A CONTRIBUTION TO THE BENTHIC BIOLOGY OF SOME SOUTHERN AFRICAN LAKES

by

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PREFACE AND ACKNOWLEDGEMENTS

The first part of this work consists of a series of reports on the benthic fauna of a number of South African coastal lakes. The work was initiated under the auspices of the Institute for Freshwater Studies, Rhodes University, which is under the direction of Professor B.R. Allanson. The work forms part of a broader study principally of Lake Sibayi, and to a lesser extent Lake Nhlange in the Kosie Estuary system, which is being undertaken by the Institute. The first two papers are reprints from the Transactions of the Royal Society of South Africa. They were published as soon as possible after the major initial studies had been completed, so as to provide incentive for the further continuation of work on Lake Sibayi and the Kosi lake system.

Part 1 of the benthic papers has been included in this thesis, since it completes the description of the benthos of Lake Sibayi, although it also deals with other matters not entirely relevant to the principal arguments of the thesis.

The second part of the thesis deals with an initial experimental study which it is hoped will introduce a phase of more vigorous experimental studies into the biological problems of lakes, particularly the coastal lakes of Southern Africa.

I would like to express my sincere thanks to Professor B.R. Allanson for the encouragement advice given during the course of the work, and for the many constructive discussions we have had over problems that were raised. I would also like to thank him and Mr R. Hart for the pH measurements that they collected for me from Lake Sibayi in September and October, 1969, when I was

not able to visit the lake.

I would like to thank the many people who at one time or another helped wash samples from the lakes, acted as "scribes", helped in sorting animals, and wound winches; to name but a few, Miss G. McFarlane, Miss M. Roeleveld and Miss J. Garrett.

My sincere thanks are due to Dr B.J. Hill who instructed me in the techniques of SCUBA diving and who, with Mr A.T. Forbes and others, helped collect data from the substrate using underwater methods we evolved together.

Thanks are also due to Mrs P. Eva for interpreting manuscripts into drafts and final copies.

Financial assistance has been given by the Research Committee of Rhodes University, by the South African Council for Scientific and Industrial Research, and in kind from the Institute for Freshwater Studies of Rhodes University. I would like to express my gratitude to these organisations for their help.

RESUMÉ

The benthos of Lake Sibayi, Lake Nhlange, Lake Shengesa and Lake Sifungwe has been sampled by means of a van Veen grab of bite area 0.225 m^2. Except for Lake Shengesa, the substrates of the lakes have been investigated visually by SCUBA divers. Some quantitative sampling, using underwater techniques, has been carried out in Lake Sibayi.

In general, the substrate of the lakes consisted of two types, either fine sand with an average particle size of 3.2 phi, or mud with a median phi value of less than 6. The sandy substrata was usually rich in fauna, and the mud usually poor.

Lake Sibayi, which is freshwater (135 ppm Cl$^-$) has estuarine and freshwater species in the benthos. Some of the species, notably *Grandidierella lignorum* and *Apseudes digitalis*, show an uneven pattern of distribution with respect to depth.

Lake Nhlange is a brackwater lake (3.4°/oo salinity) and has mainly an estuarine fauna. Many of the species are different from those found in Lake Sibayi. There is some evidence that the benthic fauna of the lake has increased its range in depth in the years following a flood in January 1966.

Only chironomid larvae were found in the benthos of Lake Shengesa. The greater part of the substrate of this lake consisted of a highly organic mud mainly derived from burnt grassland around the periphery of the lake.

Lake Sifungwe, which is connected to Lake Nhlange, has a halocline at about 9 metres. The fauna of the benthos has more species than lake Nhlange. There is some evidence to suggest that the fauna invades deeper water in summer, and is driven into shallower water in winter because of the fluctuating level of anaerobic conditions below the discontinuity layer.

The benthic fauna of the lakes has been compared with that of the estuaries reported on by the Cape Town Ecological Survey.

Experimental studies on _G. lignorum_, a species present in Lake Sibayi, suggested that the physiological responses of material from an estuary normally at $35^{\circ}/oo$ salinity (Kowie River estuary) and from lakes which are normally of low salinity (Groenvlei $2.5^{\circ}/oo$ salinity, Lake Sibayi) were not different.

Studies on substrate choice, and pressure, indicated that these factors do not affect the distribution pattern of _G. lignorum_ in the substrate.

Studies on light orientation responses and the effects of rates of carbon dioxide increase of about 20 mm $HgCO_2$/hr or more, suggest that these may be the principle factors affecting the distribution of _G. lignorum_ in the benthos of Lake Sibayi.

Some supporting evidence for the presence of changing pCO_2 has been found. A discussion of the importance of dynamic events at the substrate/water interface is given.

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EXPERIMENTAL INVESTIGATIONS OF THE RESPONSES OF *GRANDIDIERELLA LIGNORUM* TO CERTAIN PHYSICAL AND CHEMICAL CHANGES UNDER LABORATORY CONDITIONS

GENERAL INTRODUCTION

Work on benthic fauna is generally of two types, either faunal surveys, or experimental work on benthic organisms. In the first case, attempts are usually made to correlate physical and chemical characteristics of the water body with the distribution of the fauna. Very often the data is claimed to afford some biological knowledge about the fauna, or is used in an attempt to classify habitats. For example, at a very simple level lakes are put into the categories of productive and unproductive. Attempts at further classification very often employ data about the presence or absence of particular animals, generally known as indicator species. However, as Brinkhurst (1964) and Macan (1963) have clearly pointed out, efforts in this direction have not been very successful. Brinkhurst has shown that as far as the oligochaetes are concerned, not only is there inadequate information, but it has been obtained from too restricted a geographical region. Macan (1963), when discussing the work of Mackereth, Lund and Macan (1957) on the English Lake District tarns, points out that no satisfactory explanation of the occurrence of various species was forthcoming from chemical considerations, although correlations with calcium and total ion content were present. Brinkhurst (1964) suggests that

> "We should not be surprised if a scheme based on the ratio of ion A to ion B does not tally with that based on the presence of species X, Y or Z. It is difficult to see why it should."

Within the framework of survey studies, and taking a lake as a whole, this is perfectly correct. Clearly an all too embracing result has been expected from this sort of approach.

When benthic animals become the subject of experimental studies, it is often shown that although effects of factors on an animal can be very subtle, it is often possible to show that there are certain limits beyond which an animal cannot cope, and which will exclude it from a particular environment. These limiting factors must be expressed in the distribution of a species, and indeed can sometimes be inferred from the survey data. But there are two complications which make it difficult to tie up generalised survey work with the experimental data. Different limiting factors affect different species; species A might be sensitive to substrate types of a certain character, and species B to some chemical quality in the water. Furthermore, the microenvironment around a benthic animal is often very different from the macroenvironment generally measured by traditional limnological methods. The distribution of an animal will therefore certainly indicate something about the lake, but depending on the species under discussion, it may not relate very accurately to the macroenvironment and, depending on the limits of tolerance of the species, may not indicate anything precise even about the microenvironment. On the other hand, if these limits are reasonably accurately known for the animal, from experimental data, then surely a greater understanding of the lakes themselves must accrue.

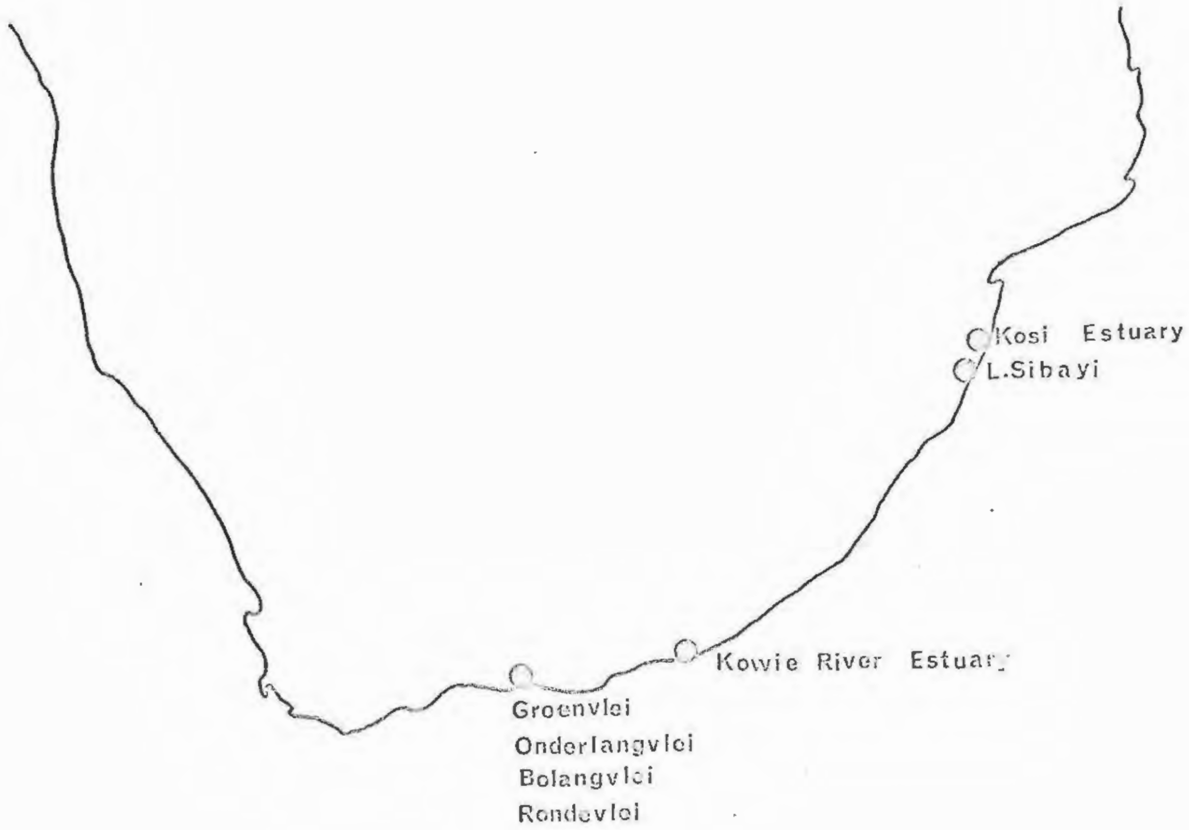
Real progress towards understanding the nature of a lake may therefore be had if data from the two sources is combined. An even better understanding may be achieved if data from the one source can be fed back into directing research into the other, in concurrent studies. Such an approach has been adopted in this preliminary work on the South African coastal lake benthic fauna.

The lakes on the eastern coastal region of Southern Africa south of the Zambezi show, in the majority of cases, unmistakable signs of an estuarine origin. The valleys of the estuaries were cut at a time when the sea level was some 30 to 40 metres below its present level (du Toit 1954). On the Tongoland plain of Mocambique and Zululand (Northern Natal), the valleys were cut in tertiary sands. Further south, the valleys were cut into the underlying bedrock. As the sea rose there tended to be filling in of the valleys by silt from the landward side, and blockage of the mouth of the valleys from the seaward side. A wide basin of variable depth often occurs just behind the mouth, which may open over a shallow sand bar. In a few cases only has infilling from the landward side been so meagre that the basin behind the mouth remained as a lake, with depths of water well below sea level. The best documented at present are the Kosi estuary lakes (Hill 1969). Swartvlei in the Knysna District is another whose depth of 11 metres goes well below sea level. The Knysna estuary has not formed a lake behind the opening of the mouth, but the channel is 16 metres deep. In a very few cases the lakes no longer have connection with the sea. The largest in South Africa is Lake Sibayi which, as Hill's (1969) charts show, has a maximum depth of 40 metres. The two other lakes about which a little is known are both small and shallow. Groenvlei in the Knysna district has a maximum depth of 7 metres, and Lake Shengezi near the Kosi estuary has a depth of 4 metres.

The small lakes of the Cape Peninsular sand flats do not seem to fall into the category of being the remains of deeply cut river valleys, but rather have formed from shallow depressions in the sand which have been flooded (Harrison 1962).

Investigations into the two northerly South African lakes, Lake Sibayi and Lake Nhlange of the Kosi estuary, started

Figure 1. Map of Southern Africa, indicating the positions of the principle lakes reported in this thesis.



in 1965 when a party of staff and students from the Zoology Department of Rhodes University made a short visit (Allanson, Hill, Boltt & Schultz 1966, and Allanson 1969). Stemming from the startling discovery of estuarine animals in Lake Sibayi, further visits were made in subsequent years up to January 1969. Serious work on the fauna of the substrate in any of the lakes did not start until January 1967, although a little data was collected from Lake Nhlange in 1965 and 1966. At the same time that sampling programmes were being undertaken, many other aspects of the lakes were being studied by Professor Allanson and others (Allanson & van Wyk 1969, Hill 1969, Forbes and Hill 1969).

The early work on the benthos of the lakes was hampered by lack of experience and facilities which could be transported into the field. Many of these were overcome with passage of time and when a field station was built at Lake Sibayi in January 1968.

Very early in the work on the benthos of Lake Sibayi and Nhlange, it was decided that data should be collected from similar lakes further south. Although the data has not been entirely worked up, reference is made to it whenever this has helped with the interpretation of data from the lakes. It was decided that experimental studies into the biology of the benthic fauna might help in the solution of the problems raised. However, since not all the species could be investigated at once, a decision had to be made as to what particular problem should be tackled, taking into account the need for a supply of experimental material with which to work on in a laboratory 800 miles from the lake.

Three general areas of attack seemed to be most promising:

1. Is there any evidence that special physiological adaptations had taken place in any of the species

which were discovered in the freshwater Lake Sibayi?

2. Could the responses that any species might show to the physical and chemical characteristics of the lake help to explain some of the problems posed from an analysis of the survey data?

3. Would a better general biological knowledge of any species help to explain some of the problems?

One of the prominent but numerically not the most important species from Lake Sibayi, the amphipod Grandidierella lignorum Brnrd. was chosen as an experimental subject because the species is abundant in the Kowie River Estuary 35 miles from the laboratory, and also because an interesting pattern of distribution in Lake Sibayi was shown by the animal.

One further important question had to be considered; how far to pursue investigations into any aspect of the biology of the animal? Two considerations seemed to be relevant. Since the study was in a sense exploratory, it was felt that more would be gained at this stage in pursuing lines of investigation which would lead directly to answers to the questions asked and to drop lines which did not lead to immediate answers for the present. The second point was that study should obviously be confined to areas where simple, readily available apparatus could be used. These studies have been pursued as far as practical considerations of time and available apparatus have allowed, and form the basis for further well orientated research on the lakes.

The results of the field surveys on the fauna and substrates of Lakes Sibayi, Nhlange, Shengesa and Sifungwe are reported first. These reports are partly self-contained, and ideas that have arisen from the analyses are assembled at the end of each description. Comparative analyses of the data are dealt with as the data become available. These studies form the first part of the thesis.

The experimental studies and collateral field work form the second part, and problems and concepts which arose are dealt with at appropriate points in the work.

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THE BENTHOS OF SOME SOUTHERN AFRICAN LAKES
PART I: DISTRIBUTION OF AQUATIC MACROPHYTES AND FISH IN LAKE SIBAYI*

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(With 3 text-figures and 1 plate)

(Read November 20, 1968)

Opsomming

'n Voorlopige ondersoek van die ondergedoke makrofiete en die visse in die Sibayameer, waarin van duiktegniese gebruik gemaak is, word beskryf. Die afwesigheid van makrofiete in die ondiep water van die hoofkom kan waarskynlik aan golfwerking toegeskryf word, terwyl hul afwesigheid benede 7 meter deur verminderde ligintensiteit veroorsaak word. *Potamogeton* is die dominante geslag tussen 1·5 en 5 meter diepte en *Myriophyllum* domineer tussen 5 en 7 meter. Die broeigedrag van *Tilapia mossambica* is beskryf en die aandag word gevestig op die waargenome verspreiding van kleinere cichlides en van *Croilia mossambica*.

Summary

A preliminary investigation of the submerged macrophytes and fish of Lake Sibayi was carried out using diving techniques. In the main basin of the lake, macrophytes were absent from water shallower than 1·5 to 2 metres, apparently due to wave action. Down to a depth of 5 metres, *Potamogeton pectinatus* L. and *P. schweinfurthii* A. Benn were the dominant species. Below 5 metres *Myriophyllum spicatum* L. was dominant. Macrophytes were absent from water deeper than 7 metres, probably due to reduced light intensities.

Tilapia mossambica Peters was found to nest either in association with submerged macrophytes or in very shallow water. *T. mossambica* did not construct its nests in dense weed or in water deeper than 7 metres. Shoals of small cichlids, chiefly *T. mossambica*, were observed to feed down to a depth of 20 metres several kilometres from the shore. The burrowing goby *Croilia mossambica* Smith was found to occur in the bottom of the lake down to 20 metres.

Introduction

During the current ecological studies on the lakes of northern Zululand an opportunity arose in January 1968 for an underwater investigation of the bottom of Lake Sibayi. This investigation was carried out using self-contained underwater breathing apparatus (SCUBA). This equipment was particularly useful for studying the vertical distribution of submerged macrophytes and the pattern of cichlid nests and fish groups which are reported in this paper.

Methods

Diving was carried out at thirteen stations (fig. 1). At each station general observations were made of the flora and macrofauna. In addition details of plant distribution and density were obtained along transects, using a frame covering 0·25 square metres. In weedy areas, lines of 10 or 20 successive quadrates were counted.

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This method was only partly successful as the rhizomes of *Potamogeton pectinatus* L. and *P. schweinfurthii* A. Benn grow in long straight lines with stems emerging from the sand at regular intervals. Depths were taken with a 'La Spirotechnique' depth gauge, which was corrected for fresh water. The angles of underwater slopes were measured by means of a floating plumb bob and a protractor attached to a length of wood. *Tilapia* nests were measured from rim to rim and from the bottom of the depression to the level of the rim by means of a fibre-glass measuring tape.

SUBMERGED AQUATIC MACROPHYTES

Northern and southern areas

The northern and southern areas of Lake Sibayi shelve gently (fig. 1). These shores are exposed to heavy wave action, resulting from the strong northerly and southerly winds (Allanson & Van Wyk, 196~~8~~9). Characteristic of the edge of these shores was a ridge of sand behind which shallow pools formed. This is an indication of shoreward transport of sand (King, 1958).

A series of observations was carried out from the southern shore to 11 metres depth (Stations 1 to 5, fig. 1). The margins of the shore were lined with the rush *Typha capensis* Rohrb. The shallow water up to a depth of 2 metres was bare of vegetation other than the sedge *Scirpus littoralis* Schrad. The rhizomes of this plant were well covered by sand and the stems were fairly rigid, and thus waves apparently had little effect on it.

At a depth of 3·5 metres (Station 1, fig. 1) the sandy bottom was ripple marked, indicating that wave action is sufficient to move sand at this depth. Both *Potamogeton pectinatus* and *Potamogeton schweinfurthii* were present although the weed cover was sparse.

At Station 2 (fig. 1) in 5 metres the weed cover was completely different. Large areas of dense *Myriophyllum spicatum* L. occurred, broken by extensive areas of bare sand on which an occasional large bush of *Myriophyllum* was found (fig. 2). Nests of *Tilapia mossambica* Peters were found in association with *Myriophyllum*. This aspect will be dealt with elsewhere. The *Myriophyllum* plants were about 45 cm high, and so dense that the sand could not be seen between the plants. Only a few single isolated strings of *P. schweinfurthii* and *P. pectinatus* and a single specimen of *Ceratophyllum demersum* L. were seen. In contrast, the bottom at Station 3 in 7 metres was almost bare of weed. Along a line of 180 metres length, only one patch of *Myriophyllum* was seen and *Potamogeton* was extremely sparse. Station 4, at a depth of 11 metres, had no weed cover.

