind of microenvironment provided by the primary species in each zone. Hence, this study was an attempt to demonstrate a secondary zonation among sea-shore animals by comparing the two above-mentioned zones.

II. METHODS

(i) General Survey and Statistical Analysis

Sixty pegs, exactly ten meters apart, were laid on the beach in rows parallel to the low water level. At each station (peg) a core sample and quadrat sample were taken. The auger was 9 inches in diameter and the sample taken of 6-9 inch depth. The sample was strained through a sieve with pores 1.4 mm^2. The quadrats were 1 meter square in diameter and 2-3 inches deep. These were strained through a coarse sieve with pores 5 mm^2. All living fauna were collected.

The numbers of species and individuals of each species were counted for each sample. All the analyses were based on absence/presence of species at the stations. Numbers of individuals were used in interpretations of some of the analyses (Table 1).

Three types of analyses were made:

Normal Analysis

Here the stations were split up into groups according to the presence or absence of a particular species. This species was determined by the computer as being one which divided the large number of stations into a small number of groups of associated species. The final groups were mapped against the contours of the beach.

Inverse Analysis

This type of analysis used one computer-determined station to divide the total number of species into a number of associated groups. Similarities between these groups were put forward as an interpretation of the computer data.

Both methods 1 and 2 are "divisive analysis".

Aggregation of Like Groups

Both the core and the quadrat data were subjected to such an analysis by the computer. The computer selects cores with similar

DISTRIBUTION AND NUMBERS OF INDIVIDUALS ON SANDY SHORE

Species Occurrence

BLOCK NUMBER	A. angusta core quadrat	A. cuneata core quadrat	Clam 'C' core quadrat	Actaecea core quadrat	C. melastoma core quadrat	Hermit crab core quadrat	Polychaete 1 core quadrat	Polychaete 2 core quadrat	Polychaete 3 core quadrat	Polychaete 4 core quadrat	Polychaete 6 core quadrat	Polychaete 7 core quadrat	C. strangei core quadrat	Asteropecten core quadrat	Clam 'P' core quadrat	Amphipod core quadrat	Soldier crab core quadrat	Talorchestia core quadrat	Polychaete 9 core quadrat	Polychaete 12 core quadrat	Polychaete 14 core quadrat	Polychaete 19 core quadrat	Polychaete 5 core quadrat
1	13	144	3	228																			
2	28	287	4	31		8																	
3	4	18	4	114		20																	
4		1				2																	
5	1	3				1										1							
6	1	3	1			7																	
7	4	7	2	3		9	1																
8	6	3	3	6		5																	
9	1					2																	
10		1															3	1				1	
11		1				1			1								3	1					
12		1				1		1										1					
13	9	29		1														2					
14	5	9				1											4						
15	5	14	1	4		1				1							2						
16		5				1											1						
17					1												1						
18										1													
19							1																
20		4														3	3		1		4		
21			1	1													1						
22		1														2	5					1	
23		1																					

2700

TABLE 1 (Cont.)

Species Occurrence

BLOCK NUMBER	core quadrat A. angusta	core quadrat A. cuneata	core quadrat Clam 'C'	core quadrat Actaecea	core quadrat C. melastoma	core quadrat Hermit crab	core quadrat Polychaete 1	core quadrat Polychaete 2	core quadrat Polychaete 3	core quadrat Polychaete 4	core quadrat Polychaete 6	core quadrat Polychaete 7	core quadrat C. strangei	core quadrat Astropecten	core quadrat Clam 'P'	core quadrat Amphipod	core quadrat Soldier crab	core quadrat Talorchestia	core quadrat Polychaete 9	core quadrat Polychaete 12	core quadrat Polychaete 14	core quadrat Polychaete 19	core quadrat Polychaete 5
24							1									3	2	1				4	
25																							
26			1		1												1						
27					1												1						
28					1	1			1								2		1			1	
29						1										2	2						
30									1								9						
31							2			2			1				1						
32			1		1												1						
33					1																		
34									2		1					1							
35									1														
36					1						1					1							
37									3		2	1			1								
38						2			1										1				
39						1					1	1					4						
40						1			4														
41	1				2		2										1					4	
42								2														8	
43					1													3					
44	1				1													3					
45						2							1		1							1	
46			1						1		2							1	2	1			
47								1										7		1		1	
48																							
49						4	1																

Species Occurrence

BLOCK NUMBER	core quadrat A. angusta	core quadrat A. cuneata	core quadrat Clam 'C'	core quadrat Actaecea	core quadrat C. melastoma	core quadrat Hermit crab	core quadrat Polychaete 1	core quadrat Polychaete 2	core quadrat Polychaete 3	core quadrat Polychaete 4	core quadrat Polychaete 6	core quadrat Polychaete 7	core quadrat C. strangei	core quadrat Astropecten	core quadrat Clam 'F'	core quadrat Amphipod	core quadrat Soldier crab	core quadrat Paiorchestia	core quadrat Polychaete 9	core quadrat Polychaete 12	core quadrat Polychaete 14	core quadrat Polychaete 19	core quadrat Polychaete 5
50			1					1		3	1			1									
51						1						1 2 1					1						
52									1					1									
53					2	2		1		1				5	1								
54				1		1	4	2	3					4							1		2
55					1		20			1				1									
56							1							2									
57							2							1									
58																							
59																							
60																							

species compositions and groups them accordingly. The distance of each link between groups along the ordinate axis is a reflection of the diversity of the two groups under consideration. Similar selections are made for the quadrats.

Tide levels at the site of the general survey were measured by means of a tide staff which was erected at low tide on May 23. To determine the height of the zero on the tide staff relative to the zero datum of tide predictions, our measurements of tide heights were plotted together with corresponding predicted tide heights at Fort Denison, Sydney Harbour (Fig. 2).

(ii) Dredging

The bottom was observed through viewing glasses from a motor boat at shallow points in the Bay. The substrate type was noted and indicated on the map (Fig. 1).

This was followed by dredging from the R.V. Saga with a scallop dredge. Seven dredges were made. The first five are shown on the map; the sixth and seventh were made in the sandy bottom outside the Bay. These two were taken as representing the sandy area shown on the map by the broken dredge line (6 and 7).

(iii) Clam Distribution

A grid was established, lying across the upper and mid-littoral zones. A metre quadrat was placed on the N.W. side of each stake, and the sand was sampled to a depth of 2-3 inches and sieved through a coarse (gauge 5 mm^2) sieve. Contour lines were superimposed on the grid as shown by the water lines at fixed times; tide records were taken from a tide gauge (Fig. 2). Sand samples were collected at Stations 5, 12, 28, and 37 to determine water content.

(iv) Distribution of Conuber melastoma and Astropecten polycanthus

Each square of the quadrat was examined for signs of each species over two days. The total number was noted for each square at low tide on May 30 and 31, and a contour map drawn on the second day by following the level of the water at a known tide height (Fig. 4), the tide stick at Gunnamatta Bay being taken as a reference.

(v) Rocky Shore Survey

Transects were made at the two sites chosen. In the exposed region of the estuary, studies were made in two precipitous and one relatively flat region as well as in one crevice, while in the more sheltered inner part of the estuary one transect was made which was characteristic of the region.

All organisms were noted in these areas with special attention being paid to zone formation. Physical factors (temperature, salinity, pH, current flow, water suspensions, and the extent of oxidation or reduction of the area) were considered with respect to their effect on the abundance and distribution of the organisms.

(vi) Faunal Associations Inhabiting Galeolaria and Trichomya Zones of the Rocky Shore

Samples from each of the Galeolaria and Trichomya zones were taken at about the time of low tide (7.54 am) on May 30. At this time the water level was about 1 ft 5 in. which corresponded to the junction between the Trichomya zone and the Hormosira zone immediately above it. Dense growths of Galeolaria and Trichomya do not occur together in Gunnamatta Bay, so samples had to be taken from different points along the shore (Fig. 1). A bucketful of samples was taken from each zone and returned to the laboratory for analysis.

III. RESULTS AND DISCUSSION

(i) General Survey and Statistical Analysis

Normal Analysis

Table 1 is the raw data which were fed into the computer. In every analysis it was necessary to refer to this table to interpret the data.

Figure 4 shows the way in which the computer broke up the information presented to it. An actual graph of much the same type was received from the computer. All the numbers were then converted back to words to give more significance to the figure.

The groups of stations were then graphed against a contour map of the beach (Fig. 5).

Figure 6 shows the distribution of Group 2 on the contour map of the beach. Group 2 contained:-

Bivalve clam	C
Polychaetes	1, 2
Gastropod	Conuber melastoma
*Isopod	Actaecea sp.
Starfish	Astropecten sp.

Figure 8 refers to the distribution of Group 4 with reference to the contour map of the beach. Group 4 contained:-

Conuber melastoma
A hermit crab
Amesodesma angusta
Conuber strangei
Astropecten

All species in this group except A. angusta are usually found in wet or very wet areas. The presence of this clam in Group 4 can be accounted for in either of two ways. Firstly, since the sample is a quadrat the species could easily be washed down by the waves and still be on the top of the sand. The second theory is the more likely. This suggests that A. angusta can live in this wet area but usually is not found alive in it because this is an area of its predators. The most likely predators would be C. melastoma, C. strangei, or Astropecten. All are found in Group 4. Because all these predators need water the clam is safe in the higher parts of the beach.

Figure 8 indicates the distribution of Groups 6 and 7 with reference to the contour map. It must be noted that Group 7 were those quadrats that contained no species at all. Group 6 contained:-

Amesodesma angusta
A. cuncata
Clam C

The amesodesmas are found high up on the sand as a protection against predation. Clam C is found mainly in the mid-tide levels. The only clam C included in Group 6 was in quadrat 20. According to the contour map this was an area similar to the mid-tide level.

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* The only actaecean was found in quadrat 6. Isopods are usually terrestrial animals and are most likely to be found in the highest parts of the beach. All other species live in the lower mid-tide regions or even in the low-tide area. The only other species found in quadrat 6 was A. angusta. This clam is mostly distributed in the high-tide regions and does belong to Group 6 in the computer results. Therefore, Actaecea was taken from this Group and put in Group 6.

Figure 9 gives the same type of information as Figure 4. That is, it shows the way in which the computer has split up the mass of stations into a number of smaller associated ones. Figure 9 refers to the core samples taken from the beach.

Figure 10 gives the mapping of the stations according to their computer groups. This gives an overall picture of the distribution. By splitting up the groups and applying them to the contour map a better interpretation of the composite map is obtained.

Figure 11 shows the distribution of Group 12 in the contour map. Group 12 contained the species:-

Amesodesma angusta
Amesodesma cuneata
Actaecea, an isopod
Talorchestia, an amphipod

The distribution of this group follows the upper contours of the beach very well except for two stations, Nos 4 and 10. As shown in Figure 9 this group was determined by the presence of A. angusta. Station 4 contained one isopod only. Isopods are present in Group 12. Therefore this station can easily be considered as belonging to 12. Station 10 has no species at all. This also can be allied to 12. The geography of the area indicates it most likely does belong.

Figure 12 marks the distribution of Group 13; this seems to be confined to the mid-tide level, near or in run-off areas, and lower than mid-tide level. The group contained:-

An isopod	Actaecea
A gastropod	Conuber melastoma
Six polychaetes	1,3,4,6,7,14
A bivalve	Clam F
An amphipod	
A soldier crab	Mictyrus platycheles

All fauna in this group are mobile though mainly restricted to underwater habitats. There are two main types of feeding habits within the group. Firstly the detritus feeders - there may be scavengers like the crab or filter feeders like the clam. The second group of animals includes predators and their prey.

Figure 13 refers to the distribution of Groups 6 and 8.

Group 8 contained:-

An isopod	Actaecea
Two polychaetes	2,14
A soldier crab	Mictyrus platycheles
An amphipod	Talorchestia
A hermit crab	

Group 6 contained:-

An isopod	Actaecea
Five polychaetes	3,19,1,6,2
A soldier crab	Mictyrus platycheles
An amphipod	Talorchestia
An unidentified amphipod	
A hermit crab	

All these groups appear to have species with no specialised habitat. They are divided only by polychaete 3 which according to its wide distribution (see Table 1) is an errant polychaete.

Figure 14 refers to the distribution of Groups 2,4 and 10.

Group 2 contained:-

Conuber melastoma	
Six polychaetes	3,9,19,6,7,12
A soldier crab	Mictyrus platycheles
Clam C	
A hermit crab	

Group 4 contained:-

Clam C

Group 10 contained:-

A hermit crab	
A soldier crab	Mictyrus platycheles
Polychaete 6	
Clam F	

All these groups have much in common. Usually each species occurs in two out of the three areas. Except for the scavengers all species live under water.

For example, hermit crabs do not like being exposed. This indicates that Groups 10 and 2 are under water even at low tides. This is verified by the contour map.

From this distribution there are two types of clams, the Amesodesma spp., and clams C and F. The first group lives in the high parts of the water, the second group lives at mid-tide level. This second group is less abundant than the Amesodesma spp. A possible explanation would be the presence of predators such as C. melastoma in the mid-tide region.

Inverse Analysis

This second analysis produced groups of animals rather than stations that had common attributes.

Figures 15 and 16 refer to the methods of splitting of the animals as copied from the computer data. Table 2 shows the actual species present mapped against the computer number of the group. Compatibility between these is obvious.

TABLE 2

GROUPING OF SPECIES PRESENT IN QUADRATS AND CORES

Computer Group	Quadrat	Computer Group	Core
2	Amesodesma angusta	6	Amesodesma angusta
4	Amesodesma coneata	8	Amesodesma coneata
6	Conuber melastroma Polychaete 2 Astropectin	2	Astaecea Mictyrus platycheles (soldier crab)
7	Hermit crab Clam C Polychaetes 1,3,4,6,7, Conuber strangei	4	Polychaetes 3,19 Amphipod
	Astaecea		Clam C Conuber melastroma Conuber strangei Hermit crab Polychaetes 1,2,4,5, 6,7,9,12,14 Talorchestia

The core sampling showed that the clams A. angusta and A. cuneata were singled out. At first it appeared that these clams were distributed relatively equally. However, more detailed work on these showed that A. angusta exists in the higher parts of the beach and A. cuneata extends into the mid-tide levels. Also evidence is given that their vertical distribution is different.

The quadrat sampling also separates these two clams. This supports the theory of their mutually exclusive distribution.

Group 6 in the quadrat sampling can be justified. Conuber and Astropecten are both predators. In fact their prey is the same. It is found that although associated, Astropecten is confined to the low water levels, whereas Conuber is found in water but may be found at mid-tide levels. The polychaete in this group is always found in water.

Group 7 of the quadrat sampling can be readily aligned to Group 9 in the core. That is, it contains many polychaetes which all live in wet areas. Both groups also contain clam C. As mentioned before when discussing quadrat normal analysis (Fig. 14) clam C is limited to wet parts of the beach. Also, Figure 14 shows that hermit crabs are found in unexposed areas of the beach. Conuber is seen to be a predator of the mid-tide region (see first part of inverse analysis discussion). All these above points verify the association of both groups with the mid-tide wet areas.

The species Actaecea, however, does not seem to fit into Group 7 of the quadrat samples. Isopods are terrestrial animals and are seen in Table 1 to be distributed in the upper dry part of the beach. For this reason Actaecea is separated from Group 7.

The last two groups are limited to core samples only.

Group 2 contains two air breathing scavengers. This group can be aligned to the isolated isopod group of the quadrat sample.

Group 4 contains polychaetes 3 and 19. Reference to Table 1 shows that these polychaetes are the most widespread of all the polychaetes. The amphipod in this group is also fairly active and has a fairly wide distribution around the middle part of the beach.

These animals can therefore be considered active animals with a widespread distribution.

Aggregation of Like Groups

The outcome of these analyses is shown in Figures 17 and 18. In the aggregative treatment of the core data (Fig. 17) there are three distinct groups having different characteristics. The 115 group is linked with the 111 group to form Group 116. The characters of the two former groups are not as diverse as say those of Groups 114 and 117.

Theoretically, it should be possible to provide some sort of ecological explanation to account for the computer's groupings. Time has not permitted an exhaustive treatment of the results, but two examples, one from each of the quadrat and core data, have been worked out in an attempt to provide an ecological interpretation of the computer's treatment of these examples.

The first of these ecological interpretations applies to the core data (Fig. 17 and Table 3). Table 3 shows an analysis of the species composition of each of the computer Groups 117 and 114. The most significant feature of this analysis is that the clam species Amesodesma angusta and A. cuneata, and the isopod Actaecea sp. seem to be entirely confined to Group 117, whereas the majority of polychaete species appear confined to Group 114.

The distribution of quadrat groupings 117 and 114 relative to the survey grid is shown on the right-hand side of Table 3. Group 117 represents a relatively drier area of the grid than does Group 114. This distribution difference is reflected in the ecological preferences of the major species within each group.

It is interesting to note that the distribution of the soldier crab. Mictyrus platycheles, does not provide any basis for discrimination between the two computer groups (114 and 117). In spite of this it is clear that the differences in the area occupied by the clams and the isopod v. the polychaetes are sufficient to provide a basis for discrimination between the computer groups. This discrimination is also borne out by an examination of the distribution of tide levels and seepages within the area of the survey grid.

The four basal groupings of the core data were examined for any signs of correlation with the tide levels and the distribution of seepages within the grid, but apart from the fact the Group 100 is best distinguished from other groups along the abscissa by the fact that it represents the area of the grid furthest from the low water mark, no good correlations could be found.

A second ecological interpretation applies to the computer's discrimination between Groups 116 and 117 in the quadrat data (Table 4). An examination of the species composition of each group shows that Group 116 accounts for the hermit crab species and also for the starfish, Astropecten, whereas Group 117 accounts for the distribution of the two species of clam.

Again the distribution of the two groups relative to the survey grid is shown on the right-hand side of the table. Group 117 encompasses a drier region of the grid than does Group 116. As with the core data analysis this difference in surface water distribution is correlated with the ecological preferences of the species concerned; the clams prefer drier sand than do the hermit crabs and starfish.

From an overall point of view this method of aggregative computer analysis appears very promising as a tool in studying seashore zonation. The computer analysis provides a set of groupings made on the basis of similarities in species composition of the faunal

TABLE 3

CORE SAMPLE ANALYSES

COMPARISON OF GROUPINGS 114 AND 117

Group	Quadrat Number	A. angusta	A. cuneata	Clam C	Actaecea	C. melastoma	Hermit Crab	Polychaete 1	Polychaete 3	Polychaete 6	Polychaete 7	Polychaete 9	Polychaete 12	Polychaete 14	Polychaete 19	Soldier Crab
117	1	13	3		8											
	2	28	4		20											
	3	4	4		2											
	6	1	1		9											
	7	4	2													
	8	6	3		5											
	15	5	1		1											
	4				1											
	9	1			2											
	10															
	18															1
	19							1								1
	22															1
	25															
	26															1
	27															1
	32															1
	33															1
	39															
	51															
	52												2			
	53											1	1			
	54															
	59															
	60							1								
Group 114																
	28					1	1		1			1			1	2
	38						2		3	1		1			1	
	30								1							9
	35								1	1					8	
	40						1			1						4
	43								4							
	45													1		3
	48															7
	50									1						
	55								1					1		1
	57								3							
	46			1					1	2		2	1			1

TABLE 4

QUADRAT SAMPLE ANALYSES

COMPARISON OF GROUPINGS 116 AND 117

Group	Quadrat Number	A. angusta	A. cuneata	Clam C	Actaecea	Hermit Crab	Polychaete 3	Polychaete 4	Polychaete 6	Polychaete 7	Polychaete 1	Astropecten
116	32											
	33											
	41	1								1		
	44	1								1		
	60										2	1
	28					1						
	49					4						
	53					1						
	31											
	45					2						
	54											1
	57											4
	59											2
	56											5
	51			1								1
117												
	58	3			1			1				1
	6	1			1							
	20			1								
	38						1					
	40									1		
	52								1			
	1	144	228									
	2	287	31									
	3	18	114									

sample between the quadrats within the survey grid. These groupings may then be explained in terms of the ecology of the species concerned.

Other promising comparisons between groups which could have been made are: core data, Figure 17: 107 v. 100, 111 v. 115 and 118 v. 116; quadrat data, Figure 18: 114 v. 113, 95 v. 118, and 114 v. 115.

(ii) Dredging

	SAND (Clear)
Dredge 1	A few Crustacea Bryozoa
Dredge 2	Anadara trapezia plus Sargassum Astropecten vappa Scallop Conuber eggs
	MUD
Dredge 3	Echinocardium Two species of polychaete 1. Sandy tube 2. Maldarni sarsi Coralline algae Ascidians
Dredge 4	Mud swallow Maldarni sarsi Astropecten polycanthus Clam
	WEED + SAND
Dredge 5	Sea urchin Luidia Anadara Two Fortescue Bryozoa Tube case Aplysia egg mass Sabellid worm Ophiuroid Pipe fish Spider crabs Green leatherjacket

WEED + SAND

Dredge 6	Catfish
	Astropecten
	Aplysia
	Spider crabs
	Conuber eggs
	Sponge
	Echinoderms

WEED OFF SAND BEACH

Dredge 7	Two species of urchins
	Thalaciea
	Holothurian
	Kelp shells
	Clams
	Tubeworm case
	Crabs
	Polychaete worm
	Isopod

The Bay is divided roughly into areas of clear sandy bottom with weed such as Zostera growing in patches, muddy bottom in the middle, a sandy shore on the south side with weed further from shore and formed by inwash from the ocean, and a rocky shore opposite with patches of mud washed down by streams. Dredgings sampled each of these areas.

Anadara trapezia, often with Sargassum growing from its anterior lip, Astropecten vappa, Conuber eggs, and Bryozoa are characteristic of the clear sandy bottom of the channel dredged in 1 and 2. The occurrence of a scallop indicated the area is not polluted. Maldarni sarsi and Astropecten polyacanthus, two clam species, are typical of muddy substrates as in dredges 3 and 4. Echinocardium and the sandy tubeworm are characteristic of sandy substrates and probably were dredged from the sand before the mud was reached. The coralline algae and ascidian show there is some solid substrate such as bricks in the mud. There are no scallops as they are sensitive to oxygen concentration. Sea urchins, Luidia, Anadara, green leatherjackets, fortescues, Aplysia egg mass, pipe fish, crabs, and sabellid worms are characteristic of sand with mud areas. The results of dredges 6 and 7 are probably characteristic of the weed and sand of area 6 on the map.

(iii) Clam distribution

Total numbers of each species were plotted over the grid (Fig. 20) and stations with A. cuneata present were shaded. Percentage size distribution of both species at Station 13 was plotted (Fig. 21).

Size classes		
	1	5- 7 mm
	2	8-10 "
	3	11-13 "
	4	14-16 "
	5	17-19 "
	6	20-22 "

The distribution of both species along a transect line through Stations 9-16 was plotted (Fig. 22); size distribution of A. angusta and A. cuneata was plotted for three and two stations respectively down the beach (Figs. 23 and 24). The water content of the sand samples at 5, 12, 28, and 37 was plotted along with number of animals found at each station (Fig. 25).

A. angusta was described as yellowish, elongated, with the umbo close to the anterior end. A. cuneata is pinkish white, less elongated, with the umbo slightly off centre.

The grid of the area as in Figure 20 showed that A. angusta had a higher frequency and wider distribution; A. cuneata was present in a definite band as seen by the shaded area which is correlated to the contour of the land. This suggests that A. cuneata is more limited by this particular environment. While samples were being taken it could be seen that A. cuneata lay deeper in the sand than A. angusta.

The percentage size distribution at Station 4 (Fig. 21) shows that the modes of A. angusta and A. cuneata were in the 3rd (11-13 mm) and 4th (14-16 mm) classes respectively, i.e. the overall size of A. cuneata was larger.

The distribution along the transect line (Fig. 23) indicates the correlation between the number of individuals and the contour of the land. More individuals of both species occurred in the lower, wetter part.

Size distribution of A. angusta along the line, including Stations 13, 19, and 40 (Fig. 23), showed that the mode of the distribution decreased down the beach i.e. the animals further down the zone were smaller. Assuming that size is directly correlated to age, the younger animals were on the seaward side. This may be due to the settling of the veliger larvae in the damper part of the zone. The predator pressure of Conuber and Astropecten on the lower part of the Amesodesma zone may be concentrated on the larger animals, causing an apparent high density of smaller animals. Figure 24 shows a similar situation for A. cuneata. Although Stations 11 and 13 are on the same north-south transect, Station 11 is actually lower because of the contour of the land (Figs. 19 and 20).

The percentage of water in the sand samples (Fig. 25) showed a sharp increase at Station 12 which was directly correlated with the contour of the zone. The largest number of individuals was found at the steep part of the zone.

An interesting point for further investigation is how the population is affected by the sampling and how long it takes to recover afterwards.

(iv) Distribution of Conuber melastoma and Astropecten polycanthus

Figure 26 shows the number of both species in each 10 m square.

Feeding experiments were conducted to determine if either or both would eat the small bivalves Amesodesma angusta and A. cuneata. None of these was eaten, but this means little, and many drilled shells were seen at the sampling points in the initial survey.

The period of activity of Conuber was at low tide as long trails, up to 10 ft, can be seen on the tide flats. The incoming tide washes these away, so obviously they are made at low tide, and observations suggest they are inactive at high tide.

Astropecten is inactive at low tide, and a brief search at high tide could show none active.

Figure 26 indicates that the distribution of Conuber is limited by the degree of dryness of the sand. In the upper point of their distribution they are found below the 4 ft contour line, which marks distinctly the boundary of the wet and dry sand.

Density of population increases as the sand increases in wetness. This may be due to risk of desiccation in dry sand, as many molluscs are limited in distribution by this; or their distribution may reflect that of their prey.

Figure 27 shows the overlap of distribution of Amesodesma angusta, A. cuneata, and Conuber melastoma. Many drilled Amesodesma shells were seen, but Conuber did not feed in the laboratory. The results of the general survey indicate that the predator of these bivalves must have been Conuber (by a process of elimination). Those drilled Amesodesma shells found outside the Conuber distribution would probably have been washed up from the regions in which Amesodesma and Conuber overlapped.

The distribution of Astropecten polycanthus seems restricted to the lowest level of the littoral zone (Fig. 28) and it seems that they are more common sub-littorally. Their period of feeding was not determined; they were inactive at low tide, and probably at high tide. This suggests they feed at night during high or low tide, but this was not verified.

(v) Rocky Shore Survey

The rocky shore localities studied in the estuary are shown in Table 1. The results of the transects are shown in Figures 29, 30, and 31 in which the indicator species for each zone are shown. Table 5 shows all additional species found in each zone in the transect areas. Table 6 gives the physical conditions considered in the two localities.

TABLE 5

ADDITIONAL SPECIES FOUND IN THE VARIOUS ZONES OF EACH TRANSECT AREA

	Exposed Area in Estuary						Mangrove Area	Comments
	Precipice	Comments	Crevice	Comments	Flat Rock	Comments		
Ecklonia	Unidentified brown alga	Abundant	No Ecklonia zone		Unidentified alga	Abundant	No Ecklonia zone	
	Colonial ascidian (Family Polyclinidae? Synoicium sp.)	Very numerous						
	Pyura pachydermatina	Very few						
Algal (Subdivided into: (a) fucoid (b) brown encrusting (c) coralline in the precipice transect)	Balanus	Isolated	Balanus	Only 1 seen	Balanus	Isolated	Zone was very small and too far below water level for any additional species to be identified.	
	Galeolaria	Sparse	Sponge	1 unidentified species which was fairly frequent	Dicathais	One small specimen		
	Chamaesipho	A few small groups	Dicathais	several; wedged right into the crevice	Galeolaria	A few small patches		
			Barnacles covered with coralline alga	Unidentified due to alga	Hormosira	In a small depression		
			Notoacmaea	Few				
			Anemone	Two of 1 species unidentified				

TABLE 5 (Cont.)

Zone	Exposed Area in Estuary						Mangrove Area	Comments
	Precipice	Comments	Crevice	Comments	Flat Rock	Comments		
Galeolaria	Oysters	Few isolated specimens	1st zone: below oysters		Few additional species		Small band 3" wide with few additional species	
	Austrocochlear		oysters	Few				
	Bembicium nanum		barnacles					
	Dicathais		Cellana					
			Dicathais					
			2nd zone: above oysters					
			Notoacmaea					
			Oysters					
			Austrocochlear					
			Bembicium nanum					
Oysters	Morula	Very many on the oysters; bore through shells	Morula		Zone small with few additional species		Oysters spread back from just below water level for about 8 yards into mangrove mud. Some were also on the mangrove pneumatophores.	
	Bembicium nanum		Cellana				Bembicium auratum	in mud
	Notoacmaea	On oysters; browse on algae on shells	Notoacmaea				Bembicium nanum	on rock
	Barnacles (Chamaesipho, Tetraclita)	A few appear towards the top of the zone	Austrocochlear				Austrocochlear	
							Morula	many, in clumps
							Hormosira	in pools

TABLE 5 (Cont.)

Zone	Exposed Area in Estuary						Mangrove Area	Comments
	Precipice	Comments	Crevice	Comments	Flat Rock	Comments		
Oysters	Chiton	1 only, in a sheltered area; small in size					Galeolaria	Scattered
							Melanerita	
	Cellana	At upper limit of the zone					Lasea australis	Found in association with the oysters
							Velacumantus australis	In mud
							Chitons	Very small and few; in rock pools
							Cellana	Few
							Patiriella exigua	Near rock pool
Melaraphe	Tetraclita, Chamaesipho, Catophragmus	Scattered	Dicathais	One, had wandered from usual area between oysters and Galeolaria	Cellana, Notoacmaea, Dicathais	Covered rocks sparsely	No Melaraphe zone	
	Cellana				Barnacles	1 small patch only consisting most of catophragmus		
	Melanerita	Few isolated and in groups	Morula	Groups where rock very moist				
			Barnacles encrusted with brown algae	Unidentified; only a few				
			Austrocochlear					
			Cellana					
			Melanerita	Only 1 which				

TABLE 6

PHYSICAL FACTORS IN THE EXPOSED AND SHELTERED AREAS OF THE ROCKY SHORE

Variable	Exposed Area	Sheltered Area
Temperature	15-16°C	15-16°C
Chlorinity (‰)	19.54	19.59
pH	7.7	7.8
Current flow and waves	Strong currents with large tidal fluctuation and waves	Little current or wave action
Suspended matter	Water clear with little suspended matter	Suspended mud and other material
Eh	+335 mV	+265-345 mV

The main differences between the exposed and sheltered areas are the presence of distinct Galeolaria and Ecklonia zones in the former and their absence in the latter. Ecklonia tends to be found in areas where there is considerable wave action even although it is usually below low tide level. Its absence in the sheltered area could be due either to an unfavourable environment or to competition with another weed for the rocky environment, or to both. With regard to competition, Sargassum v. Ecklonia is the most probable suggestion as both these weeds live attached to rock below low tide level.

One explanation for the absence of Galeolaria in the sheltered area is the absence of spray. Another is the possible competition between Galeolaria and oysters, the latter being very abundant in sheltered areas and less so in more exposed regions.

On the sea shore (not included in this study), there is a distinct barnacle zone above that of Galeolaria, while the number of oysters is small. On the exposed part of the estuary, oysters form a distinct zone above the Galeolaria, while barnacles only occur as small groups on isolated individuals. In the sheltered estuarine area, no barnacles were observed, and the number of oysters was further increased. One possible explanation for these differences is competition for space between the oysters and barnacles. Also, physical factors may affect the distribution of organisms, as different organisms have different tolerance to environmental variations.

TABLE 7

PRINCIPAL COMPONENTS OF THE FAUNAL ASSOCIATIONS IN THE BYSSAL HAIRS OF THE MUSSEL, TRICHOMYA HIRSUTA

Phylum	Species	Frequency*
NEMATODA	One unidentified species	VC
ANNELIDA	Siponculid	F
	Galeolancer caespitosa (tubes)	R
	Polychaetes - polynoid species	VC
	nereid "	VC
	glycerid "	VC
ARTHROPODA (Crustacea)	Barnacle, sp. A	R
	Corophium sp. - burrowing amphipod	F
	Amphipod, sp. A	R
	Anomura - small hermit crab inhabiting shells of the gastropod Velecumantis australis	C
	Brachyurans - Pilumnis rutopunctatus	C
	Halicarcinus rostratus	R
(Insecta)	Collembola sp.	C
MOLLUSCA	One erycinid clam species	F
ECHINODERMATA	One asteroid	R

* VC : very common
C : common
F : few
R : rare

TABLE 8

PRINCIPAL COMPONENTS OF THE FAUNAL ASSOCIATION AMONGST THE MASSES OF THE SERPULID POLYCHAETE GALEOLARIA CAESPITOSA

Phylum	Species	Frequency
PORIFERA	Fragments of a small yellow sponge	F
BRYOZOA	Watersipora cucullata	R
PLATYHELMINTHES	Polyclad	R
ANNELIDA	Polynoid polychaete	C
	Siponculid	R
MOLLUSCA	Chiton (Mesoplacophora)	C
	Bivalves: Crassostrea commercialis	F
	Erycinid clam, Lasea australis	F
	Trichomya hirsuta	F
	Mytilus sp.	R
	Gastropoda: Limpet, Montfortula conoidea	F
Crustacea	Barnacles: Tetraclita purpurescens	F
	Ibla quadrivalis (stalked)	R
	Brachyura: Eriocheir spinosus	R
	Pachygrapsus transversus	R

Bembicium nanum is the normal rock dwelling species of Bembicium. The mud in the mangrove area contained another species, B. auratum, living in close proximity with B. nanum. Velacumantus australis, a whelk, also appears with the introduction of mangrove mud- it was not found in any other areas studied.

From Table 6 it is apparent that the main factors which are likely to affect the abundance and distribution of the organisms are currents, wave action, and water suspension. The other physical factors considered were not variable in the two areas studied.

It is obvious that the rocky shore differs from the sandy shore in that the latter does not show apparent zonation. It was necessary to utilise the information from a computer to determine whether or not there was any type of pattern of the organisms on the sandy shore at all, whereas the rocky shore showed visible zonation.

(vi) Faunal Associations Inhabiting Galeolaria and Trichomya Zones of the Rocky Shore

The composition of the faunal communities in each zone is shown in Tables 7 and 8.

The study was not made quantitative because of a lack of time. References to the abundance of species are therefore in relative terms.

At the outset it is clear that certain of the species listed in the results tables are intruders from adjacent zones. This accounts for the presence of a few Trichomya in the Galeolaria zone and vice versa.

Few species were present in sufficient numbers to indicate a natural distribution which includes both the Galeolaria and Trichomya zones. This appears to apply to the species of polynoid polychaete.

Taking into consideration instrusions from adjacent zones the Galeolaria and Trichomya appear to be characterised by the following faunal associations:

Galeolaria association:

Bryozoan, Watersipora cucullata
Crab, Pachygrapsus transversus
Polynoid polychaete
Chiton (undetermined species)
Erycinid clam, Lasea australis

Trichomya association:

Nematode (undetermined species)
Polynoid, nereid, and glycerid species of polychaete
Collembolla
Crab, Pilumnus rufopunctatus
Hermit crabs inhabiting discarded shell of Velecumantis australis

IV. CONCLUSIONS

The results suggest that:-

(i) Methods of statistical analysis can prove very useful in determining patterns and their significance in faunal distribution in seemingly unzoned areas.

(ii) Dredging showed that there was distinct variation in the organisms found, this variation being the result of variations in the nature of the substrate.

(iii) A. angusta was more widely distributed than A. cuneata. More individuals of A. angusta were present. Smaller individuals of both species were found on the seaward side of the zone. Number of individuals of both species was correlated to the percentage of water in the sand.

(iv) Methods of study in this project were shown to be inadequate. In future it is suggested that all experiments be done by marking specimens in the field and observing their movements and feeding habits in their natural environment.

(v) Zoning in the estuary showed variations depending on the degree of exposure of the locality to wind, currents, and wave action.

(vi) Variations were seen in the faunal associations inhabiting the Galeolaria and Trichomya zones on the rocky shore.