A series of dives in the region of Station 5, in 3 metres, 5 metres and 7 metres showed a well-spaced-out community of *P. pectinatus* and *P. schweinfurthii*. Large, although isolated, bushes of *Myriophyllum* were interspersed with smaller specimens. Thus, although the species of plants were the same, this area differed from Station 2 at the same depth in the southern areas where the *Myriophyllum* cover was much more dense and the amount of *Potamogeton* far lower.

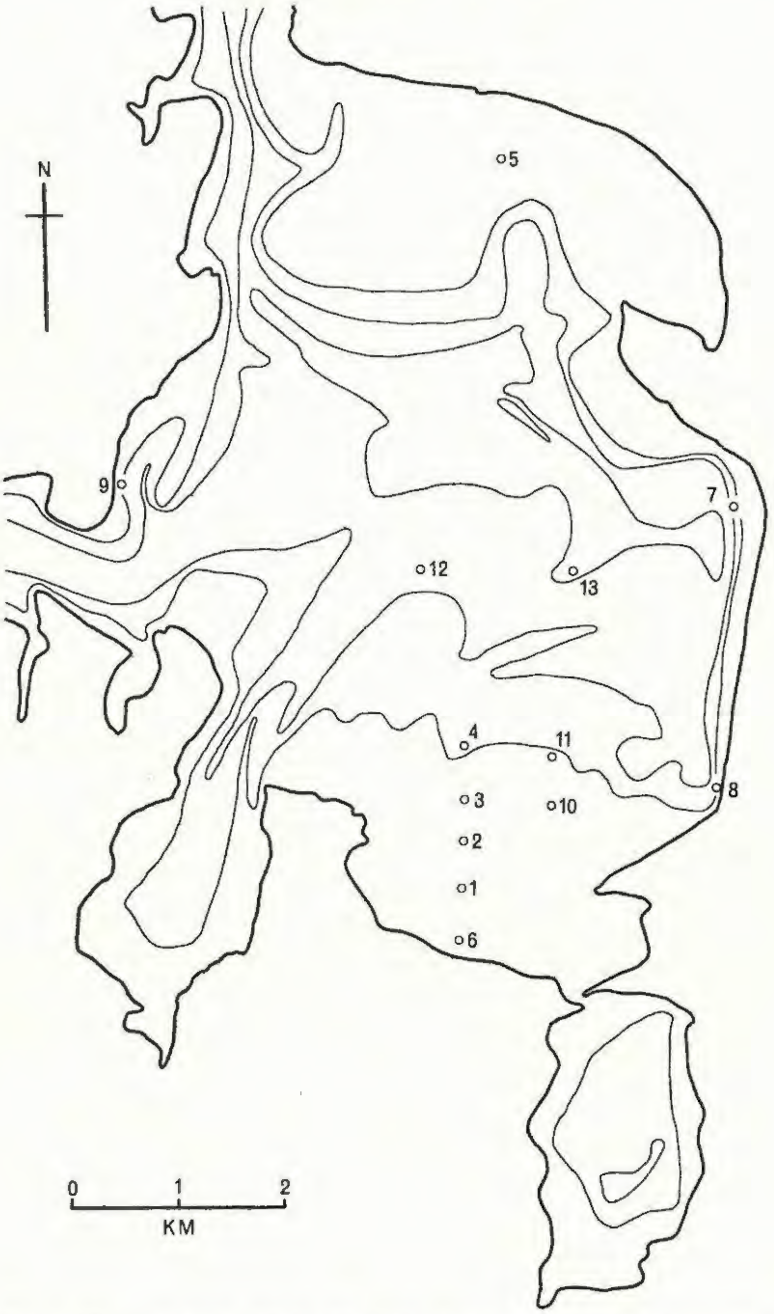

FIG. 1. The main basin of Lake Sibayi showing the position of diving stations (numbered). Depth contours at 10 metre intervals from Hill (1969).

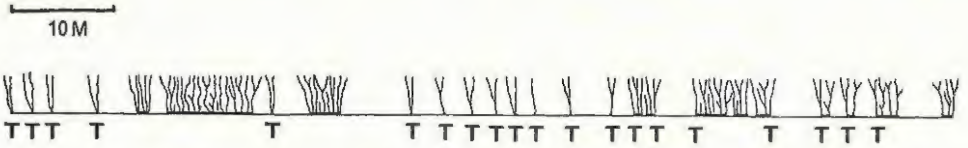

FIG. 2. Results of a traverse at station 2 showing distribution of *Myriophyllum spicatum*. The presence of nests of *Tilapia* is indicated by 'T'.

Eastern and western areas

In the east and the west of the lake, deep water occurs close inshore (fig. 1). A diagrammatic cross-section of the type of shore found in this area is shown in figure 3. This shore profile is almost certainly a result of wave action and represents a shore-face terrace resulting from the erosion of an originally steep sandy shore (King, 1958). The maximum depth of water over the terrace was 1·5 to 2 metres. Wave action could be felt at this depth and the terrace was therefore an exposed environment composed of rather loose shifting sand.

The slope at the end of the terrace (fig. 3) was extremely steep. Measurement indicated that its angle varied between 25° and 31° from the horizontal. This appeared to be the maximum angle at which sand could stand underwater and as a result the slopes tended to be unstable.

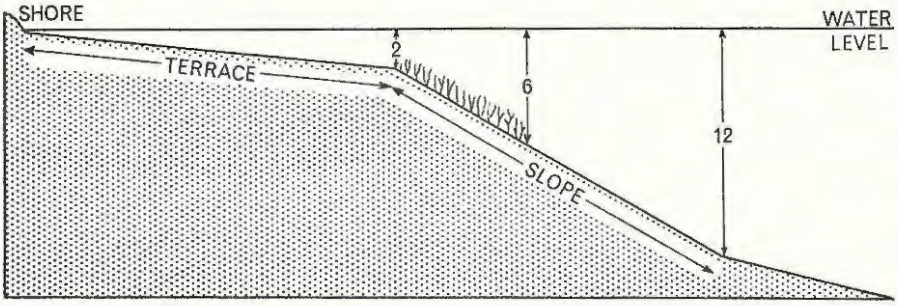

FIG. 3. Diagrammatic section of the terrace and slope showing distribution of plants. Depths are indicated in metres.

At Stations 7 and 8 (fig. 1), most of the weed on the slope was *P. pectinatus* and *P. schweinfurthii*. *P. schweinfurthii* was generally found at deeper levels than *P. pectinatus*. This may be as a result of the more delicate broad leaf of the former species, which probably makes it more susceptible to wave action. At Station 7, scattered plants of *Myriophyllum* also occurred on the slopes. However, at Station 8, both *Myriophyllum* and *Najas pectinata* (Parl) Magnus were extremely rare.

Weeds ceased abruptly on the slope at a depth of 7 metres on the eastern shore. However, on the western shore at Station 9 (fig. 1), the slope ended at a depth of about 4 metres, from which the bottom shelved gradually. At this station the slope was devoid of vegetation, probably as a result of the heavy wave action which was observed during both south-easterly and north-easterly winds. At a depth of about 4 metres, however, stands of both species of *Potamogeton* were found and *Myriophyllum* appeared below 5 metres. All weed cover ceased abruptly at about 6·5 metres, making the region below the terrace comparable to the gentle slopes of the northern and southern areas.

FISH

The majority of fish seen were either small species or juveniles. It appeared that adults fled from the presence of divers. The barbel *Clarias gariepinus* (Burchell) was only seen a few times and then always swimming away from divers. Important exceptions were the adults of *Tilapia mossambica* Peters and *T. sparrmanii* A. Smith which were frequently sighted, usually in association with nests or young. Divers approached within a few centimetres of these adults. The species of fish seen are dealt with separately as follows:

Tilapia mossambica Peters

T. mossambica constructs a nest by scooping out sand to form a pit (Jubb, 1967). In Sibayi the diameter of these nests varied between 0·5 and 1·5 metres in width with a maximum depth of 30 cm. The mean diameter of 30 nests measured near Station 8 was 1 metre. The nests were either directly associated with weed beds or were in close proximity to them (fig. 2). In areas where both *Potamogeton* and *Myriophyllum* were found, *T. mossambica* nests were always associated with *Myriophyllum*. However, nests were never found in the middle of extensive *Myriophyllum* beds, but only on the periphery of the beds and more especially alongside isolated bushes of weed (fig. 2). On several occasions *T. mossambica* was seen to hide in the overhanging weed on the edge of the nest. The only nests seen which were not associated with plants were in shallow water (less than 1 metre) on the terraces.

Although nests were confined to weed beds or shallow water, *T. mossambica* was not. Shoals of small *T. mossambica* were seen at depths down to 20 metres (Station 12). These fish appeared to be feeding and faecal ropes were common on the bottom. The presence of large faecal ropes suggested that bigger fish also lived and fed in the depths although they were not seen.

Tilapia sparrmanii A. Smith

Adults, together with shoals of young were seen on several occasions. However, they were only recorded on the slopes of the terrace and near plants.

Hemihaplochromis philander (M. Weber)

According to Crass (1964), this species is usually associated with small streams and the edges of lakes. However, in Lake Sibayi shoals of this species were positively identified in 7 metres and tentatively identified in 20 metres depth at Station 12—more than 2 kilometres from the shore. The shoals consisted of three or four males in breeding dress and up to twenty females.

Croilia mossambica Smith

When diving, holes about 1 cm in diameter were commonly seen in the sand. These proved to be the burrows of the goby *Croilia mossambica* (Plate XII). This fish has previously only been recorded from Lake Poelala in Moçambique. *C. mossambica* is most common on the terrace and slope below the terrace, but its distribution extends to at least 20 metres. Visibility at 30 metres (Station 13) was too poor to permit a search for holes.

Glossogobius giuris (Hamilton & Buchanan)

This goby was seen in all depths down to 20 metres. It was seen to inhabit holes which were apparently started by the activities of other animals. These holes had been untidily enlarged. This fish also took advantage of any other cover available such as empty tins.

Silhouetta n.sp.

This is a well-camouflaged fish which conceals itself on the open bottom, only swimming away when disturbed. They were recorded down to 20 metres.

Barbus natalensis Castelnau and *Gilchristella aestuarius* (Gilchrist & Thompson)

These species were only seen in shallow water, *B. natalensis* in less than 0·5 metre among reed stems, and *G. aestuarius* on the terrace. However, this latter species may also inhabit open water since the larvae were caught in plankton nets.

Discussion

Wave action appeared to be the chief factor limiting growth of macrophytes in shallow water. Submerged macrophytes were completely absent from the terrace which is a wave-washed area. *Ceratophyllum*, *Myriophyllum* and both species of *Potamogeton* grow at or near the surface in the sheltered arms of the lake. However, the present survey failed to reveal these plants in water shallower than 1·5 metres on the open shores of the main basin. Indeed on the exposed northern and southern shores, weed only commenced at a depth of 3·5 metres. The effect of shelter in permitting weed growth was shown by the presence of *P. pectinatus* at a depth of 1 to 2 metres in an underwater bay which occurred on the southern shore (Station 6). Presumably the shallower water surrounding this bay afforded the plants some protection from wave action. However, on the edges of the bay *P. pectinatus* was stunted. These observations agree well with those of Beadle (1932) on Lake Rudolph where *Potamogeton* was found in sheltered places behind sand bars and not at all in exposed regions.

Myriophyllum and *Ceratophyllum* were mainly found on nearly level bottoms. They were usually absent from the steep slopes below the terraces. If the terrace was exposed to heavy wave action as at Station 9, even *Potamogeton* was absent.

The lower limit of macrophyte distribution in the lake may be determined by limited light availability although pressure may in part also be responsible. Gessner

(1952) and Ferling (1957) (in Ruttner, 1963) have shown that submerged vascular plants show curtailment in various physiological functions in pressures in excess of 1 atmosphere (equivalent water depth 10 m). However, the macrophytes in Lake Sibayi ceased at a depth of 7 metres or less, indicating that light may be more important than pressure. Although no suitable light metre was available, divers gained the impression that at 6 to 7 metres the light became noticeably reduced. The Secchi disc reading in the lake was 3 metres (Allanson 1969). Beadle (1932) has shown that in Lake Naivasha, *Ceratophyllum* does not photosynthesise significantly below 5 to 6 metres. Correlated with this there was a lack of *Ceratophyllum* below 5 metres. Preliminary experiments in Lake Sibayi indicate a similar reduction of photosynthetic activity that occurs before depths at which pressure is likely to be the major factor inhibiting weed growth completely. Macrophytic vegetation in Lake Sibayi was therefore limited to a relatively narrow zone around the shores and except in sheltered areas, weed did not occur in shallow waters less than 1 to 2 metres in depth, nor did it penetrate into water deeper than 6 to 7 metres.

Observations on the nests of *T. mossambica* indicated that they were associated with cover, either in the form of plants or very shallow water. Since plants only occurred down to 7 metres in Lake Sibayi, the breeding-area of *T. mossambica* was limited to a relatively narrow band of shallow water around the shores of the lake.

The bottom of the lake at depths greater than 7 metres, although devoid of macrophytic vegetation, was rich in both animal and plant life. It was characteristically covered by fine organic detritus which was rich in diatoms and blue green algae. In addition, Boltt (1969) has reported that the sandy bottom of the lake has a large population of amphipods, isopods, tanaids and gastropods. This rich fauna and flora is probably an important source of food for the small cichlids which have been observed to feed down to 20 metres. Thus in Lake Sibayi the shallows are utilized as breeding-grounds and, in addition, the deep water substrates are included in the feeding-ranges of these fish species.

Prior to 1968 the only method which had been used in Lake Sibayi for sampling submerged macrophytes was a grappling hook operated either from the shore or from a small boat. This method suffers from the disadvantage of giving extremely little information about distribution. In addition a negative result does not necessarily indicate an absence of weed. The use of SCUBA techniques rapidly established not only the pattern of distribution of submerged macrophytes but also the relative abundance of the various species. It was also possible to relate the distribution to environmental factors which cannot normally be easily observed, such as the depth to which wave action could be felt.

SCUBA techniques also proved to be invaluable in regard to observations on fish. It is doubtful if any other method could have shown the striking association between *Tilapia* nests and weed in water deeper than 3 metres. In addition, divers discovered the presence of small fish in deep water, a fact that was entirely unsuspected and emphasizes the need for the inclusion of the small species in a consideration of the economy of the lakes. Clearly sampling methods for small mobile forms in deep water must be developed if a complete set of data for a lake system is to be obtained.

ACKNOWLEDGEMENTS

We wish to express our appreciation to the Research Grants Committee of Rhodes University for providing funds to purchase a portable compressor for filling aqualungs. In addition the South African Council for Scientific and Industrial Research through the I.B.P. has financed certain ancillary sections of the general research programme on Lake Sibayi which have greatly facilitated the work. Our thanks are also due to our colleagues for advice and assistance during the work and for constructive criticism of this paper. Finally we wish to gratefully acknowledge the encouragement of Professor B. R. Allanson who, as leader of the expedition, made this work possible.

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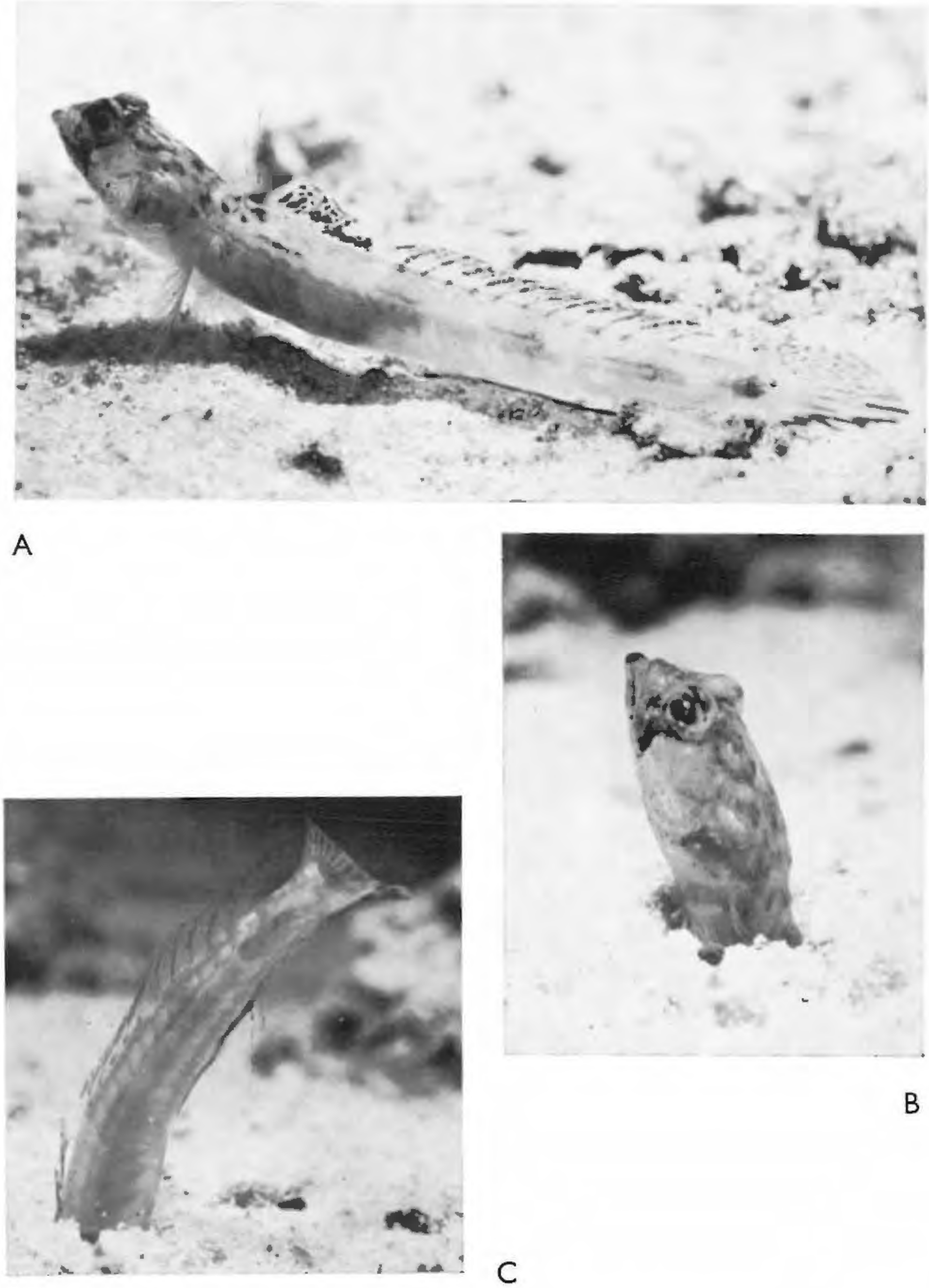

(A) The burrowing goby *Croilia mossambica*, (B) emerging from its burrow and (C) entering the burrow.

THE BENTHOS OF SOME SOUTHERN AFRICAN LAKES
PART II: THE EPIFAUNA AND INFAUNA OF THE BENTHOS OF LAKE SIBAYI*

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(With 2 text-figures)

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OPSOMMING

Die bodemlaag van die Sibayameer bestaan hoofsaaklik uit fyn sand, terwyl slik voorkom in die diepere dele wat vanaf die westelike en noordelike takke van die meer lei. Die fauna sluit 'n estuariene skaaldierkomponent en 'n polichaet in. Die varswaterkomponente bestaan hoofsaaklik uit chironomidelarwes, oligochete en molluske. Die vertikale digheidsverskille word vir drie soorte gegee; die ander diere is meer gelykmatig versprei wat diepte betref. Die bentiese ingegrawe diere is hoofsaaklik tot die sandige substraat beperk, terwyl die epifauna meer algemeen versprei is. Die fauna van die seegrasbeddings ver van die wal, in 5 meter diepte, is soortgelyk aan die van die plantegroei aan die rand. 'n Deel van die fauna vertoon seisoenvariasies in digheid. Die waarde van verskillende by die opname gebruikte metodes word bespreek.

SUMMARY

The bottom substrates of the freshwater Lake Sibayi are mainly fine sands. Silt substrates occur mainly in the valleys leading from the western and northern arms of the lake underwater in the main basin of the lake.

The fauna included an estuarine crustacean component and a polychaete. The freshwater component consisted principally of chironomid larvae, oligochaetes and molluscs.

Differences in density of animals were shown for three species in depths up to 20 metres and depths below 20 metres. Other animals are more evenly distributed with regard to depth.

The infauna of the bottom sediments is mainly confined to sandy substrates. The epifauna is more generally distributed. The fauna of weed beds far from the shore of the lake in 5 metres of water is like that of the marginal vegetation.

Some of the fauna showed seasonal changes in density.

The value of different methods that were used in the survey are discussed.

INTRODUCTION

The presence of essentially estuarine animals co-existing with freshwater animals in the freshwater Lake Sibayi has been reported by Allanson, Hill, Boltt & Schultz (1966). The presence of estuarine forms in fresh water in South Africa is apparently confined to Lake Sibayi, although at least one other brack-water lake which is closed off from the sea has similar animals (Boltt, in preparation).

An understanding of the biology of benthic animals depends on both field and laboratory observations. The present paper is a preliminary account of the results of field observations. Although only a limited amount of sampling has been possible, some clear, if unexplained, patterns have appeared from the results thus far obtained. Laboratory experiments may ultimately lead to some explanation of these field observations.

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This study has been confined to animals larger than 1 mm, and therefore has ignored the benthic microfauna. The animals reported on are therefore intermediary in any food chain since they feed on the microfauna and microflora. However, they afford food for fish and are therefore important in the economy of the lake.

METHODS

Since the benthic fauna of Sibayi may be roughly divided into closely spaced small animals such as the Crustacea and worms (the infauna) and larger widely spaced animals of the surface (the epifauna), two essentially different methods of sampling were used. These were, respectively, grab sampling from a boat, and belt transects by SCUBA divers.

Grab sampling. A Van Veen grab of sampling area 0·0225 m^2 was used during January and July 1967, and January and July 1968. Rubber flap valves were introduced into the grab for January 1968 and July 1967, since it had been shown that Van Veen grabs tend to wash away light material from the surface of the substrate (Wigley, 1967). The positions of the stations are plotted on a depth contour map (fig. 1) compiled by Hill (1969). These are only approximations, since it was not possible to define the position of the boat accurately in relation to the shore with the equipment available. Depths were measured with a weight lowered on a calibrated winch.

Wigley (1963) examined the efficiency of sampling by the Van Veen grab, and showed that it samples well to a depth of 4 cm. He also showed that at least for the estuary the great majority of animals live in the first 4 cm of substrate. In Lake Sibayi, with the possible exception of the polychaete, the fauna lives only a few millimetres below the surface of the sand, and therefore the grab is likely to catch almost all the animals under its nominal surface area.

Animals were washed out of the samples by swirling the water and decanting through a net, leaving sand behind. Checks on the recovery were done by adding formalin to washed samples and repeating the washing procedure. Such trials yielded no further recovery of animals.

In January 1967 and July 1967 material caught in the net was put in a white tray and the animals picked out by hand. In order to check the effectiveness of removing the animals by hand a number of such samples were inspected for overlooked animals by means of a scanning binocular microscope. Such inspection showed that in large samples approximately 2% to 3% of juvenile forms were overlooked. In samples with small numbers none were overlooked. The forms overlooked were always juvenile animals belonging to the numerically more abundant species. A binocular scanning microscope was used in the sorting of the material obtained in January 1968 and July 1968. The Crustacea were identified to the specific level as were the Polychaeta and Mollusca. Chironomid larvae, chaoborid larvae, oligochaete worms, and Nematodes were recorded as such. Other insect larvae were identified at least to the order.

As a check on the reliability of using the results of single grab samples from a

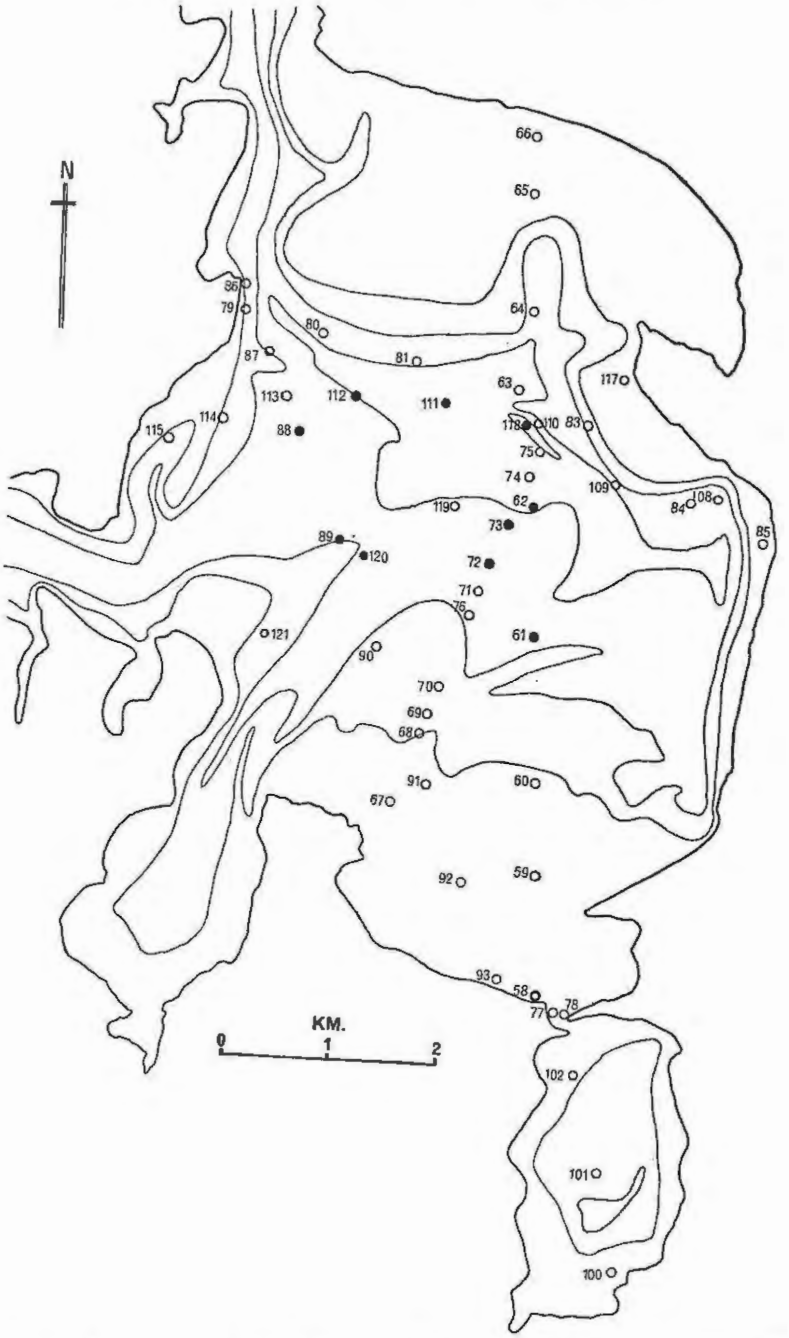

FIG. 1. Contour map of Lake Sibayi from Hill (1969). The sampling stations are marked and correspond with the SIB numbers of Tables 1, 7, 10, and 11. The positions of the stations is only approximate. Solid circles indicate silt substrates and open circles indicate fine sand substrates. The contour lines are marked in 10 m intervals.

single station, an alternative method of sampling the substrate was used. This method I have termed 'lines of boxes'. Two divers using SCUBA took down a 12 m rope marked at 0·5 m intervals. Clipped on to the rope were ten or twenty boxes with an opening of 10 × 10 cm. The line was laid straight out from a lead shot and samples taken by pressing the boxes gently into the substrate at each 1 m interval. A flat metal spade was pushed under the box to cut off the substrate sample in the box. The box was then turned over and a lid was slid over the box as the metal spade was withdrawn. The samples so collected were processed as described for grab sampling. The results of this procedure are reported with those of the grab samples.

Analysis of particle sizes in the substrate

An attempt was made to analyse the grain size of the substrate using the method of Lenard (1966), utilizing the author's apparatus. It was found that the results could be as much as 50% in error by dry weight for any fraction of grain size. Analysis of grain size was therefore carried out by the following method. A sample of about 15 g wet weight was thoroughly washed with distilled water and the washings were strained through a 0·04 mm aperture sieve and collected. The particles carried through were then settled with acetic acid and alum as recommended by Morgans (1956). This fraction of the substrate was then termed the subsieve fraction. The cleaned sands were then washed through a series of sieves with apertures of 0·5 mm, 0·25 mm, 0·20 mm, 0·16 mm, 0·08 mm, and 0·04 mm. All the fractions thus separated were dried at 105°C for 12 hours and weighed. A curve of cumulative percentages was then plotted for each sample and, once again following Morgans (1956), the median phi, phi quartile deviations and the phi skewness values were obtained. The subsieve fraction was also measured as a settled volume with a fresh aliquot of the substrate sample. Since the volume of the subsieve fraction is not necessarily proportional to its mass this was measured separately to give an indication of its importance in the composition of the sandy substrates.

RESULTS OF SUBSTRATE SURVEY OF LAKE SIBAYI

The results of the analysis of substrate from twenty-five stations are reported in Table 1. The stations from which the analysed samples were taken are shown in figure 1. The bottom sediments of Lake Sibayi may be divided into either fine sand or silt on the Wentworth scale.

The substrate is predominantly sandy. The muddy substrates are found in the submerged valleys leading from the western and the northern arms of the lake. The silt is not necessarily found in the deepest water as there is sand to be found in many of the samples from depths greater than 30 m.

The sand substrates were all extremely similar. The mean median ϕ value of twenty sand samples was 3·203 with a minimum value of 3·53. All these sand samples were well sorted with a mean ϕ quartile deviation, 0·28. However, varying quantities of fine subsieve fractions were present. Much of this material was sorted out as the subsieve fraction, although small amounts remained with the larger fractions, since they

TABLE 1

Table of analysis of substrate samples of Lake Sibayi. The phi values were calculated graphically according to the method of Morgans (1956). The positions of the Station numbers are indicated in fig. 1.

Station No.	*Depth*, m.	$Md\phi$	$Qd\phi$	$SKq\phi$	*% Subsieve dry weight* 0·04 *mm*	*Field Description*	*% Fine Material by volume*
SIB 86	0·9	3·25	0·18	+0·18	2·0	—	—
SIB 99	2	3·03	0·22	−0·12	1·0	Clean Sand	0%
SIB 67	5	2·95	0·51	−0·24	0·2	Clean Sand	—
SIB 102	8·3	3·16	0·33	0·06	1·6	Clean Sand	6%
SIB 91	9·4	3·00	0·22	−0·07	0·8	Clean Sand	8%
SIB 68	10·9	3·25	0·39	+0·18	1·9	Clean Sand	—
SIB 95	13·0	3·01	0·50	−0·1	1·6	Grey/Brown Sand	13%
SIB 79	13·3	3·22	0·20	−0·10	0·5	Clean Sand	10%
SIB 69	14·4	3·08	0·31	+0·21	0·6	Clean Sand	—
SIB 70	15·4	3·22	0·21	+0·15	1·2	Clean Sand	—
SIB 90	16·4	3·03	0·18	+0·06	1·3	Brown Sand	13%
SIB 76	20·6	3·0	0·20	−0·09	2·8	Grey Sand	—
SIB 71	23·6	3·17	0·37	+0·11	1·8	Grey Sand	20%
SIB 89	24·2	6	—	—	83·2	Mud	100%
SIB 87	24·5	3·31	0·17	+0·17	10·9	Grey Sand	8%
SIB 72	25·5	3·10	0·29	+0·03	1·0	Clean Sand	6%
SIB 80	27·8	3·30	0·21	+0·03	1·9	Grey Sand	26%
SIB 96	27·4	6	—	—	—	Black Mud	100%
SIB 73	28·6	6	—	—	—	Mud	100%
SIB 81	29·2	3·36	0·23	+0·15	2·3	Grey Sand	13%
SIB 62	33	4·5	—	—	63·5	Fine grey brown mud	—
SIB 82	35·5	3·56	0·45	−0·02	0·5	Grey/Brown Sand	36%
SIB 74	36·2	3·30	0·28	−0·03	0·9	Clean Sand	—
SIB 63	38	3·46	0·21	+0·01	2·5	Grey Sand	24%
SIB 75	39·3	3·53	0·45	+0·27	6·9	Grey/Brown Fine Sand	—

formed conglomerates. After drying, the subsieve fraction forms a very small percentage of the total weight of substrate (Table 1). This fine material may, however, form a considerable part of the substrate by volume. On wave-washed terraces the subsieve fraction is usually absent, but in general increases in importance with depth (fig. 2). This fine fraction consists of flocculent material containing large quantities of blue-green algae, green filamentous and unicellular algae, diatoms, and protozoa.

TABLE 2

Check list of animals from the bottom sediments of Lake Sibayi. Forms as small as ostracods were not counted due to the difficulty of sorting out these cryptic forms from amongst the sand grains with the unaided eye. Juveniles of the groups named were picked out, it being easy to distinguish them in the samples. *Denotes animals normally associated with estuaries.

Crustacea		
*Amphipoda:	*Grandidierella lignorum* Brnd.	Abundant in the sand.
	Corophium triaenonyx Stebb.	Common in the sand.
*Tanaidacea:	*Apseudes digitalis* Brown	Very abundant especially in deep water.
	Paratanais sp.?	Very rare.
*Isopoda:	*Cyathura carinata* (Kröyer)	Common.
	Pseudosphaeroma barnardi Monod.	Uncommon in grabs and never taken deeper than 5 m.
	Paramunna laevifrons. Stebb.	Very rare.

Branchyura	*Argulus?*	Single specimen from grab sampling.
*Decapoda:	*Hymenosoma orbiculare.* Desm.	Common.
Insecta	Chironomid larvae.	These were not identified beyond the family. The genera *Cryptochironomus* and *Clinotanypus* were identified from samples in July 1968 as was a Tanytarsini.
	Chaoborid larvae.	Very rare.
Annelida		
*Polychaeta:	*Ceratonereis keiskama* Day	Common in water less than 20 m deep.
Oligochaetes:	Recorded as such.	Common at all depths.
Mollusca:		
Prosobranchiata:	*Bellamya capillatus* Frnfd.	Common.
	Melanoides tuberculata (Müll.)	Common.
Pulmonata:	*Bulinus natalensis* (Küst.)	Common in weed fringe.
Lamellibranchiata:	*Corbicula africana* (Krs.)	Rarely taken in grabs.
Coelenterata		
Hydroidea:	Hydra sp.?	Single individual.

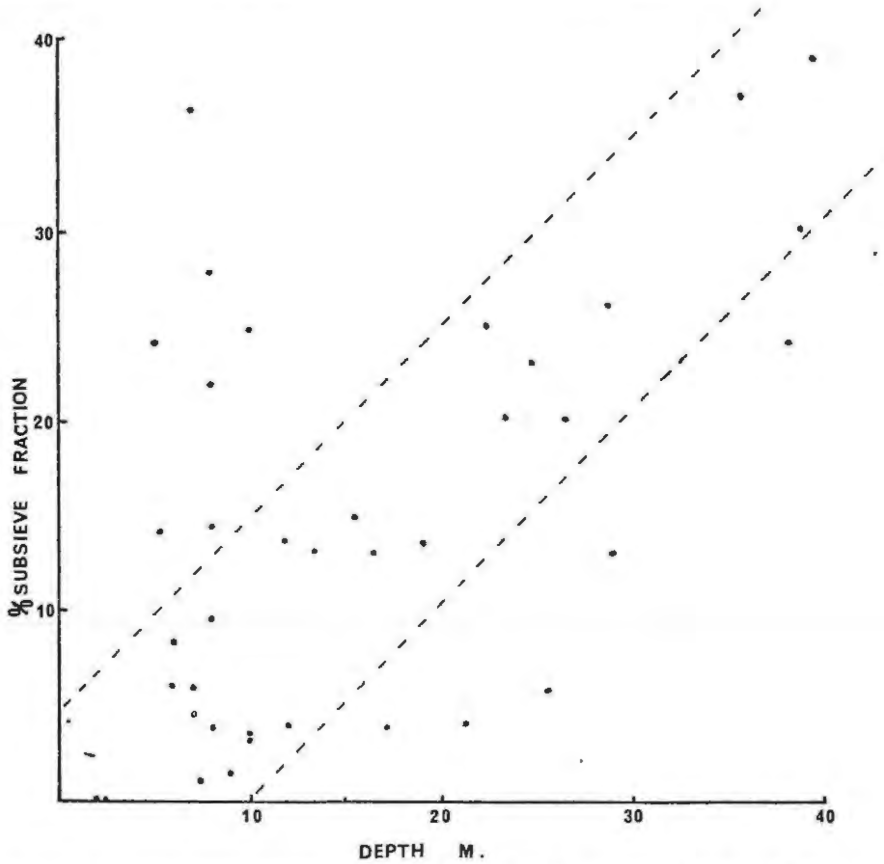

FIG. 2. The percentage subsieve fraction in sand samples is plotted against depth. The dashed lines are arbitrarily placed. Points above the line indicate samples from shallow water which might have high subsieve fraction contents because of special features on the bottom profiles. Those below the dashed lines may have reduced subsieve contents because the samples were from relatively steeply sloping profiles in the lake bottom. In general there is some indication that sediment content increases with depth in Lake Sibayi.

The silt bottoms form a sharp contrast to the sandy bottoms of Lake Sibayi and are best described as oozy, grey mud. Very little sand was found in the grabs, amounting to less than 1 % of the material. The median ϕ value was greater than 6, but could not be measured, since the particle sizes smaller than 0·04 mm were all measured together as a subsieve fraction.

RESULTS OF FAUNAL SURVEY OF LAKE SIBAYI

The faunal survey of benthic fauna has been limited to those animals which can be easily picked out from sorting trays and animals which could be seen by SCUBA divers on the bottom sediments. It should be noted that SCUBA divers would tend to miss very juvenile forms of the animals on the top of the substratum.