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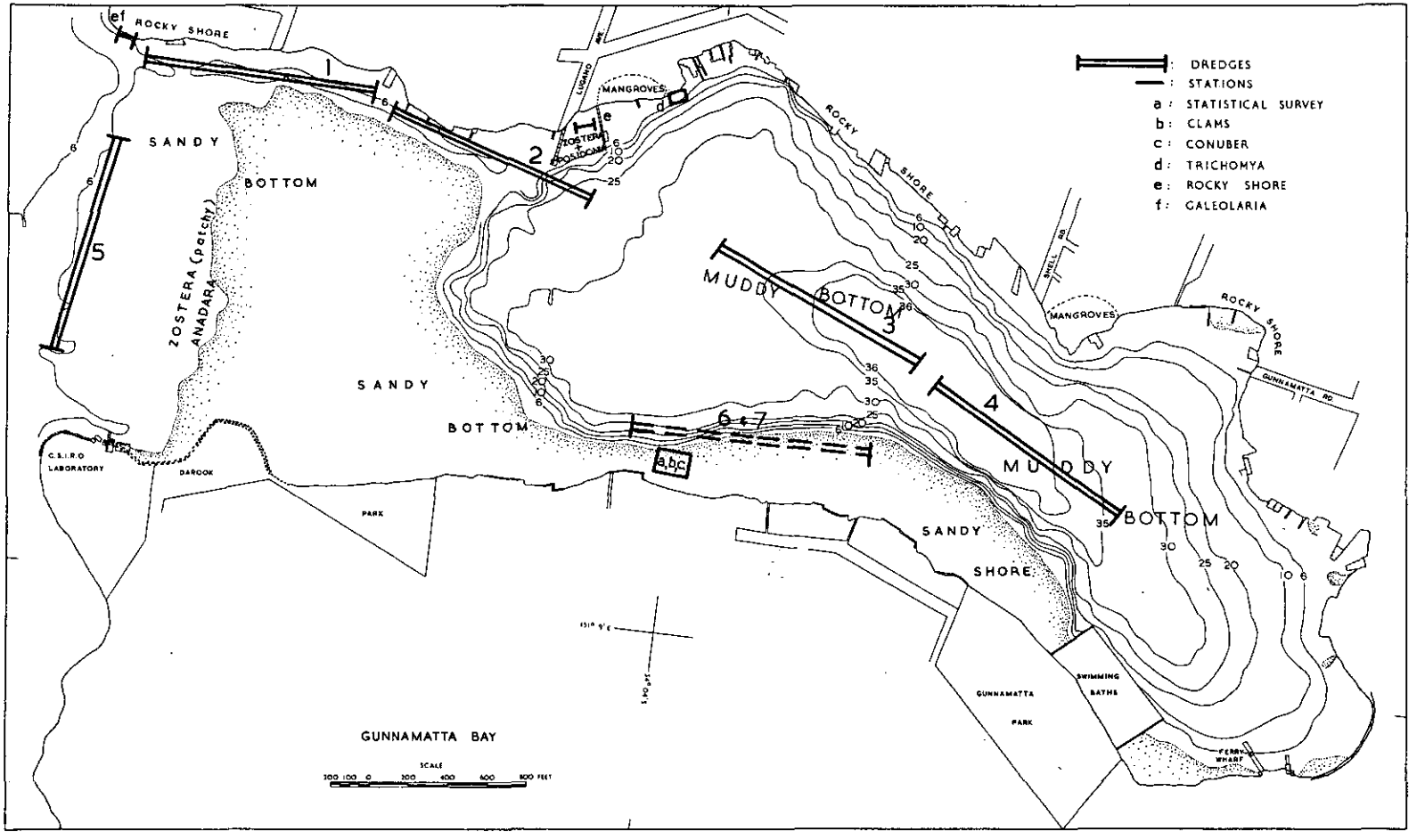

Fig. 1 - Locations of benthic sampling areas in Gunnamatta Bay.

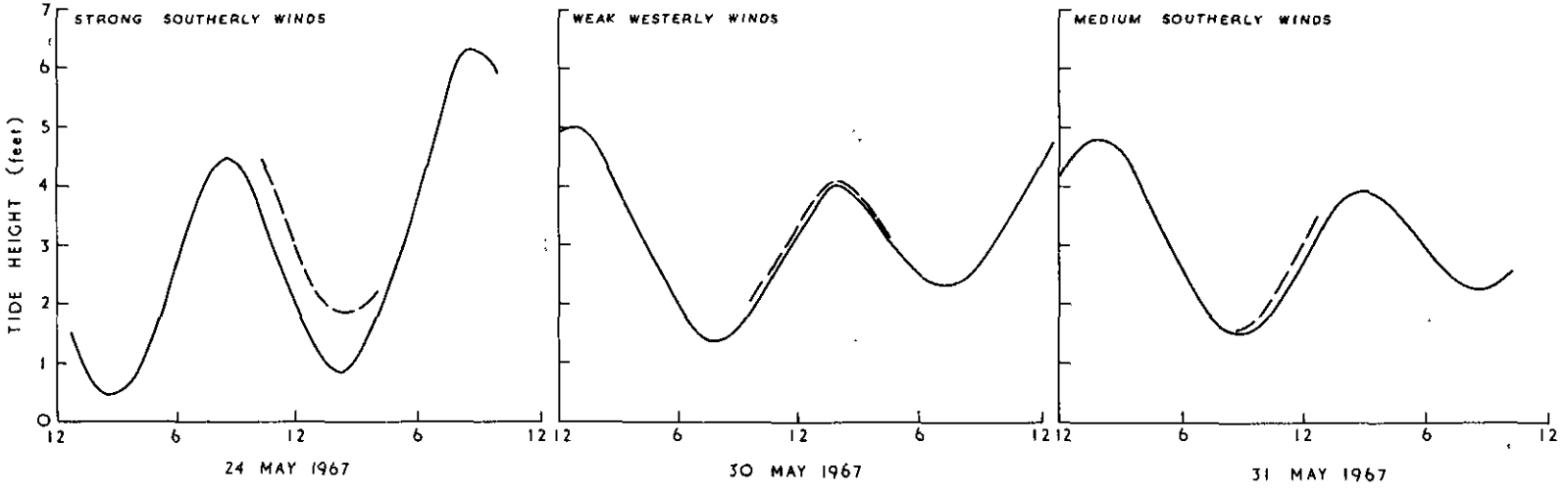

Fig. 2 - Predicted Fort Denison tides (solid line) and observed Gunnamatta Bay tides (broken line), together with wind state, on May 24, 30, and 31, 1967.

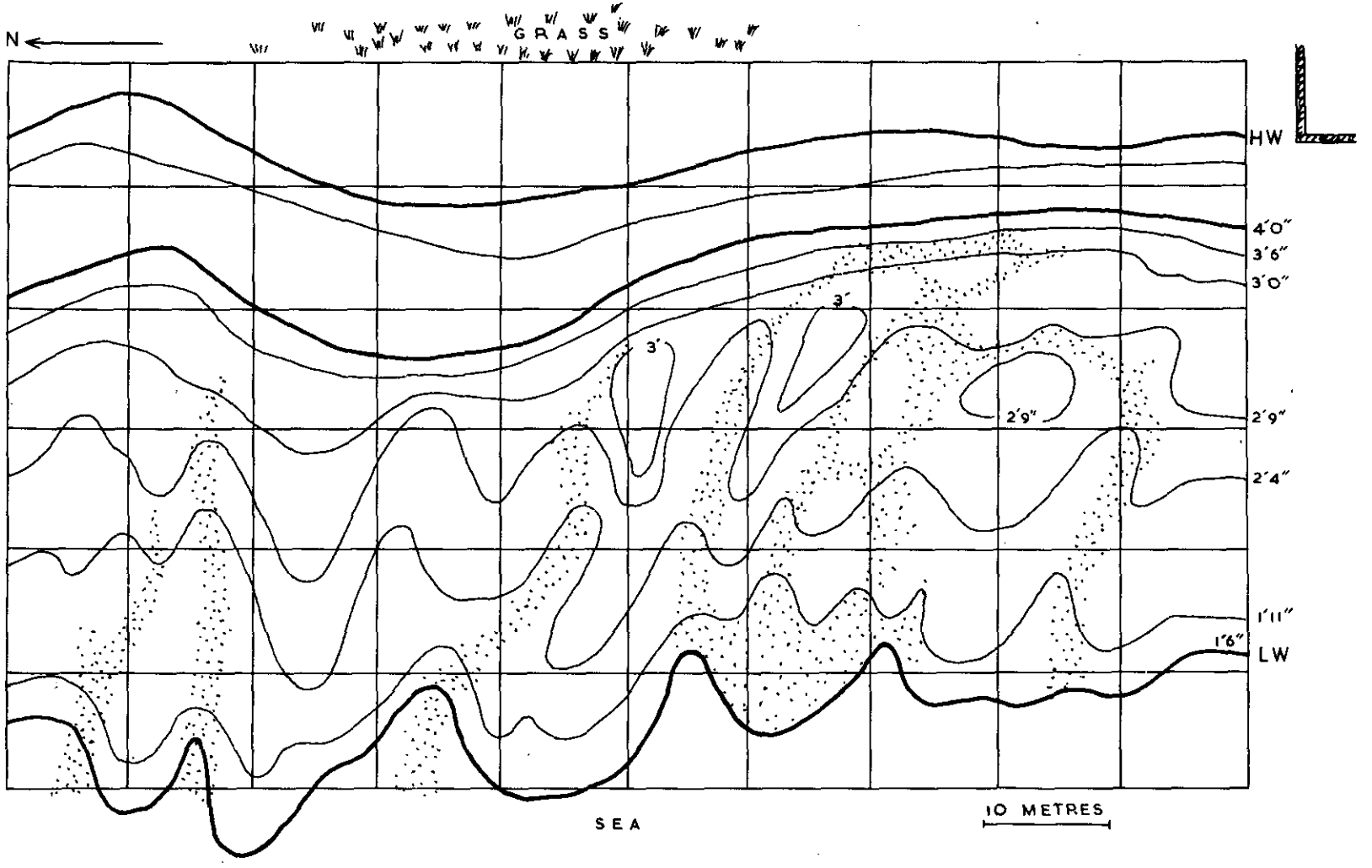

Fig. 3 - Sampling area and grid on the sandy shore (see Fig. 1). The stippled area indicates the lines of drainage, covered by a thin film of water, on May 25 and 31, 1967.

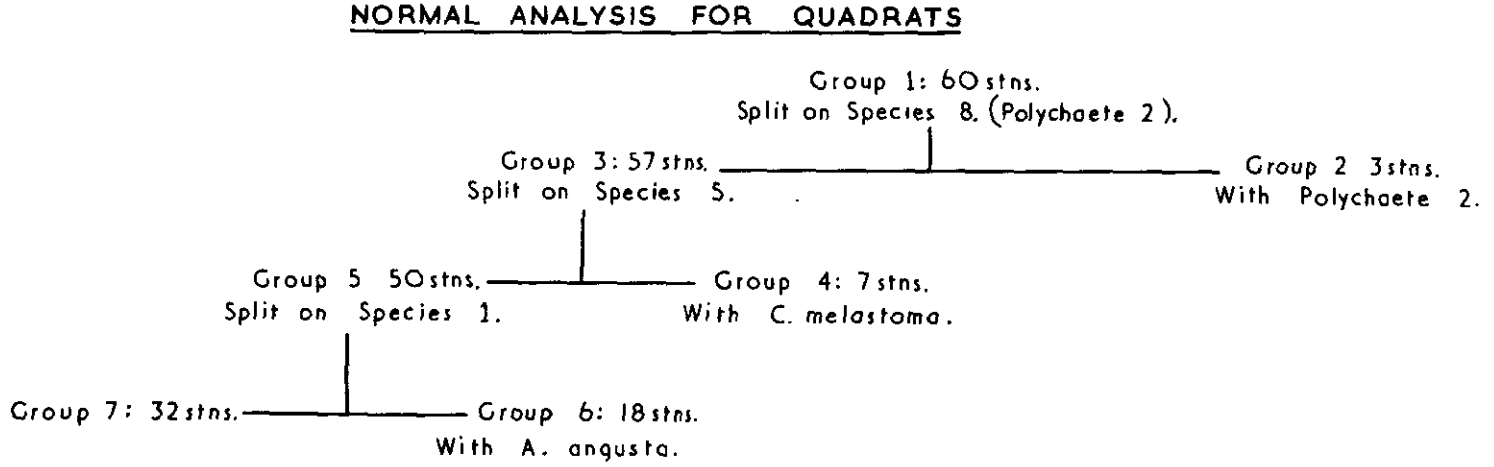

Fig. 4 – Diagrammatic representation of computer classification during normal analysis of quadrat data.

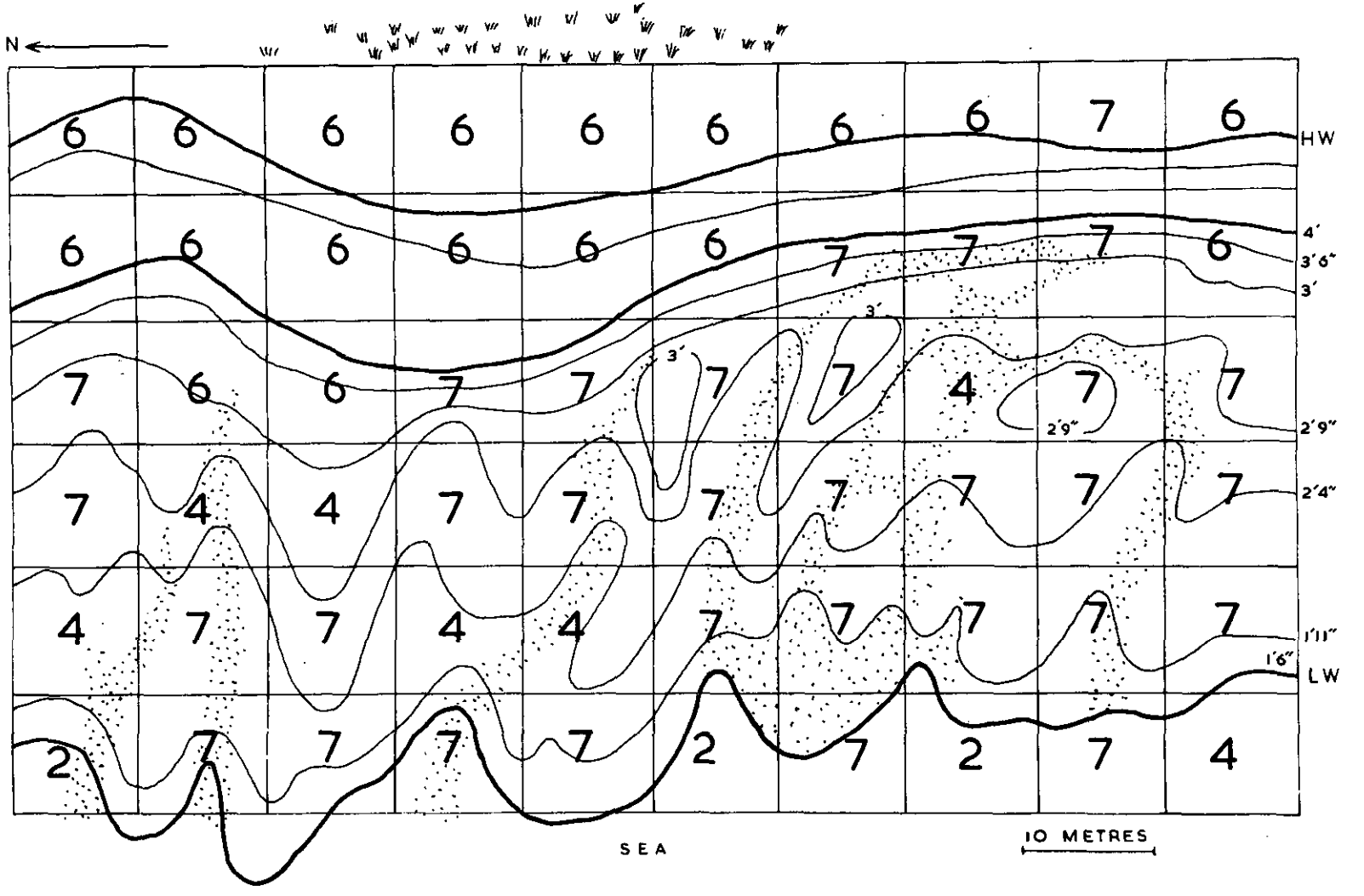

Fig. 5 - Occurrence of the computer groupings on the sandy shore quadrats.

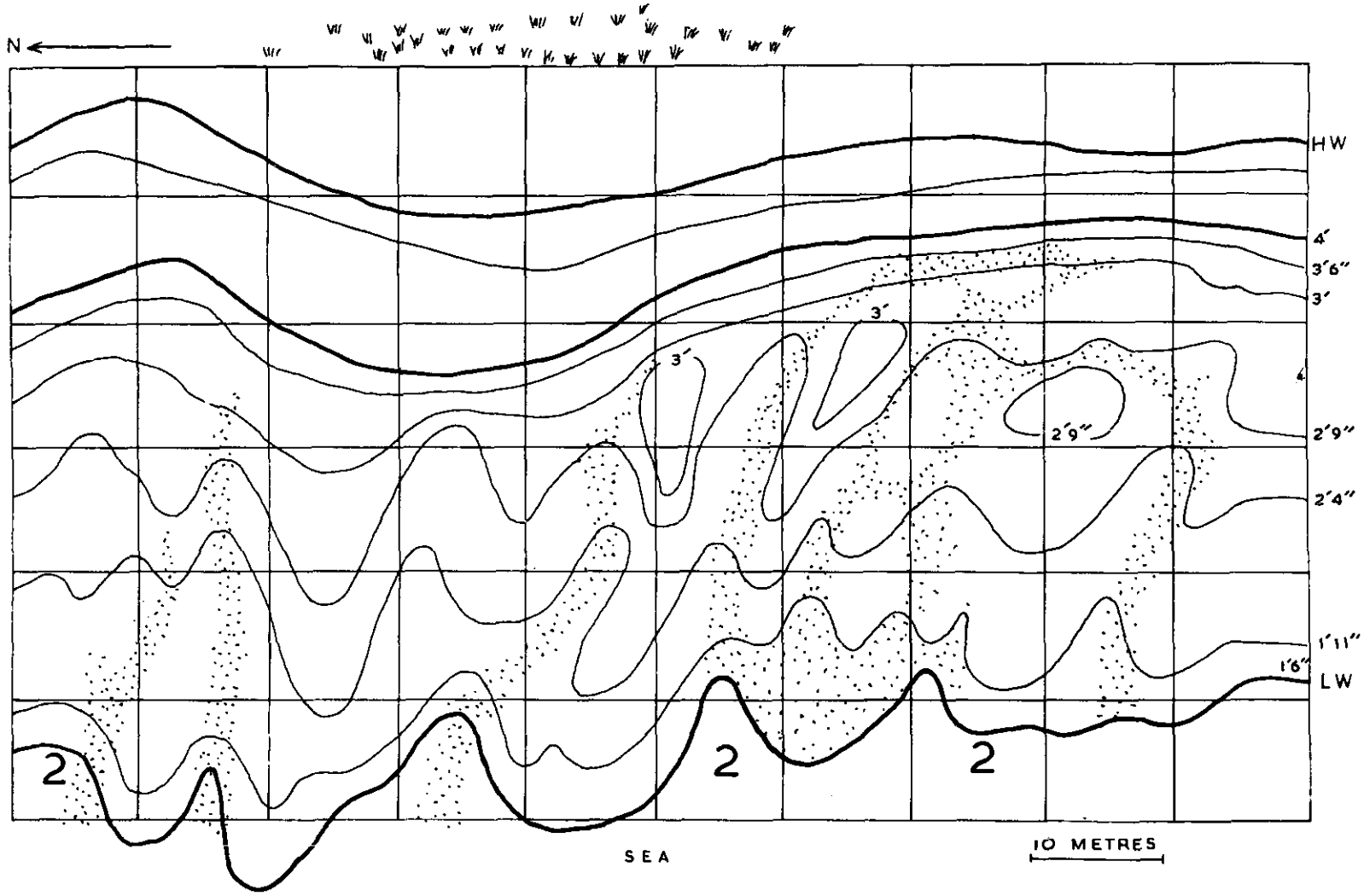

Fig. 6 - Distribution of Group 2 on the sandy shore.

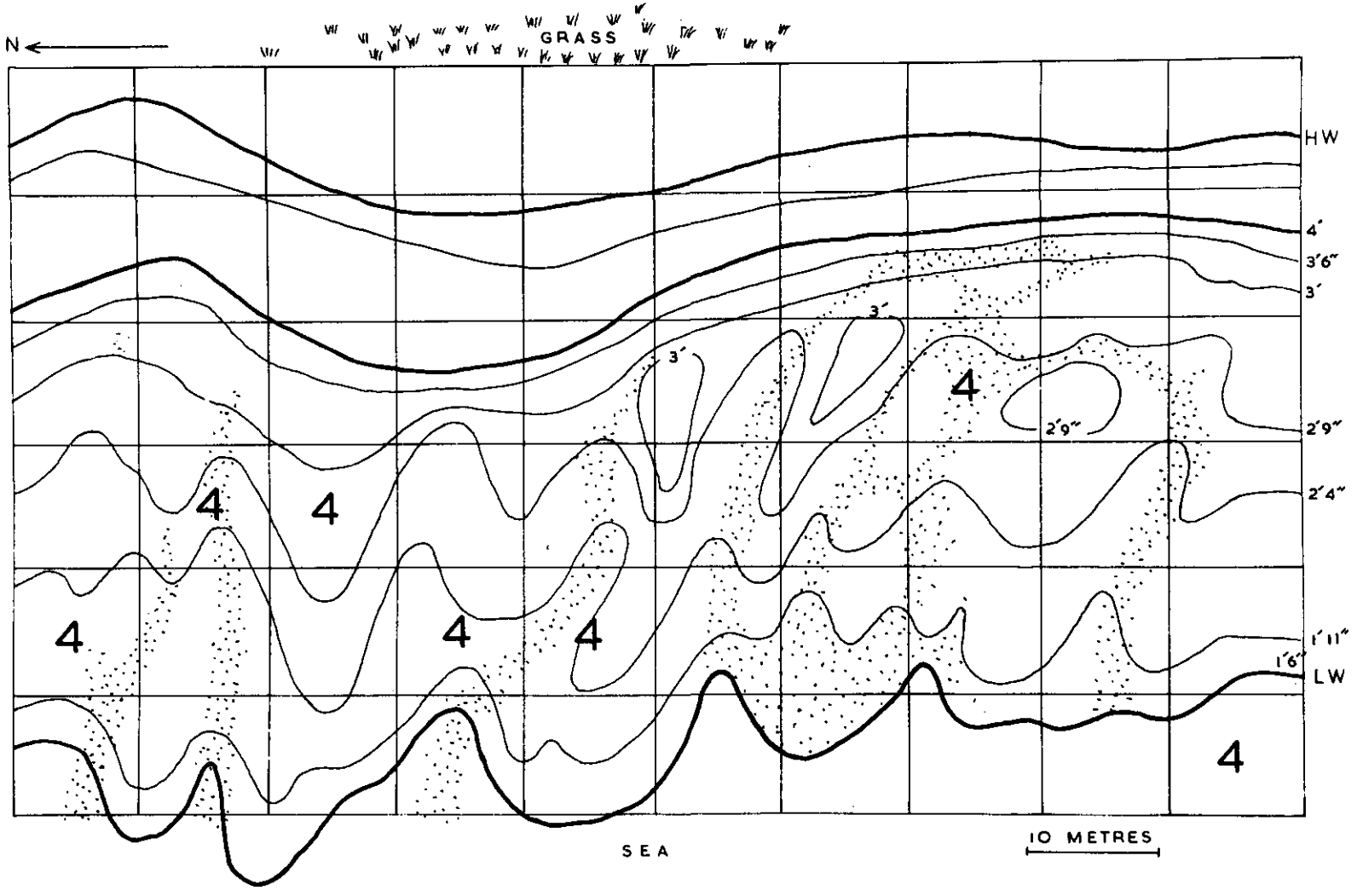

Fig. 7 - Distribution of Group 4 on the sandy shore.

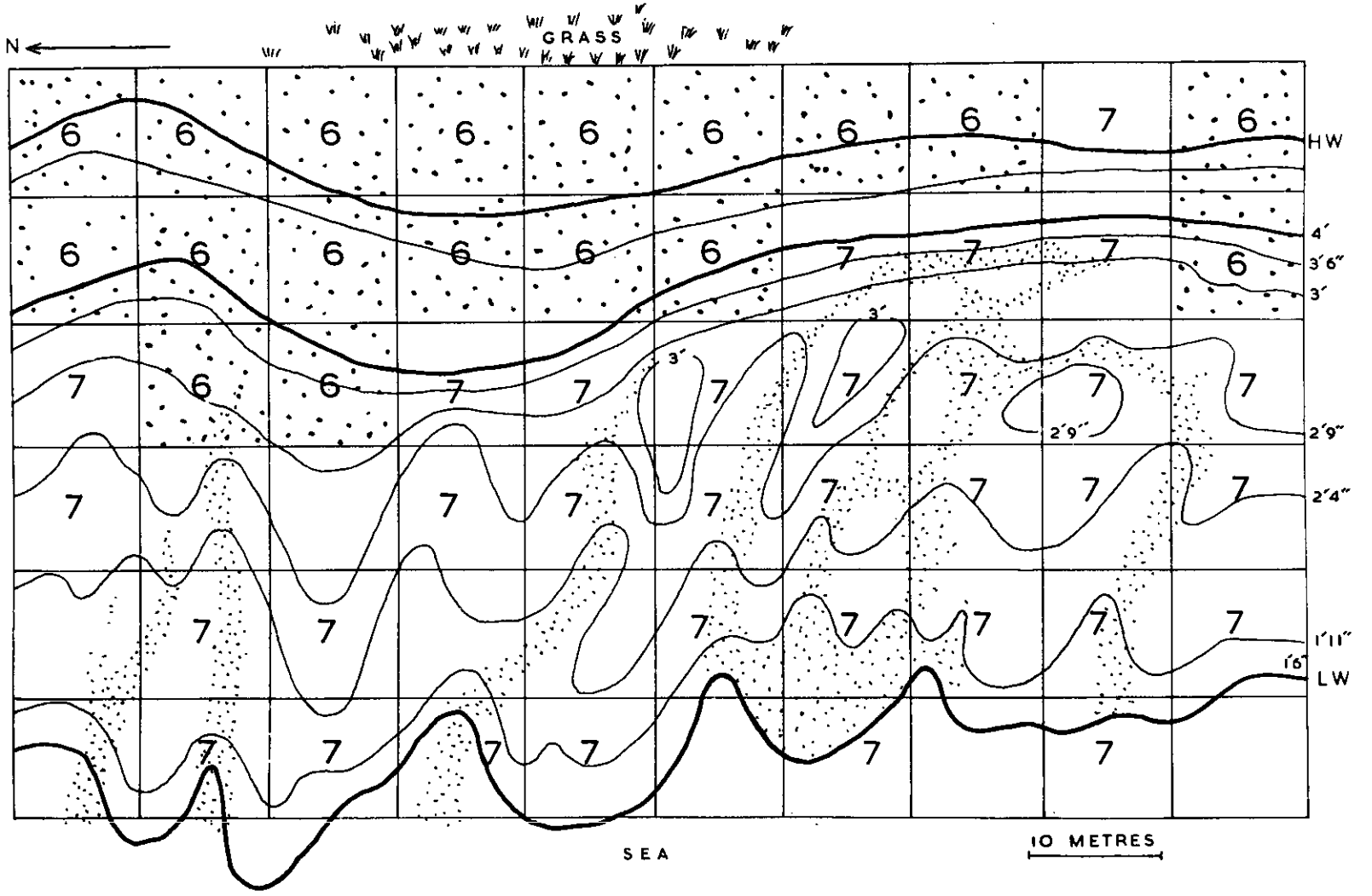

Fig. 8 - Distribution of Groups 6 and 7 on the sandy shore.

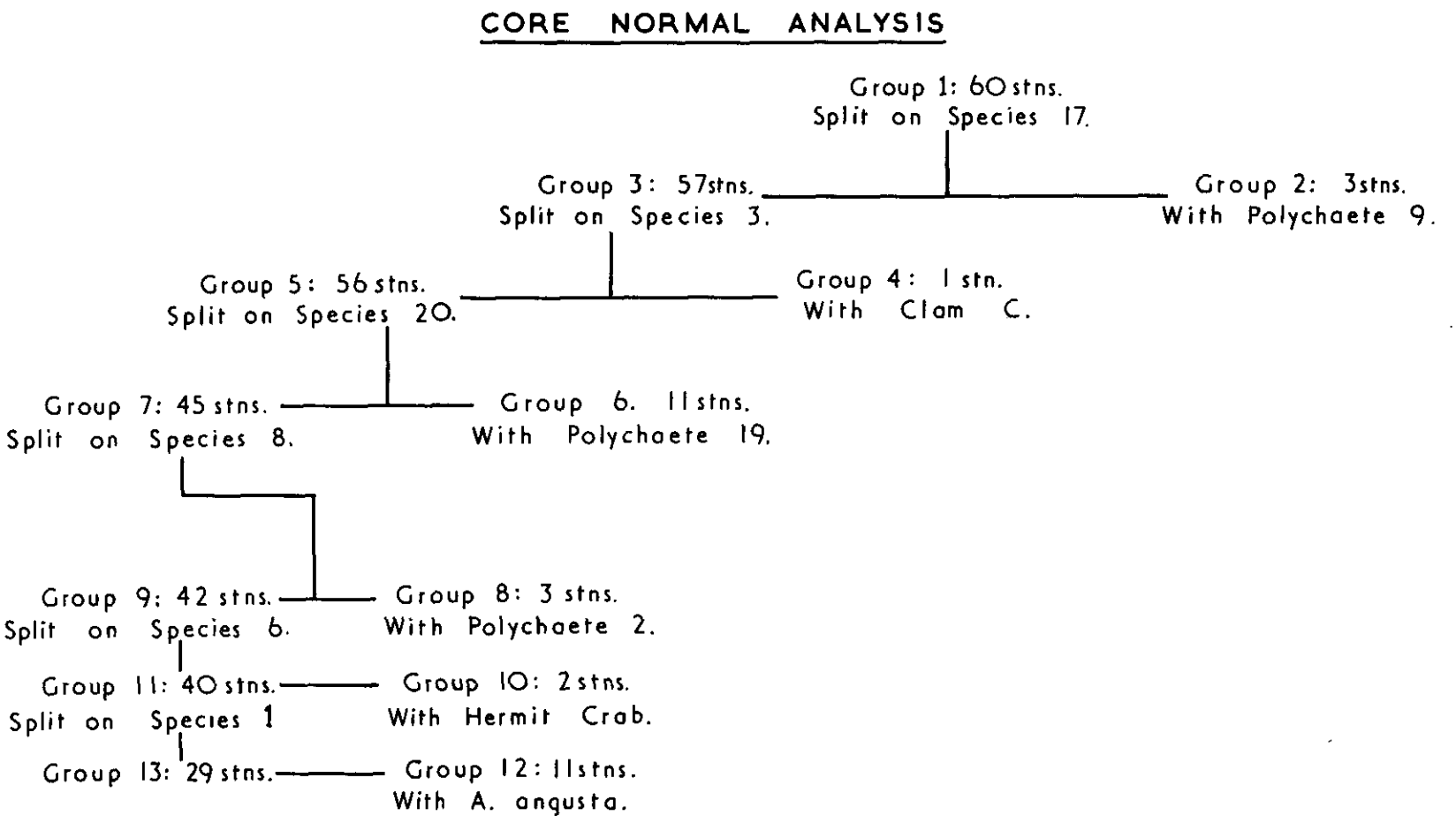

Fig. 9 - Diagrammatic representation of computer classification during normal analysis of core data.

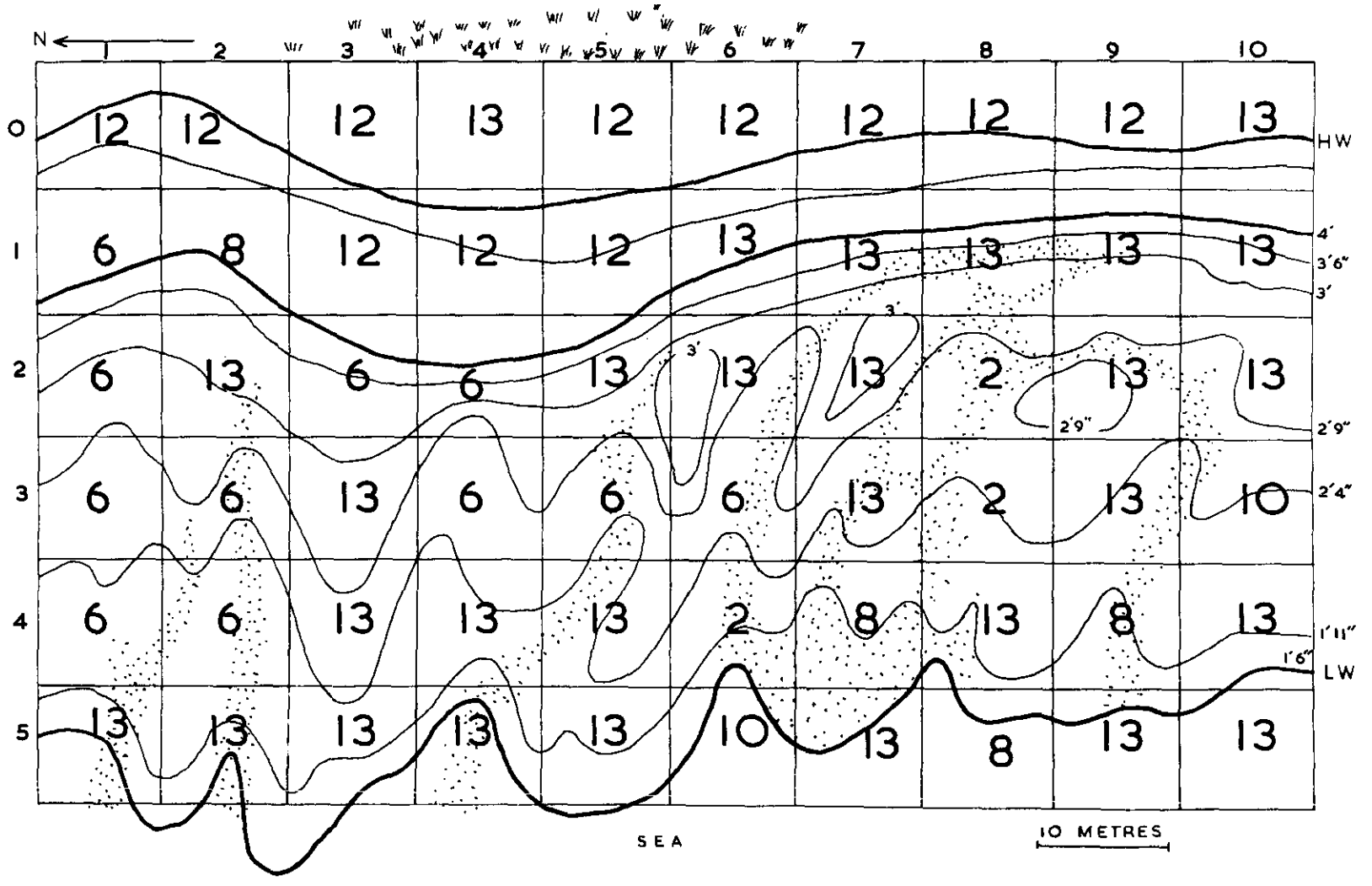

Fig. 10 - Distribution of core groups on the sandy shore.

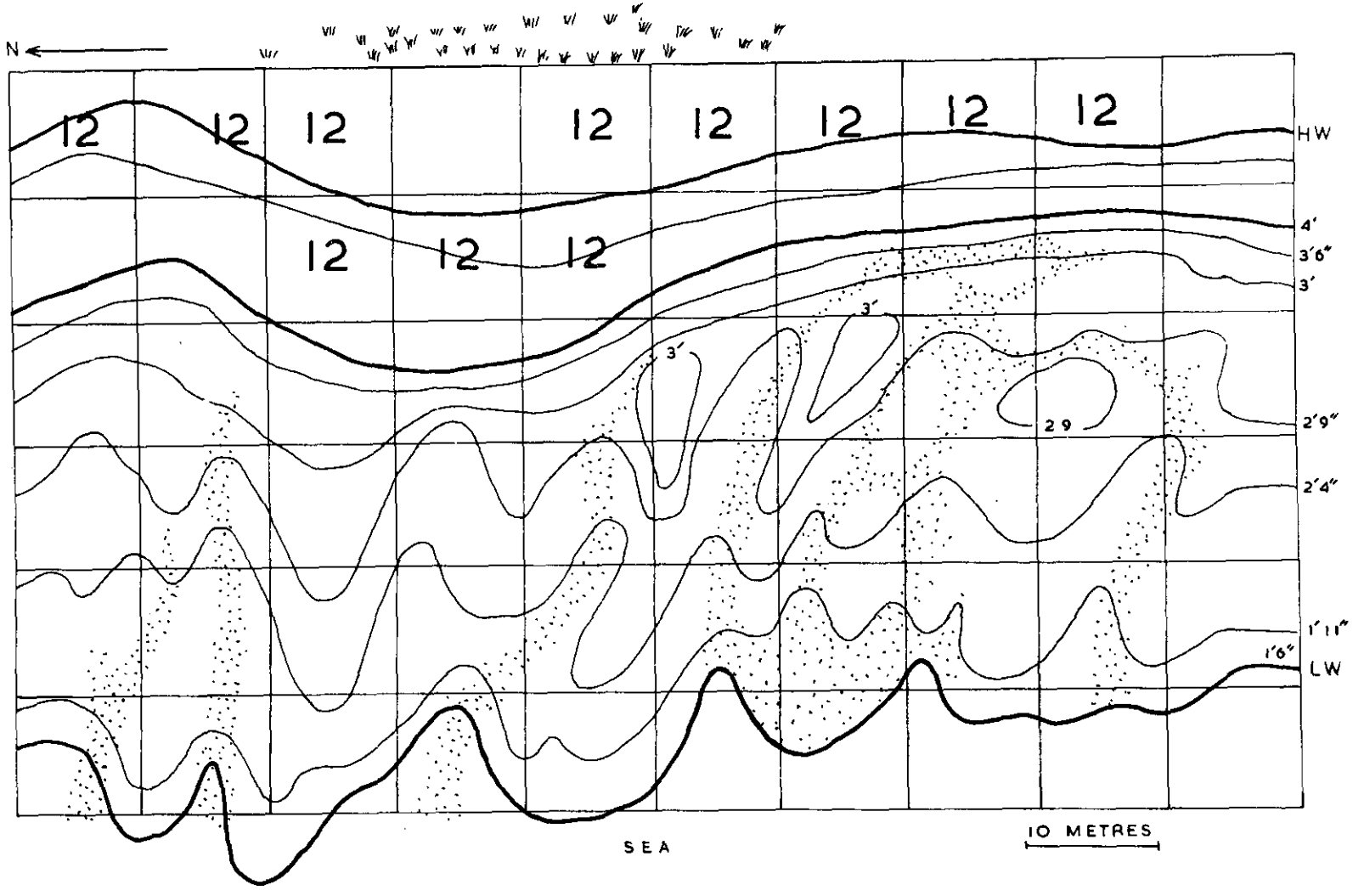

Fig. 11 - Distribution of core Group 12 on the sandy shore.

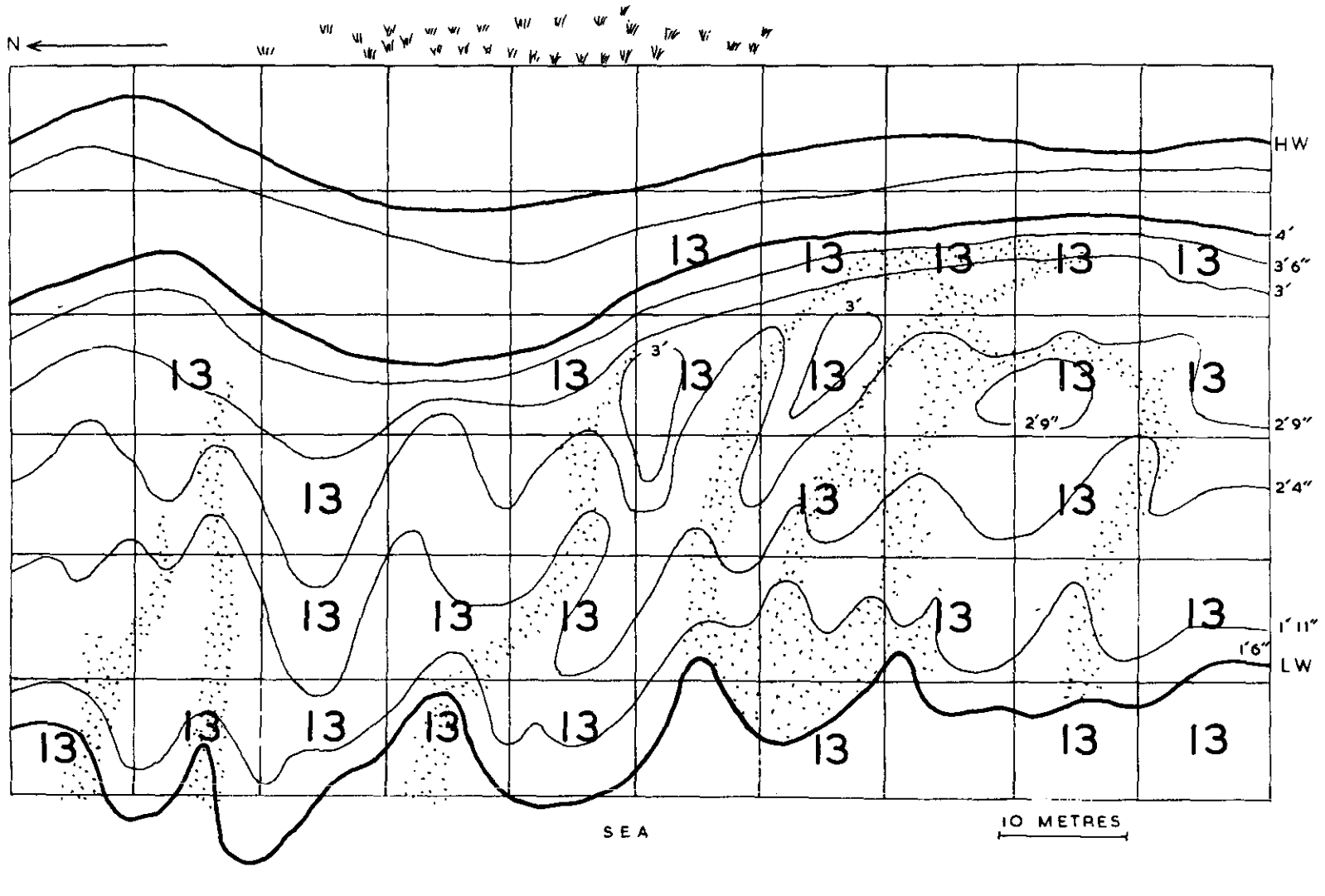

Fig. 12 - Distribution of core Group 13 on the sandy shore.

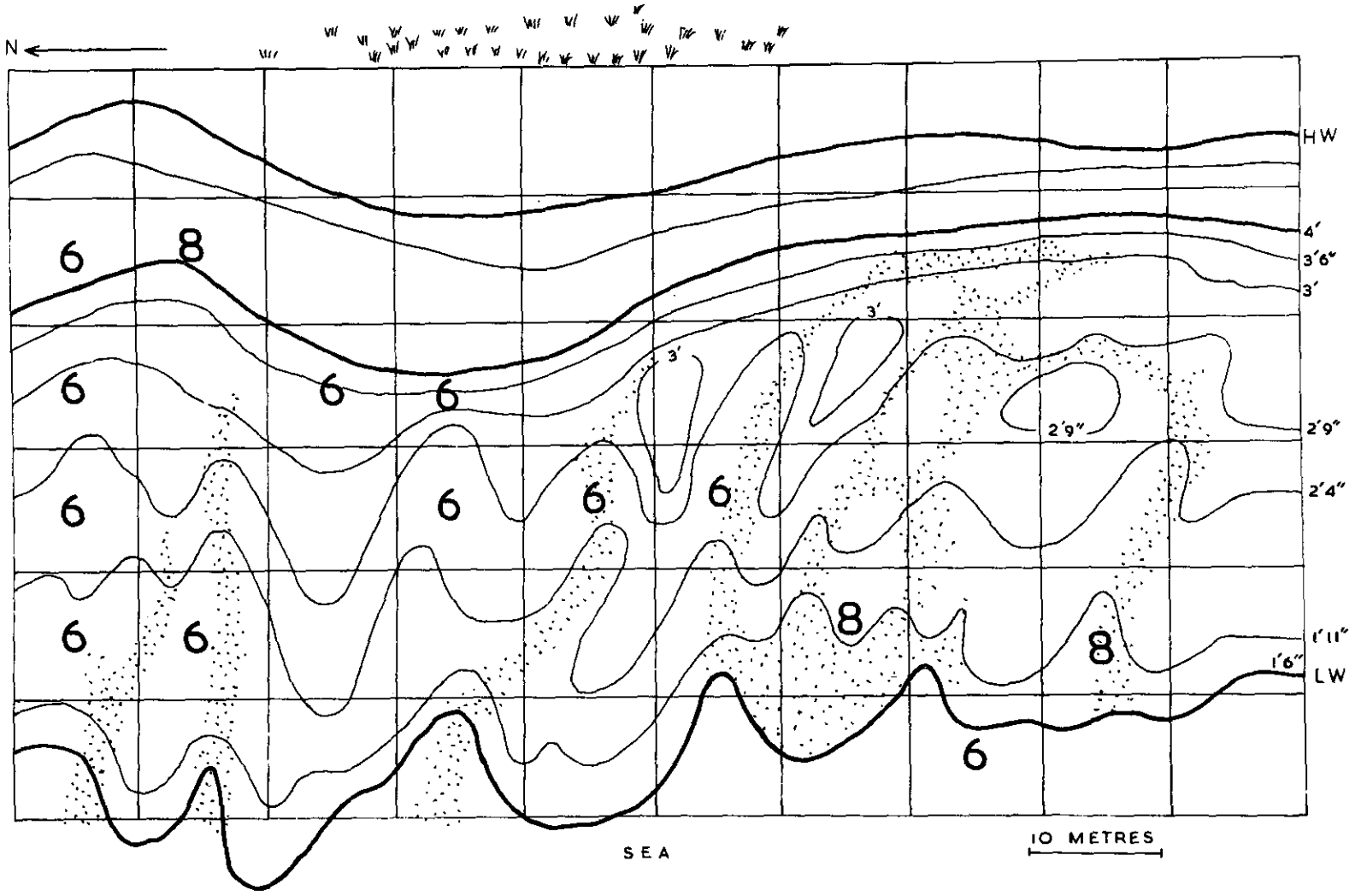

Fig. 13 - Distribution of core Groups 6 and 8 on the sandy shore.

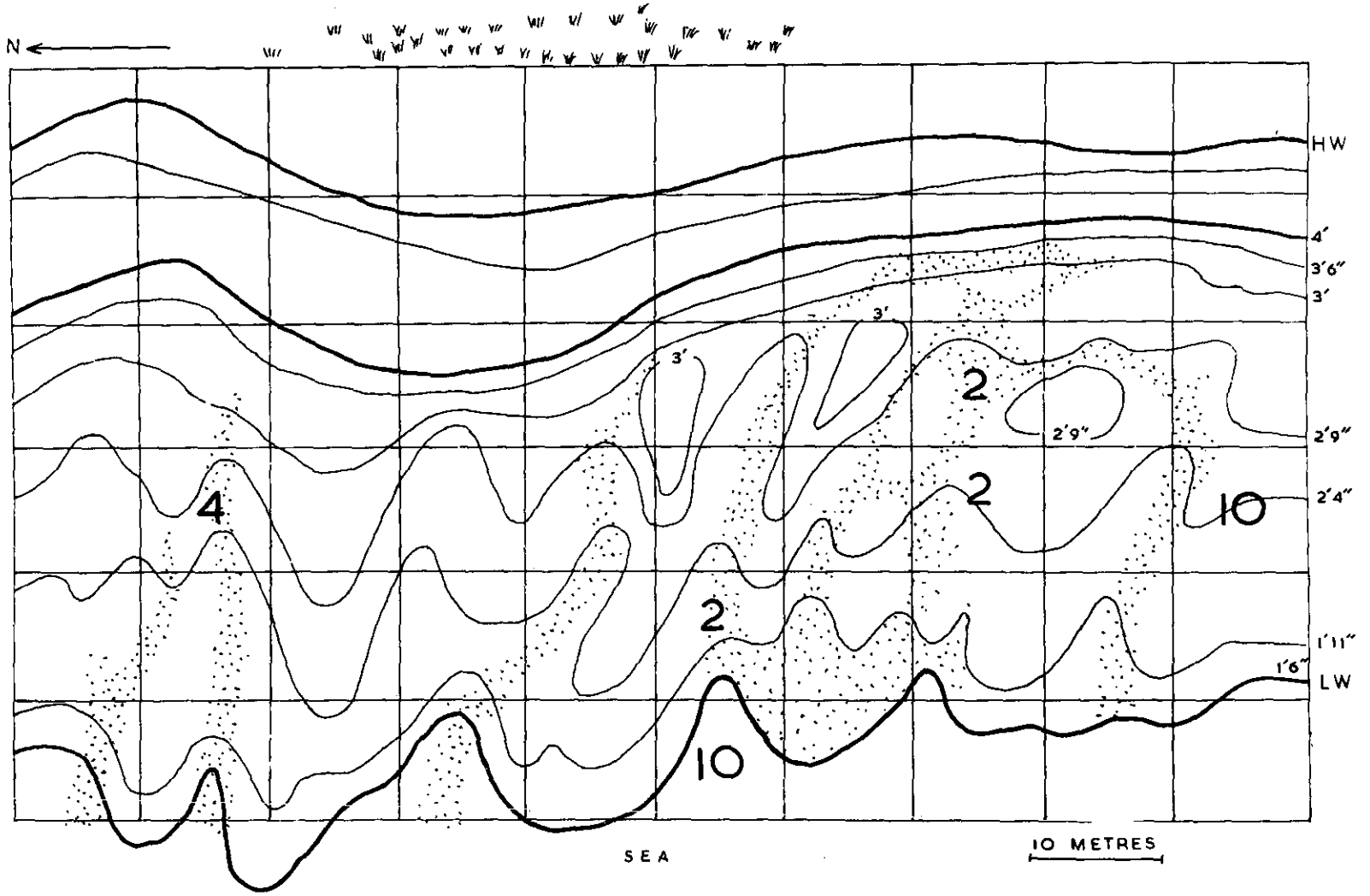

Fig. 14 - Distribution of core Groups 2, 4, and 10 on the sandy shore.

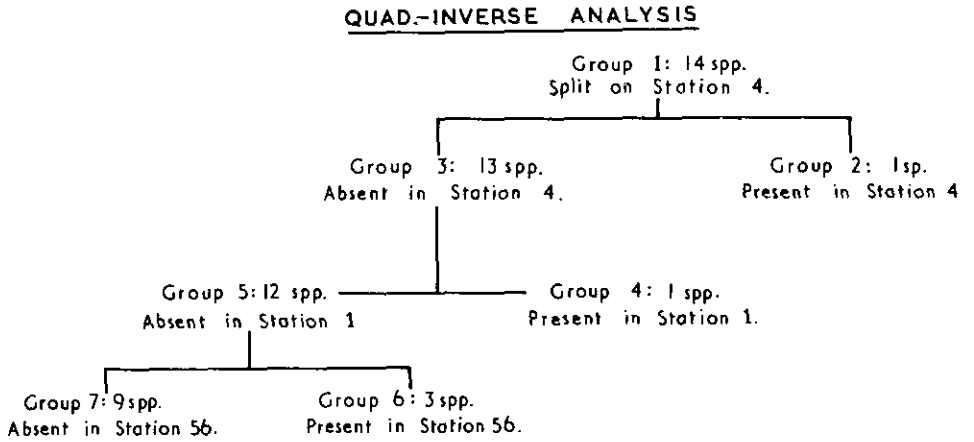

Fig. 15 - Diagrammatic representation of computer classification of quadrat data during inverse analysis.

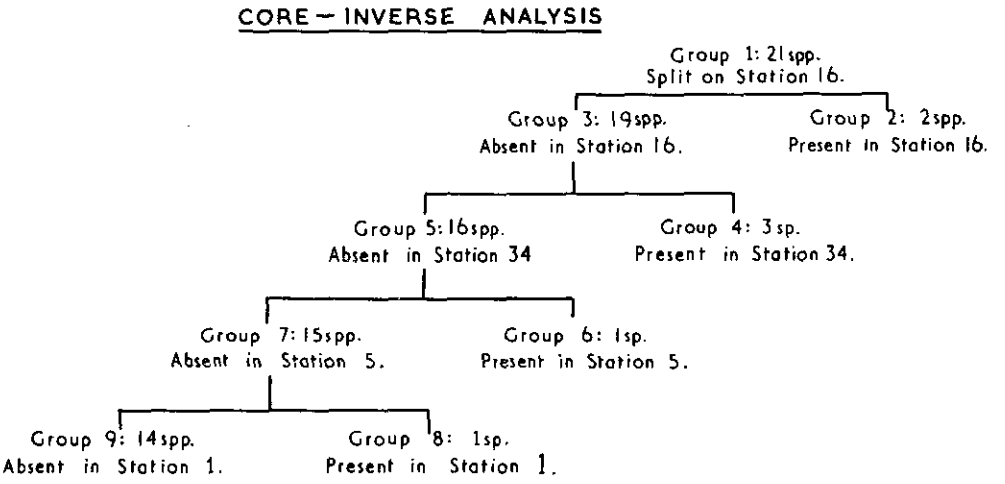

Fig. 16 - Diagrammatic representation of computer classification of core data during inverse analysis.

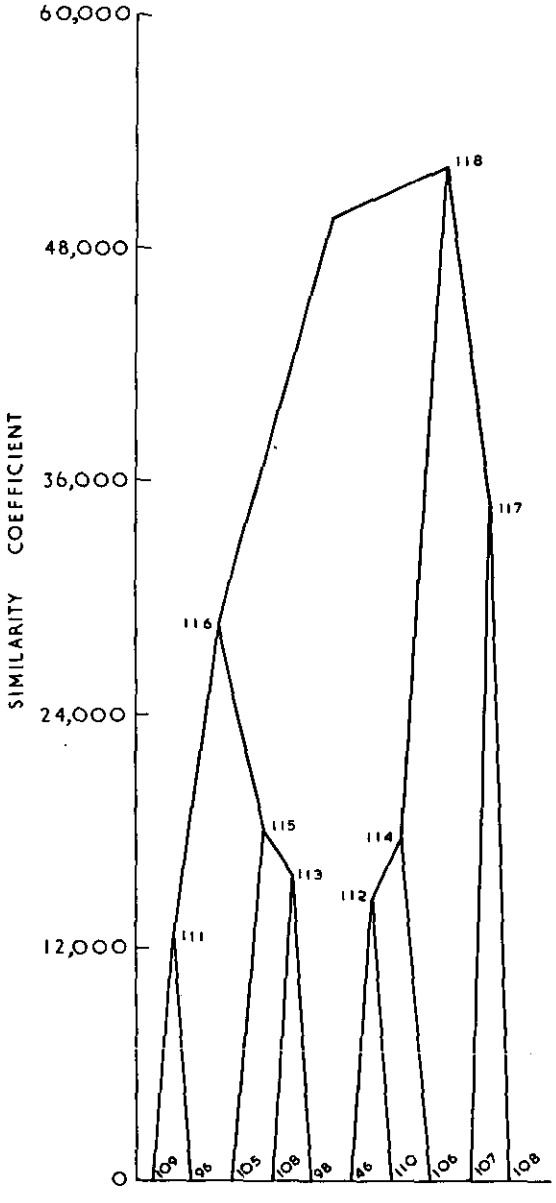

Fig. 17 - Computed representation of aggregative analysis of core data.

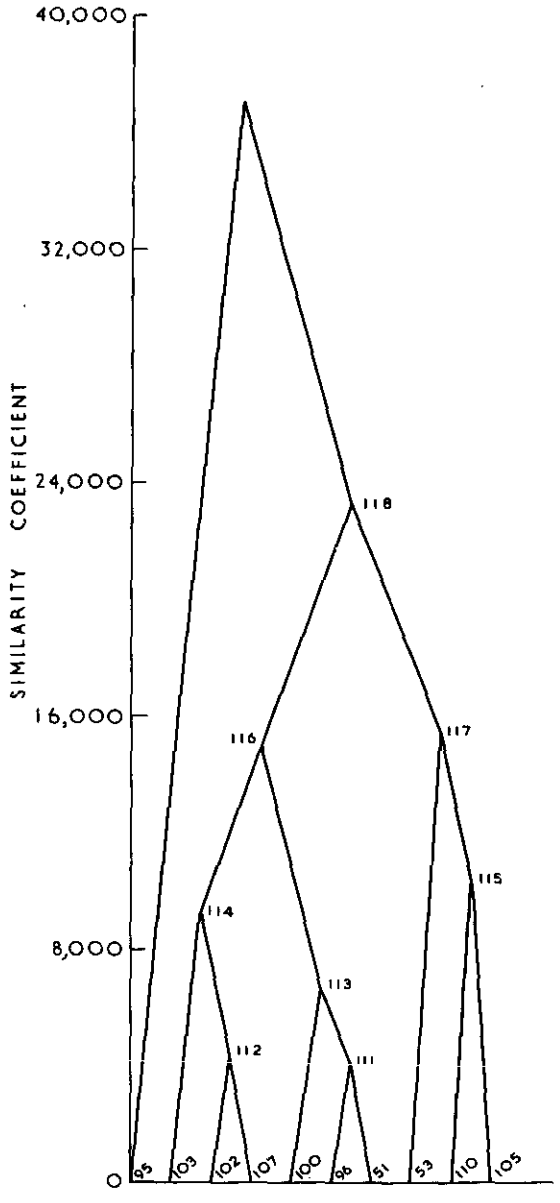

Fig. 18 - Computed representation of aggregative analysis of quadrat data.

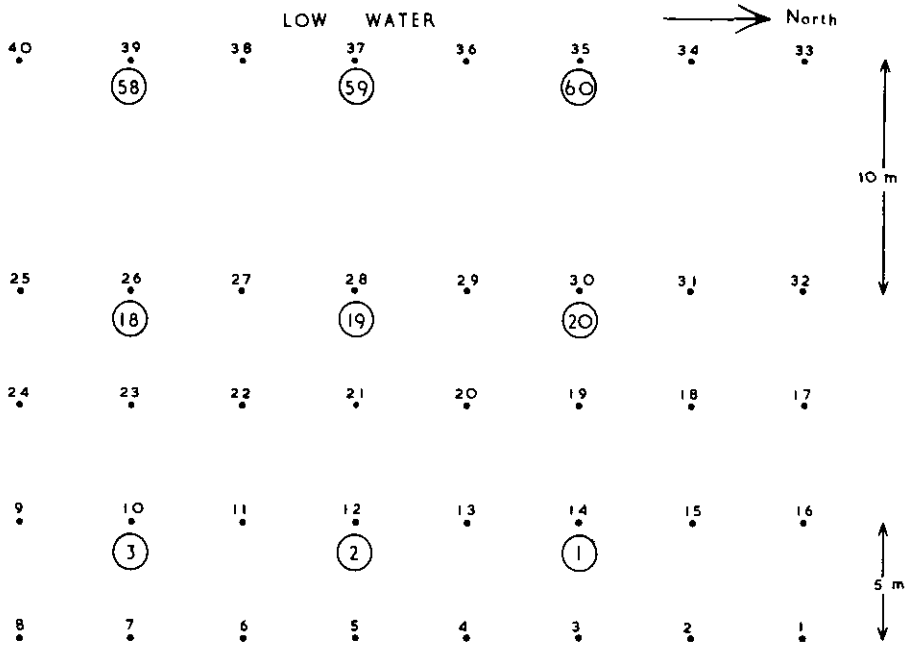

Fig. 19 - Sample grid showing its relation to the general survey grid. Uncircled numbers are general survey grid stations.

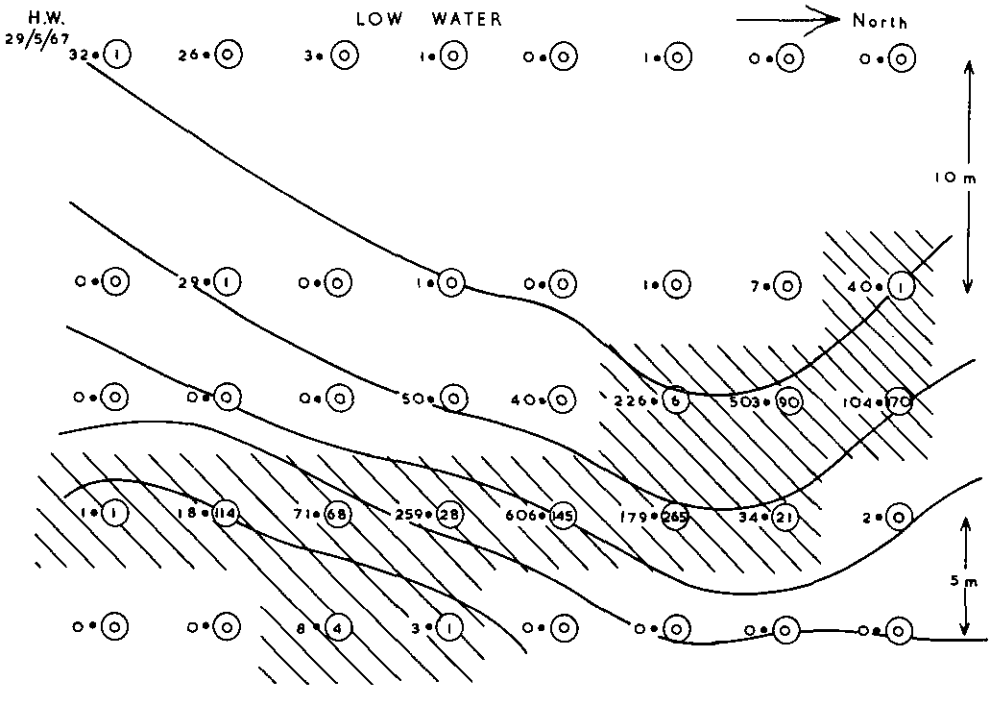

Fig. 20 - Distribution of *A. angusta* (uncircled) and *A. cuneata* (circled) in relation to the beach contours. Hatching denotes areas of occurrence of *A. cuneata*.

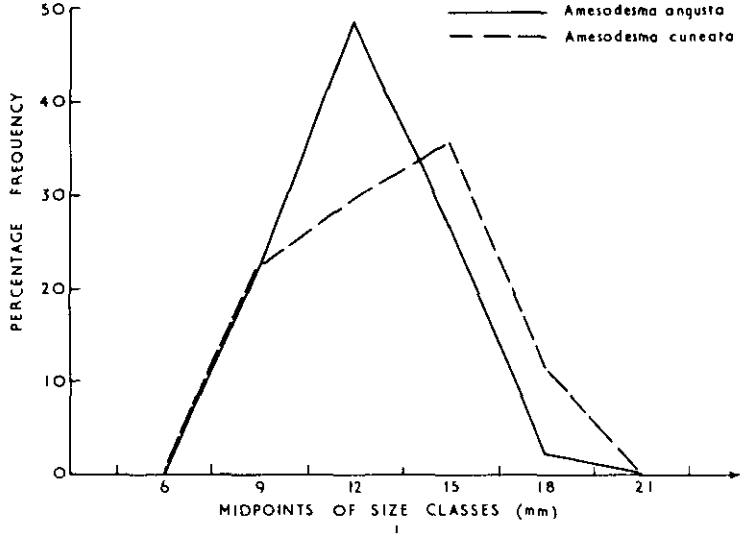

Fig. 21 - Percentage size distribution of *A. angusta* and *A. cuneata* at Station 13.

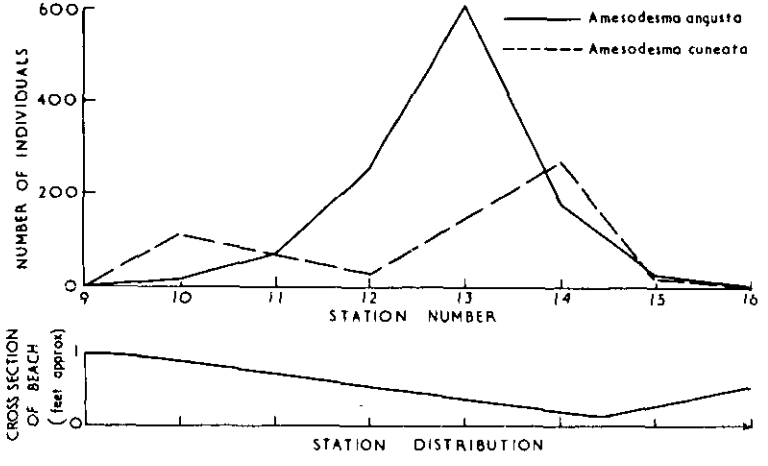

Fig. 22 - Abundance of *A. angusta* and *A. cuneata* at Stations 9-16.

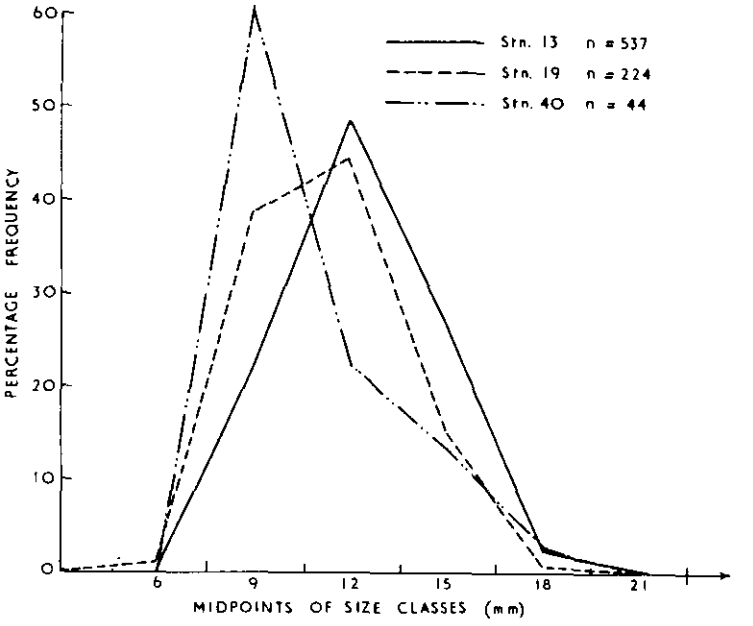

Fig. 23 - Size distribution of *A. angusta* at Stations 13, 19, and 40.

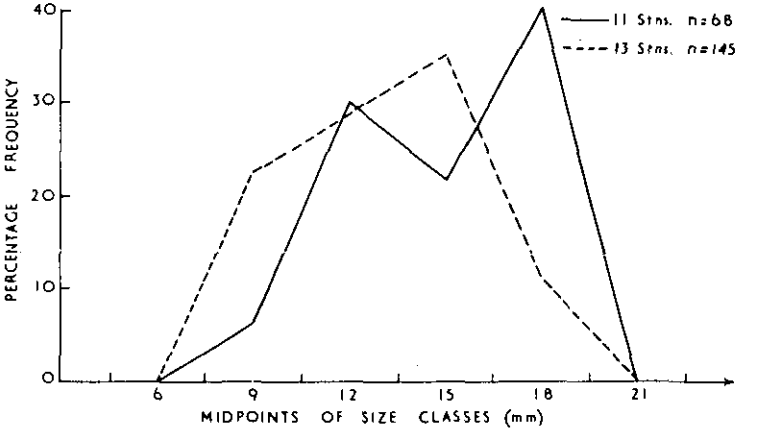

Fig. 24 - Size distribution of *A. cuneata* at Stations 11 and 13.

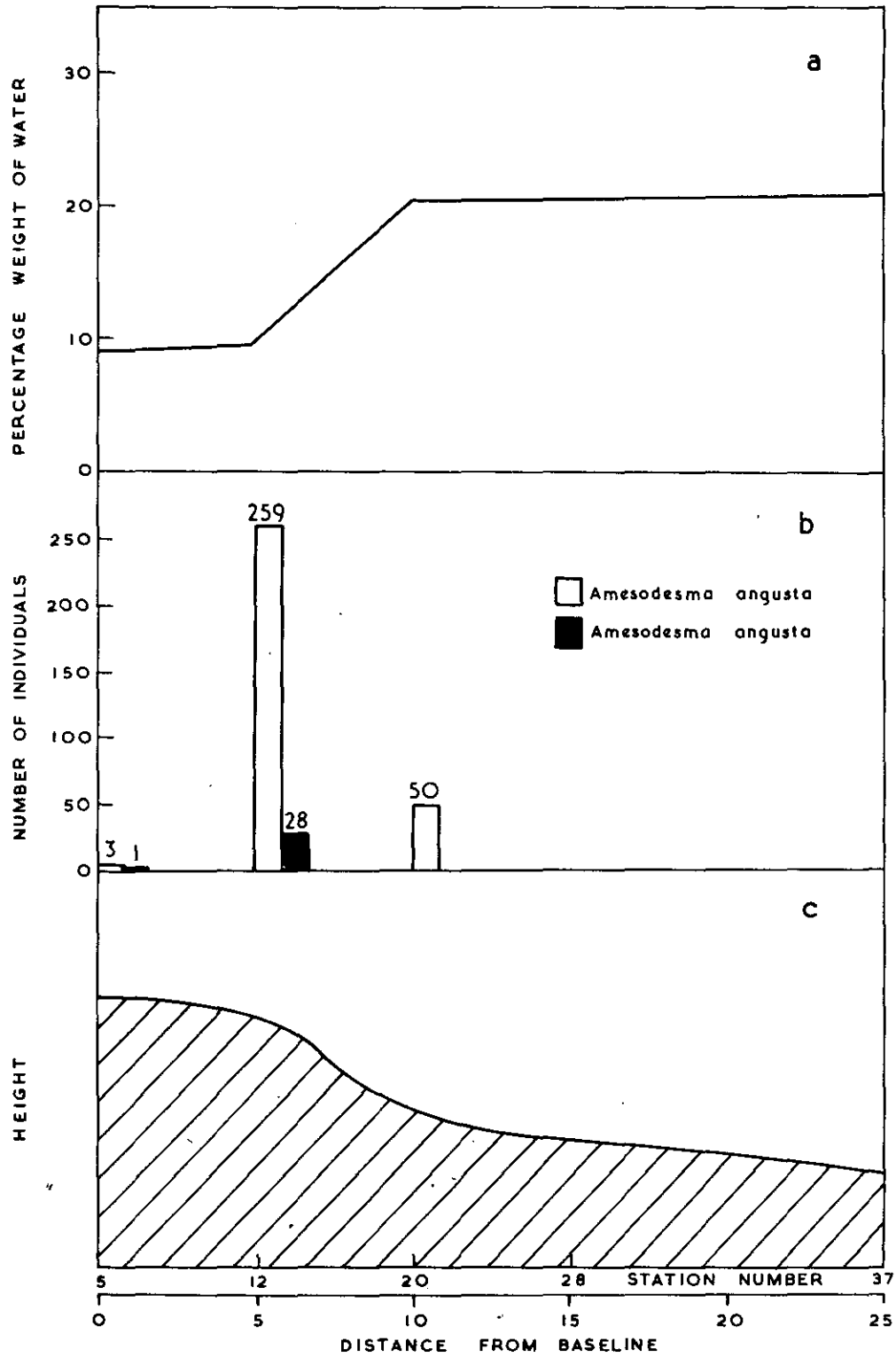

Fig. 25 - Water content of sand, total numbers of *Amesodesma angusta* and *A. cuneata* at each station, and cross section of the beach.