A check list of the animals found in the benthos of the lake is given in Table 2, from which it can be seen that both freshwater and estuarine components are present. Certain of the animals were rarely encountered in the grabs and in subsequent analysis of the data these have had to be ignored. *Paratanais?* (Tanaidacea); *Pseudosphaeroma barnardi*, *Paramunna laevifrons* (Isopoda); *Argulus?* (Branchyura); chaoborid larvae (Insecta) and *Hydra?* (Coelenterata), compose this group of animals and little can be said about them at this stage. A few are more characteristic of marginal vegetation, for example *Paramunna laevifrons*, *Hydra?* and *Paratanais?*. The isopod, *Pseudosphaeroma barnardi*, is more common on the shelf but this region has not been well sampled and hence only a limited amount of data is given in the tables.

Discounting strictly marginal vegetation, the main basin of the lake has been divided into the following habitats:

(*a*) Wave-washed terraces (Boltt *et al.*, 1969) up to 2 m deep.
(*b*) Weed beds from 2 m to 7 m.
(*c*) Sandy substrates.
(*d*) Fine silty muds.

TABLE 3

Results of dividing all the samples from fine sandy substrata into depth intervals of five metres except for the range 0 m to 2 m. This has been removed from the range 0 m–5 m since it represents the terrace region of the main basin. The samples have been lumped and a simple arithmetic mean of combined samples is recorded for each increase in depth. Only numerically important species have been recorded.

Depth	No. of samples	*Grandidierella lignorum*	*Corophium triaenonyx*	*Apseudes digitalis*	*Cyathura carinata*	*Ceratonereis keiskama*	*Oligochaetes*	Chironomid larvae
0–2	4	0·8	0·22	—	—	—	12·2	6·2
2–5	7	86·1	70·1	8·9	5·4	6·4	8·3	6·3
5–10	9	116·1	35·0	16·4	2·6	4·4	4·4	3·3
10–15	6	71·5	41·4	31·3	8·0	2·8	9·0	4·6
15–20	2	87·0	35·0	26·5	5·5	0·5	5·0	3·5
20–25	9	12·3	83·3	655·5	9·9	—	20·8	6·5
25–30	6	20·5	9·0	399·3	12·0	—	25·1	7·3
30–35	0	—	—	—	—	—	—	—
35–40	4	24·5	3·2	499·2	6·75	—	4·3	6·0

(*a*) *Wave-washed terraces*

The fauna of the terrace of the main basin in general is poor (Table 3). The infauna consists of oligochaetes, a few chironomid larvae and the amphipod, *Corophium triaenonyx.* Solid objects such as old tree trunks on the shelf harbour the tubes of the amphipods, *C. triaenonyx* and *Grandidierella lignorum.* The surface epifauna of the sands in summer consists of scattered individuals of the pulmonate, *Bulinus natalensis*, and the prosobranch, *Melanoides tuberculata.* A few individuals of another prosobranch, *Bellamya capillatus*, may also be found. In winter the shelf region is virtually barren of epifauna and numbers of dead shells litter the shore line, especially those of *M. tuberculata.*

Wave action on the shelf region is considerable, reaching to an average depth of over 2 m (Boltt *et al.*, 1969), and is responsible for the exceptionally clean sands of the terrace region. The sands of the terrace region show little or no measurable subsieve fraction by weight or settled volume. This may offer an explanation for the paucity of infauna in the terrace sands since they are so disturbed as to offer an unsuitable, and relatively barren place in which to live.

(*b*) *Weed beds from* 2 *to* 7 *metres*

It has been shown that, in the main basin, weed beds of *Potamogeton pectinatus*, *P. schweinfurthii* and *Myriophyllum spicatum* extend from 2 m to 7 m in Lake Sibayi (Boltt *et al.*, 1969). The weed beds are of two sorts, open stands of *Potamogeton* with *Myriophyllum* and dense stands of *Myriophyllum*, separated by patches of sand.

The fauna living on the weed is very different from that living on or in the sandy substrate beneath it. It may be termed the 'epiphytic' fauna, which is more like that inhabiting marginal vegetation. The density of the fauna is difficult to determine since it must depend not only on the amount of weed but also on the type of weed bed. Thus in the open stands of *Potamogeton* the commonest large invertebrate, *Bulinus natalensis*, gave a count of twenty-four snails/m^2 in thick weed and four snails/m^2 in a widely spaced community.

The *Potamogeton* weed beds are much poorer in both species and number of animals, while the close stands of *Myriophyllum* which occur in 5 m about 1 km off the southern shore of the main basin bears a fauna both rich in species and number (Table 4). Thirteen types of animals are recorded for the *Myriophyllum* beds and only six for the *Potamogeton.*

TABLE 4

Species lists with relative abundance of epiphytic fauna from open stand of Potamogeton and dense bed of Myriophyllum. + indicates present, ++ indicates common, +++ indicates abundant. The Trichopteran nymphs were *Orthotrichia* sp., *Leptoceris* sp., *Oecetis* sp. and *Ecnomus* sp.

	Potamogeton	*Myriophyllum*
Pseudosphaeroma barnardi	+	+++
Mayfly nymphs	+	+
Bulinus natalensis	+++	+++
Chironomid larvae	+++	++

	Potamogeton	*Myriophyllum*
Paramunna laevifrons	—	++
Corophium triaenonyx	++	+++
Grandidierella lignorum	—	+
Ceratonereis keiskama	—	+
Trichopteran nymphs	—	+
Hydra sp.	—	+
Ancylus sp.	—	+
Apseudes digitalis	—	+
Tanaid (undescribed SIB 98)	—	+++
Leech	+	—

(*c*) *Sandy substrates*

The sandy-substrate fauna may conveniently be divided into epifauna, living on top of the sand, and infauna, living in tubes constructed of sand (Amphipoda) or burrowing a few millimeters beneath the surface.

(i) Epifauna: The invertebrate epifauna of the sands consists of the larger forms; the gastropods, *B. capillatus*, *M. tuberculata*, *Bulinus natalensis* and the lamellibranch, *Corbicula africana*, as well as the crown crab, *Hymenosoma orbiculare*.

TABLE 5

The distribution of large invertebrate epifauna on the sand as recorded by SCUBA divers from belt transects. 'Present' indicates animals in the vicinity of the transect. + indicates animals common on the plants but not counted. The diving stations are those reported by Boltt (~~1968~~). Hill & FORBES (1969)

	Dive Station No.	Depth m	Area sampled m²	*M. tuberculata*	*B. capillatus*	*B. natalensis*	*C. africana*	*H. orbiculare*
Jan. 1968	1	3·5	2·5	8·0	Present	+	—	—
	5	5	2·5	39·2	Present	8·8	—	—
	8	9	2·5	23·2	0·3	—	—	Present
	3	9·0	5·0	4·4	Present	—	—	—
	6	9·0	9·0	13·4	1·4	—	Present	—
July 1968	5	3·3	12·5	1·8	—	—	0·1	0·1
	8	4·0	5·6	1·4	—	1·3	—	1·8
	5	5·2	5·9	4·7	1·4	2·6	0·2	1·7
	5	7·0	7·5	9·4	1·9	0·3	—	0·4
	8	15·0	5·6	—	0·2	—	—	2·0
	12	18·0	7·4	—	0·3	—	—	1·8
	7/8	24·0	5·6	0·9	0·7	—	—	0·2

The only form which was restricted in vertical distribution was *Bulinus natalensis*. This snail is only recorded from depths within the weed belt which reaches down to 7 m (Boltt *et al.*, 1969). Although it was not confined to the weeds themselves, since it was often taken crawling free on the sand, it may be restricted to the weed zone since it lays its eggs in a mass of jelly which seems to require a firm substrate on which to be placed. The weeds do not reach the surface in the main basin and the problem of how the animals replenish the gas bubble in the mantle cavity, if indeed they carry one, is exposed.

All the other forms of the epifauna have been collected by SCUBA divers down to 24 m (as deep as we have yet investigated). Evidence that members of this fauna occur in deeper water has been occasionally afforded by grab samples. Thus *Hymenosoma* has been taken in 36 m (SIB. 74), *Melanoides* in 35 m (SIB. 82) and *Corbicula* in 33 m (SIB. 62).

TABLE 6

Numbers of animals collected in 10 or 20 boxes of 0·01 m² along a straight line at 1 m intervals in 10 m (A) and in 21 m (B) depth of water.

		Grandidierella lignorum	*Corophium triaenonyx*	*Apseudes digitalis*	*Cyathura carinata*	*Ceratonereis keiskama*	Chironomid larvae	*Hymenosoma orbiculare*	*Corbicula africana*
A at 10 m									
Box No.	1	19	1	8	2	10	16	—	—
	2	15	7	3	2	2	17	—	1
	3	20	4	2	2	6	14	—	—
	4	23	6	2	—	3	20	—	—
	5	21	3	2	4	4	16	—	—
	6	21	2	5	3	6	10	—	—
	7	13	—	4	2	7	24	—	—
	8	13	1	3	2	4	30	1	—
	9	15	2	1	3	2	18	—	—
	10	22	—	2	1	1	13	—	1
Mean		18·2	2·6	3·2	2·1	4·2	17·8		
Standard deviation		±3·6	±2·2	±1·36	±1·04	±3·1	±5·4		
B at 21 m									
Box No.	1	7	4	225	4	—	12	—	1
	2	6	4	306	15	—	14	—	—
	3	2	11	258	8	—	12	—	—
	4	—	—	139	5	—	12	—	—
	5	—	6	139	9	—	11	—	—
	6	—	5	165	5	—	6	—	—
	7	—	3	113	7	—	1	1	—
	8	1	7	99	5	—	5	—	—
	9	—	6	156	7	—	3	—	—
	10	—	3	99	3	—	5	—	—
	11	3	7	141	7	—	4	—	—
	12	—	6	224	3	—	14	—	—
	13	—	7	153	3	—	5	—	—
	14	1	10	201	2	—	9	—	—
	15	2	5	215	7	—	18	—	—
	16	1	4	101	1	—	3	—	—
	17	1	10	157	5	—	8	—	—
	18	—	7	172	3	—	13	—	—
	19	—	1	77	2	—	11	—	—
	20	—	3	296	9	—	9	—	—
Mean		1·2	5·5	171·8	5·0	—	8·7		
Standard deviation		±0·1	±2·7	±45·2	±3·2	—	±4·5		

Seasonal differences in number are apparent for *Melanoides* and *Hymenosoma*, the former with decreased numbers in winter and the latter with increased numbers in winter. The numbers recorded for *Bellamya capillatus* are too small to estimate the changes in number during the seasons. The areas measured by SCUBA divers in summer were inadequate and in winter barely so. *Corbicula* is altogether too uncommon on the bottom for its density to be easily or confidently measured. The figures for *Bulinus* are difficult of interpretation since the animals occur both on weed and on sand and, as has been indicated, seem to be related to weed density. Weed density moreover varies with season.

(ii) Infauna: Some measure of the distribution of animals in the bottom sediments is afforded by the 'lines of boxes' method. The results are reported in Table 6 which are from samples taken at 10 m and 20 m.

Over relatively short lengths, at least, the results show that single samples will give a reasonably accurate indication of the numbers of any one type of animal occurring in the bottom. The area covered by each 'box' (0·01 m^2) is less than that covered by the grab (0·0225 m^2) and hence the grab is likely to be slightly more reliable than single 'box' samples. Although there seem to be small variations over small areas, as Table 3 and Table 7 indicate, there is considerable variation over wider areas of the lake. Nevertheless, at least some of the variation is systematic and leads to a pattern of distribution in the lake which at the moment is discernible only from the common animals in the benthos of the lake.

TABLE 7

The individual results of a line of Stations shown on fig. 1. This line was an attempt to take samples at approximately 5 m increments in depth. The single muddy sample had a median value of 4 and was devoid of animals. Sample size 0·0225 m^2.

Station No.	Depth	Description of substrate	*Grandidierella lignorum*	*Corophium triaenonyx*	*Apseudes digitalis*	*Ceratonereis keiskama*	*Cyathura carinata*	Oligochaetes	Chironomid larvae
SIB 67	5	sand	131	12	2	3	0	11	0
SIB 68	10·9	sand	40	73	25	4	1	8	14
SIB 69	14·4	sand	79	69	96	7	1	0	12
SIB 70	15·4	sand	69	31	30	0	0	6	5
SIB 76	20·6	sand	15	5	35	0	2	0	1
SIB 71	23·6	sand	2	0	714	0	15	9	12
SIB 72	25·5	sand	0	0	77	0	1	5	0
SIB 73	28·6	mud	—	—	—	—	—	—	—
SIB 74	36·2	sand	2	0	117	0	11	8	1
SIB 75	39·3	sand	0	0	26	0	6	4	0

Those animals which are found consistently in the lake are the amphipods, *Grandidierella lignorum*, *Corophium triaenonyx*, the tanaid, *Apseudes digitalis*, the isopod, *Cyathura carinata*, the polychaete, *Ceratonereis keiskama*, the oligochaetes and chironomid larvae. Of these the first three are most numerous. The isopod, although less numerous, is important in view of its larger size.

Two types of distribution are shown in Tables 3, 6 and 7: firstly a 'discontinuous' distribution of particular animals which were found in higher numbers in different depths of water, and secondly other animals which were evenly distributed throughout the depth of the lake. The discontinuously distributed forms are the amphipods, tanaids and *Ceratonereis*. *Grandidierella* appears in greater numbers in depths of 2 to 20 m than in water 20 to 40 m deep. The mean for all samples from sandy substrates from 2 to 20 m is 98·1 (± 48·8) and for 20 to 40 m is 16·65 (± 24·9)/0·0225 m^2. Using Fisher's method for small uncorrelated samples, 't' was calculated and found significant at $p = 0 \cdot 01$. A comparison of density means for *Corophium* using the same technique was not significant. However, there is some reason to believe that with a larger set of samples the distribution of this species may prove to be also discontinuous.

High numbers of *Apseudes digitalis* were taken from sandy substrates between 20 m and 40 m depth. The average number of individuals was 515 per grab sample for water deeper than 20 m, which contrasts markedly with 190 for sands at shallower depths of 2 m to 20 m. A summary of the range of number of individuals per sample is given in Table 8.

TABLE 8

The frequency with which certain ranges of density of *A. digitalis* occurs in samples drawn from depths of 2 m to 19 m and 20 m to 40 m.

Density of animals/ 0·0225 m^2 grab sample	No. of samples 2 m–19 m	20 m–40 m
0–9	11	1
10–49	11	1
50–99	2	1
100–499	—	9
500–999	—	5
1000–1500	—	3
Total No. of Samples	24	20

Differences in the numbers of oligochaetes with depth cannot be dealt with as there are several genera in the samples which have not been taken into account.

Perhaps the most strikingly discontinuously distributed animal is the polychaete, *Ceratonereis*, which was never found below 20 m.

Evenly distributed animals are the isopod, *Cyathura*, oligochaetes and chironomid larvae. There may be some suggestion of increased number in deeper water but this is almost certainly due to a few samples which had high numbers of juvenile forms in them as compared with the great majority which had only adults present. The chironomid larvae have usually been lumped together. In the survey of July 1968 some attempt was made to identify to lower systematic categories. *Cryptochironomus* and *Clinotanypus* were found as was a tanytarsini and *Tanytarsus?*. Too few individuals were found in grabs to decide on patterns of distribution, if any.

Although only one summer sampling has been taken (January 1967) and two winter samplings (July 1967 and July 1968) there is some slight evidence of seasonal

TABLE 9

Samples from January 1968 (Summer S) and samples from July 1967 and 1968 (Winter W). No. of samples in brackets after S.D.

	Mean 2 m–20 m	S.D.	Mean 20–40 m	S.D.
G. lignorum				
S	79·9	±36·5 (14)	2·7	± 3·9 (10)
W	116·3	±55·4 (10)	30·5	± 29·3 (10)
C. triaenonyx				
S	37·8	±32·8 (14)	2·1	± 4·2 (10)
W	31·5	±29·3 (10)	12·1	± 7·1 (10)
A. digitalis				
S	17·6	±25·2 (14)	340·7	± 80·1 (10)
W	21·1	±26·7 (10)	689·1	±451·7 (10)
C. carinata				
S	6·1	± 5·8 (14)	4·4	± 4·0 (10)
W	3·4	± 2·0 (10)	14·4	± 7·7 (10)

variations in number. The numbers have been averaged by season and for depth for each of the more numerically abundant species (Table 9). In the winter, *Grandidierella* showed increases in number in both shallow and deep water. Increases in number of individuals of *Apseudes* was only apparent in deep water. The apparent increase in number of *Cyathura* in deep water was due to an increase in small juveniles in some of the samples.

TABLE 10

Table showing the results from samples with 100% silt contents. For a discussion of the results see text.

Station No.	Depth m.	*Melanoides tuberculata*	*Bellamya capillatus*	*Apseudes digitalis*	*Grandidierella lignorum*	*Corophium triaenonyx*	Chironomid larvae	*Cyathura carinata*	Remarks
SIB 101	17·5	2	—	—	—	—	—	—	100% silt.
SIB 88	24·6	6	—	1	—	—	—	—	,,
SIB 73	28·6	—	—	—	—	—	—	—	,,
SIB 106	9·0	—	1	27	—	—	3	1	,, Western Arm
SIB 96	27·4	1	—	3	—	1	—	—	,,
SIB 61	25·0	—	—	1	—	—	—	—	,,
SIB 62	33·0	1	—	1	1	—	1	—	,,
SIB 5	22·3	2	—	1	—	—	—	—	,,
SIB 112	22·6	1	—	1	—	—	—	—	,,
SIB 89	24·2	—	—	399	29	20	—	4	,, Western Arm
SIB 111	25·7	2	1	9	—	—	—	—	
SIB 110	40·2	—	—	178	—	1	1	10	a little sand.

(*d*) *Silt substrates*

Twelve stations with substrates having a median ϕ value greater than 4 have been sampled from Lake Sibayi. These samples generally showed a poor infauna (Table 10) with the exception of two stations from the western arm and SIB 89 from the main basin. Of the epifaunal elements *Melanoides tuberculata* appeared in four samples and *Bellamya capillatus* in one.

Discussion

Some understanding of the distribution of the essentially fine sand and the essentially silt substrates in Lake Sibayi may be gained from a consideration of how the lake was likely to have formed and its subsequent history. Hill (1969) has shown that the lake was formed from the blocking up of an old estuary originally cut in Tertiary sands. This accounts for the similar median ϕ value in all depths of the sandy substrate. The lake has not been subject to serious silting since its formation, and what silt has been found occurs in the valleys leading into and carrying on under the surface of the main basin. It is, however, curiously absent from the deepest portions of the lake to which these valleys lead. This silt may have come from either the streams leading into the lake or from generally wind-blown material falling on the lake. The presence of the material in the valleys, but not in deeper water, tends to suggest that it has come down the valleys from the streams of the western and northern arms of the lake and is slowly proceeding towards the deep portion of the lake.

The origin of the subsieve fraction found in the sandy samples may well come partly from wind-blown material and partly from the detritus falling on to the bottom from plankton, and also fish faecal ropes which were seen by the SCUBA divers on the bottom during many dives. There may be a tendency for this light material to move slowly into deeper water due to water disturbances generated by the wind in the shallower water, and hence the tendency for the subsieve fraction to increase with depth.

The benthos of Lake Sibayi was originally sampled exclusively by means of a Van Veen grab of rather small bite. During the analysis of the material collected there was a constant feeling that many problems would be much clearer if one could go down and 'see for oneself'. The introduction of a short SCUBA diving programme in January and July 1968 showed that many of the ideas which had evolved about the distribution of the animals in the lake had to be greatly altered.