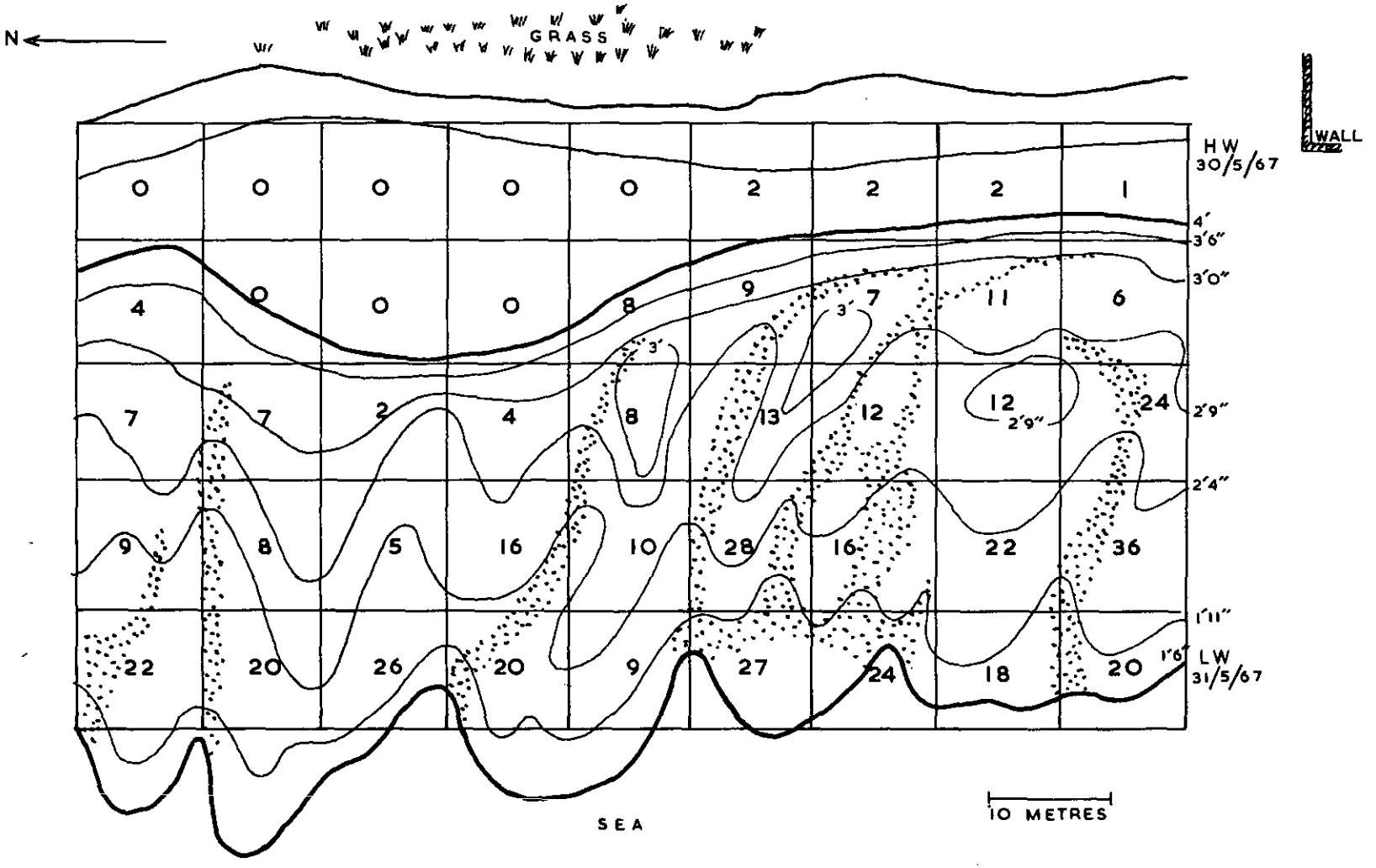

Fig. 26 - Distribution of *Conuber melastoma* on the sandy shore. Values give the number found in each square. The stippled area shows the drainage system.

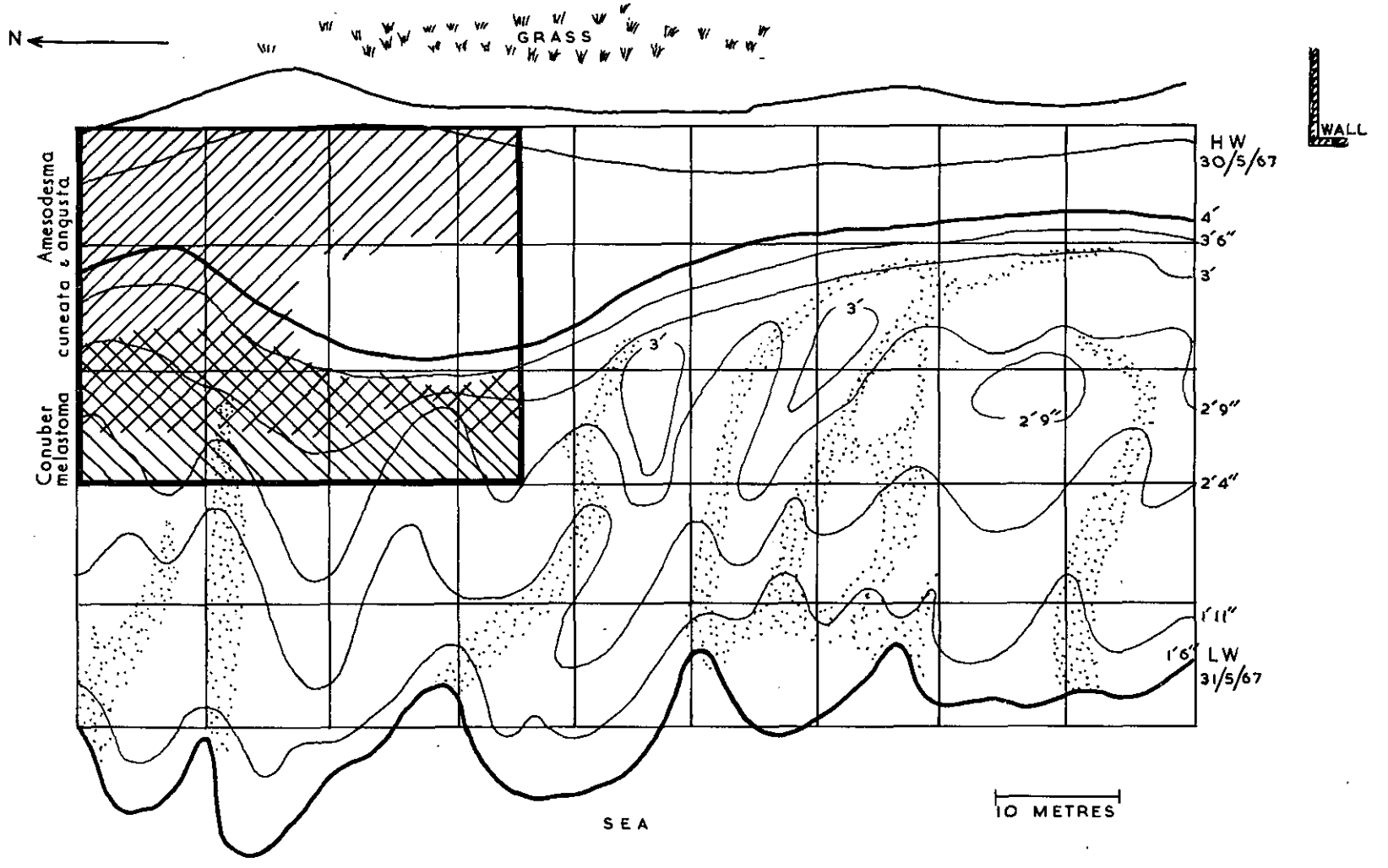

Fig. 27 - Approximate overlap of the distributions of *Amesodesma angusta* and *A. cuneata* and *Conuber melastoma* on the sandy shore.

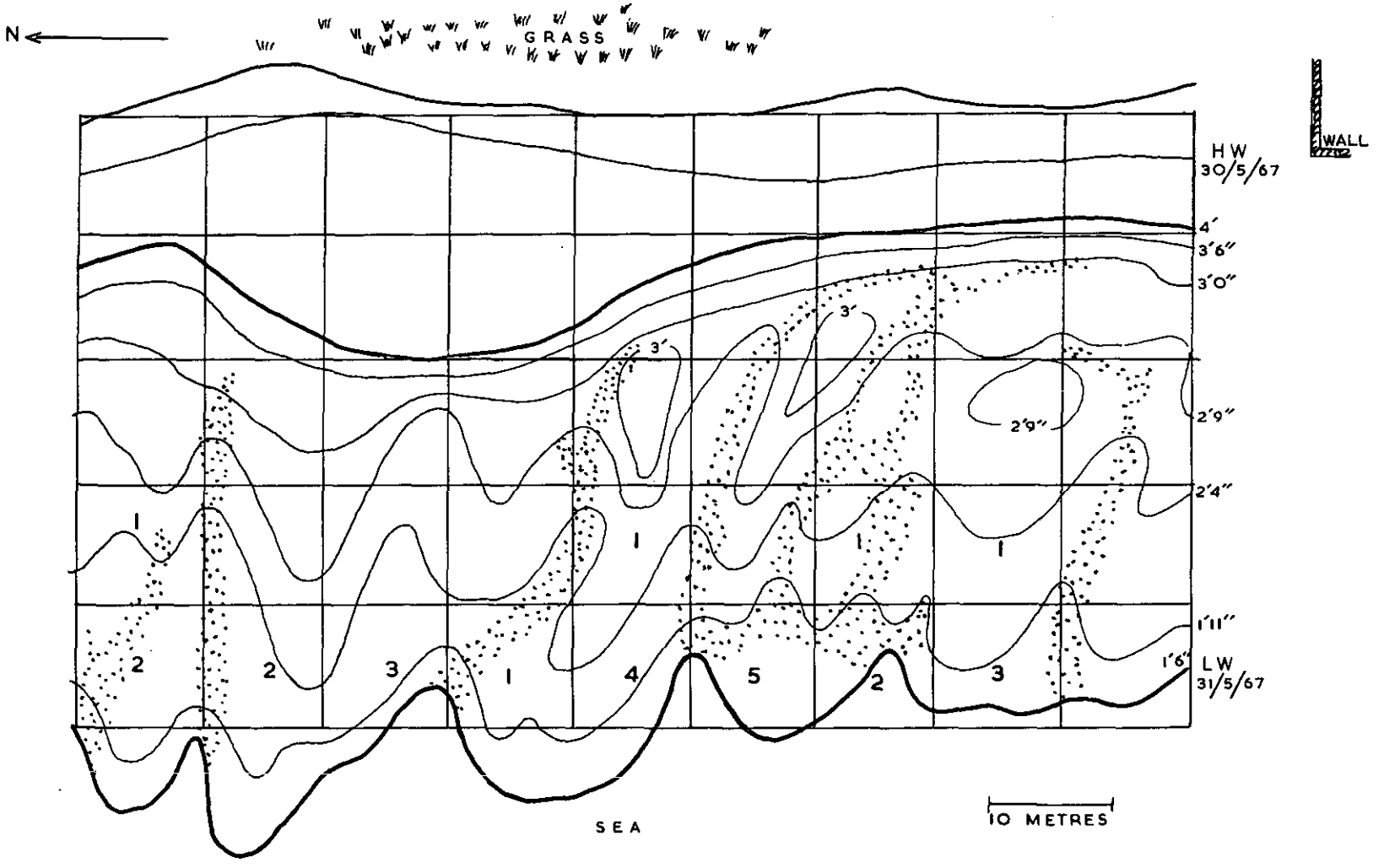

Fig. 28 - Distribution of *Astropecten polycanthus* on the sandy shore. Values give the number found in each square.

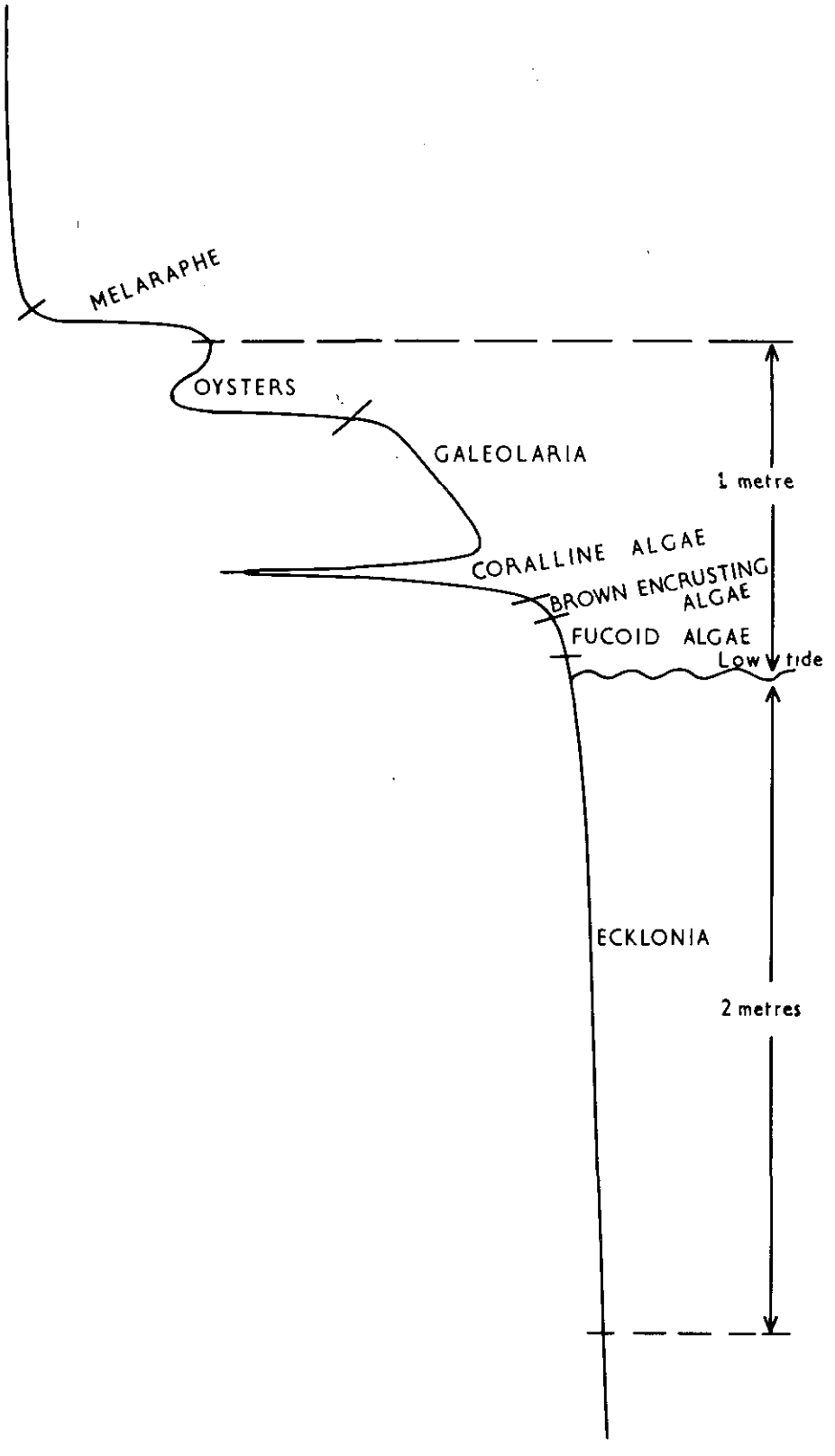

Fig. 29 - Profile of transect across precipice on the rocky shore.

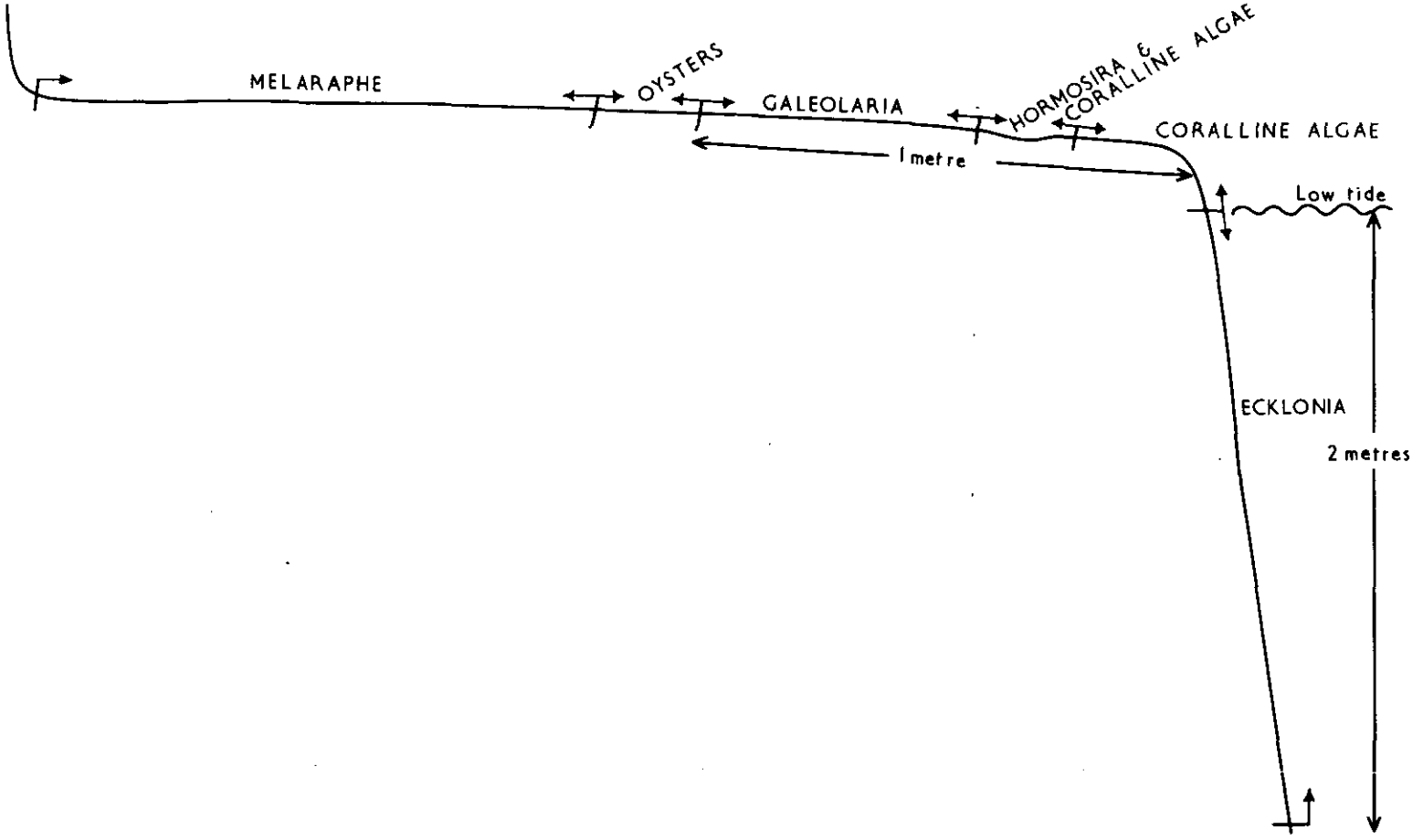

Fig. 30 - Profile of transect across flat rock on the rocky shore.

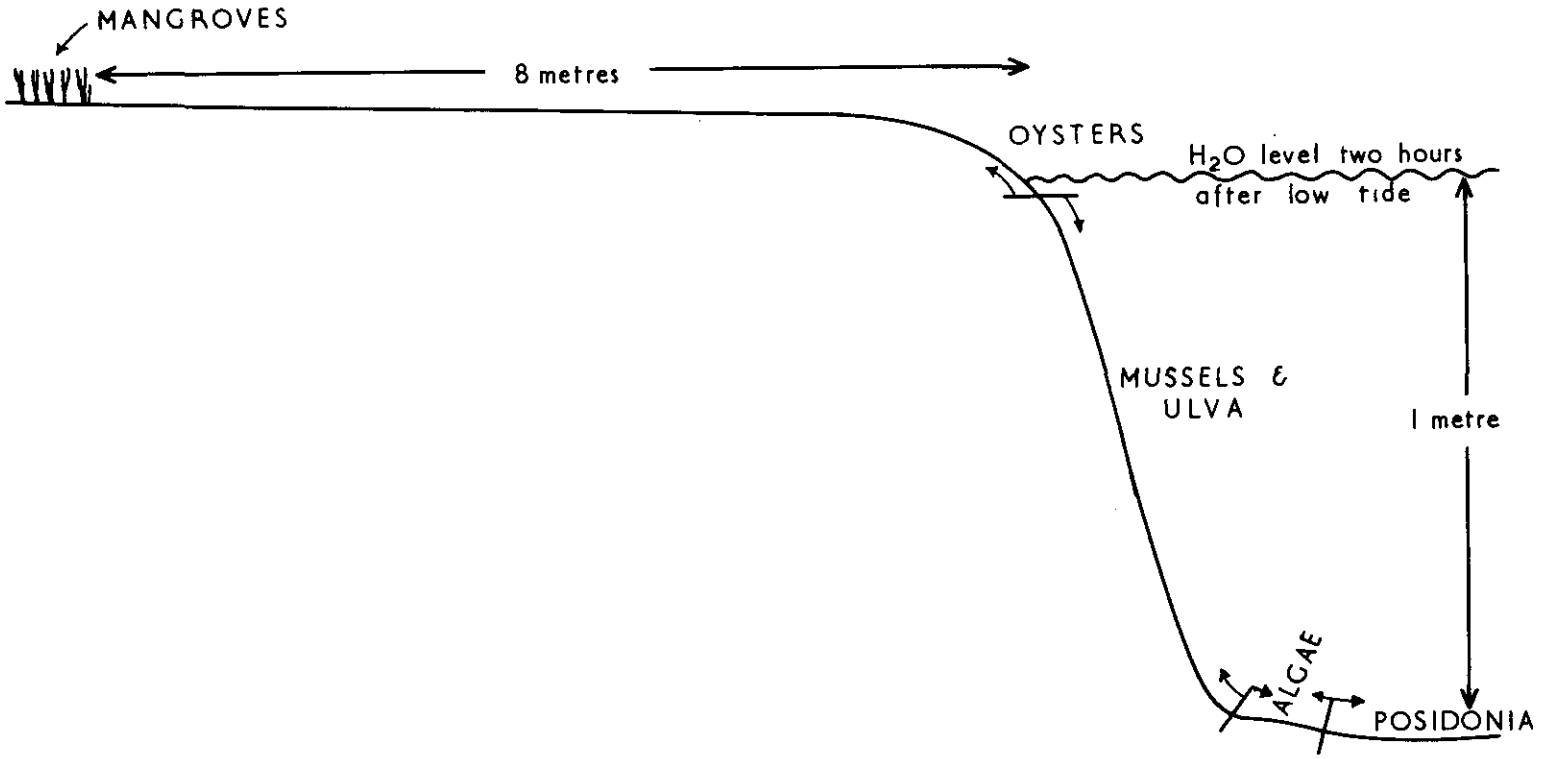

Fig. 31 - Profile of transect across mangrove area in the exposed area of the bay.

DISTRIBUTION OF FORAMINIFERA IN GUNNAMATTA BAY

I. INTRODUCTION

This group investigated the distribution and relative abundance of the benthic Foraminifera in Gunnamatta Bay, and their relationships to the local environment.

Foraminifera are invaluable as a tool for stratigraphic dating, as well as being good indicators of local environmental conditions. They are characterized by an alternation of sexual and asexual generations in their life cycles, the possession of filopodia, and the secretion of a test.

II. METHODS AND MATERIALS

Samples of about two litres were taken at 10 stations (Fig. 1) along a north-south traverse in Gunnamatta Bay, using a Phipps bottom sampler. Dredge samples were taken at various depths, to sample a variety of bottom environments.

Samples were divided into three parts - one to be examined for Foraminifera, one for sediment type distribution, and one for reference. Foraminifera samples were boiled with sodium carbonate to flocculate clay particles, and to free organic matter from the tests. These boiled samples were then passed through a stack of three sieves, and the fractions retained on the 30 and 85-mesh sieves dried. The dried samples were then treated with carbon tetrachloride to concentrate the Foraminifera. The concentrated fractions were classified and relative abundances determined. Total abundances were determined from dried samples weighing 100 g.

III. RESULTS

Table 1 shows foraminiferal distribution. Figure 2 shows the distribution of the whole population, Figure 3 the distribution of walled types, Figure 4 the distribution of rare species, Figure 5 the distribution of some interesting genera, and Figure 6 shows areas where some genera are abundant.

IV. DISCUSSION

Figure 2 shows two distinct peaks in population number - one on the southern slope and the other on the northern slope of a deep basin in the middle of the bay (see Fig. 1). The sample from the southern slope contained the larger number of coarser

foraminiferans. The lowest numbers occurred in the channel, on the sand bar, and in the deep basin, the deep basin having the highest numbers of the three areas.

Further discussion relates only to coarse species.

The tests of foraminiferans are of three different types - arenaceous, calcareous porcelaneous, and calcareous hyaline. The arenaceous test is composed of sand grains cemented together by calcium carbonate. In the calcareous porcelaneous type, the calcite crystals making up the wall lie with their 'c' (long) axes parallel to the test wall, and form several layers. This makes a solid and relatively heavy shell. The calcareous hyaline type, however, has the calcite crystals with their c axes lying perpendicular to the test wall. This produces a lighter and more fragile test with numerous pores.

Figure 3 shows that arenaceous types are relatively abundant at Stations 4, 10, and 11, all of which are at the northern end of the channel. It is interesting to note the small numbers of this type at Station 5 which is close to Stations 4, 10, and 11.

The porcelaneous Foraminifera have a greater relative abundance on the southern slopes of the basin, while the hyaline types were most abundant at the northern end of the channel.

The relative distribution of the more common species (those making up more than 5% of the total foraminiferans counted) showed little variation between stations. In the case of the rare species, peaks of abundance are shown at Stations 4 and 11 (Fig. 4). It seems therefore, that a study of the rare species might be more useful for the identification of slight environmental differences.

From Table 1, Discorbis dimidiatus, Triloculina, Quinqueloculina, and Spiroloculina can be seen to have localized increases in density over the study area and thus might be used as species indicators of environment.

Discorbis dimidiatus was characteristic of the sand bar and the entrance to the channel. Possibly, its distribution is related to the conditions prevailing in the main part of Port Hacking.

Triloculina trigonula was the dominant species on the southern slope and in the channel, while T. affinis was predominant on the northern slope of the deep basin.

Quinqueloculina lamarckiana was the predominant species at the northern end of the channel, and also on the northern slope.

Spiroloculina communis and S. lucida, which were two of the most abundant species seen, were characteristic of the deep basin. S. communis is widespread in its distribution, whereas S. lucida was particularly abundant in the deep basin area (see Figs. 5 and 6).

TABLE 1

DISTRIBUTION OF FORAMINIFERANS IN GUNNAMATTA BAY

SPECIES (from 100 gms dry matl.)		STATIONS									
		1	2	4	5	10	11	8	7	6	3
	coarse	15	93	75	1.760	2.700	1.093	200	800	1.033	47
	fine	530	1.665	3.950	28.000	37.300	28.300	6.900	17.400	38.252	276
	total	545	1.758	4.025	29.760	40.000	29.393	7.100	18.200	39.285	323
Textularia											
Candeiana		R	R	F		R	F	R	R		
Pseudogramen				R	R	R		R		R	
Sagittula atrata				R			F				
Spiroloculina											
Antillarium			R	R	R	R	R	R		R	
Communis				R	F	A	C	A	A	F	R
Lucida		R	F	R	F	R	F	A	C	R	R
Vertebralina											
Striata					R	R	R	R	R		
Quinqueloculina											
Baragwanathi							R				
Costata			R	R	R	R	R				R
Lamarckiana				F		R			R	A	
Pseudoreticulata		R	R	R			R				R
Seminula				R	R		R				
Striata		R									
Subpolygona		F	R	F	R		R	R			F
Massilina											
Secans tropicalis			R		R						R

2700

TABLE 1 (Cont.)

SPECIES (from 100 gms dry matl.)		STATIONS									
		1	2	4	5	10	11	8	7	6	3
	coarse	15	93	75	1.760	2.700	1.093	200	800	1.033	47
	fine	530	1.665	3.950	28.000	37.300	28.300	6.900	17.400	38.252	276
	total	545	1.758	4.025	29.760	40.000	29.393	7.100	18.200	39.285	323
Triloculina											
Affinis		R		R		F	F	R	R	C	
Oblonga							R				
Striatotrigonula				R			R				
Trigonula		F	V.A.	C	C	F	F	R	R		F
Milionella											
Labiosa									R		
Flintina											
Sp.					F	R	R	R	F	R	
Pyrgo											
Depressa		R	R	R		R	R				
Peneroplis											
Planatus		R		R	F	F	R	R	R	R	R
Spirolina											
Cylindracea							R				
Sigmoidella											
Elegantisima		R		R							
Guttulina											
Lactea			R	R		R		R	R	F	
Pacifica				R	R		R	R			
Regina				R	R		R		R	R	

TABLE 1 (cont.)

SPECIES (from 100 gms dry matl.)		STATIONS									
		1	2	4	5	10	11	8	7	6	3
	coarse	15	93	75	1.760	2.700	1.093	200	800	1.033	47
	fine	530	1.665	3.950	28.000	37.300	28.300	6.900	17.400	38.252	276
	total	545	1.758	4.025	29.760	40.000	29.393	7.100	18.200	39.285	323
Globulina											
Gibba globosa											R
Lenticulina											
Reniformis				R							
Vaginulina											
Patens				R							
Vertebralis				R							
Amphicoryna											
Scalaris				R	R		R				
Lagena											
Flatulenta				R							
Baggina											
Philippinensis				R		R				R	R
Pileolina											
Australensis				R							
Rosalina											
Australis				R							
Bertheloti				R							
Bradyi				R							

TABLE 1 (Cont.)

SPECIES (from 100 gms dry matl.)		STATIONS									
		1	2	4	5	10	11	8	7	6	3
	coarse	15	93	75	1.760	2.700	1.093	200	800	1.033	47
	fine	530	1.665	3.950	28.000	37.300	28.300	6.900	17.400	38.252	276
	total	545	1.758	4.025	29.760	40.000	29.393	7.100	18.200	39.285	323
Discorbis											
Dimidiatus		A	R	R	R	R	R	R	R	R	A
Asterorotalia											
Inflata						F	F	R	R	R	
Ammonia											
Beccarii			R	R	F	R	R	R	R		
Cibicides											
Cygnorum Regulgens		R		R							
Cibicidella											
Variabilis		R	R	R	R		R	R	R		F
Dyocibicides											
Biserialis		R	R	R	R						R
Planorbulina											
Mediterranensis		F	R	R	R	F	F	R	R	R	R
Anomalina											
Nonionoides			R			R					R

2700

TABLE 1 (Cont.)

SPECIES (from 100 gms dry matl.)		STATIONS 1	2	4	5	10	11	8	7	6	3
	coarse	15	93	75	1.760	2.700	1.093	200	800	1.033	47
	fine	530	1.665	3.950	28.000	37.300	28.300	6.900	17.400	38.252	276
	total	545	1.758	4.025	29.760	40.000	29.393	7.100	18.200	39.285	323
Elphidium											
Advenum				R			R			R	
Craticulatum		F	R	R		R	R	R	R	R	R
Crispum				R	R	R	R	R	R	R	R
Depressulum				R			R				
Discoidale multiloculum					R						
Imperatrix				R	F	R	F	R	R	R	R
Poeyanum				F							

0- 5%	-	rare	R
5-20%	-	frequent	F
20-35%	-	common	C
35-50%	-	abundant	A
> 50%	-	very abundant	V.A.

V. CONCLUSIONS

By the distribution of various genera, different environments in Gunnamatta Bay were recognizable.

1. The sand bar and entrance to the channel, typified by Discorbis dimidiatus.
2. The deep central region of the bay, typified by Spiroloculina lucida.
3. The northern and southern slopes of the basin, typified by Quinqueloculina spp. However, these two slope areas might reveal differences in environmental conditions after more detailed study.

The limited time available prevented more extensive sampling, and ruled against more specific conclusions.

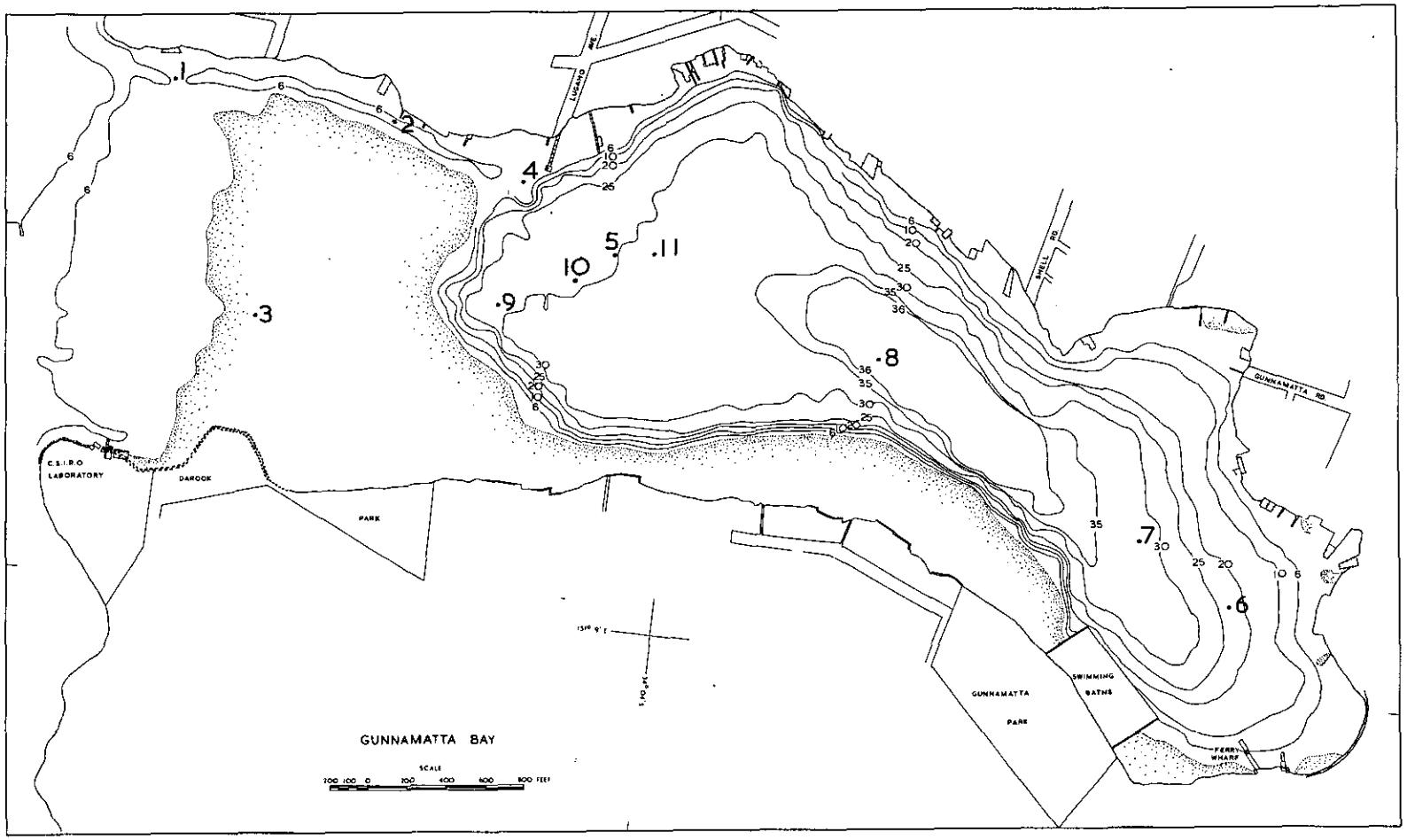

Fig. 1 - Sampling sites in Gunnamatta Bay. Depth contours in feet.

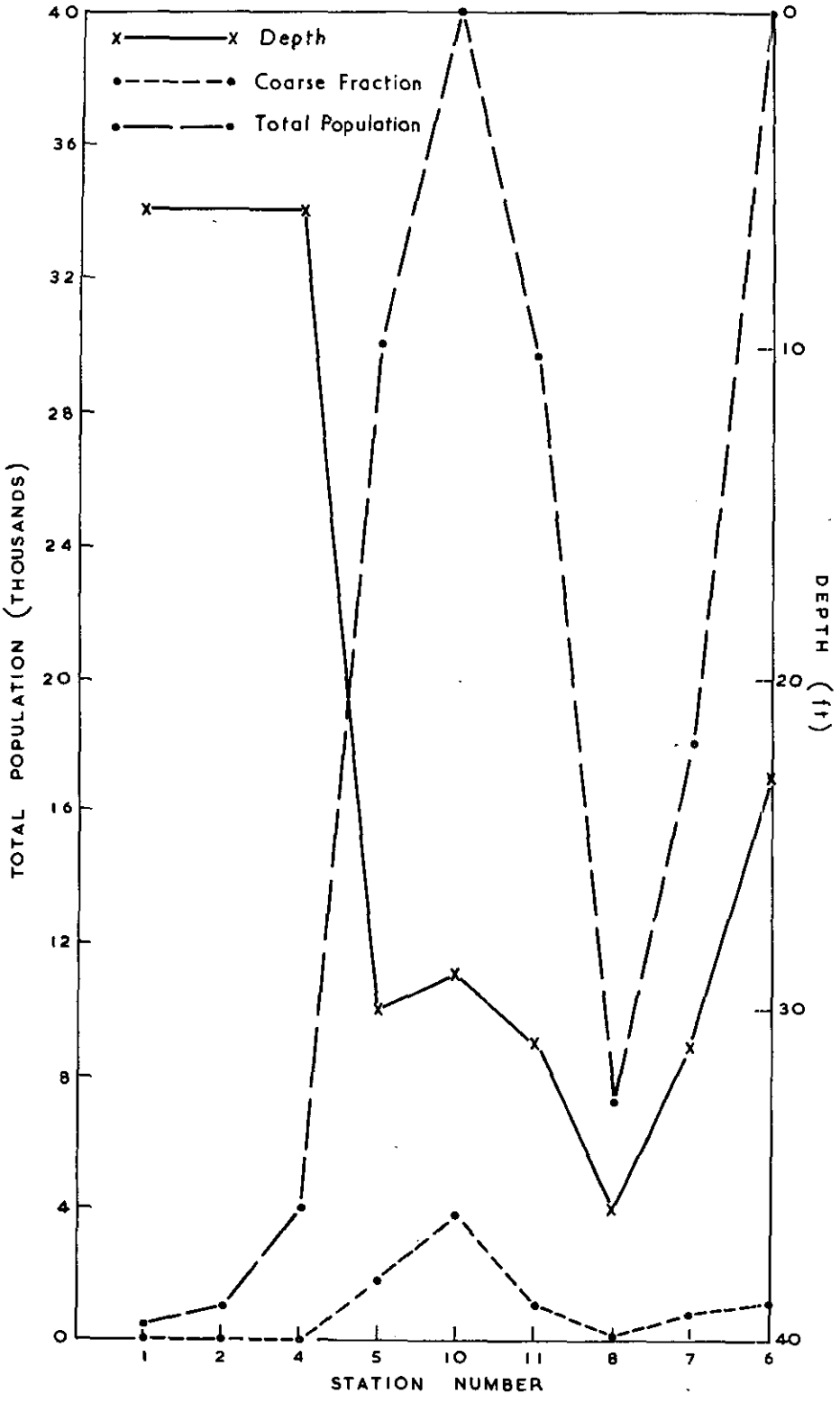

Fig. 2 - Variation with depth of the abundance of the coarse fraction and the total population of foraminiferans at stations in

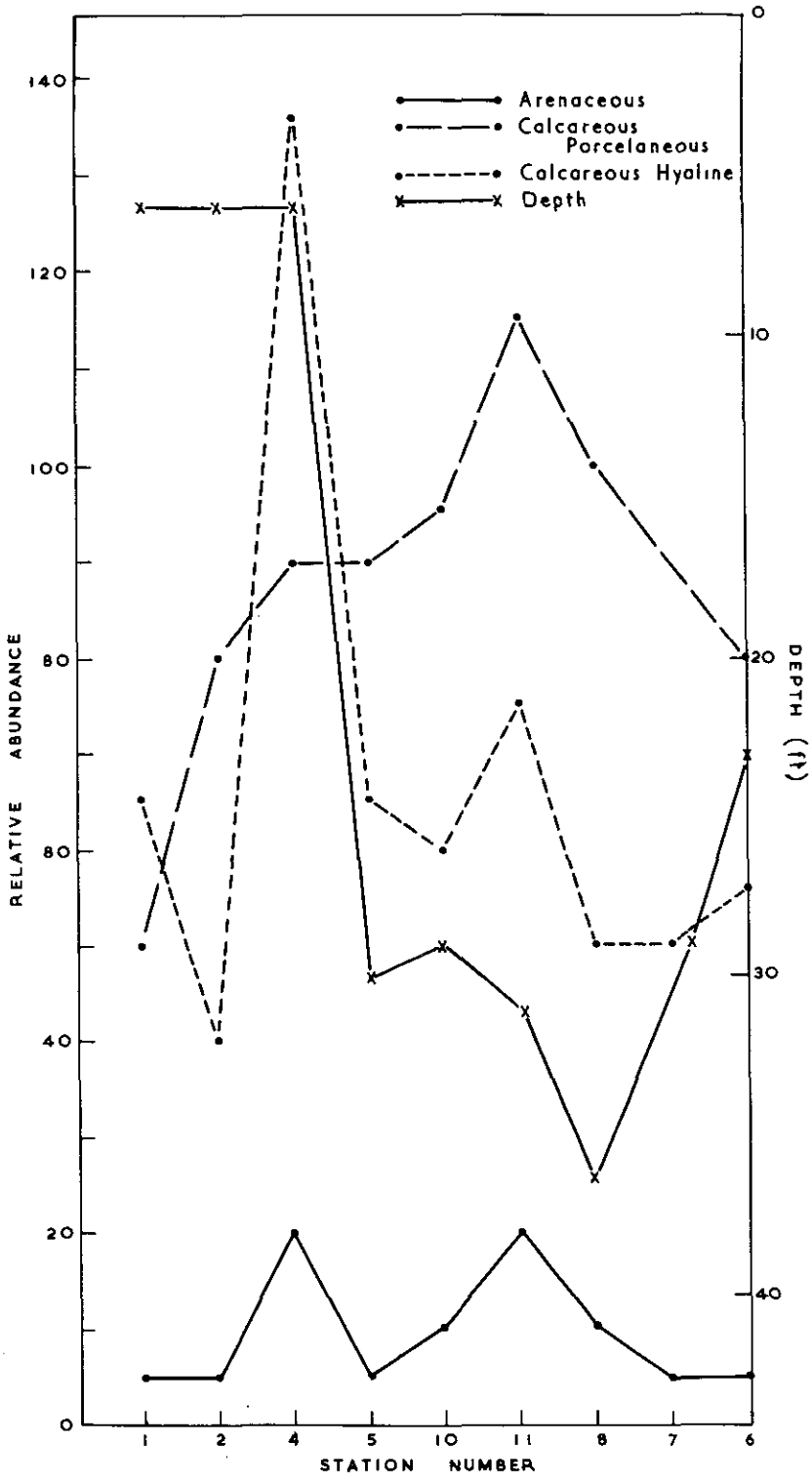

Fig. 3 - Variation with depth of the abundance of walled type foraminiferans at stations in Gunnamatta Bay.

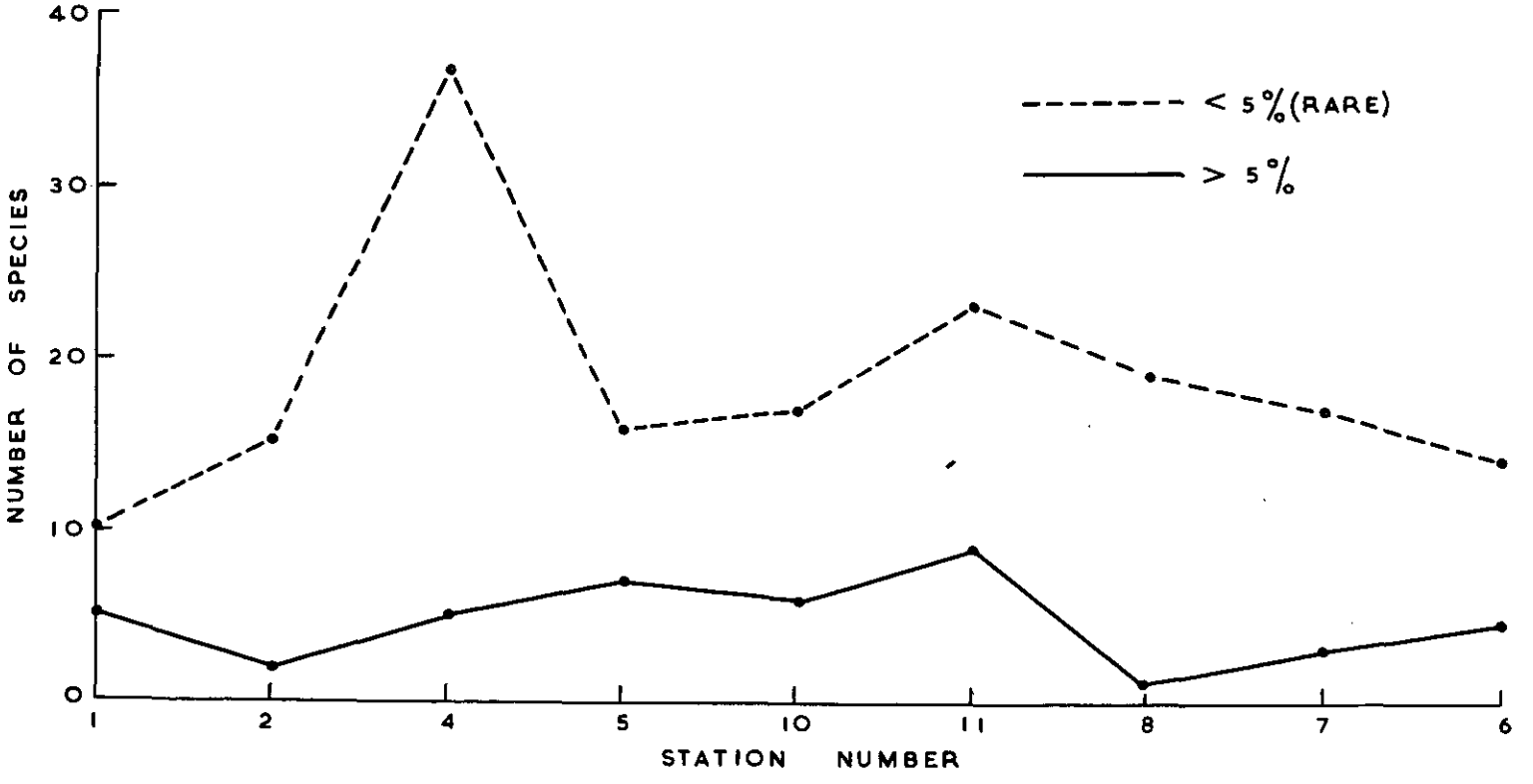

Fig. 4 - Numbers of rare foraminiferan species at stations in Gunnamatta Bay.

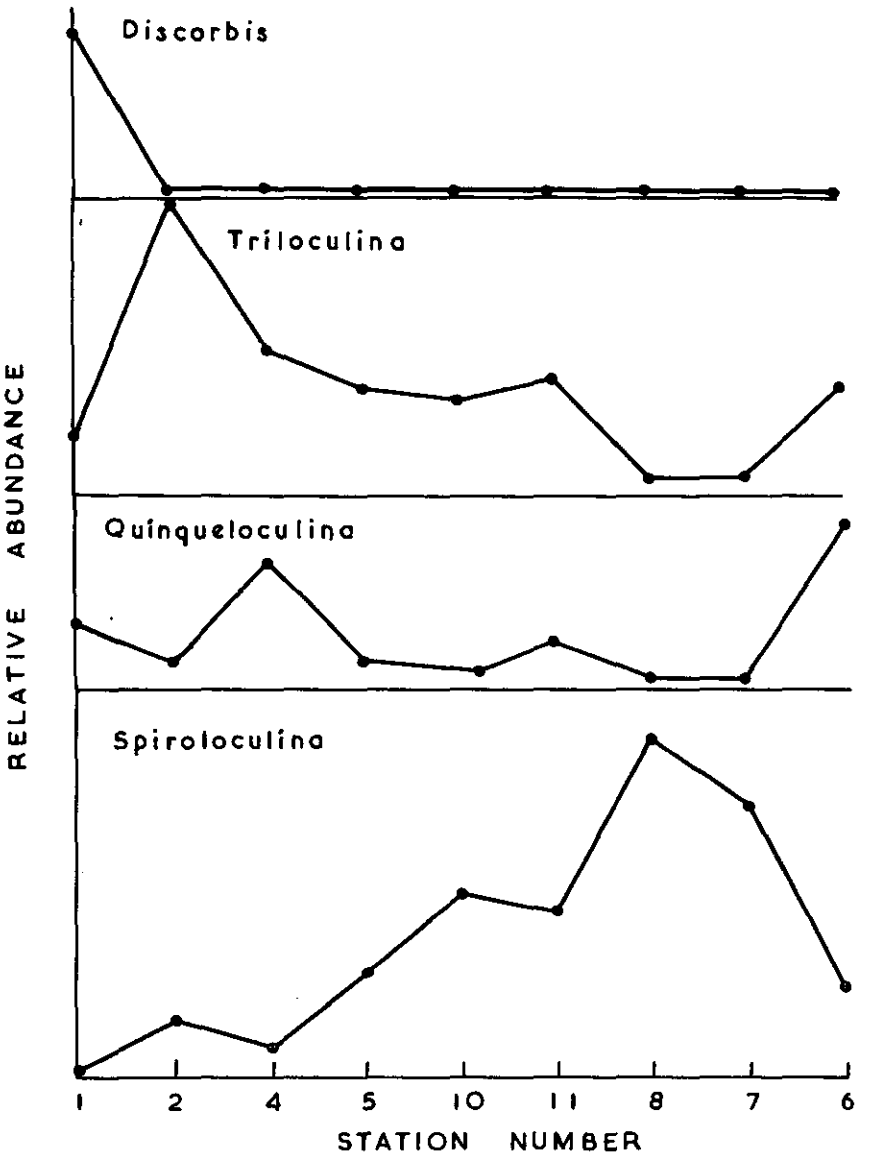

Fig. 5 - Abundance of *Discorbis*, *Triloculina*, *Quinqueloculina*, and *Spiroloculina* at stations in Gunnamatta Bay.

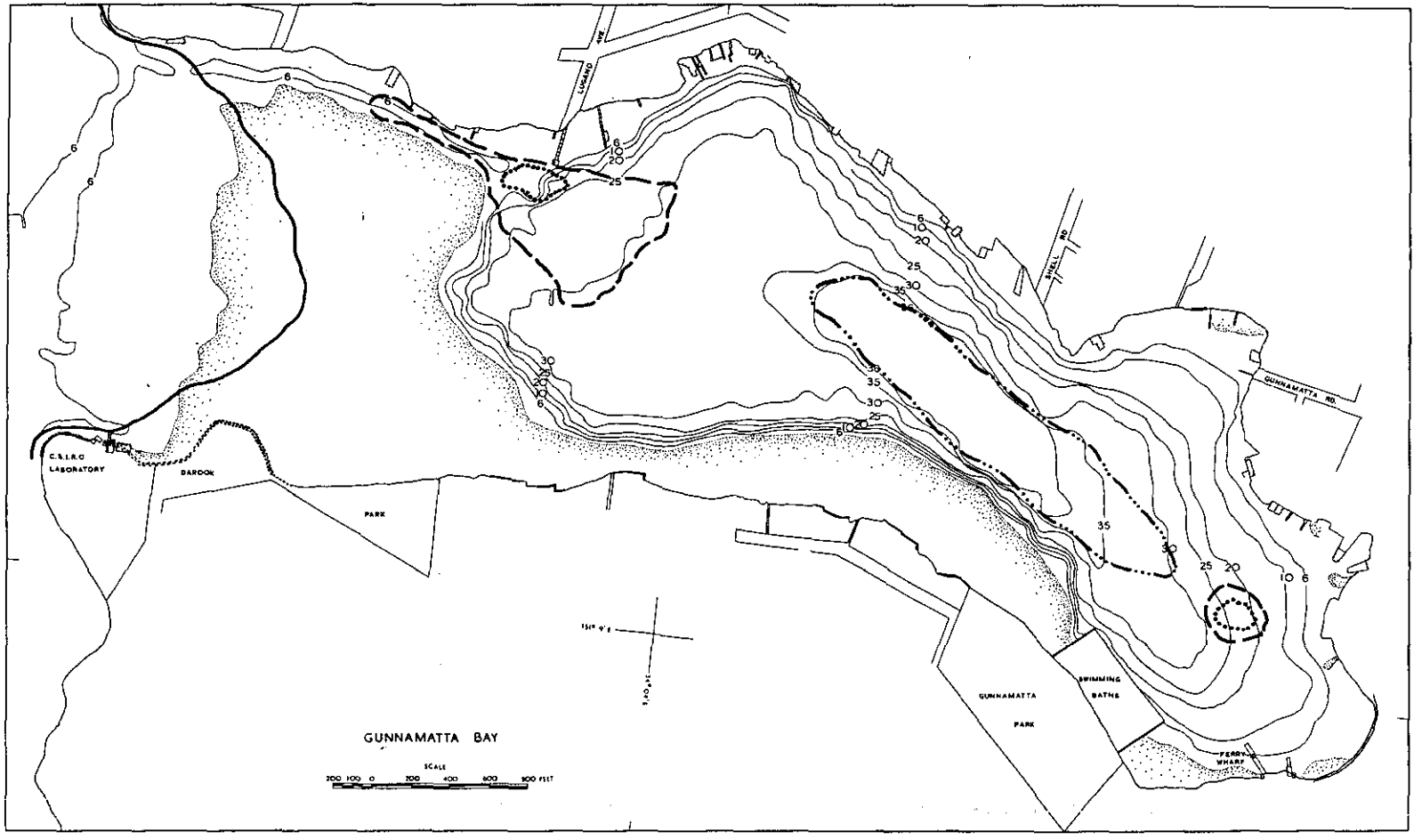

Fig. 6 - Distribution of *Discorbis*, *Triloculina*, *Quinqueloculina*, and *Spiroloculina* in Gunnamatta Bay.
Discorbis _____ *Triloculina* _ _ _ _ *Quinqueloculina* ..... *Spiroloculina* _ ... _

ALGAL PHYSIOLOGY

PART 1. PRIMARY PRODUCTION IN MARINE ALGAE

I. INTRODUCTION

This group investigated the effects of cell density, culture age, and incubation period on rate of production in three species of algae - Dunaliella tertiolecta, a green alga, Isochrysis galbani, a chrysomonad, and Phaeodactylum tricornutum, a diatom.

II. METHODS AND MATERIALS

The method used was that of Jitts (1957), but with some modifications.

The light source for photosynthesis was fluorescent light of 1100 f.c. Millipore and charcoal-filtered seawater was used for all dilutions.

Production was estimated by measuring the amount of $^{14}CO_2$ (supplied as bicarbonate) fixed in a set time, under constant conditions. Bicarbonate initially present in the seawater used was estimated by titrating with 0.01N HCl. Thus, the ratio of labelled to unlabelled CO_2 was known. One microcurie of ^{14}C was used in each experiment. Samples were incubated in light and dark bottles. The dark bottles were controls, the difference in production between light and dark bottles giving net production due to photosynthesis.

After incubation, the cells were harvested by Millipore filtration and the amount of $^{14}CO_2$ fixed determined in a liquid scintillation counter with diphenyl oxazole as the primary scintillator (Jitts and Scott 1961).

Effect of Cell Density on Rate of Production

Samples of Dunaliella culture (in log growth phase) containing 2.37 x 10^7 cells per ml were diluted to the required cell densities and incubated in light and dark bottles for 3 hr.

Effect of Culture Age on Rate of Production

Samples of cultures of different ages - 2, 4, and 6 days - were diluted to 80-200 cells/ml and incubated for 3 hr in light and dark bottles. This experiment was done in triplicate.

Effect of Incubation Time on Rate of Production

A sample of Phaeodactylum culture was diluted to a concentration of 74 cells/ml and aliquots incubated for various times in light and dark bottles.

III. RESULTS

Cell Density

Figure 1 shows that the maximum rate of ^{14}C-fixation occurred at concentrations between 10^4 and 10^5 cells per ml. Other concentrations gave lower rates.

Culture Age

Figure 2 shows that dark bottles frequently gave a higher ^{14}C-fixation than corresponding light bottles. Differences between light and dark bottles were generally very small and the results might have little significance. Cell concentrations used were of the order of 100 cells per ml. The cell density experiments showed that optimum concentrations were between 10^4 and 10^5 cells per ml. These results were not available at the time of the cell age experiments.

Dunaliella and Isochrysis showed a steady decrease in rate of ^{14}C-fixation as the cultures became older. Phaeodactylum, however, showed a steady increase in fixation with age.

The results obtained for each alga depended probably on their growth stage immediately after inoculation. Because a fairly large inoculum (10 ml in 150 ml media) was used, the resulting concentrations of Dunaliella and Isochrysis could have been such that initially they were in the log phase. This would explain the decrease in ^{14}C-fixation, indicating cells passing through log to stationary phase. Phaeodactylum, however, might have been at very low concentration at the time of inoculation. The increase in ^{14}C-fixation with culture age would then correspond to the alga passing from stationary to log phase.

Incubation Time

Figure 3 shows that an incubation time of 30 min gave the maximum rate of ^{14}C-fixation. Rate decreased with time of incubation.

IV. CONCLUSION

The experiments were repeated under better conditions after the School. The results given above were not reproducible and further investigations are needed.

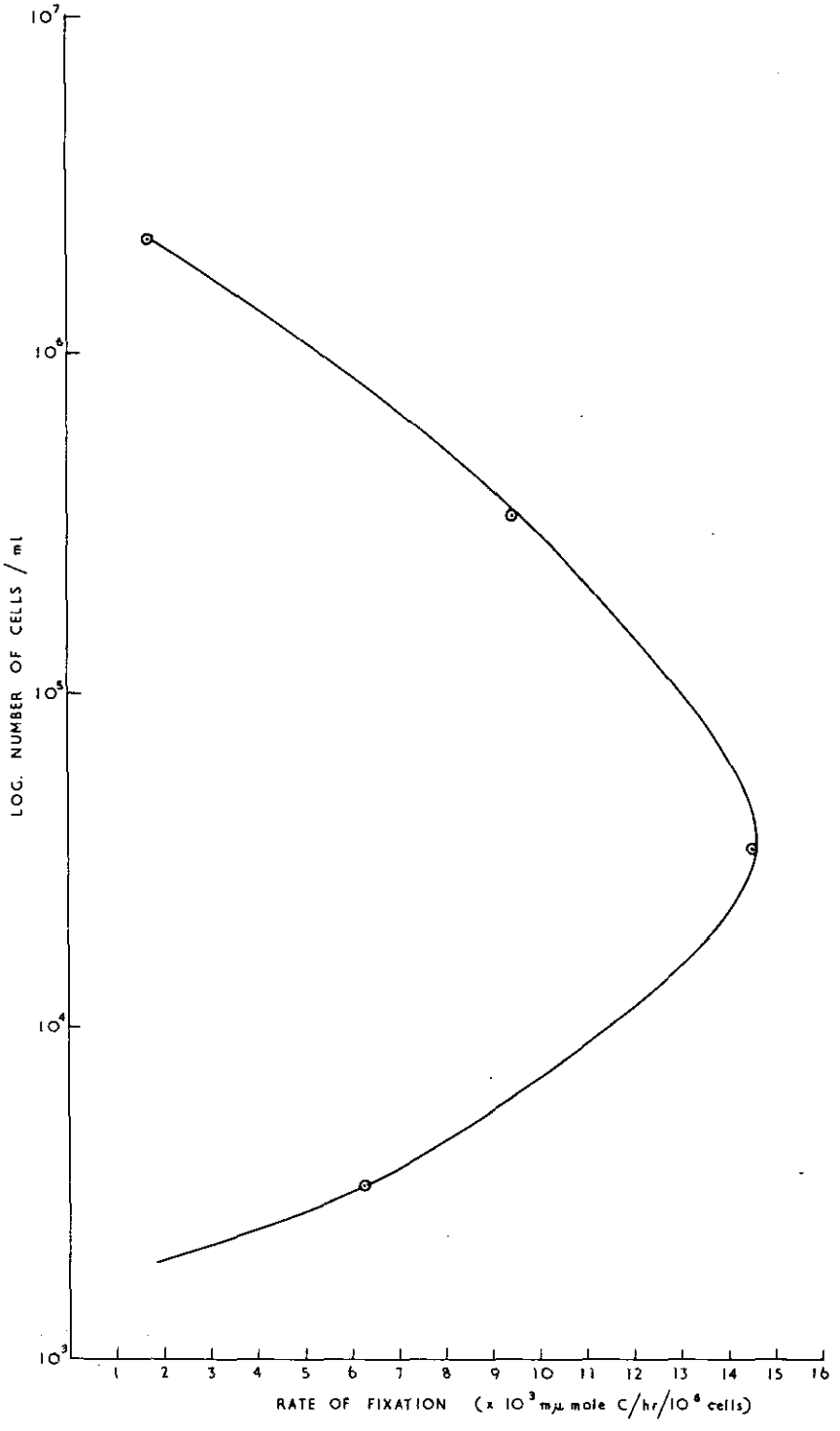

Fig. 1 - Relation between cell density and rate of ^{14}C-fixation.

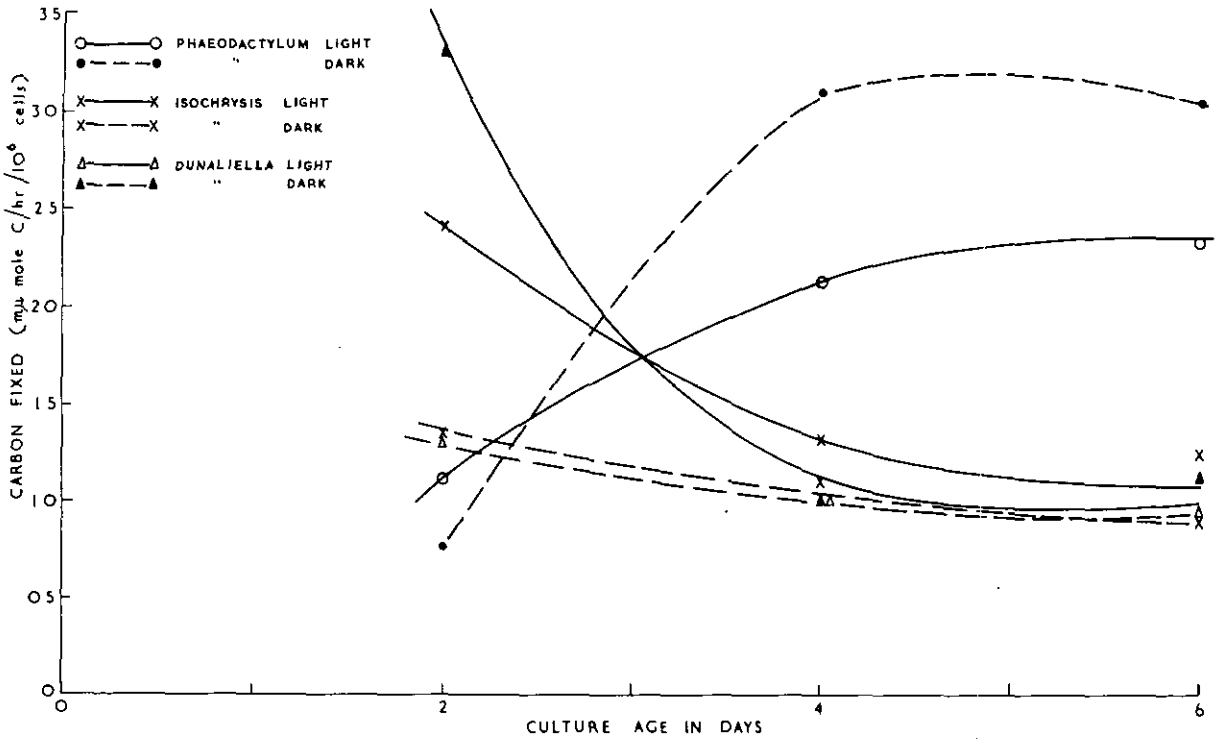

Fig. 2 - Relation between culture age and rate of ^{14}C-fixation.

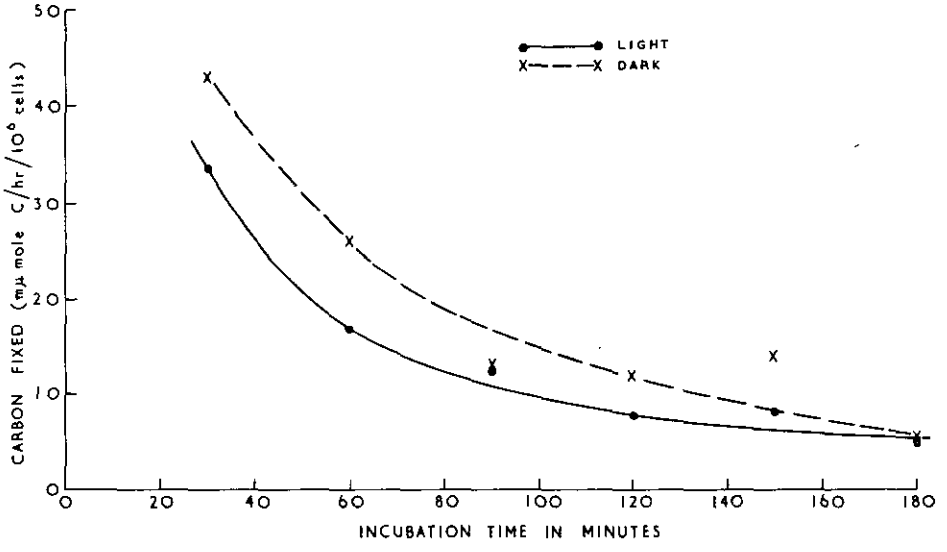

Fig. 3 - Relation between incubation time and rate of ^{14}C-fixation.

V. REFERENCES

JITTS, H.R. (1957).- ^{14}C method for measuring CO_2 uptake in marine productivity studies. CSIRO Aust. Div. Fish. Oceanogr. Rep. 8.

JITTS, H.R., and SCOTT, B.D. (1961).- The determination of zero-thickness activity in Geiger counting of C^{14} solutions used in marine productivity studies. Limnol. Oceanogr. 6, 116-23.

ALGAL PHYSIOLOGY

PART 2. EFFECTS OF pH, BUFFER SYSTEM, AND NITROGEN SOURCE ON THE PHYSIOLOGY OF TWO SPECIES OF PHYTOPLANKTON

I. INTRODUCTION

There are many buffer systems now in common use in biological studies, including some recently developed types. It is very important that when using any one of these, its effects on the physiology of the organism are understood. It is also likely that different organisms will respond differently to different systems. Similarly, the nitrogen source in a substrate will probably influence the physiology of cultured organisms.

The aim of this project was to determine these effects in the green alga Dunaliella tertiolecta, and the diatom, Phaeodactylum tricornutum.

II. METHODS AND MATERIALS

(a) The Effects of Buffer System and pH

The following range of buffer solutions was prepared:-

1. Seawater, internally buffered with HCO_3/CO_3 at pH 8.6 and containing 8.6 g $NaHCO_3$ and 0.16 g K_2CO_3 per litre.
2. 0.5M tris/HCl at pH 7.6, 8.6, and 9.1.
3. 0.5M phosphate buffer at pH 6.4 and 7.5.
4. 0.5M tricine buffer at pH 7.6, 8.6, and 9.1.

The age of cultures used varied as indicated in the Results.

The following measurements were obtained:-

Packed cell volume was measured by the standard method - centrifuging at 3,500 g for 1 hr. Chlorophyll was estimated in a methanol extract spectrophotometrically at 668 and 635nm. Estimates of cell numbers were made using the Petroff Hauser counting chamber.

Gas uptake and evolution were determined by Warburg manometry under both light (2,500 f.c.) and dark conditions, at 20°C. In each case, 1 ml of cells (number and volume noted) plus buffer and artificial seawater were prepared. In the case of phosphate, tris/HCl, and tricine buffers, an external source of CO_2 was enclosed in the flask - viz., 1 ml of 75/25 solution HCO_3 and CO_3 (2.0M), and 0.02 ml of cartase enzyme solution to increase the rate of CO_2 evolution, thus ensuring that the organisms were not inhibited by lack of CO_2.

(b) Nitrogen Source

Three concentrations of each of two commonly used nitrogen sources - NO_3 and NH_4 - were used. One ml of 10^{-3}, 10^{-4}, and 10^{-5} M KNO_3 or NH_4CL was added to the cell solution, buffered with HCO_3/CO_3 buffer at pH 8.6, and measurements taken, manometrically in both the light and the dark. The nitrogen was estimated in duplicate samples at the time of equilibration of the Warburg manometer, and at the completion of the experiment, to give an indication of the nitrogen uptake during the experiment. NH_3-N was estimated by the ninhydrin method, and NO_3-N by catalytic reduction to NO_2, and estimation by the sulphanilamide/N-1 naphthyl ethylene diamine dihydrochloride method.

The aim was to determine the optimum range of concentrations, but time did not permit further studies.

III. RESULTS

(a) Buffer Systems

Table 1 shows the rate of O_2 evolution by D. tertiolecta and P. tricornutum during the light period of measurement.

TABLE 1

THE EFFECTS OF pH AND THREE BUFFER SYSTEMS OF THE PHYSIOLOGY OF DUNALIELLA TERTIOLECTA AND PHAEODACTYLUM TRICORNUTUM

Buffer System	µg O_2 evolved/l.							
	Dunaliella				Phaeodactylum			
	Age of culture	per 10^6 cells	per mm^3 cells	per µg Chl a	Age of culture	per 10^6 cells	per mm^3 cells	per µg Chl a
HCO_3/CO_3 pH 8.6	3 days	3.76 3.51	14.85 12.20	5.50 4.52	3 days	0.52 0.45	3.68 3.17	5.17 4.44
Tris/HCl pH 7.6	"	3.82 3.76	15.10 14.90	5.60 5.50	"	0.59 0.59	4.16 4.10	5.82 5.77
Tris/HCl pH 8.6	"	3.64 3.70	14.40 14.70	5.34 5.43	"	0.56 0.59	3.96 4.16	5.57 5.82
Tris/HCl pH 9.1	"	3.14 2.18	12.40 8.60	4.60 3.20	"	0.60 0.56	4.27 3.98	5.98 5.60

Buffer System	µl O_2 evolved/l							
	Dunaliella				Phaeodactylum			
	Age of culture	per 10^6 cells	per mm^3 cells	per µg Chl a	Age of culture	per 10^6 cells	per mm^3 cells	per µg Chl a
Phosphate pH 6.4	1 day	2.14 3.06	8.46 12.10	1.19 1.72	*5 days	0.24 0.25	1.72 1.74	1.32 1.35
Phosphate pH 7.5	"	4.41 3.32	17.50 13.20	2.46 1.86	"	0.47 0.54	3.31 3.81	2.56 2.95
Tricine pH 7.6	"	6.80 4.03	27.00 16.00	3.81 2.26	"	0.66 0.93	0.46 0.66	0.37 0.51
Tricine pH 8.6	"	7.30 6.35	28.80 24.20	4.07 3.54	"	0.24 0.19	1.72 1.32	1.32 1.02
Tricine pH 9.1	"	5.80 5.80	23.00 23.00	3.24 3.24	"	0.24 -	1.72 -	1.32 -

* 5 day Phaeodactylum in HCO_3/CO_3 buffer - 1.44 µl/mm^3, 0.2 µl/10^6 cells, and 1.11 µl/µg Chl a.