One of the most striking results of the diving programme was the discovery that a number of benthic animals, previously only rarely captured in the grabs, were of considerable importance in the description of the benthos of the lake. Furthermore, some of the species found were completely unsuspected from grab results, namely typically benthic fish such as *Croilia mossambica* and *Silhouetta* nov. sp. It is doubtful whether any sampling method without visual inspection would have given meaningful results, quite apart from the conceptual difficulty of sampling for animals which might never have been there. A combination of techniques has, on the other hand, produced a pattern of faunal distribution in the bottom of the lake. However, the techniques used have also revealed shortcomings. The 'lines of boxes' showed that for certain small forms a single grab at any one spot in the lake was likely to give a fairly representative number of animals at any one site in the benthos of the lake, but not for widely spaced stations. It would seem that although the present data are inadequate to give a completely quantitative picture of the density of animals in the lake, a large number of reasonably closely spaced grab samplings would yield reliable information for small animals. The grab samples are, however, completely unrepresentative of the density of larger forms, the molluscs, the crown crab and fish. In order to try to overcome this problem, a number of methods using diving apparatus were tried. One such method involved turning a rigid frame over and over on the substrate and was the principal method used in January 1968. It soon became apparent that *Melanoides* was now being sampled adequately, but that *Bellamya*, *Corbicula* and *Hymenosoma* were not. It was also apparent that with such active forms as fish there was no hope of even estimating their numbers with the techniques available. One exception was the counting of holes of *Croilia mossambica* Smith. Even this is fraught with difficulty since it has been shown in the laboratory that the animal may cover its hole temporarily and hence the holes are no measure of the fish's abundance. This also points to the importance of knowing the biology of an animal so as to help interpret data from the field. In January a square covering 9 m^2 was laid out on the bottom (Dive station 6, Table 5). The results of this indicated that some measure of the density of *Bellamya* was now being gained. However, the technique was too clumsy for routine sampling by divers. During July 1968 a further method of laying out parallel tapes was used and larger areas were being covered with better measures of at least *Bellamya* and *Hymenosoma*. However, the problem is not yet solved due to the poor visibility in the lake and the fact that disturbance of the bottom of the lake by the activities of the divers stirred up the fine material over the sand on the bottom, which tended to decrease visibility. The divers were therefore obliged to detect the material partly by touch and partly by sight. It was noticeable that in the collections of epifauna made by divers no really small juvenile forms were present. This is in contrast to the grab-sampling which had a good recovery of small juvenile forms.

It is with these difficulties in mind that one must view the data at hand. The recovery of animals from the substrate has not been even, in the sense that the area from which a sample is taken while being demonstrably adequate for some species, is barely adequate for others.

The density of animals in the benthos of Sibayi has been measured by counting the numbers of animals from known areas. While this method gives some insight into certain biological problems and raises others, it is not a good method for assessing the relative importance of different species utilizing the food resources of the bottom sediments. Thorsen (1957) has pointed out the importance of relative size of different species affecting their density on a flat marine bottom. The same is probably true for the benthos of lakes.

In Lake Sibayi high numbers of small animals are characteristic of the infauna, while low numbers are characteristic of the epifauna. The former might be measured in mm while the latter in cm. The question of the relative importance of the epifauna must therefore be answered with reference to whether they are consistently found on the bottom of the lake or not.

General observation by SCUBA divers showed that the molluscs *Bellamya capillatus* and *Melanoides tuberculata* were the commonest large invertebrates of the epifauna. Data presented in Table 5 show that *Bellamya* occurs in somewhat low numbers whereas *Melanoides* was found in higher numbers, at least in summer.

However, it is believed that both are at least equally important in the lake since *Bellamya* is larger (approx. 1·14 gm) than *Melanoides* (approx. 0·37 gm). The difference in size might imply that the larger animal is more widely spaced, since it requires a larger area over which to forage for food.

Quantitative data are available for *Bellamya* and *Melanoides* for depths of only up to 24 m. The fact that these animals were occasionally obtained in grabs from deeper water on both types of substrate must indicate that they are generally distributed throughout the lake. If this is so these molluscs are of at least equal importance with the infauna, since they must account for considerable quantities of the food resources in the bottom sediments.

These same arguments apply to a consideration of the distribution of the crown crab, *Hymenosoma orbiculare*, which is also large relative to the infauna. These crabs are difficult to sample since they are capable of swift escape movements and are highly active. Water disturbances caused by the descending grab may well have stimulated most crabs to escape. Divers noted that when disturbed the crabs swam away, and it was often only this escape movement which revealed their presence, since they are very cryptically coloured, and dig into the sand, leaving only their eyestalks visible. This crab was present in the grab samples from 5 stations (SIB 58, SIB 55, SIB 74, SIB 78, SIB 77), ranging from shallow water on the shelf down to 36·2 m depth. One must conclude therefore that as with the mollusca, *Bellamya* and *Melanoides*, these crabs are an important element in the fauna of the benthos.

The presence of actively moving forms associated with the invertebrate epifauna of the lake has already been reported by Boltt *et al.* (1969). These are the Gobiid fish, *Silhouetta* nov. sp., *Croilia mossambica* Smith and *Glossogobius giurus* (Hamilton-Buchanan). Since these animals have a reduced swim-bladder they cannot be pelagic and are necessarily always on or near the bottom. At least the first two species are prominent members of the benthos as judged by divers working on belt transects.

It has been shown that the infauna of the fine sand substrates is made up of a discontinuously distributed component, and an evenly distributed component in terms of depth. It has also been shown that in general there tends to be an increase in the quantity of fine material in the sandy substrate with increasing depth (fig. 2). It is tempting to suggest that there might be a causal relationship between the increase in fine material in deeper water and the reduction of numbers of Amphipoda and *Ceratonereis keiskama*, and the increases in number of Tanaidacea. However, inspection of the data in Table 11 shows that fine sand bottoms of high subsieve fraction content in water less than 20 m depth carry a pattern of fauna much like that of sediments from shallow water with less fine material. Conversely, stations with low fine material content in the fine sands from deep water still show a high tanaid number similar to substrates from similar depths with a high fine material content. Therefore no good explanation is forthcoming as to why there is a discontinuous distribution with relation to depth in these forms. There may of course be qualitative differences in the fine material which is found in the sandy substrate of the lake. It has been shown that macrophytic plants cease in a band around the lake at some 7 m depth and that this might be correlated with light penetration (Boltt *et al.*, 1969). Microscopic

TABLE 11

Although generally there is an increase in fine material in the sandy substrates with increasing depth, there are exceptions. These have been emphasized in the table by placing a box around them. The species distribution appears to be more a question of depth rather than of the proportion of fine material in the sand. Area sampled was 0·0225 m².

Station No.	Silt %	Depth m	*Grandidierella lignorum*	*Corophium triaenonyx*	*Ceratonereis keiskama*	*Apseudes digitalis*	*Cyathura carinata*	*Oligochaetes*	Chironomids
SIB 117	2	2	3	—	—	—	—	—	15
SIB 99	0	2	44	1	—	—	—	—	2
SIB 64	0	15	52	1	—	10	9	6	12
SIB 92	1	7·5	160	13	7	4	2	—	9
SIB 100	4	6	181	5	—	20	2	—	—
SIB 85	4	0·7	—	1	—	—	—	17	3
SIB 76	4	20·6	15	5	—	35	1	2	—
SIB 72	6	25·5	—	—	—	77	—	1	5
SIB 102	6	8·3	148	5	7	17	4	5	1
SIB 74	7	36·2	2	—	—	117	1	11	8
SIB 55	7	5·9	59	2	—	1	—	1	1
SIB 93	7	4·4	94	103	24	1	2	2	6
SIB 69	8	14·4	79	69	7	96	12	1	—
SIB 91	8	9·4	206	33	15	5	3	12	4
SIB 78	8	3·7	34	3	3	12	4	15	11
SIB 66	9	1·4	—	—	—	—	—	32	2
SIB 79	10	13·3	159	30	—	18	2	1	5
SIB 67	10	13·1	5	131	12	3	—	2	11
SIB 87	10	24·5	30	17	—	1214	14	33	—
SIB 121	12	12·2	286	19	—	8	1	6	6
SIB 81	13	29·2	—	4	—	521	7	19	9
SIB 68	13·5	10·9	40	73	4	25	14	1	8
SIB 95	13	13·0	43	24	6	21	7	20	—
SIB 80	13	16·4	136	39	1	29	6	10	1
SIB 56	14	5·2	77	100	—	1	5	—	6
SIB 70	15	15·5	69	31	—	30	5	—	6
SIB 58	17	3·7	135	49	12	3	20	9	—
SIB 98	20	25·5	10	10	—	279	11	16	2
SIB 71	20	23·6	2	—	—	714	12	15	—
SIB 60	22	8·8	45	25	2	—	2	—	6
SIB 119	23	24·9	14	2	—	1431	18	27	—
SIB 65	24	5	97	5	—	1	1	2	2
SIB 63	24	38	1	—	—	488	2	5	14
SIB 83	25	22·5	2	3	—	675	7	32	39
SIB 80	26	27·8	1	—	—	396	9	5	26
SIB 118	30	39·3	95	12	—	1241	24	1	1
SIB 82	37	35·5	—	—	—	5	—	—	1
SIB 75	39	39·5	—	—	—	26	—	6	4
SIB 59	63	7·8	74	9	—	4	1	1	1
SIB 84	63	29·6	—	—	—	2	—	—	1

examination of the subsieve fraction from 10 m shows that it is rich in diatoms, monofilament blue green algae and protozoa. Light may affect the type of species found in different depths of the lake and hence the fauna. There may, however, be other factors of which, as yet, we are ignorant. Thus Enright (1962) has shown that amphipods may react to changes of pressure by swimming up into the plankton. However, Enright does not report any ability in these animals to register absolute pressure.

Discontinuous distributions have been shown for the fauna of at least one lake in Canada. Rawson (1930 in Welsh, 1935) has shown that in Lake Simcoe the amphipoda, trichoptera, and ephemeroptera do not extend below 20 m. While this is perfectly understandable as far as the insects with an aerial adult are concerned, this is less explicable in terms of the amphipoda.

In summary, therefore, we must leave this problem largely unexplained.

The contrast in animal numbers from fine sandy substrates and the number taken from silt substrates is marked. Although it must be borne in mind that it is only from grab samples that we have data, certain conclusions seem valid. The larger epifaunal elements are present in some of the grab samples. In the light of previous argument it would seem that these animals are equally distributed over the sand and the silt substrates. The number of animals belonging to the infaunal elements is, however, much reduced when compared with the results from sandy substrates. Although definite conclusions cannot by any means be drawn, some at least of the absenteeism can be partly explained. Both the tube building amphipods use a reasonably undisturbed substrate on which to construct their tubes. Although *Corophium triaenonyx* uses only fine silt particles cemented together with a tanned protein (Rao & Shyamasundari, 1963) these amphipods are found on solid obstacles on the substrate, or even on plant stems and poles for harbours. They furthermore appear to be entirely filter feeders and it is possible that an increase of fine material reduces the efficiency of the feeding or respiratory mechanisms due to clogging.

Grandidierella lignorum cements larger sand particles together to build a tube which is lined with fine silt particles. Although it can cope with only silt if this material will conglomerate into lumps of the size of a fine sand grain, they appear only to use this method when sand particles are unavailable. They feed both by filtering particles out of the water by means of their hairs on the antennae and also by working the mouth parts over sand grains in a kind of scrubbing motion. The legs, especially peraeopod 5, have a dense brush of setae on the posterior border. It seems likely that these brushes and other setose parts may become clogged by silt particles in the water.

The lack of chironomids in this type of substrate is harder to understand, save for the fact that they appear to belong to genera associated with sand rather than pure silt substrates. Similarly the tanaids and other generally burrowing forms are difficult to interpret since we are very ignorant of their biology.

The seasonal variation in the numbers of *Melanoides* and of *Hymenosoma* is very intriguing, as is the absence of animals from the terrace regions of the main basin in winter. Allanson & Van Wyk (1969) has reported on the temperature régime of terrace region of the lake margin. In summer it was shown that although the day temperatures rose some 4°C above the ambient lake temperature (± 28°C) at night there was little difference between lake temperatures and temperatures occurring over the terrace. This is not true in winter when general lake temperature may fall to 17°C (the water on the terrace region has fallen on very cold nights to as little as 13°C). It may well be that low temperatures at the margin of the lake drove away or killed the animals in that region of the lake. There were numbers of empty gastropod shells and a general absence of life on the shelf in winter which bore evidence to this.

The large fluctuations between the numbers of *Melanoides* counted in summer and in winter from the deeper waters of the lake are harder to explain. The animals may have burrowed into the substrate and thus were not visible to SCUBA divers. This is unlikely since, although they show the burrowing behaviour quite freely, they usually leave a disturbed track in the sand. Furthermore collection of animals by divers was carried out both by sight and by feeling through the sand so as to disturb *Hymenosoma*, and many snails were discovered at the same time.

The contrast in the numbers of *Hymenosoma orbiculare* between summer and winter was most striking, and somewhat more easily explained. Forbes & Hill (1969) and Broekhuysen (1955) have shown that the breeding of *Hymenosoma* is distinctly seasonal and that the crabs do not live for much more than a year. Although animals are to be found in berry all the year round in Sibayi, the major breeding period is the spring and the crabs begin to perish a little over a year later. In summer the population may still be mainly juvenile and in the plankton and only later does it descend and swell the population in the bottom of the lake in winter.

Some evidence for seasonal fluctuations in number is presented for infaunal crustacea. While the egg production of the crab is markedly seasonal the same is not true for the amphipods and tanaids. Egg-bearing females are always present in samples from the benthos of Lake Sibayi. The fluctuation in numbers in the benthos must be looked for in other phenomena. Principal among these in the physical environment of the lake is temperature. The reported drop in temperature of some 10°C between summer and winter would delay the development of eggs in the brood pouches of the amphipods and the tanaids. Generation times would therefore be extended in winter relative to summer. Further the warming and cooling of the lake will inevitably lag behind mean air temperatures although this lag has not yet been measured. Increased numbers of generations in the warmer months of the lake may lead to a build up in population which would be highest as the lake begins to cool down. Thus the population may be highest at the time when the generation time is longest. This, however, does not account for a drop in the numbers encountered in the summer. Two observations may be relevant. *Hymenosoma* which is in its highest numbers in winter feeds by probing the sand with its chelipeds. Presumably it will eat any small crustacean that it comes across. These animals would then be preying heavily on the amphipods and tanaids when the recruitment to these populations is slowest, due to increased time for development of eggs. They might then reduce the population. Conversely, the crab is least common in summer. They in turn are heavily preyed on by fish, in particular *Clarias gariepennis*. Quantities of *Hymenosoma* have been recovered from gut contents of these fish, and the crab populations may be reduced by *Clarias*.

The second factor which may well be of importance is the sensitivity, which can be shown experimentally of amphipods, to small increases in CO_2. It may be that the increased summer temperatures of the lake increase the oxygen demand of the fine material of the bottom with consequent increases in CO_2. The amphipods react to an increase in CO_2 by abandoning their burrows and swimming off into the water. They may thus migrate in the warmer weather to shallower regions of the lake.

For all the shortcomings of the methodology and the scope of sampling in the lake, the field results have raised a number of problems of which the following are the most important: (i) the discontinuous distribution of *Grandidierella*, *Apseudes* and *Ceratonereis*; (ii) the problem of seasonal fluctuations in the density of some of the benthic fauna; (iii) the lack of animals as infauna in silt substrates; (iv) the role in the food web played by the subsieve fraction, the snails, and *Hymenosoma*, in the lake, and (v) the problem of the presence of estuarine forms living in fresh water in Lake Sibayi but apparently nowhere else in southern Africa. Some of these problems can be solved by more extensive sampling in the lake, but difficulties of interpretation of others are due to a lack of knowledge on the biology of the animals concerned. It is felt that critical experimental investigations into the biology of these animals will undoubtedly help to solve some of the problems of distribution of animals in the lake.

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THE BENTHOS OF SOME SOUTHERN AFRICAN LAKES

III. The Benthic Fauna of Lake Nhlange

The presence of two South African lakes on the coastal plain in Tongaland led to the hope that a study of the benthic fauna of one might lead to a better understanding of the other. The benthic fauna of Lake Sibayi has already been reported on (Boltt 1969), and although the water is fresh, estuarine animals occur in the lake. Lake Nhlange, which is some 30 miles north of Lake Sibayi, is brack with a salinity of 3°/oo to 4°/oo (Fig. 1). Lake Nhlange is connected by channels to the Kosi estuary system which is normally open to the sea (Fig. 2). The fauna of Lake Nhlange is mainly estuarine, but the species from the benthos of the lake were markedly different from those occurring in Lake Sibayi. Comparisons between the two lakes has raised more problems than it has solved, and has drawn attention to the importance of detailed investigations into the biology of the animals in the coastal lake systems of South Africa before an understanding of the problems raised is likely to be gained.

Most unfortunate was the fact that the first detailed sampling programme of Lake Nhlange benthos was done shortly following a heavy flood in January 1966. The conditions of the lake were not stable and the significance of dynamic changes that were taking place in the bottom of the lake was not fully appreciated until late in the period of investigation. Although the broad chemical characteristics of the lake are well documented by Allanson & van Wyk (1969) adequate chemical measurements were not always taken in the lake at the bottom sediment/water interface. This has made interpretation of the conditions pertaining on the bottom of the lake largely a matter of educated guesses.