(b) Nitrogen Source

Table 2 shows oxygen assimilation by D. tertiolecta grown on two nitrogen sources.

TABLE 2

OXYGEN UPTAKE BY DUNALIELLA ON TWO NITROGEN SOURCES

Concentration	O_2 evolution µl/hr/10^6 cells	
	NO_3^- Substrate	NH_4^+ Substrate
10^{-3}	0.845	0.825
10^{-4}	1.08	0.825
10^{-5}	0.88	0.808

Table 3 shows variations in NO_3 and NH_4 concentration during the course of the experiment.

TABLE 3

VARIATIONS IN NO_3 AND NH_4 CONCENTRATION DURING THE COURSE OF THE EXPERIMENT

N Source*	Light or Dark	Molarity of Nitrogen Source		
		Before Experiment	After Experiment	Change
$NO_3 10^{-3}$	Light	2.84×10^{-4}	1.9×10^{-4}	-0.94×10^{-4}
	Dark	3.4×10^{-4}	2.6×10^{-4}	-0.8×10^{-4}
$NH_4 10^{-3}$	Light	-	1.4×10^{-4}	?
	Dark	-	4.2×10^{-4}	?
$NH_4 10^{-4}$	Light	1.4×10^{-4}	1.6×10^{-4}	$+0.2 \times 10^{-4}$
	Dark	1.5×10^{-4}	1.8×10^{-4}	$+0.3 \times 10^{-4}$
$NH_4 10^{-5}$	Light	1.8×10^{-4}	1.4×10^{-4}	-0.4×10^{-4}
	Dark	2.1×10^{-4}	1.5×10^{-4}	-0.6×10^{-4}

* 10^{-4} and 10^{-5} NO_3 levels undetectable

IV. DISCUSSION

(a) Buffer Systems and pH

From the results, the following trends are apparent:-

Dunaliella

1. Activity in PO_4 buffer - pH 7.5 > pH 6.4.
2. Activity in tris/HCl buffer — pH 8.6 and pH 7.6, which give about the same activity, > pH 9.1.
3. Activity in tricine buffer - pH 8.6 > pH 7.6 and pH 9.1, which give similar results.
4. Activity in HCO_3/CO_3 seawater is comparable to that in tris/HCl, pH 8.6 and pH 7.6, and PO_4 buffer at pH 7.6.
5. Comparing all systems used, on the basis of activity per cell number and per cell volume, tricine buffer produces the greatest activity with the optimum pH 8.6. With

tris/HCl and PO_4 buffers, activity is reduced, but optimum pH is again 8.6. Moreover, on the basis of oxygen evolved per µg chlorophyll a, tris/HCl is the better system.

Phaeodactylum

1. Activity in PO_4 buffer - pH 7.5 > pH 6.4.
2. Activity in tris/HCl buffer - does not vary with pH.
3. Activity in tricine buffer - pH 7.6 > pH 8.6 and pH 9.1.
4. The activity in buffered artificial seawater (HCO_3/CO_3) was much less in 5-day cultures than in 3-day cultures.
5. It is likely that, because 3-day cultures were more active, the apparent preference for tris/HCl buffer over tricine and PO_4 buffers was simply an age effect.

Comparing the two species, Dunaliella appears to be the more photosynthetically active:- O_2 evolution/10^6 cells 10:1, Dunaliella: Phaeodactylum; O_2 evolution/mm^3 cells 4:1, Dunaliella : Phaeodactylum. However, in O_2 evolution/µg chlorophyll a there was a ratio close to one except using the tricine buffer system (tricine might actually inhibit Phaeodactylum). This might be coincidence but it could have been worthwhile investigating activity in relation to chlorophyll concentration more closely, since it might be that activity can be assessed purely on differences in chlorophyll concentration between cultures of different age.

An important source of error was that cultures were not of the same age. The 1 and 3-day old Dunaliella cultures were both early in the log phase and therefore comparable, but the 5-day old Phaeodactylum culture was approaching the stationary phase. Therefore, when the culture was diluted by half the day before use, it is possible that it had not returned to the log phase as assumed.

(b) Nitrogen Source

No conclusions could be drawn from the results. Nitrate 10^{-4} and 10^{-5} were present in such small quantities that they could not be accurately estimated. For NH_4 10^{-4} there was a gain in NH_4 during the experiment and this might have been due to leaching of nitrogen from the cells during centrifugation thus casting doubt on any further interpretation of data. To overcome this a blank containing no NH_4 or NO_3 substrate should have been run.

ALGAL PHYSIOLOGY

PART 3. GROWTH OF MARINE ALGAE

I. INTRODUCTION

To estimate productivity and changes in the distribution and abundance of marine algae, the potential for rapid growth under suitable conditions must be known. Many of the previous estimates of the growth rates of marine algae have been conflicting.

This project investigated rates of growth of three species of marine algae, and tested different methods of estimating changes in growth that occur in algae, under controlled conditions.

II. METHODS AND MATERIALS

Three species were studied - Dunaliella tertiolecta, a green alga, Phaeodactylum tricornutum, a diatom, and Isochrysis galbani, a chrysomonad.

Inoculations were taken from established cultures in a state of rapid growth and placed into 30 250 ml conical flasks containing 150 ml of sterile "medium f" (Wisely and Purday 1963). The cultures were illuminated continuously at a temperature of 20°-23°C with "daylight" fluorescent light (1100 f.c.). The atmosphere was not enriched with CO_2. The flasks were shaken several times a day to keep cells in suspension.

Increase in growth of the cultures was measured by taking estimates of cell number per ml twice a day with a Petroff-Hauser counter. Three culture flasks of each species were counted in this way for a period of 150 hr.

Chlorophylls and packed cell volumes were measured by combining several of the cultures, and concentrating the cells by centrifuging and resuspending in buffered artificial seawater (pH 8.5). This suspension was divided into several portions, so that chlorophyll, photosynthesis, and packed cell volumes could be estimated.

Chlorophyll was extracted with methanol, and the optical density of this solution was measured in a Unicam spectrophotometer. The proportions of chlorophylls a, b, and c, in µg chlorophyll per ml, were derived from the equations -

For chlorophylls a and b

$$\text{chl } a = 13.71 \times e_{665} - 2.12 \times e_{645} \qquad \text{Dunaliella}$$

$$\text{chl } b = -4.4 \times e_{665} + 21.05 \times e_{645}$$

For chlorophylls a and c

$$\text{chl } a = 13.31 \times e_{663} - 0.27 \times e_{630}$$

$$\text{chl } c = -8.37 \times e_{663} + 51.72 \times e_{630}$$

Phaeodactylum and Isochrysis

The photosynthetic rate was measured by Warburg manometry, suspending the cells in seawater-HCO_3/CO_3 buffer at 20°C and measuring O_2 exchange during light and dark pulses of 15 min duration.

III. RESULTS

Growth curves were plotted for all three species of algae, on log/linear paper. Dunaliella and Phaeodactylum (Fig. 1) showed a long period of logarithmic growth, with generation times of 20 hr and 12 hr, respectively. Isochrysis (Fig. 1) however, stayed in a lag phase for most of the period, undergoing a short period of rapid growth with a generation time of 14 hr, followed by a decline in cell numbers.

Due to a slight precipitation of salts in medium f, the packed cell volumes could not be measured accurately.

Chlorophyll concentration was estimated in Dunaliella (Fig. 2) and Phaeodactylum (Fig. 3), but Isochrysis did not grow sufficiently. A general increase in absolute chlorophyll per ml was observed in Dunaliella (Fig. 3), while the chlorophyll content per 10^6 cells was difficult to interpret on the basis of the few readings that were made (Fig. 3).

The estimation of photosynthetic and respiratory rates was performed on all species. However, Phaeodactylum at 44 and 92 hr and all Isochrysis cultures were too low in cell numbers to be measured by this technique. An increase in photosynthetic rate per 10^6 cells for Dunaliella was observed. However, the respiration rate was at a minimum for the cells in the log phase, indicating that the cells were smaller than those of the original inoculum or those in the stationary phase. Therefore, these results are not presented.

IV. DISCUSSION

The growth curves for each species indicate a different response by the cells to inoculation into a new medium. All species were expected not to go through an appreciable lag phase after inoculation. However, Isochrysis did stay in a lag phase, indicating that the medium differed from the old environment. While the rapid period of cell growth was short, it did show that the medium was capable of supporting growth. Apparently, Isochrysis was slower to adapt to these new conditions.

V. REFERENCES

WISELY, B., and PURDAY, C. (1963).- A culture method for marine diatoms and flagellates. Tuatara 11, 20-6.

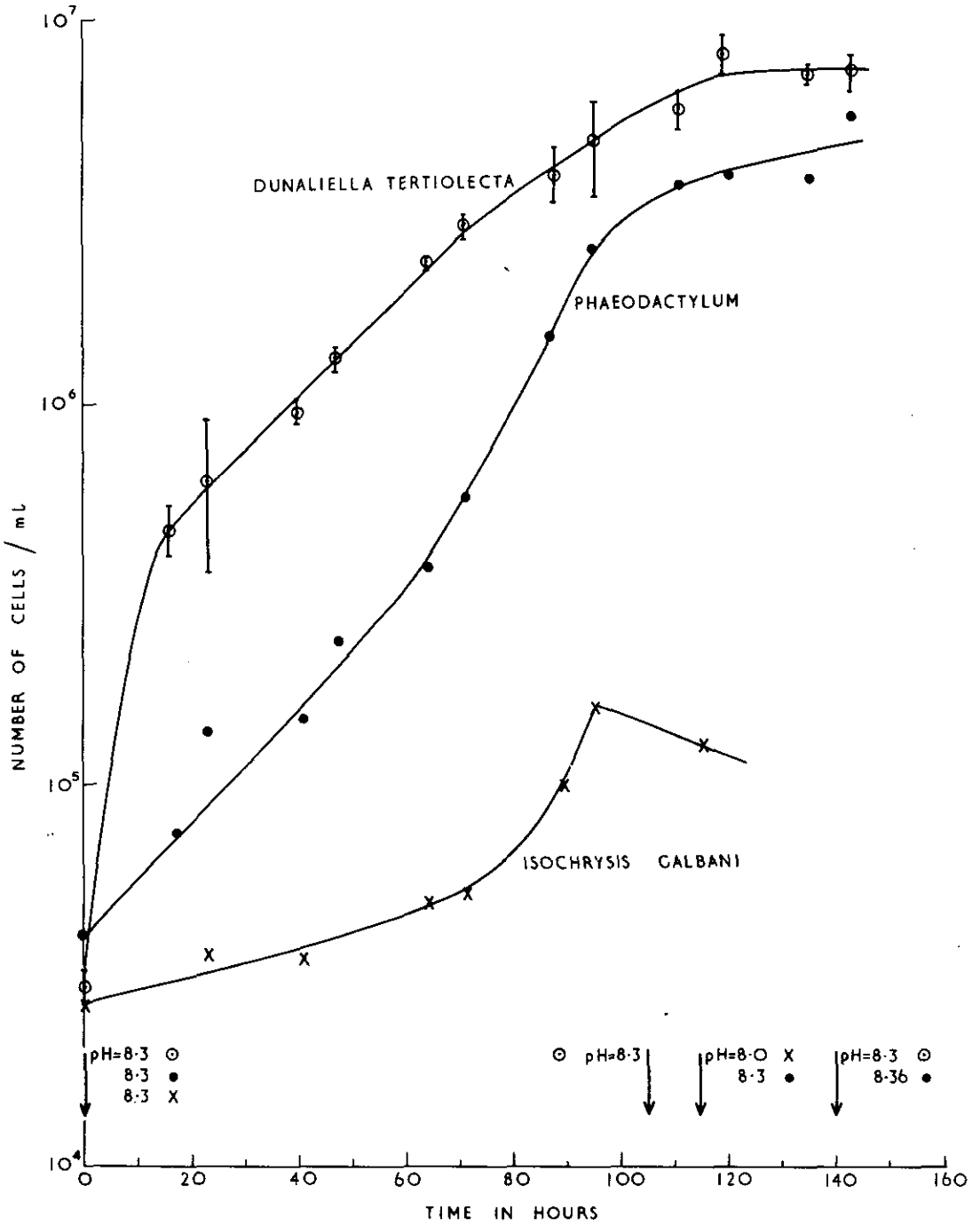

Fig. 1 - Increase in cell number with time in *Dunaliella tertiolecta*, *Phaeodactylum tricornutum*, and *Isochrysis galbani*.

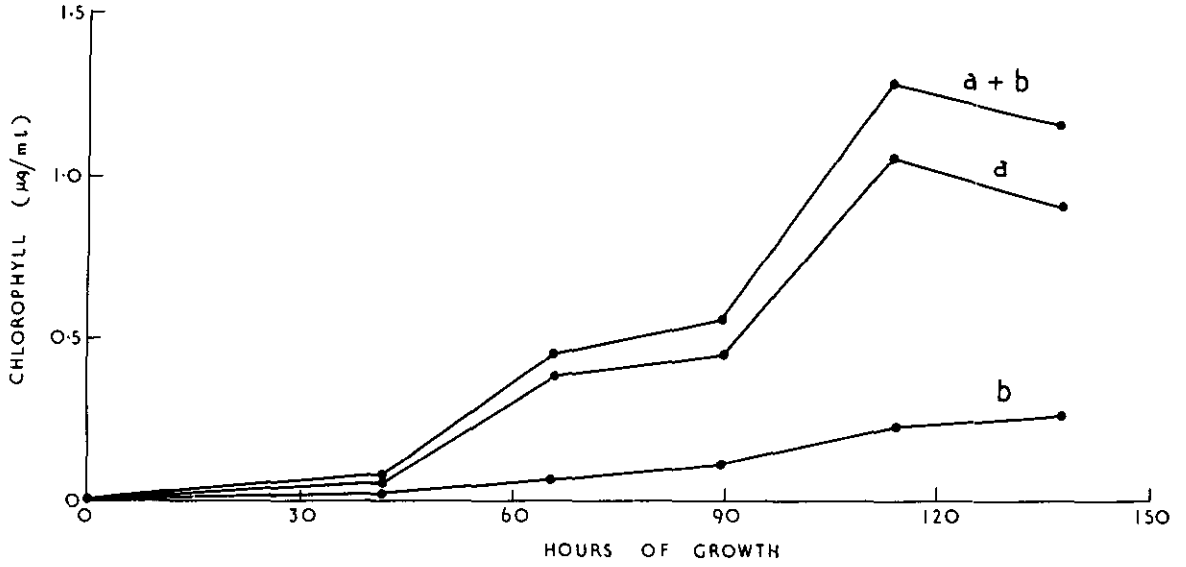

Fig. 2 - Increase in concentration of chlorophylls with time in *D. tertiolecta*.

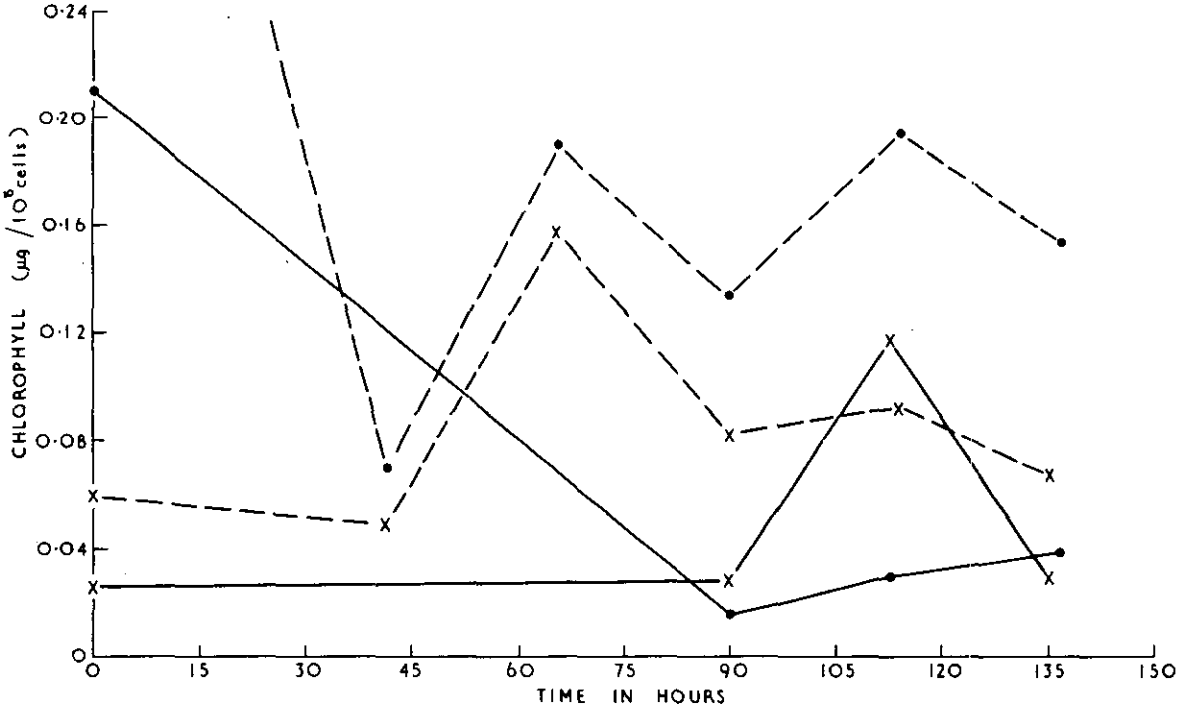

Fig. 3 - Changes in chlorophyll,concentration with time in cultures of *D. tertiolecta* and *P. tricornutum*.

●———● *Phaeodactylum* total chlorophyll
●- - - -● *Dunaliella* total chlorophyll
X———X *Phaeodactylum* chl *a*/ chl *c*
X- - - -X *Dunaliella* chl *a*/ chl *c*

PIGMENTS IN MARINE ALGAE

I. INTRODUCTION

Spectroscopic analysis of photosynthetic pigments is often used to assay plant biomass and phytoplankton productivity. However, this technique has failings; it does not distinguish between either the major pigments, or the major pigments and their breakdown products. A technique using thin-layer and paper chromatography which overcomes these problems has been developed.

This group, in a series of field and laboratory experiments, used thin-layer and paper chromatography to study the characteristics of the main groups of chlorophylls, carotenes, and their degradation products.

II. METHODS AND MATERIALS

(a) Extraction of Pigments

1. Unicellular algae were harvested by either spinning at 1500-2000 g for 5 min in a Sorvall RC2B refrigerated centrifuge, or in a GSA Rotor, or for larger volumes, in the continuous plankton centrifuge. The pellet was then extracted with 90% acetone until no colour remained.

Rock samples were crushed with a hammer and chisel in 100% acetone.

Multicellular algae were first soaked in methanol to soften the thallus, then extracted in 100% acetone by homogenising in a Sorvall Omnimix at speed 3 for 1 min.

2. When no further colour could be extracted, the debris was spun down at speed 9 in the large swing-out head of an M.S.E. Minor bench centrifuge.

3. The acetone extract was added to an equal volume of peroxide-free diethyl ether. Five to ten volumes of 10% (w/v) NaCl were added to the acetone/ether mixture thus transferring pigments to the ether phase. The ether fraction was taken off in a separating funnel and concentrated under a nitrogen stream (oxygen-free).

4. The concentrated extract was stored in a stoppered flask, in the dark, at -15°C.

(b) Chromatography

(i) Paper

Concentrated pigments in ether were spotted onto 22 cm^2 Whatman No. 3MM paper. Solvents used were 4% n propanol in 60-80°

petroleum ether for the first dimension, and 30% chloroform in 60-80° petroleum ether for the second dimension.

Chromatography chambers were equilibrated with their solvents for 5 min.

Chromatograms were run in the dark to prevent photolytic breakdown of pigments.

Resulting chromatograms were examined under u.v. light to locate chlorophylls a and b, chlorophyllides a, b, and c, pheophytins, and pheophorbides. Non-fluorescent pigments were located in daylight.

(ii) Thin-layer

Preparation of plates

Sucrose plates were prepared with 'Sunny Cane Superfine' icing sugar (5% cornflour). Sugar was dried at 90°C for 15-30 min, then sieved twice through a 200 mesh sieve. Thirty grams of this sugar were added to 50 ml 60-80° petroleum ether and homogenised for 15 sec at speed 3 in the Sorvall Omnimix. The plates were then spread using a glass rolling pin technique and dried in a Desaga Vacuum Desiccator.

Polyethylene plates were prepared with Dow (U.S.) polyethylene powder - 15 g to 90 ml of A.R. acetone. The slurry was homogenised and spread as above.

Cellulose plates were prepared using 15 g of Schering-Ag cellulose powder in 75 ml H_2O. Plates were prepared as above.

Solvents

Solvent systems used were -

Plate	1st Dimension	2nd Dimension
Sucrose	0.6% n propanol in 60-80° petroleum ether	12% chloroform in 60-80° petroleum ether
Polyethylene	90% acetone in water	"
Cellulose	20% acetone in 60-80° petroleum ether	"

All thin-layer chromatography chambers were lined with filter paper and equilibrated for 5-10 min.

Spotting

Pigments were spotted onto the plates in the usual manner. Examination of the plates after running was as for paper chromatography.

Uses of thin-layer plates

The sucrose plate was used to separate all the carotenoids and chlorophylls of all marine algae.

The cellulose plate was used to separate degradation products of chlorophylls a and b. With a solvent of lutidine-water (10:7) chlorophyllides a and c were separated on cellulose plates.

The polyethylene plate was used to separate chlorophylls c^1 and c^2, pheophytins c^1 and c^2, chlorophyll a, pheophytin a, pheophorbide a, and chlorophyllide a.

(c) Specific Pigment Isolation and Determination

(i) Breakdown Products

Pheophorbides and chlorophyllides were produced by extracting algae in 60% acetone and incubating in the dark for 3 hr. This allowed the action of the enzyme chlorophyllase.

Pheophytins were produced by acidifying a 90% acetone extract with two drops of 5M HCl.

(ii) Elution of Paper Chromatograms

Pigment zones were cut from completed chromatograms and shaken with an appropriate solvent.

Chlorophyll b	-	acetone
Chlorophyll c	-	methanol
Carotene	-	petroleum ether or ethanol

Eluted pigments were examined spectrophotometrically.

(iii) Elution of Thin-Layer Chromatograms

Pigment zones were scraped from plates into an appropriate solvent. After 5 min this was spun down and the supernatant examined spectrophotometrically.

(iv) Spectrophotometry

For single wavelength measurements, extinction values were determined using a Unicam SP500 spectrophotometer calibrated with a didynium filter.

Full absorption spectra were determined on a Unicam SP700 double-beam, recording spectrophotometer calibrated with a hydrogen lamp.

III. RESULTS

(a) Laboratory Studies

The pigments present in the marine representatives of the main groups of algae and in the higher plant are given in Table 1.

TABLE 1

PIGMENTS FOUND IN THE VARIOUS CLASSES OF ALGAE AND IN A HIGHER PLANT

Pigment	Organism								
	Green Alga	Diatom	Red Alga	Cryptomonad	Spinach	Dinoflagellate	Blue-green alga	Chrysomonad	Brown Alga
Chlorophyll a	+	+	+	+	+	+	+	+	+
Chlorophyll b	+				+				
Chlorophyll c		+		+		+		+	+
Carotene	+	+	+	+	+	+	+	+	+
Fucoxanthin		+		+				+	+
Neofucoxanthin		+		+				+	+
Diadinoxanthin		+		+		+		+	+
Diatoxanthin		+		+		+		+	
Peridinin						+			
Lutein	+		+		+				
Neoxanthin	+				+				
Violoxanthin	+				+				

Such a distribution serves to emphasize possible phylogenetic relationships among major taxonomic groups and in some cases permits the identification of certain types of algae in field samples. For example, the presence of peridinin and dinoxanthin implies that the sample contains dinoflagellates, and chlorophyll b implies the presence of green algae.

From paper chromatograms of the extracts from all the major algal groups the R_f values characteristic of each pigment where determined. The average values are presented in Table 2. Figure 1 shows the distribution of the pigments on a two-dimensional chromatogram and their approximate R_f values.

TABLE 2

PIGMENT AND PIGMENT BREAKDOWN PRODUCT R_f VALUES

(a) Paper Chromatography

Pigment	Colour	R_f 1st Dimension	R_f 2nd Dimension
Chlorophyll a_1	Blue-green	0.78	0.33
Chlorophyll a	Blue-green	0.76	0.06
Chlorophyll b_1	Olive-green	0.65	0.14
Chlorophyll b	Olive-green	0.61	0.02
Chlorophyll c	Light green	0.10	0
Carotene	Orange	0.97	0.98
Lutein	Yellow	0.78	0.74
Diatoxanthin	Yellow	0.71	0.56
Diadinoxanthin	Yellow	0.70	0.47
Violaxanthin	Yellow	0.67	0.40
Fucoxanthin	Deep orange	0.65	0.33
Neofucoxanthin	Deep orange	0.65	0.09
Peridinin	Red	0.58	0.29
Neoperidinin	Red	0.58	0.28
Neoxanthin	Yellow	0.49	0.09
Unknown xanthophyll	Pink	0	0
Unknown xanthophyll	Red	0.83	0.25
Breakdown Product			
Pheophytin a	Grey-green	1.0	0.95
Chlorophyllide a	Blue-green	0.25	0
Chlorophyllide b	Blue-green	0.17	0
Pheophorbide a	Grey	0	0
Pheophorbide b	Grey	0	0

TABLE 2 (Cont.)

(b) Thin-Layer Sucrose Plate

Pigment	R_f 1st Dimension	R_f 2nd Dimension
Chlorophyll a	0.64	0.27
Chlorophyll b	0.58	0.10
Carotene	1.00	0.93
Lutein	0.55	0.60
Violaxanthin	0.55	0.45
Neoxanthin	0.50	0.20

(c) Thin-Layer Polyethylene Plate

Pigment	R_f 1st Dimension
Lutein	0.75
Chlorophyll c^1	0.37
Chlorophyll c^2	0.27
Chlorophylls a and b	0.77
Fucoxanthin	0.50
Peridinin	0.97
Breakdown Product	
Pheophytin a	0.62
Pheophytin b	0.70
Pheophytin c	0
Chlorophyllide a	0.95
Chlorophyllide b	0.95
Pheophorbide a	0.90
Pheophorbide b	0.89

The chromatograms obtained using sucrose plates correspond to those obtained on paper, but the separation of pigments by the former method is more complete and very small amounts of extract can be separated effectively.

Thin-layer polyethylene chromatograms of some of the breakdown products of chlorophylls were prepared. The R_f values of these products and their colours under white and u.v. light are given in Table 3.

TABLE 3

R_f VALUES AND COLOURS OF SOME CHLOROPHYLL DECOMPOSITION PRODUCTS

Decomposition Product	R_f	Colour	u.v. Colour
Pheophytin a	0.80	Grey	Red
Pheophytin b	0.70	Yellow	Pink
Pheophytin c	0	Grey-green	-
Pheophorbide a	0.25	Grey	Dark
Pheophorbide b	0.22	Dark	Orange

The absorption spectra of many of the algal extracts were determined and on these curves, peaks and inflexions corresponding to the constituent pigments could be distinguished. One example (the absorption spectrum of the brown alga, Hormosira) is shown in Figure 2.

Many of the major pigment zones were eluted from paper and polyethylene chromatograms for identification of absorption spectra. Figure 3 shows the absorption spectra for these pigments; knowledge of the absorption spectra of individual pigments enables a rough estimation to be made of the relative contribution of the constituent pigments to the gross absorption spectrum of an algal extract. In addition, this can be done quantitatively by means of equations, from the extinction coefficients and absorptions of pure pigments in the visible spectrum. The concentrations of these pigments in an extract can be determined from the absorption spectrum.

Iodine Isomerization of Fucoxanthin

Pure samples of leucoxanthin were prepared from Sargassum sp. by paper chromatography. A solution of approx. 15 µg/ml in hexane was prepared and an absorption spectrum determined.

An iodine equivalent of 0.75% of the concentration of fucoxanthin was added and the solution exposed to diffuse light.

The reaction was followed by determining the extinction of the solution at 448.7 nm every 10 min. Equilibrium was reached within 60 min and the absorption spectrum of this equilibrium solution determined.

Figure 4 shows the resultant changes in absorption spectrum, i.e. a slight shift of the maximum absorption wavelengths towards the blue end of the spectrum and the appearance of a characteristic cis-peak at 329 nm.

(b) Field Studies

(i) Brown Algae on a Rock Platform

A survey of the pigments of all the brown algae found on a rock platform was carried out to discover if all the algae contained the same pigments and, if not, whether differences in pigment composition could be related to taxonomic position and/or zone on the rock platform. Eight species of brown algae were studied.

1. Since similarities in pigment composition between closely related major groups of algae have been observed, it is conceivable that variations in pigment distribution within groups if any, might reflect phylogenetic relationships. The names and broad classification of the brown algae studied are as follows:

Phylum Phaeophyta
- Class Isogeneratae
  - Order Dictyotales
    - *Dictyopteris* sp.
    - *Padina* sp.
    - *Pocockiella* sp.
- Class Heterogeneratae
  - Order Dictyosiphonales
    - *Colpomenia* sp.
  - Order Laminariales
    - *Ecklonia radiata*
- Class Cyclosporeae
  - Order Fucales
    - *Hormosira* sp.
    - *Sargassum* sp.
    - *Phyllospora* sp.

2. Differences in pigment composition between major groups of marine algae have been related to their tendency to be located at different depths in the water and consequently to the amount and nature of light reaching them. Within the brown algae, variations in type or proportion of photosynthetic pigments, if they exist, may be associated with their position in the littoral or sub-littoral zone. The distribution of brown algae on the rock platform is shown in Figure 5.

Paper chromatograms of extracts from the eight algae were prepared. The distribution of their constituent pigments is given in Table 4.

TABLE 4

DISTRIBUTION OF PIGMENTS IN BROWN ALGAE

Pigment	Dictyopteris	Padina	Pocockiella	Colpomenia	Ecklonia	Hormosira	Sargassum	Phyllospora
Chlorophyll a	+	+	+	+	+	+	+	+
Chlorophyll c	+	+	+	+	+	+	+	+
Carotene	+	+	+	+	+	+	+	+
Vialoxanthin	+	+		+	+	+	+	+
Fucoxanthin	+	+	+	+	+	+	+	+
Neofucoxanthin	+	+	+	+	+	+	+	+
Unknown orange	+	+	+					
Unknown yellow	+	+						

All the algae have the same pigments except for Pocockiella which lacks violaxanthin and has an unknown orange pigment, and Dictyopteris and Padina which both have additional unknown orange and yellow pigments. The R_f values for the unknown orange compound are approximately the same for the three species (R_f D_1 = 0.45, R_f D_2 = 0.14). Similarly, the R_f values for the unknown yellow compound correspond in the two species (R_f D_1 = 0.52, R_f D_2 = 0.17). The orange compound may be a third type of fucoxanthin. It is interesting that these additional compounds are found only in the order Dictyotales.

Absorption spectra were obtained for the eight extracts. Superficially all absorption curves appeared very similar. Using equations based on the absorption of pure chlorophylls a and c, the relative concentrations of these pigments was calculated for Sargassum. The ratio of the concentration of chlorophyll a to the concentration of chlorophyll c was found to be 7.23.

(ii) Rock Samples in the Intertidal Zone

Limpets and other molluscs feed on algae living on rock surfaces in the intertidal zone. Pigment studies were made to determine the algae and other organisms living on these surfaces.

Samples of rock were taken from four areas -

1. Main limpet zone - from horizontal rock surface of elevation about 5 ft.
2. Main limpet zone - from vertical rock surface of elevation about 5 ft.
3. Low water level - horizontal surface, elevation about 4 in.
4. Splash zone - 45° surface elevation about 7 in.

Results of analyses of these samples are shown in Table 5.

TABLE 5

PIGMENTS IN ROCK SAMPLES FROM THE INTERTIDAL ZONE

Sampling Area	Description of Sample	Pigments	Suspected Organisms
1.	Yellow sandstone with grey-green surface. Distinct green zone about 0.5 cm below rock surface. Limpets present.	Carotene, lutein, chlorophyll a, fucoxanthin, neofucoxanthin, chlorophyll c, myxoxanthin, pheophytin c, and astaxanthin.	Diatoms, blue-green algae, brown algae, crustaceans, and possibly barnacles.
2.	Sandstone with dark red layer on surface. Same distinct green layer as found at 1. Limpets and other mulluscs present.	Carotene, lutein chlorophyll a, fucoxanthin, neofucoxanthin, chlorophyll c, and pheophytin c.	Diatoms, blue-green algae, and brown algae.
3.	Finer-grained grey sandstone. Dark red and black layers on surface. No green zone. Limpets and barnacles present.	Carotene, chlorophyll a, diadinoxanthin, fucoxanthin, neofucoxanthin chlorophyll c, and pheophytin c.	Diatoms, red algae, and possibly blue-green algae.
4.	Orange sandstone with black crystalline-like layer on surface. No green zone evident.	Pheophytin a and carotene. Trace of chlorophyll a, pheophytin c, and degradation products.	Little or no living matter.

Where there were no limpets there was no evidence of living algae. Only degradation products were found. This was in the splash zone where there would be considerable dehydration in summer.

Green algae were absent.

(iii) Pigments in Seawater Samples

Samples were taken in Gunnamatta Bay, in the channel entering the Bay, and in the open sea.

Figure 6 shows pigments present in these samples. There was little active chlorophyll present in Gunnamatta Bay. This correlates well with another groups finding of low ^{32}P in the Bay. The large amounts of chlorophylls a and c and peridinin in the channel correlates well with the large number of dinoflagellates found in the sample from that station.

(iv) Pigments in Zooplankton

Zooplankton samples were collected from the open sea for pigment analysis. Copepods made up a large proportion of the catch. A small number of pteropods with green interstitial masses were also present. Larger gelatinous organisms (chaetognaths and medusae) present in the sample were removed before pigment extraction.

At least four carotenoids were detected in the copepod sample using two-dimensional paper chromatography method. Table 6 gives R_f values for these.

TABLE 6

R_f VALUES OF CAROTENOIDS IN A COPEPOD SAMPLE

Description	R_f	
	1st Dimension	2nd Dimension
Yellow	1.00	0.95
Pink	0.86	0.97
Pink - major carotenoid	0.76	0.86
Pink	0.65	0.67

The absorption maxima of both the total extract and the major carotenoid were at 472-473 nm.

The four carotenoids separated came from large numbers of salmon tinted copepods. The yellow pigment was a carotene type compound and the pink zones were probably esters of astaxanthin. A small amount of chlorophyllide was detected. This was probably a breakdown product of chlorophyll from phytoplankton eaten by pteropods present in the sample.

IV. CONCLUSIONS

Paper chromatography and sucrose thin-layer chromatography give the same type of separation for chlorophylls and carotenoids. Sucrose plates however, have the advantage that very small amounts of pigment can be used and separation of pigments is better.

Paper and sucrose plates do not separate degradation products of chlorophylls. To separate these polyethylene plates are used, and these also completely separate chlorophylls $\underline{c}^1$ and $\underline{c}^2$.

Chromatographic techniques are a tool for phylogenetic classification and identification of most types of plant chlorophylls and carotenoids.

This project has given some idea of the variety of algal pigments adapted to light absorption at the blue end of the spectrum. This correlates well with the lack of red light reaching phytoplankton in the sea.