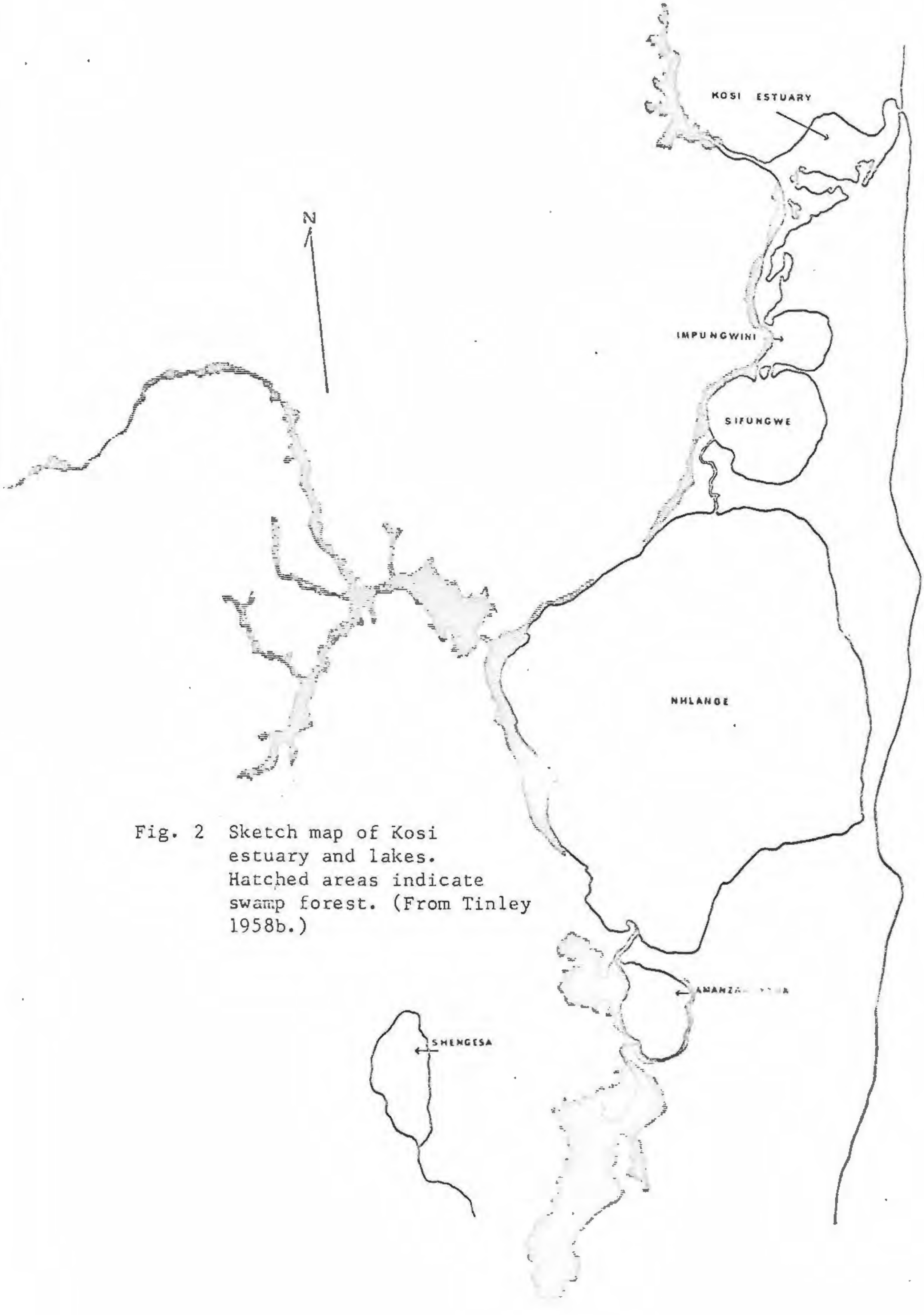


Fig. 2 Sketch map of Kosi estuary and lakes. Hatched areas indicate swamp forest. (From Tinley 1958b.)

In spite of all these shortcomings, the data is interesting in that it raises a number of problems which may be more adequately tackled in the future.

The immediate past history of the lake

Although the Kosi estuary has been known for a long time, there are few written records of conditions in Lake Nhlange. Broekhuysen and Taylor (1959) visited the Kosi estuary in 1949, but spent little time at Lake Nhlange. They described a clear water lake with a secchi disc of 14 ft (4 m). Tinley has written an unpublished report which is in the archives of the Natal Parks Board (Tinley 1958a).

The lake was first visited by us in July 1965. At this time only a few qualitative bottom samples were taken. The lake water was clear and the mouth of the estuary open. The lake was next visited in January 1966. The mouth of the estuary had closed the previous August, and the waters of the estuary and the lake had gradually risen. There was, consequent on a cyclone in January 1966, a great deal of rain that finally raised the level of the lake to some 2 metres above normal before the sand bar at the mouth of the estuary was broken and the water drained away (Breen & Hill 1969). At the time of our visit in January 1966, the water was stained almost a coffee brown, and the inundation of the margins of the lake had caused a considerable decay of vegetable material. It is also likely that much decayed plant material had been brought into the lake by the flooded rivers which drain through extensive swamps. (Fig. 2).

During the subsequent period in which the lake had been revisited, the water has gradually cleared and in June 1968 the water was hardly tinted brown at all. In January 1969 the brownish tint was only seen after heavy wind. This indicates that conditions in the lake improved over the period. The improvement has been reflected in the faunal changes which have been observed in the benthos of the lake. Measurements on oxygen conditions in the lake support the view that an unusual amount of organic detritus was deposited in the lake. However, the conditions in the benthos of the lake were not known previous to the flood, and the data must be viewed with this in mind.

A survey of the substrates, and benthic fauna, has been carried out on Lake Nhlange over the period July 1965 to January 1969. Measurements of oxygen conditions in water very near the substrate has been attempted in January 1969. Some attempt has been made to understand the pattern of faunal distribution in the lake with respect to substrate type and chemistry near the substrate/water interface.

A comparison of the benthic fauna of Lake Nhlange and Sibayi has been attempted.

Methods

Grab sampling was done using the modified van Veen grab sampler employed in Lake Sibayi (Boltt 1969). It must be pointed out that only samples carrying an adequate load of substrate were analysed; inadequate samples were rejected and retaken. Substrate was analysed in the same way as that reported for Lake Sibayi. Sample stations are plotted in Fig. 4.

A special apparatus was constructed to sample water very near the substrate surface interface. The details of construction and a theoretical analysis of the mode of action of the apparatus is given in Appendix I.

A limited SCUBA diving programme was undertaken, but only in relatively shallow water up to just less than 10 metres.

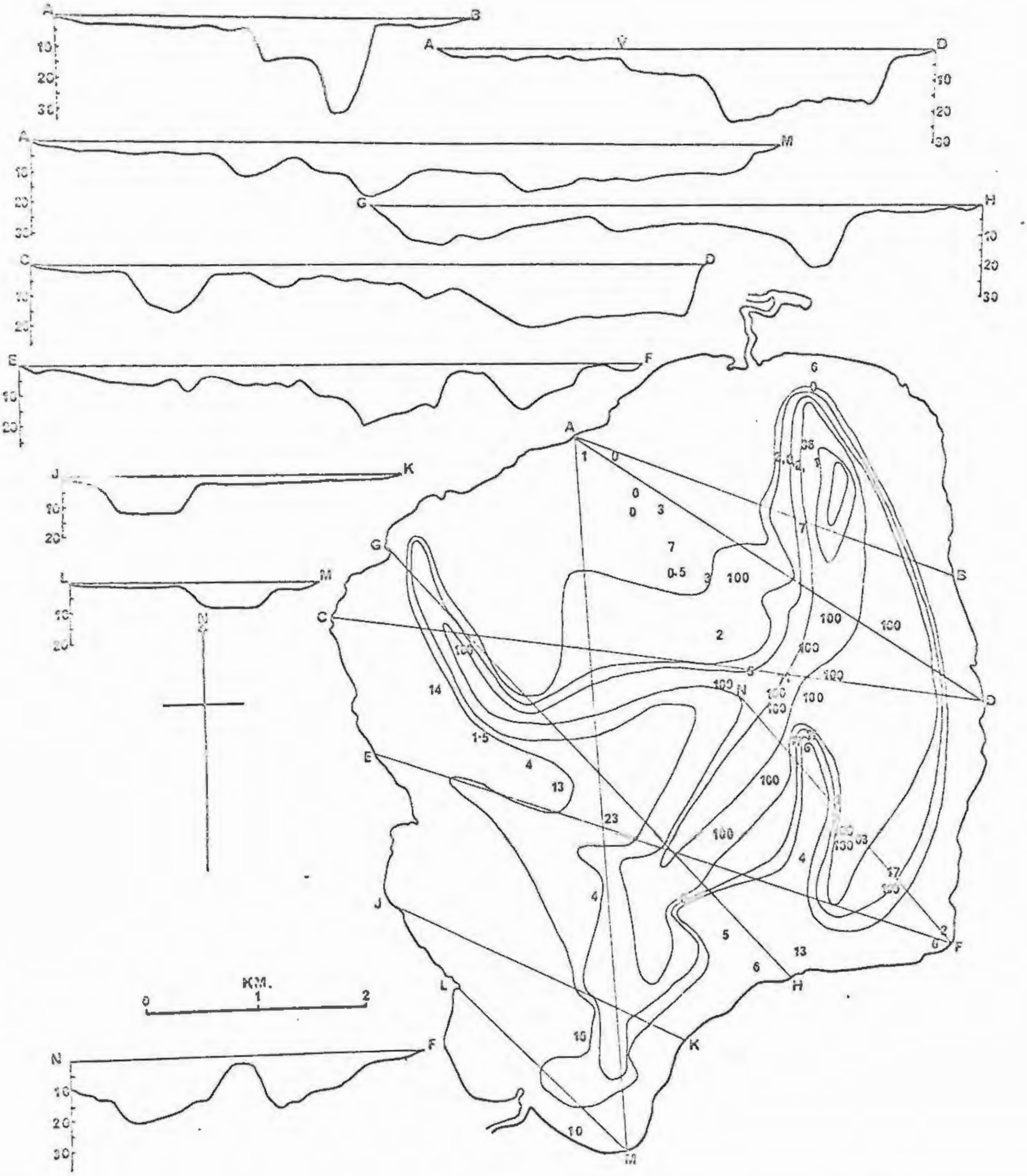


Fig. 3. Map of Lake Nhlange showing percentages of fine material in the substrate of less than 0.04 mm particle size. Profiles from the data used by Hill(1969) are shown together with the transect lines on the lake. The contours are marked at 5 metre intervals.

A list of the numbers of samples taken at different sampling dates is given in Table 1.

Table 1.

Numbers of samples taken at various sampling dates from Lake Nhlange. Station number plotted in Figure 4.

Date	No. of grabs taken	Station numbers
July 1965	0 (5 anchor dredges obtained)	
January 1966	5	NHL 1-5
January 1967	21	NHL 2-27
July 1967	16	NHL 28-44
January 1968	33	NHL 45-77
July 1968	30	NHL 78-108
January 1969	50	NHL 109-159

The distribution of samples with respect to depth has tended to be uneven. The bathymetric chart of the lake shows the greatest areas of the lake to lie above the 10 metre depth contour (Fig. 3). The system of taking samples at regular intervals along transect lines of the lake has therefore tended to favour taking more samples in shallow water than in deep water, especially at early dates in the sampling programme. In 1968 and 1969, more samples were taken below the 15 metre contour by choosing some transects to lie over deep water.

Because the profile of the substratum tended to be made up of flat plains divided by steep slopes, samples taken at depths usually occupied by steep profiles are naturally less frequent (Fig. 3).

RESULTS OF THE SUBSTRATE SURVEY

i) Bottom topography

The bottom topography of Lake Nhlange has been described by Hill (1969). The lake has two underwater valleys which join

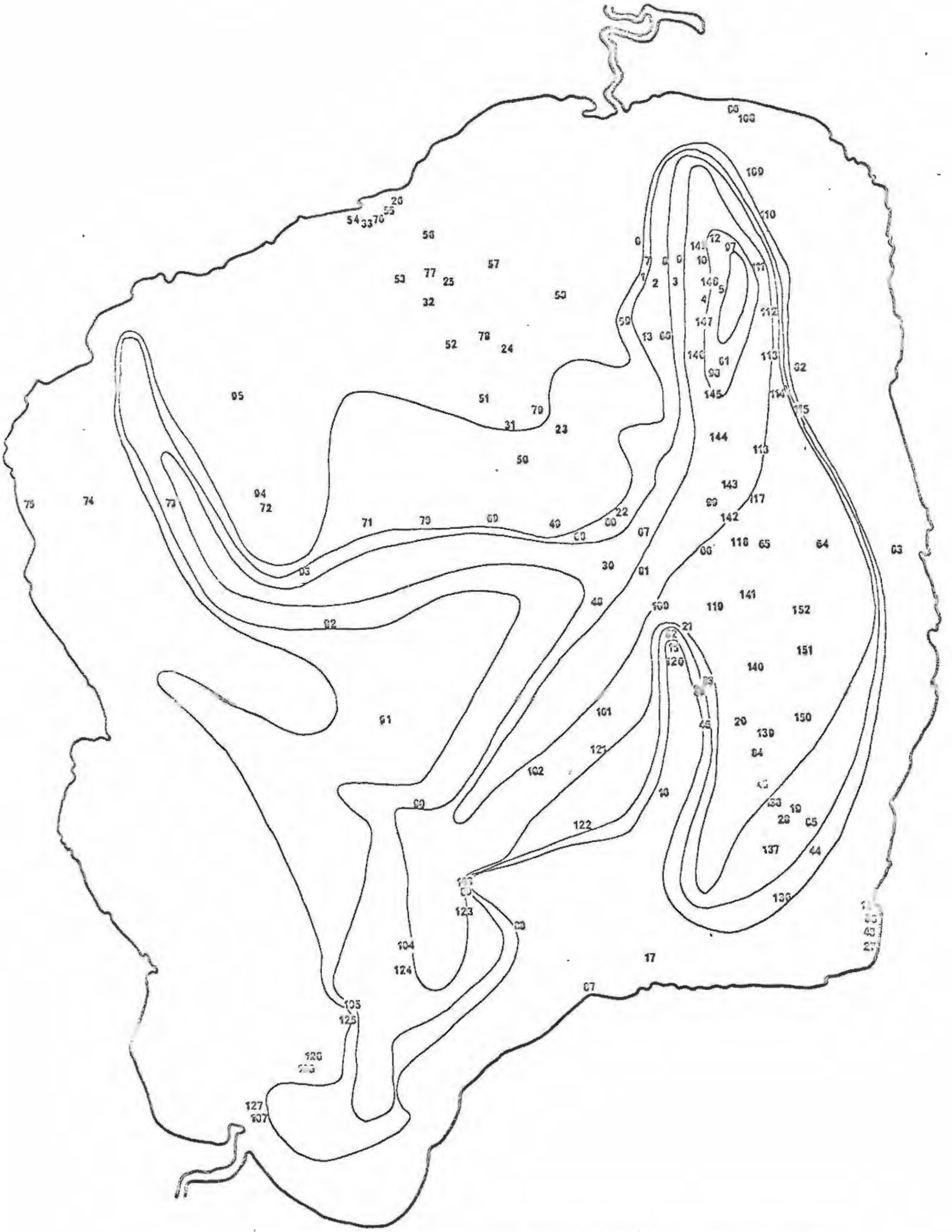


Fig.4 Contour map from data of Hill(1969) of Lake Nhlange. The contours are in 5 meter increments. Numbers on the map refer to NHL. Station No. and show the aproximate positions of the sampling stations.

Table 2

Table of sandy substrate analysis from Lake Nhlange. Analysis according to Morgan's method (1956)
% subsieve consisting of black detritus.

Station No.	Depth	Mdϕ	Qdϕ	Skqϕ	% Subsieve 0.04 mm
NHL 22	15.3	3.15	0.36	+0.09	5%
NHL 24	3.2	3.21	0.12	+0.07	0.5%
NHL 7	10.1	3.31	0.21	+0.07	0%
NHL 10	24.4	3.53	0.68	+0.43	1%
NHL 19	8.0	3.46	0.25	-0.02	17%
NHL 33	2.3	3.32	0.29	+0.05	0%
NHL 15	4.5	2.98	0.23	-0.05	6%
NHL 31	3.5	3.00	0.28	-0.03	3%
NHL 42	2.5	3.40	0.36	+0.12	5%
NHL 23	6.4	3.23	0.24	-0.11	1.4%
NHL 40	3.0	3.30	0.21	-0.06	10%
NHL 32	1.2	3.13	0.21	-0.06	3%
NHL 35	17.7	3.34	0.22	-0.42	6%
NHL 26	2.1	3.18	0.29	-0.05	1%
NHL 29	7.5	3.06	0.20	-0.02	6%

before reaching the deep area in the North Eastern corner (Fig. 3 & Fig. 4). Adjoining the main valley in the east is a gently sloping area demarkated by the 15 m contour (see Sections A-D and CD, Fig. 3). The whole of this area and the bottoms of the valleys form a deep water plain which is roughly defined by the 15 m contour. To the west of the main valley, and especially in the North West, a gently undulating plain at intermediate depths exists (the 10 - 15 m contour). The nature of this "15 metre" plain is shown in the sections A-B, A-D and A-M (Fig. 3). A shallow water plain less than 5 m deep is shown by all sections through the lake. SCUBA divers were impressed by the steepness of the slope dividing the intermediately deep water plain from the shallow water plain (arrow on section A-D, Fig. 3). Measurements have indicated a slope of 1:5 between the flat plains, with sharp clearly defined edges at the top and bottom of the slopes. A deep portion of the lake defined by the 25 metre contour in the north east of the lake descends to a depth of just over 30 metres.

ii) The Substrate

In general, two types of material were found to make up the substrate - siliceous sand particles with small quantities of shell fragments from small bivalves, foraminifera etc., and silt (Wentworth scale, Morgans 1956). The sand fraction of the substrate consisted of well sorted fine sand with an average of median phi values 3.21 on the Wentworth scale (Table 2). The silt had a median phi value of less than 6.

Essentially two types of substrate were found; sandy substrates and muddy substrates composed of silt particles.

Analysis of substrates showed that sand, when present, contributed the greatest bulk of the dry weight of material, whilst the subsieve fraction very little. Analysis by volume (Boltt 1969, p. 252) was therefore more meaningful. Sandy substrates were so called when not more than 20% by volume of silt was present, muddy sands containing from 20% to 50% silt, sandy mud from 50% to 95% silt, and mud containing no more than 5% sand.

Using these criteria, the lake bottom showed largely fine sandy substrates or mud substrates. Sandy mud, and muddy sand substrates were characteristic of regions where steep bottom profiles were adjacent to flat or gently sloping bottoms. Percentages of silt fraction are marked on Fig. 3 for stations in the lake. These data show that the areas roughly enclosed by the 15 m contour contain either pure silt and hence are mud substrates or have a very high percentage of silt fraction over sand, and are sandy mud. Slopes leading down to these deep waters have low percentages of silt in them and are therefore sand substrates. The shallow water plains in less than 5 m show the least silt fraction in the sand. SCUBA divers noted currents and wave action in 2.5 m during a dive when a force 3 wind was blowing over the lake. Small specks of material were visibly being transported by the water. It is likely that shallow water substrata are swept clear by these water movements, and the material aggregates in deep water where less movement is to be observed.

The character of the silt of mud substrates is different from silt in sand substrates. The mud is black and porridge-like in consistency. There tends to be aggregation of silt particles into soft conglomerates. A small part by volume of the mud consists of plant fragments and chironomid exuvae.

On sand substrates the silt particle size overlies the sand as a layer of fine material. In shallow water (0-5 m) this layer of fine material was dark brown, and was rich in microscopic life such as algae, protozoa, and very small crustacea, copepods and ostracods. In intermediate depths (5-15 m) the fine material was dark grey to black. Much more plant fragment was found.

Some physical and chemical characteristics of the water in the lake

The sources of enrichment by detritus in the lake are from Lake Amanzimnyama, and the swamps around the north west corner of Lake Nhlange. Lake Amanzimnyama, besides being surrounded itself by low lying densely forested swampy ground, has running into it a fairly wide river which itself also drains extensive swamps (Fig. 2). As the native name suggests, the lake water has always been deeply stained with humic material and has acted as a settling pond for the material entering from the river. The bottom of the channel connecting Lake Amanzimnyama and Nhlange is covered with a deep layer of dark brown plant material. In June 1965, deep brown water from the channel leading into Lake Nhlange was evident only at the mouth of the channel. It would appear that normally, at least some of the organic detritus passes through Lake Amanzimnyama and reaches Lake Nhlange.

The salts of Lake Nhlange come mainly from sea water. The lake has remained connected to the sea by way of a narrow winding channel between the lakes and the estuary. The series of lakes leading to the sea, of which Lake Nhlange is the furthest but one away from estuary mouth, show decreasing salinities. Lake Amanzimnyama is virtually freshwater, Lake Nhlange is brack ($3\text{-}4^{0}/oo$ salinity, Allanson & van Wyk 1969), and the hypolimnion of Lake Sifungwe shows values of nearly $12^{0}/oo$, while the epilimnion is $3\text{-}4^{0}/oo$. The water of Lake Nhlange cannot be considered

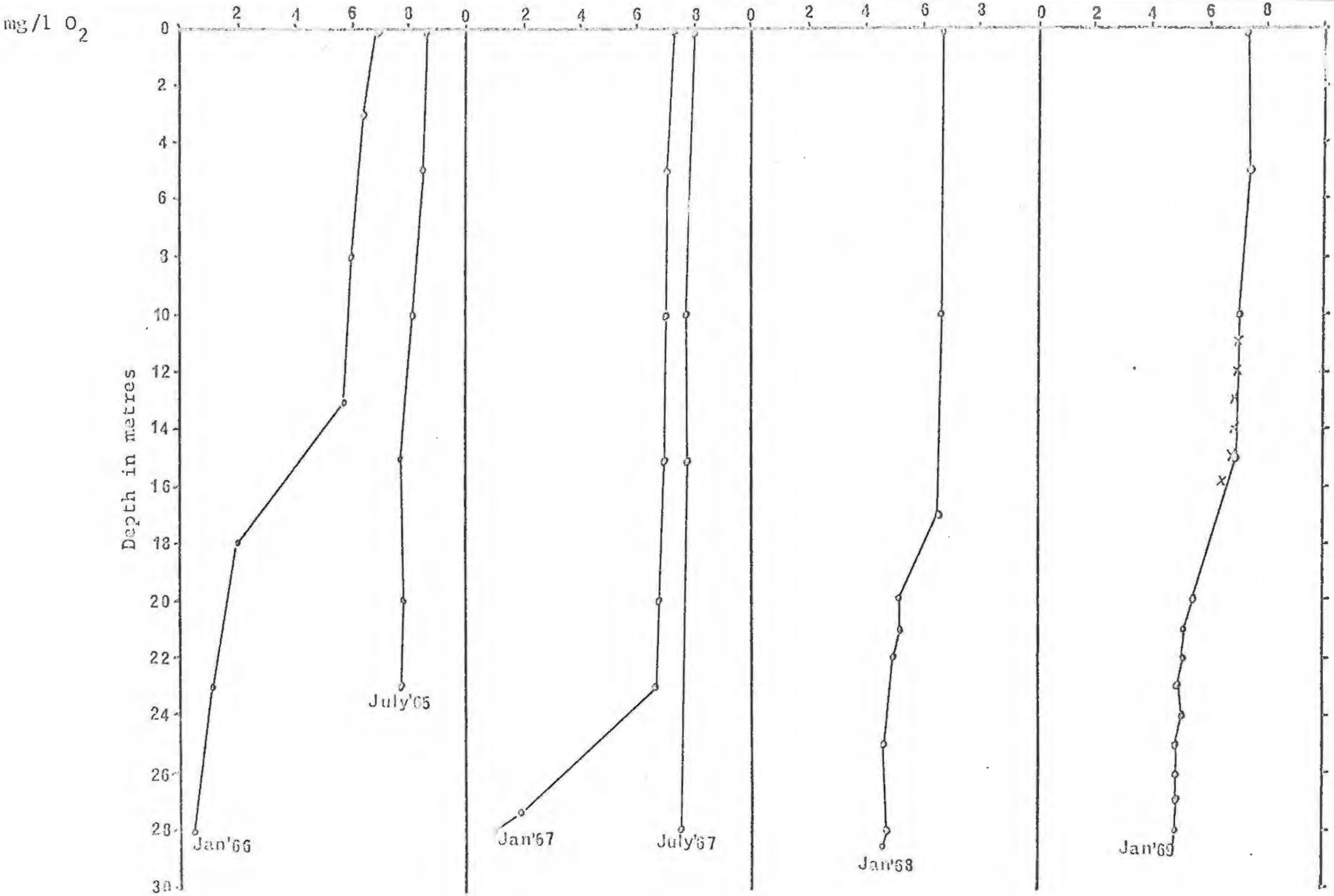


Figure 5. Dissolved oxygen profiles for different seasons on Lake Nhlange.
(x denotes samples taken from a different station over 15 metre depth in 1969).

simply diluted sea water since the ion ratios vary markedly from sea water (Allanson & van Wyk 1969).

Decolorisation of the brown waters from the swamps and Lake Amanzimnyama probably takes place in Lake Nhlange in response to the high calcium and magnesium ion content of the water. A small exchange of water between Nhlange and Sifungwe takes place regularly, especially at spring tides when the water drains backwards through the connecting channel.

The loads of organic material brought in by the inflow into Lake Nhlange seem to form a good part of the subsieve fraction of the bottom, as evidenced by the plant detritus recognizeable in it. This subsieve fraction seems to play an important part in the oxygen requirements of the bottom of the lake. Data on this point are fragmentary and complicated by the possible effects of the floods in the lake in January 1966.

The lake water was well oxygenated down to the deepest waters sampled in July 1965 (Fig. 5). In January 1966, immediately after the flood, oxygen concentrations dropped drastically below 13 m, and it is likely that water very close to the substratum below 18 m was anaerobic. In the following summer, January 1967, serious oxygen depletion was only encountered below 23 m. 2 ppm O_2 was still present 45 cm above the surface of the sediments, and immediately above the surface 1 ppm O_2 was present. By July, no significant depletion of oxygen in the deepest waters sampled was shown. However, in January 1968 there were signs of depletion of oxygen just above the bottom. Unfortunately the deepest sample taken was about 0.5 m from the bottom. Water at the substrate surface may well have contained less oxygen.

In January 1969 oxygen analyses were done on two stations on the lake with Friedinger bottles, one in 28.7 metres and one in 15.8 metres on the 15 metre plain. The results of these measurements are also plotted in Fig. 5. Small reductions of oxygen are present below 20 metres in the deep station.

Table 3

Concentrations of oxygen near the bottom taken in January 1969 with a bottom water sampler, from Lake Nhlange. For details of the apparatus see Appendix 1.

Depth	Nominal distance of sample from bottom	O_2ppm	Nature of bottom
29.4m	9.0cm	3.1	Black muddy
	5.0cm	3.3	sand
	0.5cm	2.9	
28.0	9.0cm	3.8	Black muddy
	5.0cm	2.8	sand
	0.5cm	3.4	
9.0m	9.0cm	5.9	
	5.0cm	5.6	Grey sand
	0.5cm	5.4	
14.0m	9.0cm	2.6	
	5.0cm	2.7	Mud
	0.5cm	0.3	

In January 1969 the bottom water sample was fitted with plastic disposable syringes in which to transport the water from the site of sampling to the surface. Tests with the apparatus in Lake Sibayi showed that oxygen could and would diffuse slowly through the plastic, and contaminate the sample. The trials showed that this contamination in oxygen depleted samples carried on at the rate of approximately 1 ppm per hour. Further, after liberation of iodine for Winkler determinations, iodine was also slowly lost through the apparatus. In order to obviate as far as possible these effects, and at least gain some idea of the conditions on the bottom, the samples were treated with Winkler reagents as quickly as possible after reaching the surface. The precipitate was only allowed to settle until there was sufficient space of clear fluid above it to introduce the acid safely. The samples were transported back to camp with the iodine already liberated from the precipitation. The period between sampling and determination by titration did not exceed 2 hours, the minimum time required to take samples and return to camp. The determinations therefore cannot be trusted for great accuracy.

Thus, in deep water over sandy bottoms, something of the order of 2 to 3 ppm O_2 was present even very near the bottom (Table 3). In shallow water of 9 m depth, approximately 6 ppm O_2 was available. At 14 m depth over a mud bottom, barely detectable amounts of O_2 appear to be present at 0.5 cm depth above the bottom. At 5 cm and 9 cm much more oxygen was available although still in very low concentration when compared with the water taken with Friedinger water sampling bottle. Over sandy substrates at 28 m and 29m, about 3 ppm of oxygen was present.

Other conditions were sampled at the same time. It was known from previous experience that H_2S was present in grab samples, especially from muddy stations. Some indication of the exact locality of the H_2S was furnished with the bottom water sampler. Unless placed very gently on the bottom, and allowed to settle for at least 10 minutes, disturbed material from the bottom was likely to be drawn into the syringe at 0.5 cm from the bottom. A test for H_2S, using amine sulphuric acid reagent which develops methylene blue in the presence of sulphide (Standard Methods, 12th ed. 1965), was positive for disturbed water samples from mud bottoms, but negative in samples taken after a period was allowed for the bottom to settle.

Thus, free H_2S was only present as far as could be ascertained in water below the surface of the mud, and not in water immediately above the mud. Small traces of oxygen would of course oxidise free H_2S in water immediately above the bottom.

From the data available, the following tentative picture of the events in the water near the bottom emerges. From all samplings in winter months (July 1965, July 1967) there was little diminution of O_2 throughout the depths of the lake. Homothermal conditions obtained in the lake in winter (Allanson & van Wyk, 1969), and one would not expect diminution of oxygen in the lakes even near the bottom.

Samplings for O_2 and temperature measurements in the lake in summer (January 1966, 1967, 1968, 1969) (see Allanson & van Wyk 1969 and also Fig. 5), show lowered oxygen tensions and temperature changes in deep water. Temperature discontinuities form and change over a period of hours, indicating considerable movement of water in the lake, and speak against any sort of stable thermocline with consequent complete impoundment of water in a hypolimnion. However, the discontinuities allow for temporary ping of water below certain depths and these tend to have reduced oxygen concentration. The severity of O_2 depletion has varied in different years, the most striking being obtained just after the flood in January 1966. In January 1967, severe O_2 lack was experienced, especially below 25 m. In the summers of 1968 and 1969, deficits of oxygen below 25 m are clear. The detailed investigations of water near the bottom in January 1969,

Table 4

List of animals taken from the benthos of Lake Nhlange. The list is not exhaustive since very small forms such as benthic ostracods and copepods were not considered.

Coelenterata	
Hydroidea	*Ostroumovia inkermanika* (Paltschikowa-Ostromouval 1925) (Black sea and India, brack water).
Oligochaeta	Recorded as such.
Polychaeta	*Ceratonereis keiskama* Day.
Mollusca	*Lamya capensis*. Mostly juveniles about the size of sand grains. Larger animals are found on weed.
Nematoda	Recorded as such.
Crustacea	
Amphipoda	*Grandidierella bonnieri* Stebb.
	Melita zeylanica Stebb. from weed.
	Corophium triaenonyx Stebb.
Tanaidacea	*Tanais philitraeus* Stebb.
Isopoda	*Paramunna laevifrons* Stebb.
	Cirolana fluviatalis Stebb.
Cumacea	Cumacean, rare.
Decapoda	*Hymenosoma orbiculare* Desm.
	Rhynchoplax bovis Brnrd.
Insecta	Chironomid larvae. These have only been identified partially from samples from July 1968 and January 1969. Otherwise recorded as such.
	Chironomus sp.
	Clinotanypus sp.
	Tanytarsini. *Tanytarsus* sp?

for all the shortcomings of the apparatus, indicates real deficits of oxygen less than 10 cm from a mud bottom, but not near a sandy substrate. Small deficits of oxygen in the main water column were not indicative of the nearly anaerobic conditions on the mud substrates. Therefore, anaerobic or nearly anaerobic conditions at least on the mud substrata probably occurred in the previous summer of 1968.

It would appear, therefore, that consequent upon the inflow of organic material from the streams and swamps, as evidenced by the deep brown stain in the water, the substrate in January 1966 below about 17 m was anaerobic. In successive summers, reductions of oxygen in the water at deep depths still occurred, but with less severity. Over sandy substrata, oxygen concentrations were in all probability not limiting to benthic faunal life, but severe reductions of oxygen in water less than 10 cm from the bottom was to be expected over mud bottoms in Lake Nhlange. It may well be that better mixing in the homothermal conditions obtaining in winter, together with lower temperatures and less bottom oxygen demand consequent on it, allowed oxygen to be present even in highly enriched mud bottoms. In summer, high temperatures with high oxygen demands from mud bottoms, coupled with poor circulation, may result in anaerobic conditions in water lying immediately over mud bottoms.

Results of the Faunal Survey

A check list of the fauna from the benthos of Nhlange is given in Table 4. The majority of forms are estuarine (Day 1951, 1967) and a few are more typically brackwater forms. These latter are the chironomid larvae and the oligochaetes.

Table 5

Frequency of grab samples containing high and low total numbers of animals from water shallower than, and deeper than, 15 m.

Date	Depth range m	Range of no. of animals per grab sample 0 - 99	100 - 10 000
ary 1966	0 - 14.9	4	1
	15 - 30	3	0
January 1967	0 - 14.9	5	10
	15 - 30	6	0
July 1967	0 - 14.9	5	8
	15 - 30	2	1
January 1968	0 - 14.9	2	23
	15 - 30	7	1
July 1968	0 - 14.9	0	21
	15 - 30	7	3
January 1969	0 - 14.9	3	21
	15 - 30	17	9
Total	0 - 14.9	14	83
	15 - 30	42	14

Table 6

Frequency of numbers of grabs with different ranges of animal densities from water deeper than 15 metres in Lake Nhlange. Sand and muddy sand implies substrates with less than 60% subsieve fraction. Mud and sandy mud are substrates with not more than 30% subsieve fraction by volume.

	Range of animal number per grab 0	1 - 10	11 - 100	101 - 1,000	1,000 +	Total no. of grabs.
Sandy or muddy sand	3	1	4	12	2	22
Mud or sandy mud	27	4	3	0	0	34
All samples	31	5	7	12	2	56

Members of both of these groups are known for their tolerance of high total dissolved solids content in water.

Inspection of the bottom sediments by SCUBA divers has shown that on sandy substrates there is a lack of both large epifauna and of good growths of weed in depths at which it might be expected. Diving has not been extensively carried out on Lake Nhlange, but from a series of dives in 2.5, 3.5, 8.3 and 9.5m in the north west of the lake, and 2-3m in the west, weed was only found in 2.5 to 3m. Potamogeton pectinatus was only found in a stunted growth form. Potamogeton weed beds occur in very shallow water of less than 1m around much of the edge of the lake. The only animal evident on the sand was the burrowing goby Croilia mossambica Smith, which was taken in up to 9.5m. It may well occur in deeper water. The shallow depressions in the sand were taken to be the result of feeding activities of the fish in the lake.

Distribution of the infauna with depth

At all sampling periods from January 1966, the richest samples with respect to total individuals per grab were taken from above the 15 m contour in the lake (Table 5, Figure 7).

Of 56 samples taken below the 15 m contour, 30 were barren, 5 contained 10 or less individuals and 21 had more than 10 animals (Table 6). Of these 21 samples, 14 had greater than 100 animals, and all 14 were from sandy substrates (Table 6). Of the 22 essentially sandy substrates, only 4 were barren. By contrast of the 34 muddy or sandy mud samples, 31 had less than 10 animals, and of these 31, 7 samples were barren. None contained 100 or more animals (Table 6).

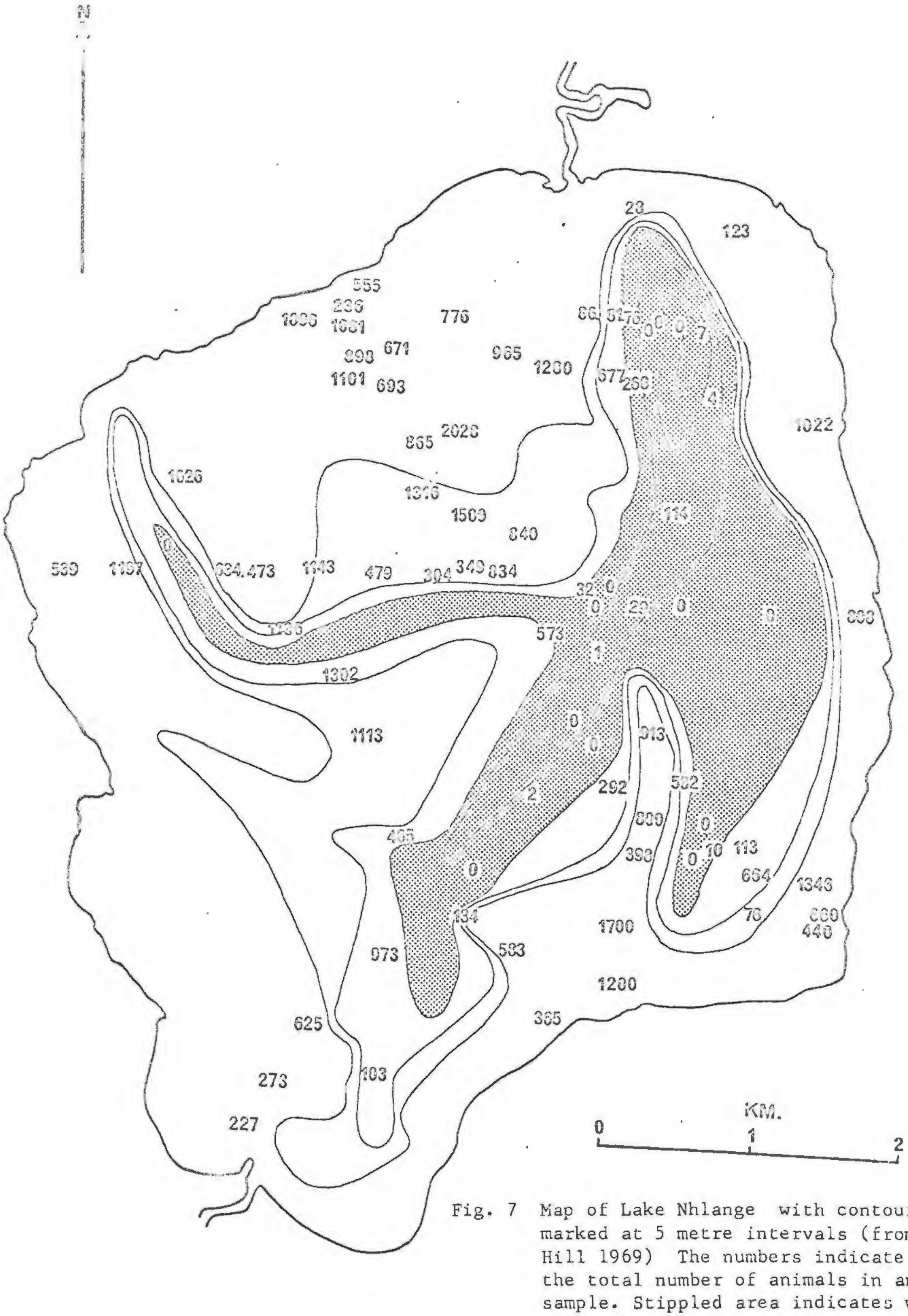


Fig. 7 Map of Lake Nhlange with contours marked at 5 metre intervals (from Hill 1969) The numbers indicate the total number of animals in any sample. Stippled area indicates water below the 15.m contour

There is, therefore, a general pattern of sandy samples with many animals and muddy samples with few or no animals in them. There are important exceptions. Sandy and muddy sand samples devoid of animals have been taken only from below 20 metres depth. The only areas where sand with very low subsieve contents has been sampled at these depths is at the north end of the central valley, around deep water of up to 30 metres (Fig. 3). Sand is to be expected in this region because of the steep profiles leading down to the 30 metre contour (Fig. 3, profile A-B). The development of anaerobic conditions in summer in the bottom of the lake accounts for these samples in a general sense, but a greater understanding of these conditions can be gained after a more detailed analysis of the species and number composition of the samples. The muddy samples containing animals similarly is better dealt with after detailed considerations.