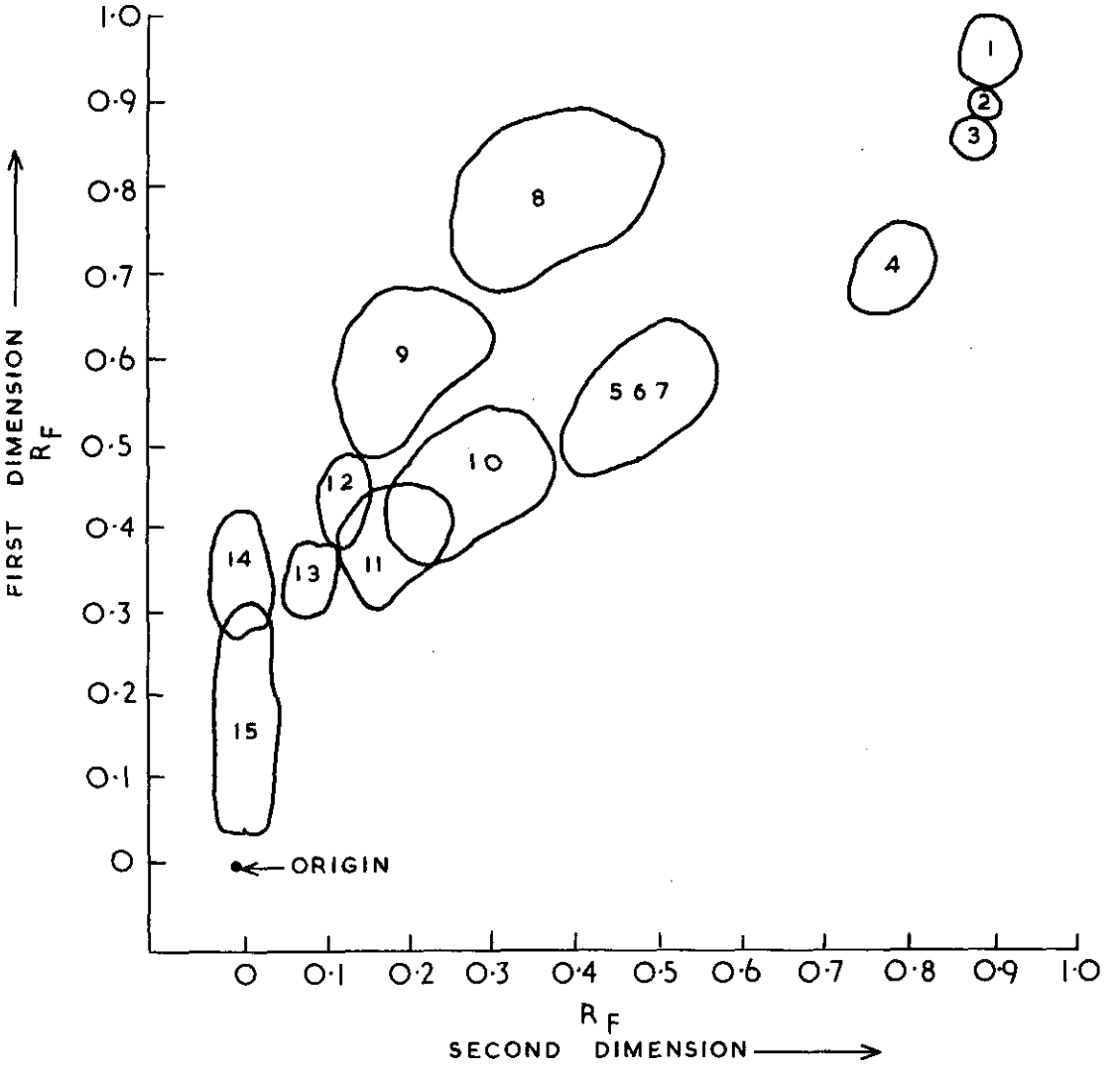

Fig. 1 - Two-dimensional chromatogram of pigments found in marine algae.

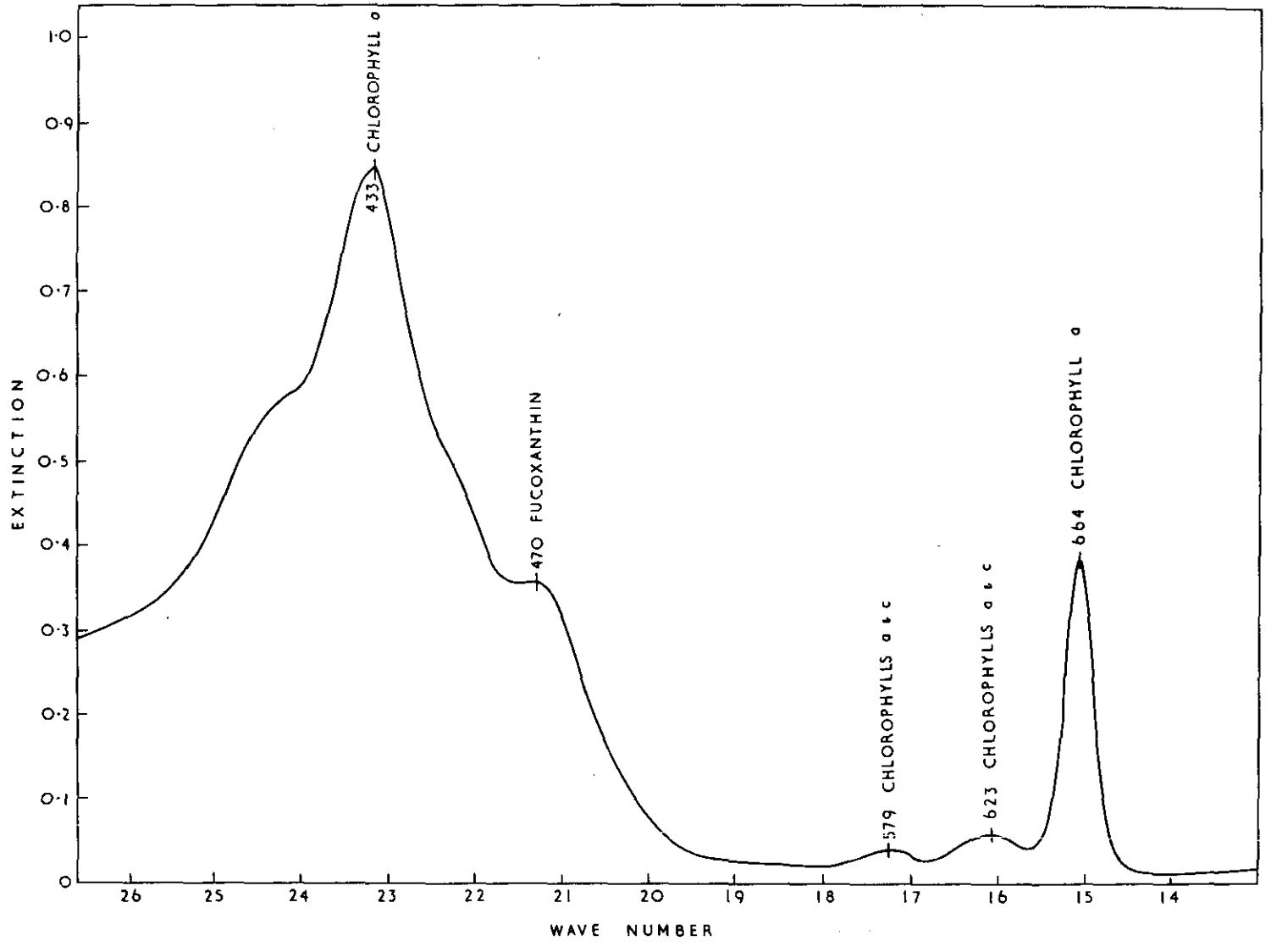

Fig. 2 - Absorption spectrum of pigments in the brown alga, *Hormosira*.

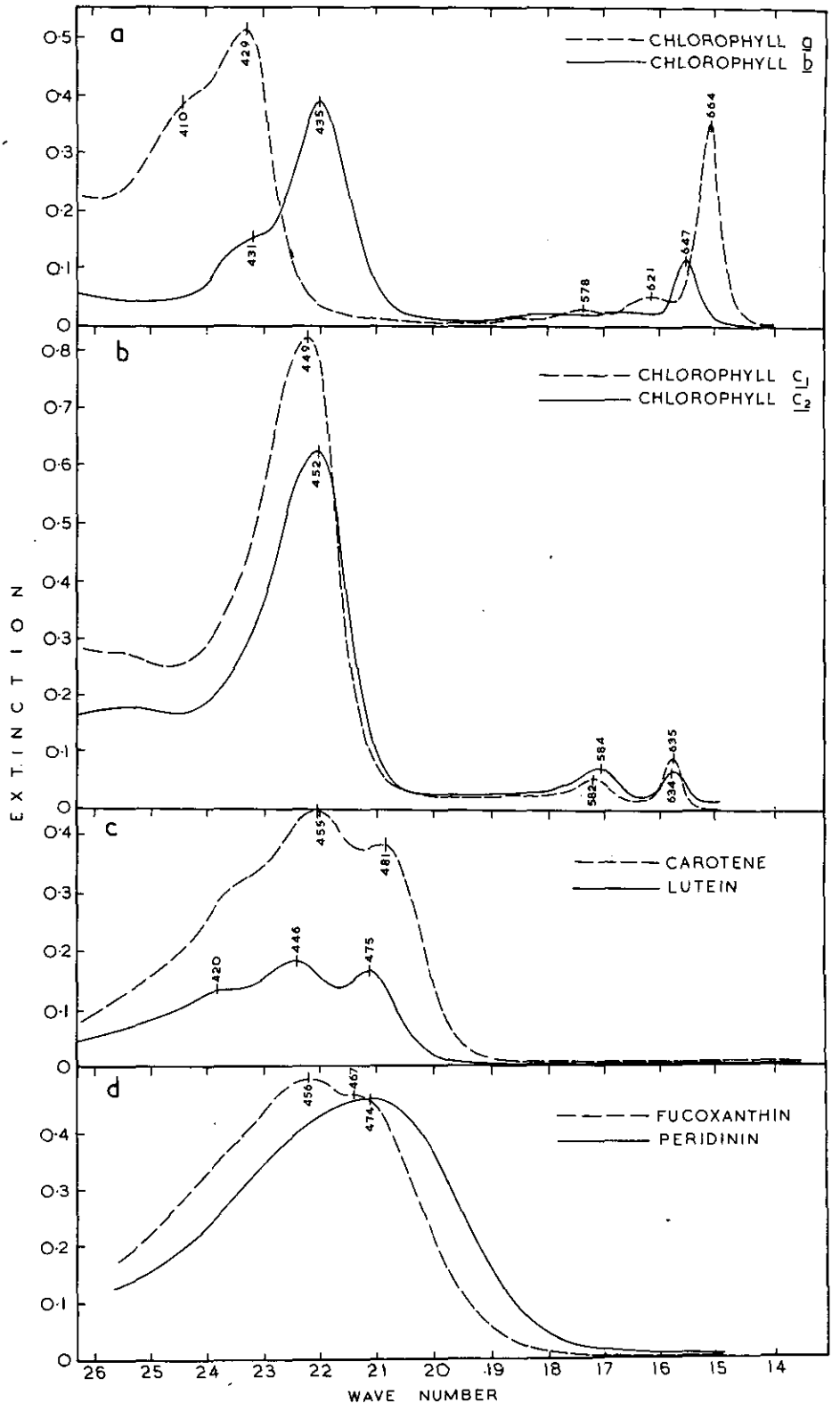

Fig. 3 - Absorption spectra of chlorophylls a, b, c_1, and c_2, carotene, lutein, fucoxanthin, and peridinin.

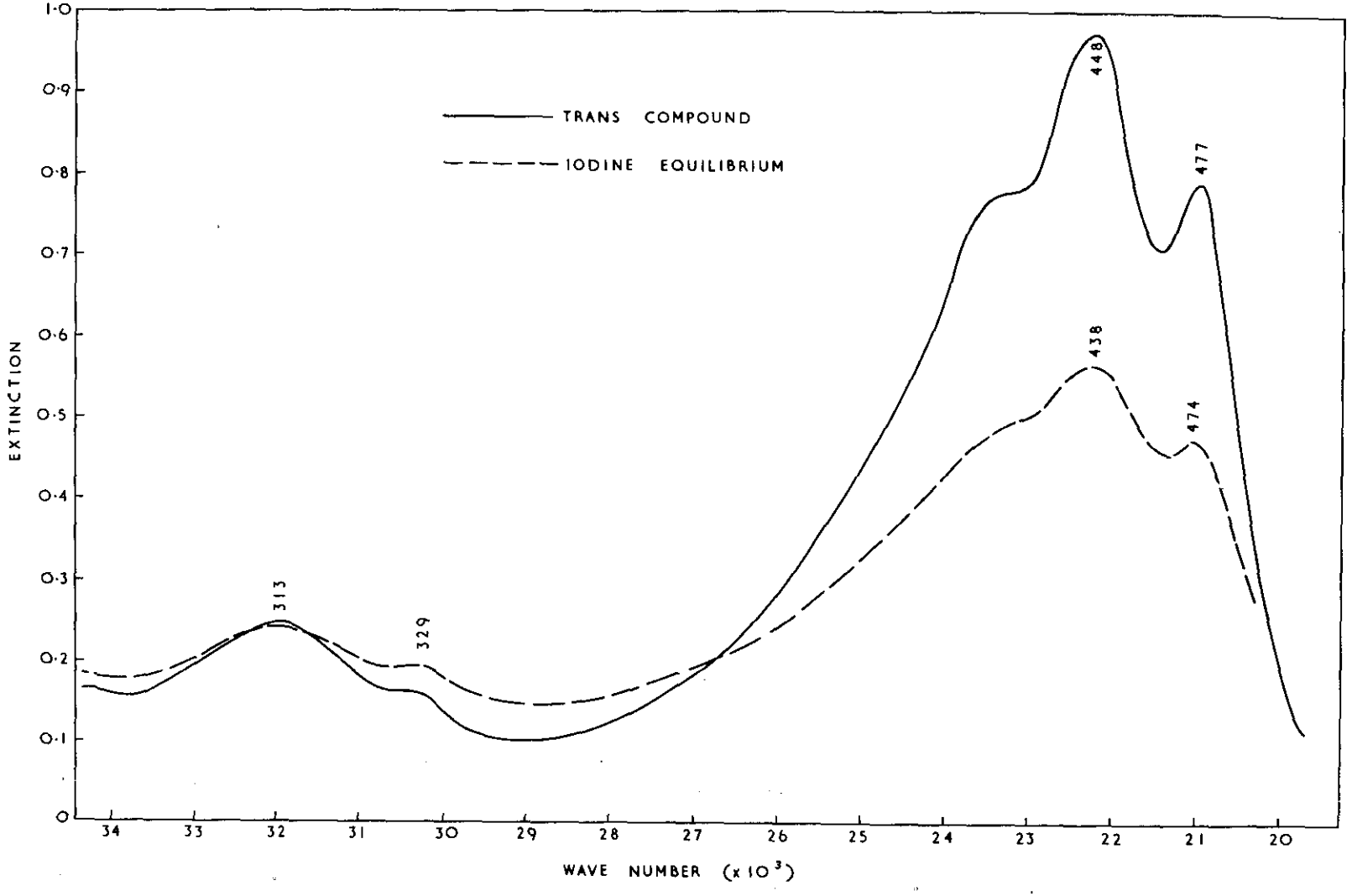

Fig. 4 - Change in absorption spectrum of fucoxanthin on iodine isomerization.

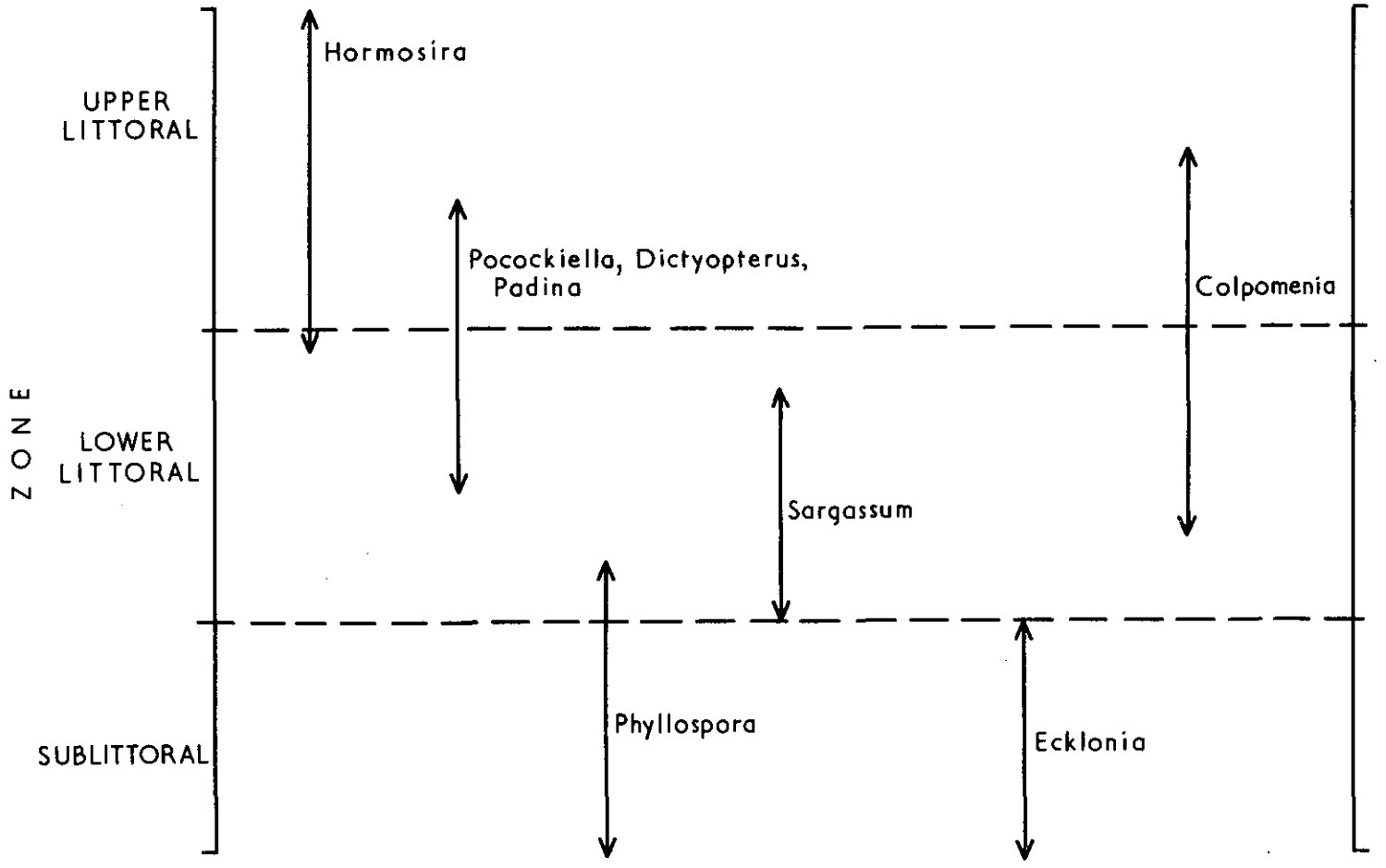

Fig. 5 - Distribution of brown algae on a rock platform.
For pigment hues see Tables 2 and 3.

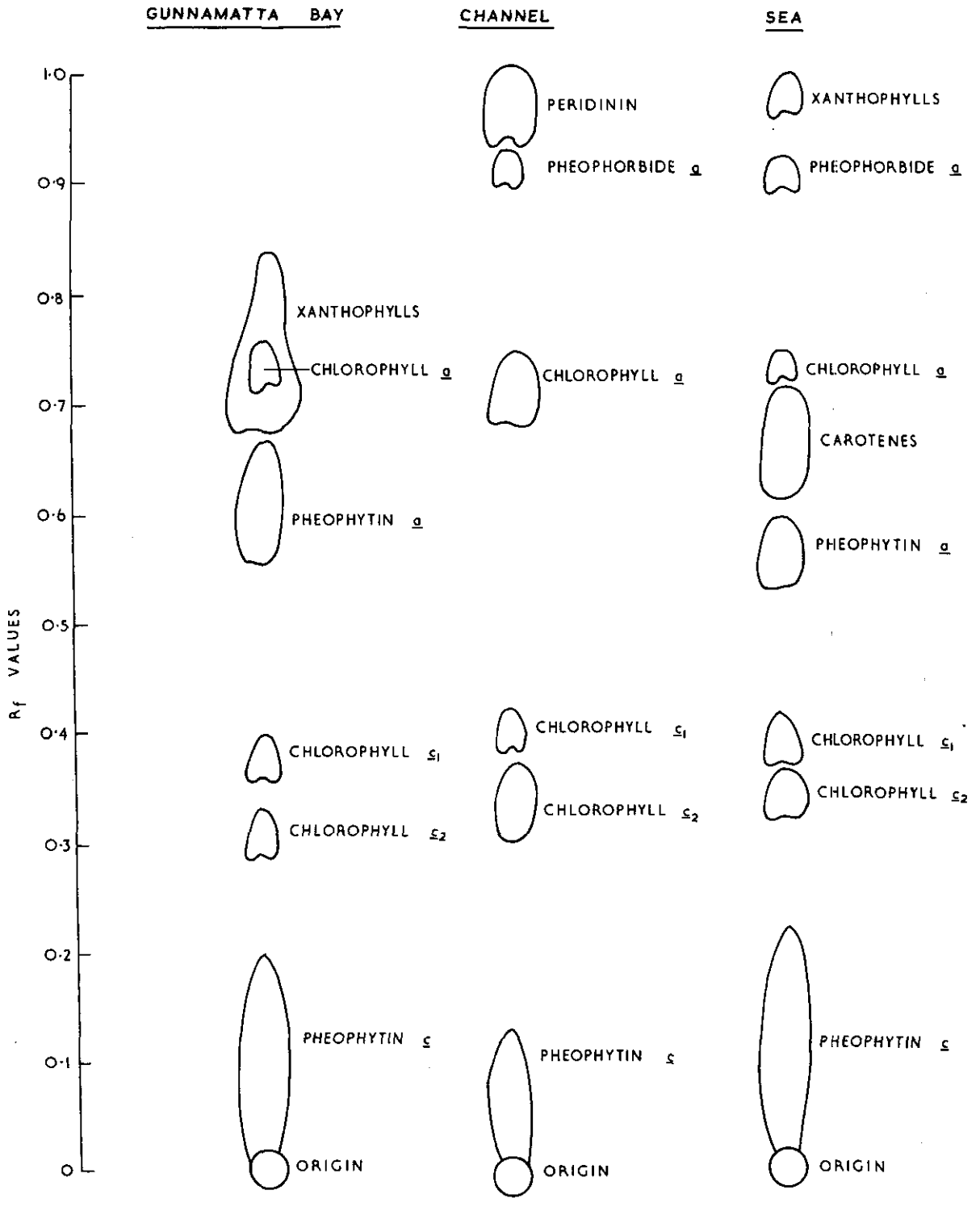

Fig. 6 - Thin-layer chromatogram (polyethylene plate; 90% acetone) of pigments in water samples from Gunnamatta Bay, the channel entering Gunnamatta Bay, and the open sea.

ZOOPLANKTON

I. INTRODUCTION

A study was made of the distribution and abundance of zooplankton in Gunnamatta Bay. The study related distribution to tidal water movement and possible vertical migration in some species. The species diversity in several areas, inside and outside the bay, was calculated using herbivore copepods as a group which occupy a well defined, wholly planktonic niche. The sampling pattern covered diurnal and tidal variations.

II. METHODS

The sampler was a conical nylon net of mouth area 0.25 m^2 and mesh aperture 0.5 mm. The net was towed horizontally, attached with a 3-leg bridle about 1.5 m above a 20 lb weight. Towing time was approx. 60 sec. A T.S.K. flowmeter (No. 694) was suspended in the mouth of the net at about one quarter of a radius from the ring. This position was used because the flow there is nearer mean mouth velocity (Tranter and Heron 1967). The volume filtered was calculated using the formula -

$$V = 0.167\ A\ t\ \left(\frac{c}{t} + 0.011\right)$$

where V = volume sampled (m^3),

A = mouth area of net (m^2),

t = time of tow (sec),

c = flowmeter revolutions, and

0.167 and 0.011 are constants appropriate to meter No. 694.

Temperature and salinity were measured using an inductive salinometer (Brown and Hamon 1961).

Species diversity was measured by the method of MacArthur (1965). If successive individuals in a census were independent of previous ones, $H = -\sum_{i=1}^{n} p_i \log_e p_i$ is an appropriate measure of the uncertainty of the specific diversity of the next individual in the census. The number of species is n, and p_i the proportion of the total number of individuals which belong to the ith species. If the exponential of H (e^H) is calculated this gives the number of equally common species, which is a measure of species diversity. In this way, changes in species diversity can be compared with environmental changes.

III. RESULTS

Figure 1 shows the sample locations in the four main sampling areas. Temperature and salinity measurements were taken both inside and outside the bay and together with data from other groups (e.g. Hydrology) seem to indicate that, on the early incoming tide, the water mass entering the bay is much the same as that which left on the previous receding tide. On the latter part of the incoming tide, there seems to be a main current from the channel along the eastern side of the bay. This current probably mixes fairly quickly.

(a) Horizontal Distribution

(i) Distribution of Salps and Doliolids

Figure 2 shows a decrease in the density of both species going into the bay. Thalia democratica and Doliolum denticulata occur in equally high densities outside the bay, at Station I. In the channel, at Station II, the density is lower than that at the end of the bay (Station IV). Table 1 shows salp and doliolid density for specific samples.

TABLE 1

ACTUAL DENSITIES OF SALPS AND DOLIOLIDS
IN SELECTED SAMPLES
(No. per 100 m^3)

Sample No.	Salps	Doliolids	Sample No.	Salps	Doliolids
3	6	99	12	300	145
4	0	18	13	4	8
6	64	64	14	0	0
7	18	12	15	8	8
8	0	30	16	0	24
9	0	13	17	0	12
10	0	7	21	12	12
11	8	68	24	115	150

(ii) Distribution of Pseudeuphausiids

All specimens collected were larval stages. At all stations a mixture of Calyptopis 2 and 3, and Furcilia ranging in length from 1.8 to 3.1 mm were collected, and all stages were included in the final count.

The raw data thus obtained were combined to give average frequencies per 100 m^3 in sampling areas I, II, and III and IV combined. The results are shown in Figure 3.

(iii) Distribution of the Larvae of a Species of Callianassidae

Table 2 and Figure 4 show the density of callianassid larvae at stations inside and outside the bay. The overall density increases further inside the bay. However, while at Station III there are more animals at the lower levels, at the end of the bay (Station IV) they concentrate nearer the surface. This was also observed during the night.

On an incoming tide the concentration was highest at the end of the bay on the surface, while on the outgoing tide there are equal levels in the middle and the end of the bay.

TABLE 2

DENSITY OF DECAPODS AT THE FOUR SAMPLING STATIONS
(No. per 100 m^3)

Date		May 24		May 25		May 25			May 29		May 30	
Time		0100-1300		0100-1200		1530-1630			1900-2000		0100-1100	
Tide		Outgoing		Outgoing		Incoming			Incoming		Incoming	
Sample Nos		2-5		6-12		13-16			17-21		22-25	
Depth		Surface	5 m	Surface	5 m	Surface	5 m	9 m	Surface	5 m	Surface	5 m
(a) Callianasid spp. Larvae												
Station	I	-	-	80	180	-	-	-	30	-	36	24
	II	700	-	70	-	60	-	-	-	-	-	-
	III	240	380	300	480	16	120	2100	220	380	30	-
	IV	100	-	400	80	-	-	-	390	140	480	-

(b) Brachyuran spp. Zoea

Station												
	I	-	-	20	150	-	-	-	110	-	30	55
	II	420	-	140	-	17	-	-	-	-	-	-
	III	140	320	250	280	100	23	450	180	140	11	-
	IV	2600	-	190	200	-	-	-	2600	430	15	-

(iv) Distribution of Brachyuran Zoea

Table 2 gives the density of zoeas in the bay for all samples. Figure 5 shows densities for selected samples. The zoea were of the same species, but at all stages. Figure 5 shows that on the outgoing tide there is an even distribution of zoea throughout the bay, with the greatest numbers in the middle of the bay. On the incoming tide, during the night, they were concentrated at the end of the bay. Although there was no significant difference between the surface and 5 m during the day, a large concentration was observed at the bottom on one occasion, and during the night a very high surface concentration was observed at the end of the bay (Station IV).

(b) Vertical Distributions

(i) Distribution of the Cladoceran, Penilia schmackeri

Figure 6 shows the densities of P. schmackeri at the various sampling stations. During a receding tide, P. schmackeri was distributed equally between the middle of the bay (Station III) and the channel (Station II) at the surface (Fig. 6a). Large numbers occurred at the bottom of the bay during the day (Fig. 6b).

Figure 6c shows that on an incoming tide during the morning or the afternoon the species was found mainly in the middle of the bay (Station III). However, a very small proportion is found outside (Station I) and in the channel (Station II).

Figure 6d shows that at night there is a vertical migration of P. schmackeri to the surface at Station III.

These figures for the distribution of P. schmackeri show that there was a vertical migration to the surface of the bay at night. This result is best demonstrated by taking the ratio of the distribution at the surface to that at a depth of four to five metres for the samples in the middle of the bay (Station III) where the numbers of P. schmackeri per cubic metre are greatest (Table 3).

TABLE 3

RATIO OF DISTRIBUTION OF PENILIA SCHMACKERI AT THE SURFACE TO DISTRIBUTION AT 4-5 M AT STATION III

Date	Tide	Surface Density (No./m^3)	4-5 m Density (No./m^3)	Surface / 4-5 m
Day				
May 23	Receding Morning	2.18	35.60	0.063
May 24	Receding Morning	7.00	25.00	0.280
May 30	Incoming Morning	15.00	20.20	0.680
Night				
May 29	Incoming Evening	79.00	39.20	2.020

(ii) Distribution of the Decapod, Lucifer hanseni

Table 4 shows the total distribution of L. hanseni at all stations. Figure 7 shows the vertical distribution at Stations III and IV (in the bay proper) in the morning, afternoon, and evening. There was a high density at the bottom on the afternoon of May 25 and a very high density at the surface on the night of May 29. Densities at other times were variable, but always low.

(iii) Distribution of a Cumacean

A species of cumacean was observed on the night of May 29 (Table 4). Density just outside the bay was greater than inside, although significant numbers appeared in the bay.

(iv) Distribution of Ostracods

A swarm of ostracods was found in night hauls on May 29. Similar swarms were found during the 1966 School. No specimens were found in daylight hauls taken prior to May 29, and relatively few were found in the haul taken on May 30 (Fig. 8).

Other groups found the ostracods living on the bottom in sediment samples.

TABLE 4

DENSITIES OF SOME CRUSTACEA AT THE SAMPLING STATIONS

Date		May 24		May 25		May 25			May 29		May 30	
Time		0100-1300		0100-1200		1530-1630			1900-2000		0100-1100	
Tide		Outgoing		Outgoing		Incoming			Incoming		Incoming	
Sample Nos		2-5		6-12		13-16			17-21		22-25	
Depth		Surface	5 m	Surface	5 m	Surface	5 m	9 m	Surface	5 m	Surface	5 m
(a) Lucifer hanseni												
Station	I	-	-	12	0	-	-	-	4	-	82	55
	II	24	-	3	-	0	-	-	-	-	-	-
	III	0	0	20	3	0	23	260	640	290	11	-
	IV	180	-	11	8	-	-	-	530	440	15	-
(b) Cumacea												
Station	I	Not found							160		Not found	
	II											
	III								18	16		
	IV								36	50		
(c) Ostracoda (Pyrocypris acuminata)												
Station	I	Not found							2320		51	208
	II											
	III								2450	7670	3.7	
	IV								1940	4730	1150	

(c) Species Diversity

Table 5 gives the density of herbivorous copepods for stations representative of the four major sampling areas. Where possible identifications were made to the specific level (Mori 1964). Calanoids x and y could not be identified even to the generic level but are different from each other and from all other species found.

TABLE 5

NUMBERS OF ALL HERBIVOROUS COPEPOD SPECIES IN SAMPLES OUTSIDE AND INSIDE THE BAY

Species	Sample Numbers											
	Outside				Inside							
	6	12	24	25	13	8	9	19	23	3	10	11
Acartia erythrea	10	23	0	0	5	6	10	17	14	200	380	7.5
Temora turbinata	8	60	308	495	2	3	6	1	1	15	40	7.5
Tortanus forcipatus	0	0	92	10	6	9	23	13	5	1	11	6
Canthocalanus pauper	13	17	12	10	0	0	0	0	0	0	0	0
Eucalanus attenuatus	6	1	4	0	0	0	0	0	0	0	0	0
Labidocera sp.	2	36	30	0	0	0	0	0	0	3	0	0
Scolecethrix danae	150	6	44	0	0	0	0	0	0	0	0	0
Euchaeta marina	13	1	118	175	0	0	0	0	0	2	0	0
Acartia danae	6	43	0	0	0	0	3	0	0	0	0	0
Labidocera acutum	0	1	0	20	0	0	0	0	0	0	0	0
Calanoid y	0	0	40	0	0	0	0	0	0	0	0	0
Pontellina sp.	0	0	16	0	0	0	0	0	0	0	0	0
Pontella sp.	0	0	4	0	0	0	0	0	0	0	0	0
Calocalanus pavo	0	12	0	0	0	0	0	0	0	0	0	0
Undinula vulgaris	0	0	72	0	0	0	0	0	0	0	0	0
Nannocalanus minor	0	0	208	150	0	0	0	0	0	0	0	0
Centropages sp.	0	0	20	3	0	0	1	0	2	0	0	0
Calanopia thompsoni	0	0	0	0	0	0	0	4	0	0	0	0
Calanoid x	0	0	0	0	0	0	0	0	5	0	0	0
Acartia clausi	0	0	0	0	0	0	0	0	2	0	0	0
Eucalanus pileatus	0	0	0	0	0	0	0	0	1	0	0	0

The order of samples in Table 5 is from the marine to the bay area. Most species except Acartia erythrea, Temora turbinata, and Tortanus forcipatus are restricted to the marine area. The densities of these three copepods were found to vary widely for different stations, and other less frequently encountered species dominated in particular samples. Scolecethrix danae, for example, far outnumbered any other species in Sample 6. This occurred, however, only in the marine situation; one or more of the three copepods always dominating in the bay. In general then, the results show that the bay has no copepods peculiar to it; except for Calanopia thompsoni, all are found in the marine environment.

This distribution is reflected in the species diversity estimates (Fig. 9).

Species numbers are higher in the marine samples and fall to between 3 and 5 for the bay and channel. One exception to this is Sample 3, taken at the surface in Gunnamatta Bay, during very rough weather. Similarly, values of $e^{H}(E)$ and H are higher outside the bay. In Sample 24, for example, there are roughly 6 species occurring with about equal frequency. In contrast to this is the average of 2 to 3 for bay samples. If the few rarely occurring species in the bay are omitted, then H and e^{H} values for Acartia erythrea, Temora turbinata, and Tortanus forcipatus are barely changed.

IV. DISCUSSION

(a) Horizontal Distributions

(i) Distribution of Salps and Doliolids

The density of the doliolids is consistent with the counter-clockwise circuit of the incoming water through the bay. On the outgoing tide, low densities in the channel are typical of bay water. This water might dilute the other stations at very low tide. On the incoming tide, the channel still contains some returning bay water.

At Station IV the salps are absent. They might detect the bay water and swim away from it, or be caught by bay predators. The doliolids are apparently more tolerant and are carried to the end of the bay. Highest densities occur at high tide. Lowest densities occur at low tide, when complete mixing with bay water has taken place.

To find when the higher densities of salps and doliolids in the outside water enter the channel, sampling over the full range of an incoming tide is necessary. Sampling on the east side of the bay would show the doliolid movement.

(ii) Distribution of Pseudeuphausiids

The general pattern of distribution agrees with the anti-clockwise pattern of water circulation suggested by movement of salps and doliolids.

Numbers in the bay showed a tendency to decrease over the period. The decrease might possibly be correlated with a change in the amplitude of the tides. The difference between maximum high and minimum low decreased from 5 ft 11 in. on May 25 to 3 ft 4 in. on May 31.

The distribution might be explained if the pseudeuphausiids in the bay were washed in from a relatively large population in Port Hacking, when the tidal amplitude was large and much of the bay water was exchanged. As the amplitude decreased the larvae became trapped in the bay, and few more were washed in. Predators and lack of food could then quickly reduce the numbers in the bay.

More detailed and precise information is necessary to support this hypothesis. To obtain such information sampling in and around the channel is necessary at all periods of the tidal cycle.

(iii) Distribution of the Larvae of a Species of Callianassidae

The callianassid larvae found probably represent the young of a permanent bay population living in the bottom sand as they are not found in significant numbers outside the bay. They move with the tidal current up and down the bay, and individuals collected outside might have been washed out from the inside population. There is evidence of an unfavourable environment at lower levels in the end of the bay, since the larvae are not found there in great numbers whereas they are found at low levels in the middle of the bay. There is no evidence of a diurnal rhythm, the distributions during the day and night being the same. Bottom samples indicate a marked preference for deep water.

(iv) Distribution of Brachyuran Zoea

The brachyuran zoeas showed similar horizontal distribution in the bay to that observed for callianassidae. They were most highly concentrated in the middle of the bay during outgoing tides. It is impossible to separate clearly diurnal movement, if it occurs, from tidal movement, but the high concentration at the end of the bay during the night, coupled with even distribution on the outgoing tide on May 30 indicate vertical diurnal migration, and some ability to maintain position in the bay.

(b) Vertical Distributions

(i) Distribution of Penilia schmackeri

During a receding tide in the morning, there is an equal distribution of P. schmackeri at the surface between the channel (Station II) and the middle of the bay (Station III) (Fig. 6a). This indicates that P. schmackeri is being carried out with the receding tide. However, the greater percentage of the species is found at the bottom of the bay (Fig. 6b). This might be the result of a diurnal response.

For an incoming tide during the afternoon or morning P. schmackeri is found predominantly in the middle of the bay (Fig. 6c). However, the presence of a significant number of P. schmackeri outside the bay (Station I), and in the channel (Station II), during these incoming tides, suggests that for receding tides during the day or during the night a considerable number of the P. schmackeri at the surface in the middle of the bay (Station III) will be carried out of the bay.

At night there is a vertical migration of P. schmackeri to the surface at the middle of the bay (Station III). This appears to be a diurnal migration.

Probably, the migration of P. schmackeri is only a diurnal response. It is not adjusted to keep from being washed out of the bay by the tides. This conclusion could be verified by taking samples during a late receding tide at night when a significant number of P. schmackeri should be found at Stations I and II. A better picture of the distribution could also be obtained by taking samples on either side of the bay.

(ii) Distribution of the Decapod, Lucifer hanseni

L. hanseni showed a marked vertical migration which must occur in the early evening. In the morning, small numbers were present at the surface, but by the afternoon they had probably all descended to the bottom of the bay, at least in the central area.

Although L. hanseni is not considered an estuarine species, large numbers were not observed outside the bay, suggesting that this might be a bay population. Further sampling would be required to clarify this. Further sampling at various depths could elucidate the diurnal movements of this species.

(iii) Distribution of a Cumacean

The unexpected appearance of the cumaceans indicated that there was a population living outside and perhaps inside the bay. The diurnal movement might not be a regular feature of this population. Co-operation with benthic studies could elucidate the behaviour of this species.

(iv) Distribution of Ostracods

Ostracod swarms were found in night samples taken by both the 1966 and 1967 schools, so it seems likely that they usually move towards the surface at night. They are rare in daylight surface samples, so presumably return to the bottom during the day.

The sample taken at Station IV on May 30 was taken relatively early in the morning and presumably caught ostracods which had not sunk to the bottom for the day.

This evidence indicates that the ostracods have a diurnal cycle of vertical migration.

(c) Species Diversity

The limited penetration of copepods into Gunnamatta Bay is reflected in the species diversity values. Only one high value for species numbers was obtained within the bay and this was probably due to outside contamination during rough weather. Low diversity can be interpreted as an indication of environmental extremes and can result in unstable populations. Longer observation would be necessary to show any extreme of either temperature or salinity. Compared to the marine region sampled, the bay exhibited less variation for both these factors. Another possibility was suggested by study on ^{32}P uptake and pigment concentration. The bay appears to have low levels of phytoplankton with low activity. Thus, the lack of food might impose limits on which species of the outside copepods which are being washed into the bay on every tide can survive inside the bay. No definite evidence of population instability was observed over the short period of observation.

V. CONCLUSIONS

Planktonic animals such as doliolids, which live mainly outside the bay, are brought into the bay by the tide, where they will die or be eaten. Other animals, which live mainly inside the bay, can be carried outside on the receding tide. Animals such as the cladoceran, Penilia, show a marked vertical migration to the surface at night. There appears to be no mechanism to stop Penilia from being carried out of the bay on the receding tides at night. This source of drain on the population must be made up by high reproduction.

The distribution of plankton favours the idea of a counter-clockwise circulation in the bay. During the later part of the receding tide and the early part of the incoming tide the

channel and the area immediately outside contain animals characteristic of the bay. It is only during the later part of the incoming tide that animals characteristic of the outside water enter through the channel. These animals then appear at the far end of the bay before the water is mixed and the animals disappear.

The low species diversity of the copepod herbivores inside the bay suggests that the bay is more difficult for this group to survive in than the outside water. The three copepods which successfully colonised the bay, could either be more efficient at living on the low phytoplankton levels existing in the bay or be more efficient at escaping the predators which would be characteristic of this type of shallow bay.

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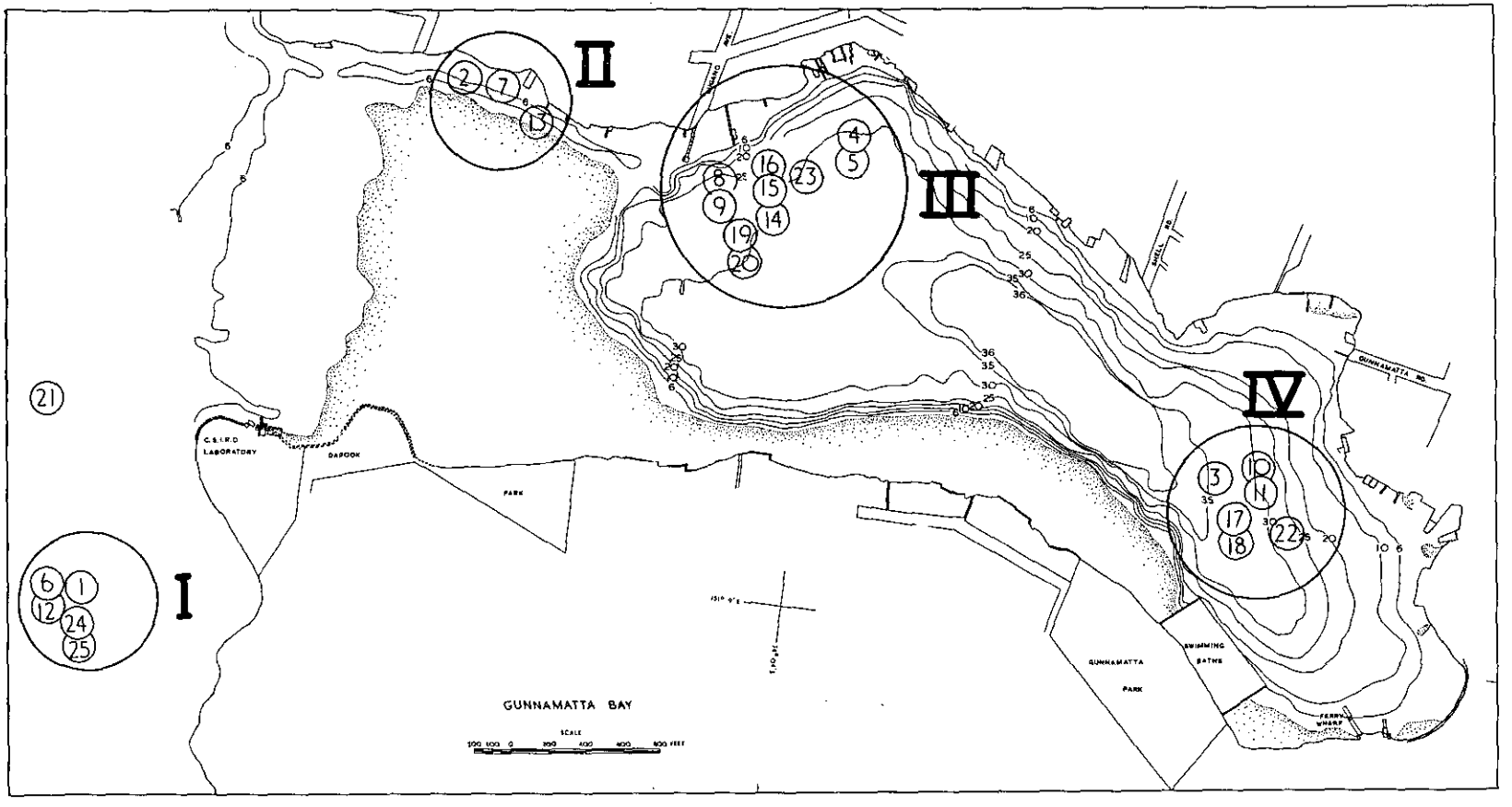

Fig. 1 - Locations of Stations I-IV and serial numbers of samples taken there. Depth contours in feet.

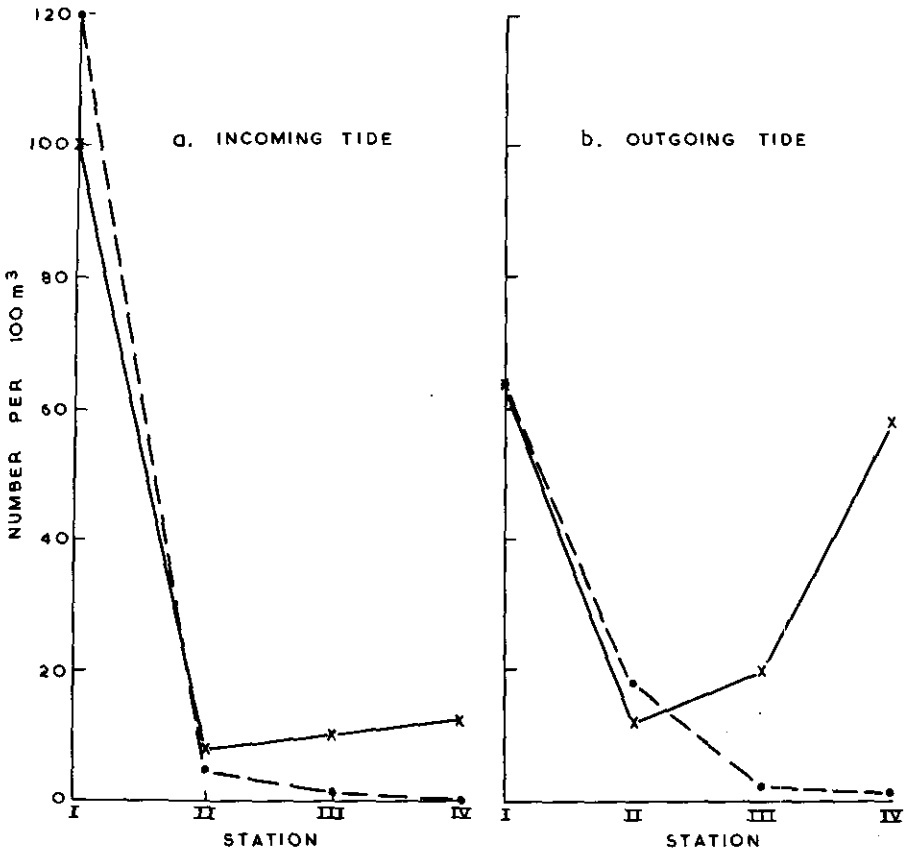

Fig. 2 - Densities of salps and doliolids at Stations I-IV.

●----● *Thalia democratica* (Salpidae)
X——X *Doliolum denticulatum* (Doliolidae)

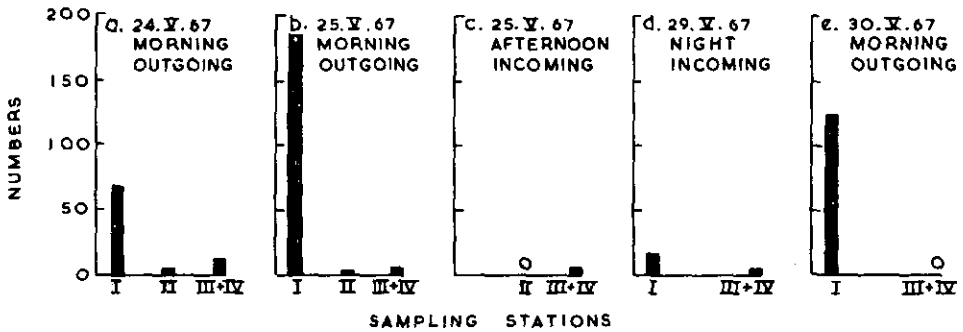

Fig. 3 - Numbers of pseudeuphausiids at Stations I-IV. Time of day and tide state given.

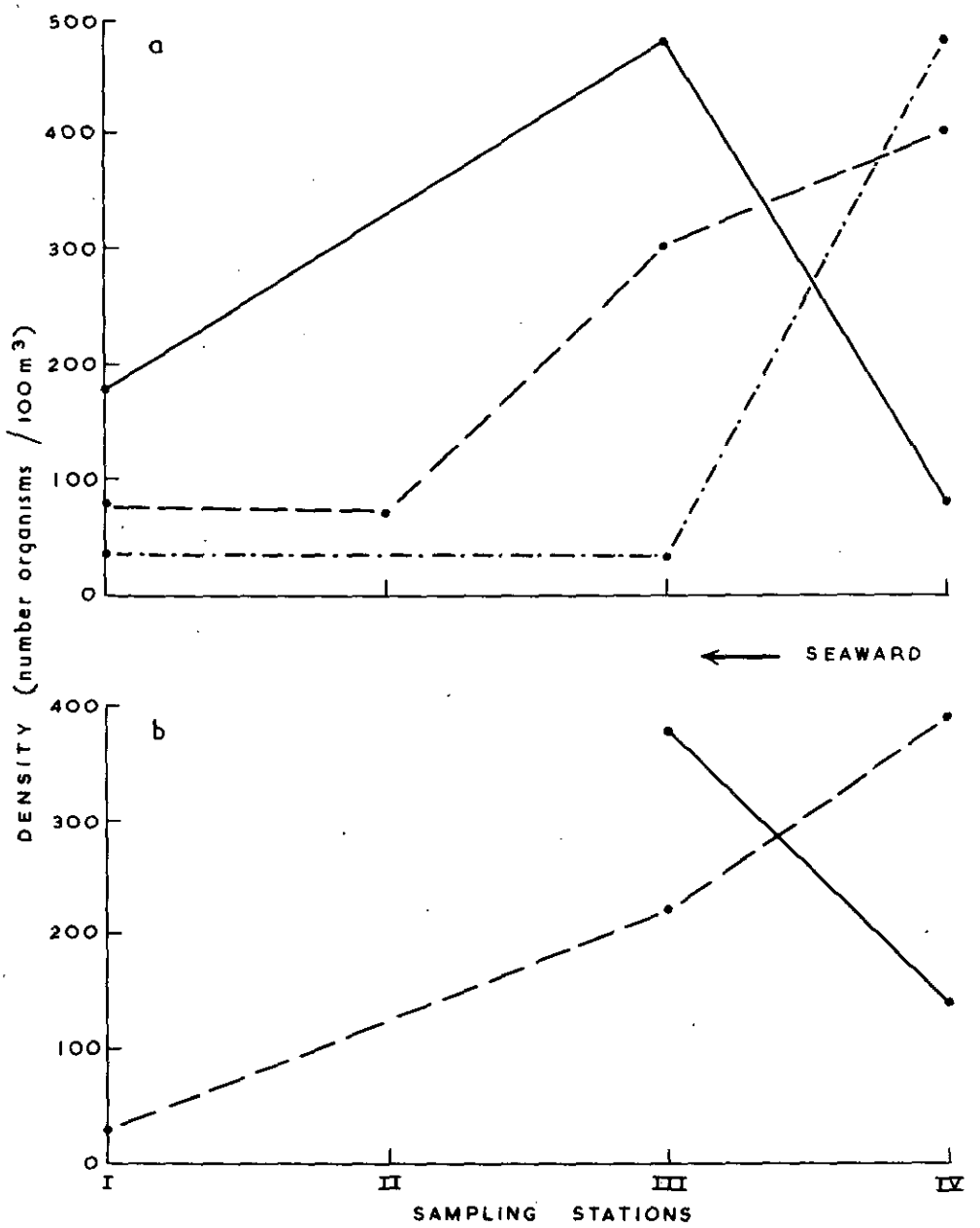

Fig. 4 - Density of the larvae of a callianasid at Stations I-IV.

a Morning
- - - - surface, outgoing tide, 25.5.67
------- 5 m, outgoing tide, 25.5.67
-.-.-.- surface, incoming tide, 30.5.67

b Evening
- - - - surface, incoming tide, 29.5.67
------- 5 m, incoming tide, 29.5.67

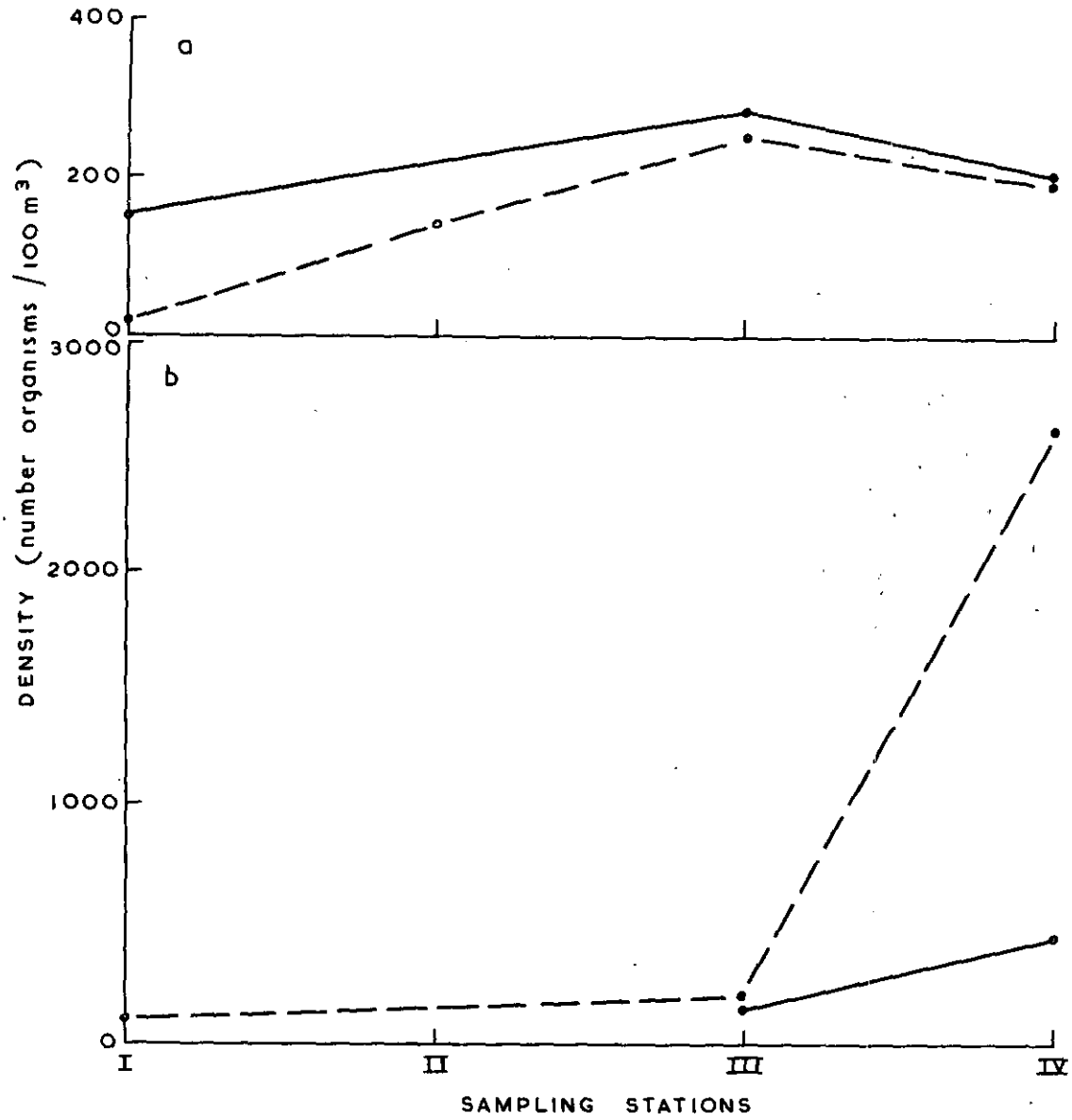

Fig. 5 - Densities of brachyuran zoea at Stations I-IV.

a Morning, outgoing tide, 25.5.67
- - - - surface
------- 5 m

b Evening, incoming tide, 29.5.67
- - - - surface
------- 5 m

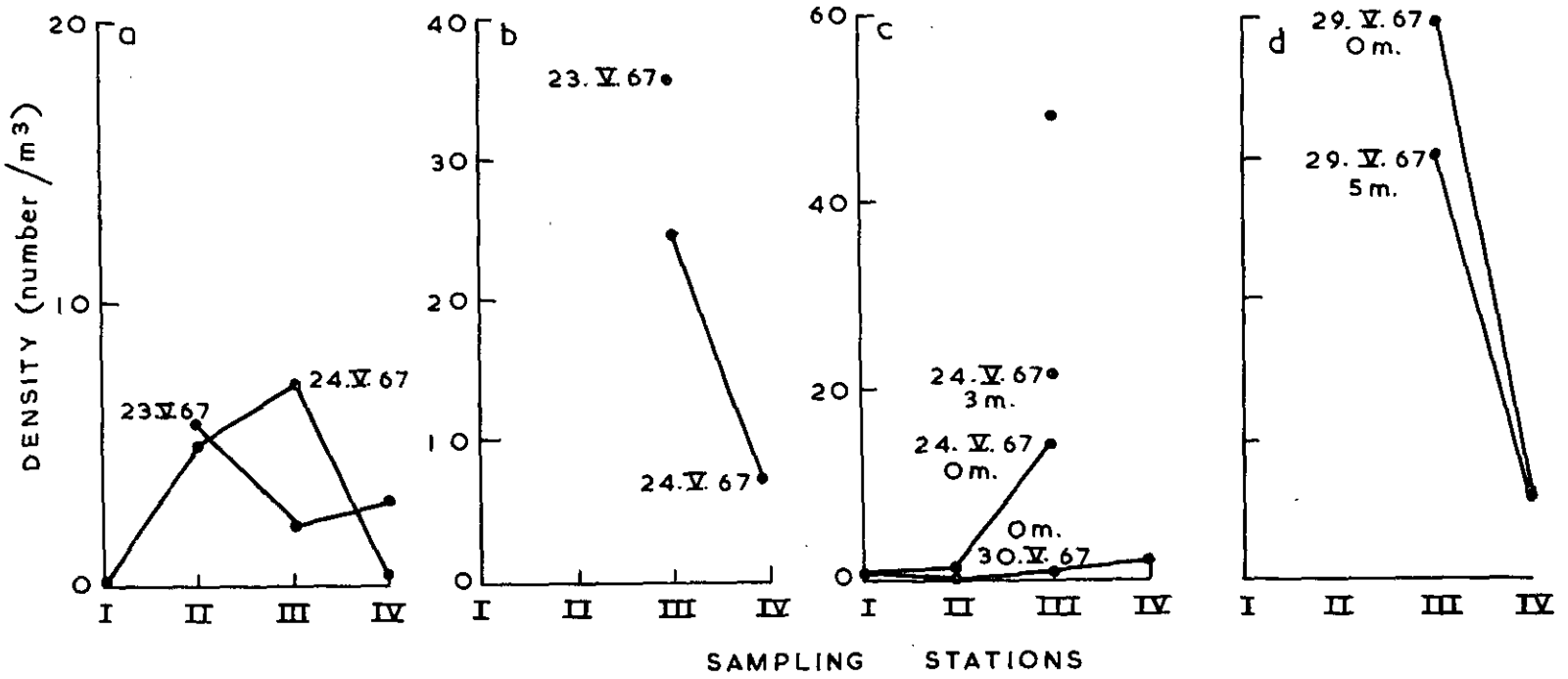

Fig. 6 - Densities of the cladoceran *Penilia schmackeri* at Stations I-IV.

a Outgoing tide, morning, surface
b Outgoing tide, morning, 5 m
c Incoming tide, morning (30.V), and afternoon (24.V)
d Incoming tide, evening

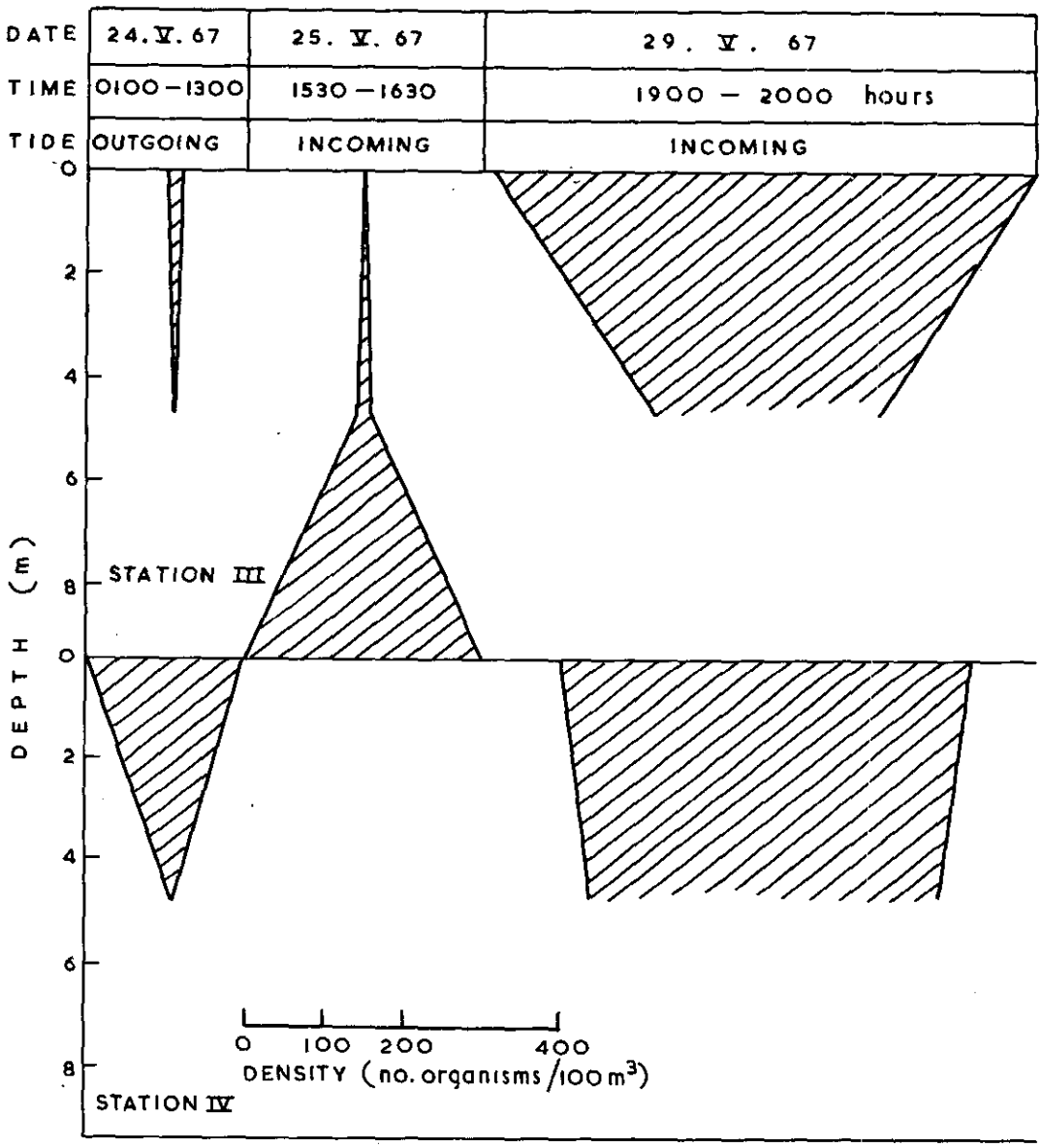

Fig. 7 - Diurnal distribution of the decapod *Lucifer hanseni* at Stations III and IV.

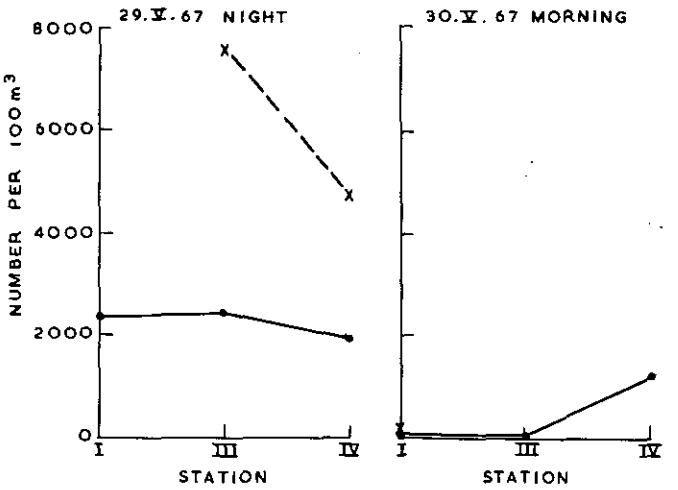

Fig. 8 - Density of ostracods at Stations I, III, and IV.
Surface ------ 5m - - - -

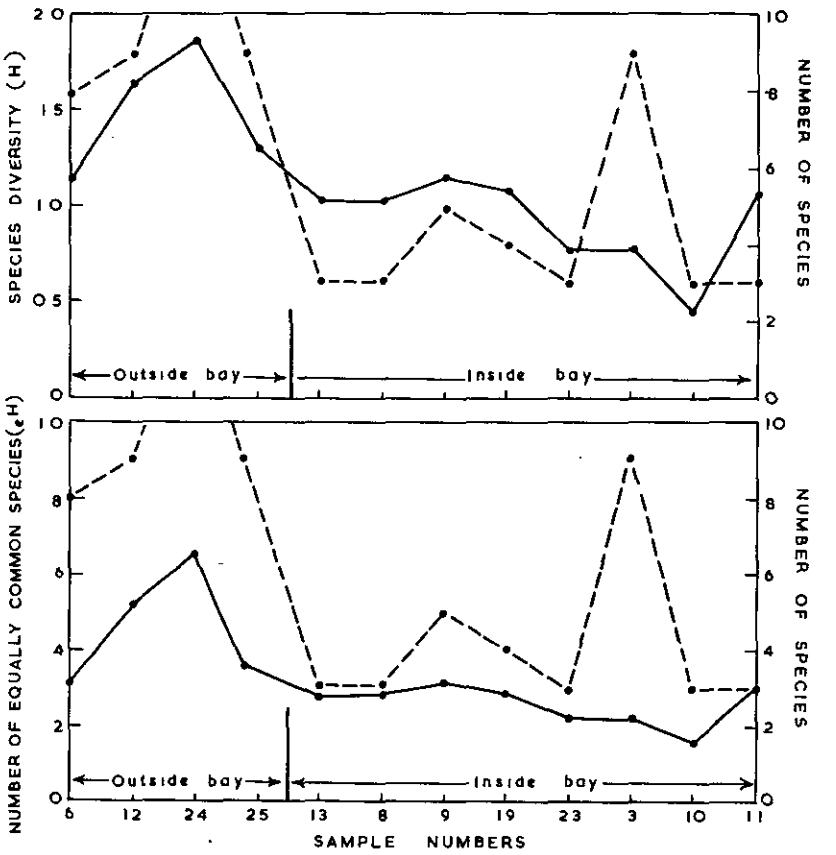

Fig. 9 - Species diversity and e^H values compared with species numbers for herbivore copepods, inside and outside Gunnamatta Bay.

a Species diversity (H) -------
Number of species - - - -

b Number of equally common species (e^H) -------
Number of species - - - -

Appendix

Algal Physiology Dr B.R. Grant
Mrs I. Turner

Miss R.M. Carey (Syd.)
D. Cheng (Tas.)
G. Gibbons (Syd.)
N.K. Howes (Syd.)
J. Peterson (Syd.)
Miss C.J. Pettigrew (Melb.)
R.W. Stephens (Syd.)

Benthos Dr R.J. MacIntyre
Dr S.B. Haven

Miss D.H. Conway (Melb.)
Miss J. Hunter (A.N.U.)
Miss S. McLaughlin (Syd.)
S.J. Miles (W.A.)
Miss L. Musgrave (Syd.)
K. Walker (Monash)
Miss J. Watson (Syd.)
L.P. Zann (U.N.E.)

Fish distribution and population studies Mr I. Munro
Mr B. Jones
Mr R. Blick

No report available
P. Baverstock (Adel.)
D.T. Brewster (N.S.W.)
T. Done (A.N.U.)
D.J. Finnegan (Adel.)
Miss S.M. Gough (Syd.)
Lee Chan Lui (Monash)
Miss J. Narbeth (N.S.W.)
H.T. Ong (Tas.)
J.W. Penn (W.A.)
Miss M. Reymer (A.N.U.)
P. Temple-Smith (Tas.)
Miss M. Williams (Melb.)

Foraminifera Dr A. Albani
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Miss I. Hergstrom (Adel.)
Miss M. McNeil (Tas.)
H.J. Marchant (Adel.)
J.W. Steen (Tas.)
A.H.M. Vandenberg (Melb.)
R.T. Wells (Adel.)
T.F. White (W.A.)

Hydrology Mr B.S. Newell
Dr R. Corsset
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B.J. Cannon (Melb.)
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Miss J. MacLean-Smith (Adel.)
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No report available
A.W. Bradshaw (Flinders)
P.F. Linden (Flinders)
D.A. Mills (Flinders)
L.P. Whitehead (Flinders)

Physical chemistry of sediments Dr M. Whitfield

D.F. Gartside (Melb.)
F.R. Higginson (Syd.)
R. Rose (N.S.W.)

Sediments Dr C. Phipps
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A. Daniel (Syd.)
Miss S.Y. Fletcher (N.S.W.)
W. Green (Townsville)
Miss H. Maggs (Melb.)
J. Matthews (Syd.)
C.D. Moyes (W.A.)

Pigments in marine algae Dr S.W. Jeffrey
Dr G. Humphrey

J.S. Hadfield (U.N.E.)
Miss W.L. Howe (Latrobe)
D. Rounsevell (Adel.)
P. Saenger (Melb.)
P. Tolstoshev (Adel.)

Zooplankton Mr D.J. Tranter
Mr A.C. Heron

G.W. Brand (Monash)
G. Burrows (Tas.)
D.R. Goodfellow (Syd.)
Miss K. Hammerston (Syd.)
S. James (Syd.)
Miss A.E. Smythe (Syd.)
D. Williams (Syd.)

Benthos/Foraminifera/Sediments
Dr A.N. Carter (Univ. of N.S.W.)

Report included with other groups
I.M. Curtois (N.S.W.
W. Harder (N.S.W.)
R.K. Ingram (N.S.W.)
D. Jongsma (N.S.W.)
F.R. Kalf (N.S.W.)
D.A. Lyons (N.S.W.)
M. Olivier (N.S.W.)
C. Somogyi (N.S.W.)
G. Winter (N.S.W.)