A series of kite diagrams showing the pattern of distribution of the common animals in the lake is given in Figures 8 - 15. The kite diagrams must be interpreted with care because of the way in which they are drawn up. The maximum densities for any particular depth have been shown on the diagrams. The data are therefore pertinent only to the best conditions that were sampled in the lake at that particular depth. For example, muddy samples from about 15 metres would of course have had few or no animals, while sandy samples would have many. These must be considered separately.

One clear result shown by the kite diagrams is a general tendency for the fauna to be found at increasing depths in the years following the flood of December 1965. Only five samples were taken in January 1966, and the two samples from 25 metres and 28 metres were void of animals (Table 7).

Table 7

Results of the five grab samples taken from Lake Nhlange in January 1968.

Station No.	Depth		
NHL 1	5 metres	G. bonnieri	41
		C. triaenonyx	410
NHL 2	12 metres	G. bonnieri	2
		C. triaenonyx	0
		Oligochaetes	95
		Chironomids	2
NHL 3	15 metres	G. bonnieri	16
		C. triaenonyx	0
		Oligochaetes	7
		Chironomids	1
NHL 4	25 metres	Nil	
NHL 5	28 metres	Nil	

A year later, in January, 1967, samples with animals were taken only down to just over 15 metres. Samples from July 1967, January 1968 and July 1968, all showed greater penetration of the fauna with respect to depth. It is true that the earlier sampling programmes were small (Table 1). However, even if we only treat with samples from January 1968 to January 1969, the conclusion that the fauna invaded deeper water seems to be inescapable. This point is also clearly brought out when the groups are inspected separately. Thus, all groups except the chironomid larvae, *Chironomus* sp. were found at depths deeper than was the case in July 1968. In January 1969 some groups had retreated to slightly more shallow water, but the polychaete *Ceratonereis keiskama* and bivalve *Lamya capensis* were found in even deeper water than the previous July.

An examination of the results with respect to species or

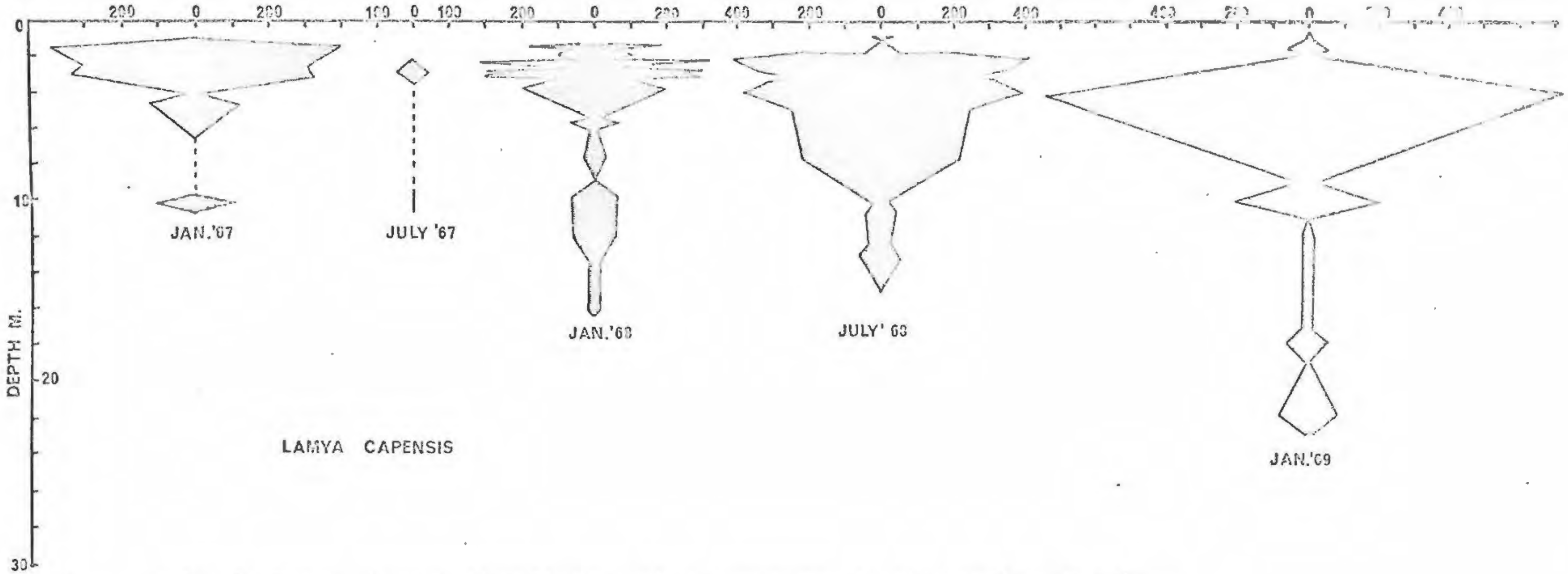


Fig. 8 Kites showing distribution of L. capensis in the benthos of Lake Nhlange with respect to depth. The number of animals per 0.0225 m^2 grab sample are indicated. It is suspected that the poor returns of animals in July 1967 may be an artifact of sorting. For explanation see text.

group follows.

The bivalve *Lamya capensis* is numerically the most important faunal element in the lake. *L. capensis* is usually associated with some hard surface on to which it attaches itself by byssus threads. These may be plants or rocks. It is not normally associated with relatively soft sandy bottoms; for example Day (1969) notes that it is attached to weeds in estuaries. Weed growth was poor in Lake Nhlange. Only in one sample, NHL 96 was weed of good size taken (0.5 m length), and on this weed a dense cover of well grown *L. capensis* was counted. In most samples *L. capensis* was found loose on the sand and in all cases only very small animals of about 2mm were present. (The bivalve normally grows to 12mm in estuaries (Day 1969).) They were, however, present in large numbers. Inspection of Fig. 8 shows that these animals were found in slightly deeper water in 1968 than in 1967 samplings. A dramatic increase in the depth in which they were found occurred in the samples taken in January 1969. The animals may be intolerant of high subsieve content in sand samples since they do not occur in samples with high subsieve volumes, even in very shallow water. This may be due to the method of mucous sheet filter feeding which may clog with silt.

The distribution of chironomid larvae with depth for each successive sampling is shown in Fig. 9. The larvae were restricted to water less deep than 15m in January 1967. In the following July chironomid larvae other than *Chironomus* sp. larvae were restricted to 17m. *Chironomus* sp larvae were found in 27m. Much the same pattern of distribution was found in January 1968. Once again in deep water *Chironomus* sp. was found in 25m. It was only in July 1968 that chironomid larvae of all types were found

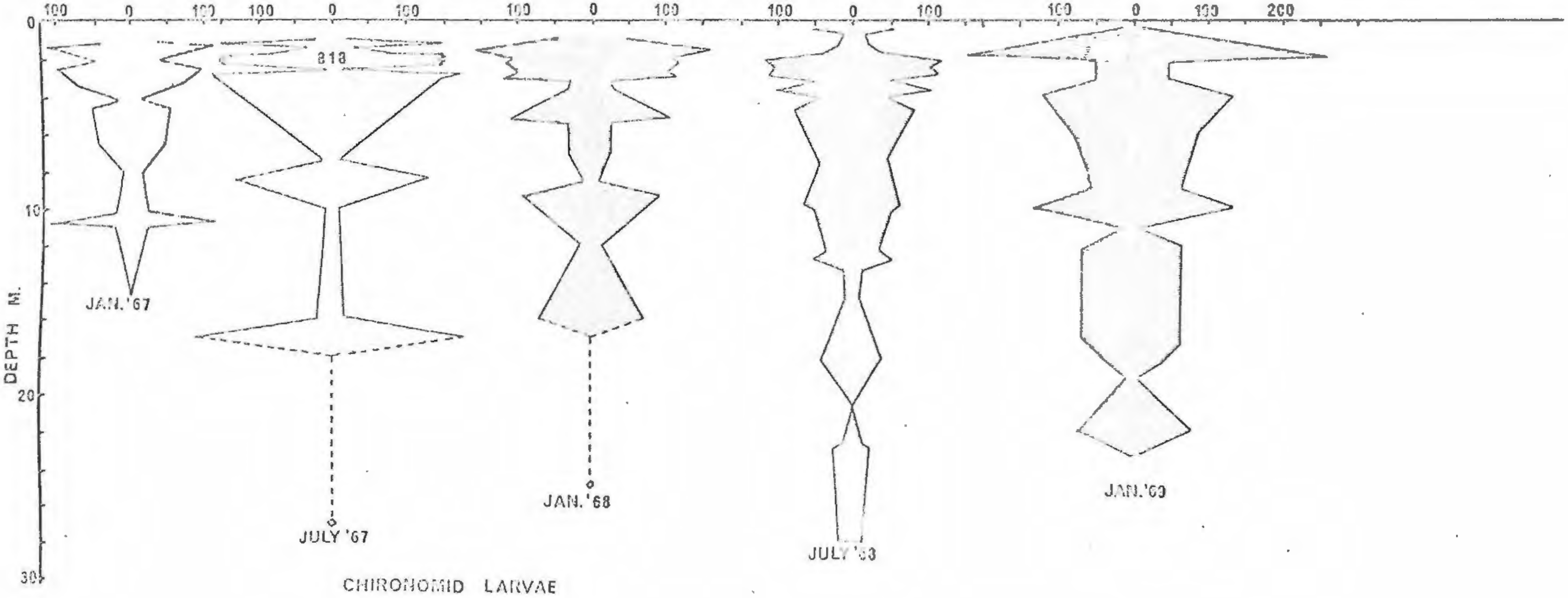


Fig 9. Kite diagrams of the distribution of all chironomid larvae with respect to depth in Lake Nhlange. The small open kites at 27m and 26 metres from July'67, and Jan '68 represent Chironomus sp. Note that the chironomids were not found as deep in Jan.'69 as they were in July 68. For further explanation see text.

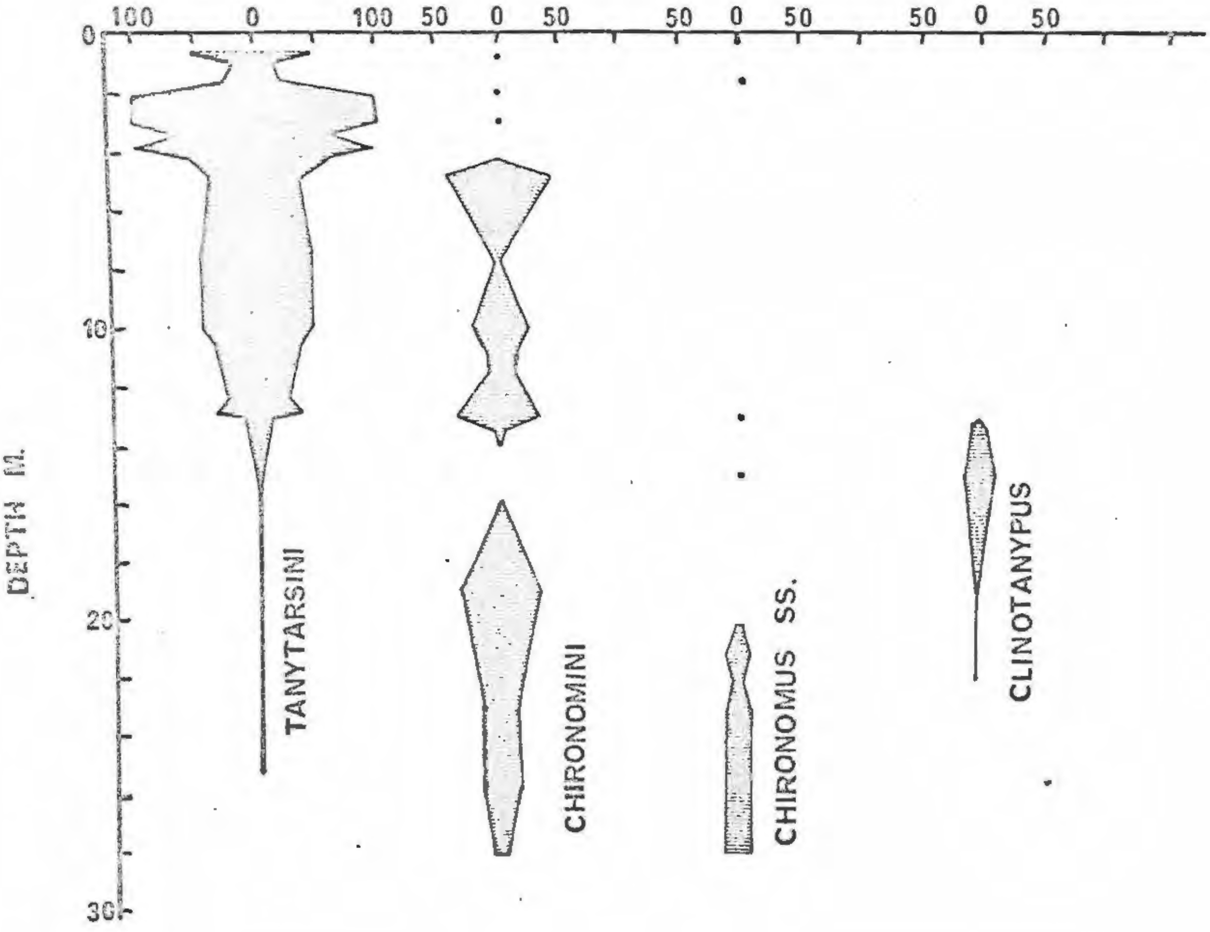


Fig. 10 Kite diagram of Chironomous larvae taken from the benthos of lake Nhlange in July 1968. There was a clear zonation of different types of larvae especially with regard to Chironomus sp. This figure should be compared with fig. 11. The number of animals is indicated on the top of each kite.

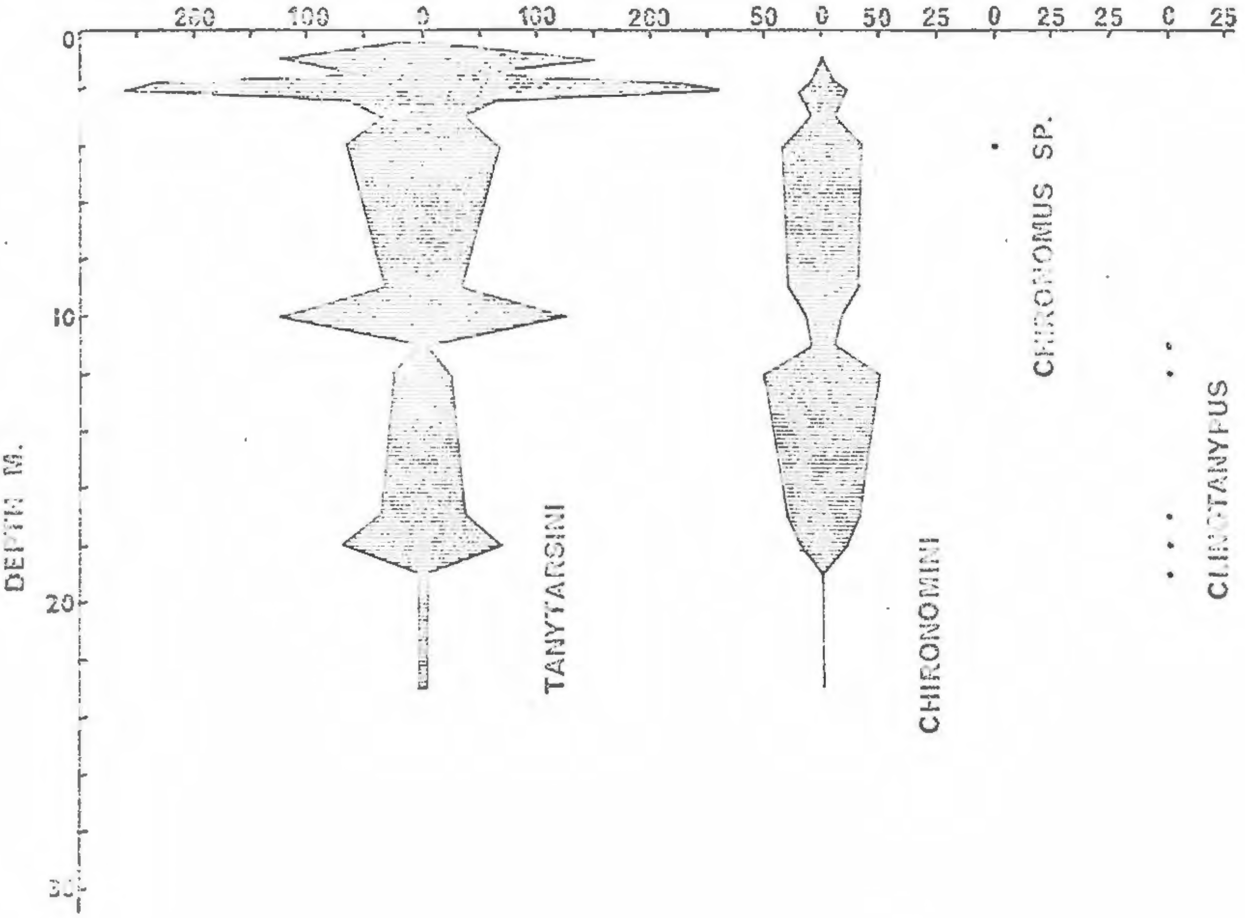


Fig. 11. Kite diagram of chironomid larvae taken from the benthos of Lake Nhlange in Jan. '69. No clear zonation was found. Chironomus sp. and Clinotanypus sp. were rare in the samples. The number of animals per grab sample is indicated above each kite.

to penetrate to 28m.

In January 1969 in spite of extensive sampling, chironomid larvae were only found down to a maximum depth of 23 metres (Fig. 9).

Prior to July 1968, all chironomid larvae except larvae of the genus *Chironomus* sp. were counted together. Only in July 1968 and January 1969 were the larvae classified as far as possible into the following groups: *Tanytarsus*, Chironomini, *Chironomus* sp. and *Clinotanypus* (the latter is a carnivorous type). The distribution of these different types of chironomid larvae in the sediments in July 1968 is illustrated in Fig. 10. *Tanytarsus* larvae were in high numbers in water of up to 15m depth, Chironomini larvae in waters from 4m to 28m depth, and *Chironomus* sp. in waters deeper than 20m depth. *Clinotanypus* which is carnivorous was only found in waters 12 to 22m depth. A clear zonation of chironomid larvae with regard to depth was shown. *Chironomus* sp. is especially important because of the known tolerance of these animals for highly enriched bottom sediments. *Chironomus* sp. was first taken in July 1967 in 27m. On that occasion and subsequently the animals were recovered from samples only [illegible] silt substrates or sand overlaid with considerable silt.

In January 1969 the chironomid pattern of distribution was greatly modified. The pattern is presented in Fig. 11. In contrast to the situation in July 1968, there were very few chironomids below 20 metres. *Chironomus* sp. was only discovered in one grab sample, and that at 4 metres.

The oligochaetes have not been identified further. Their pattern of distribution in the lake is well shown in Fig. 12. As with the chironomid larvae, the animals appeared to penetrate into deeper water with respect to later sampling dates. Unlike

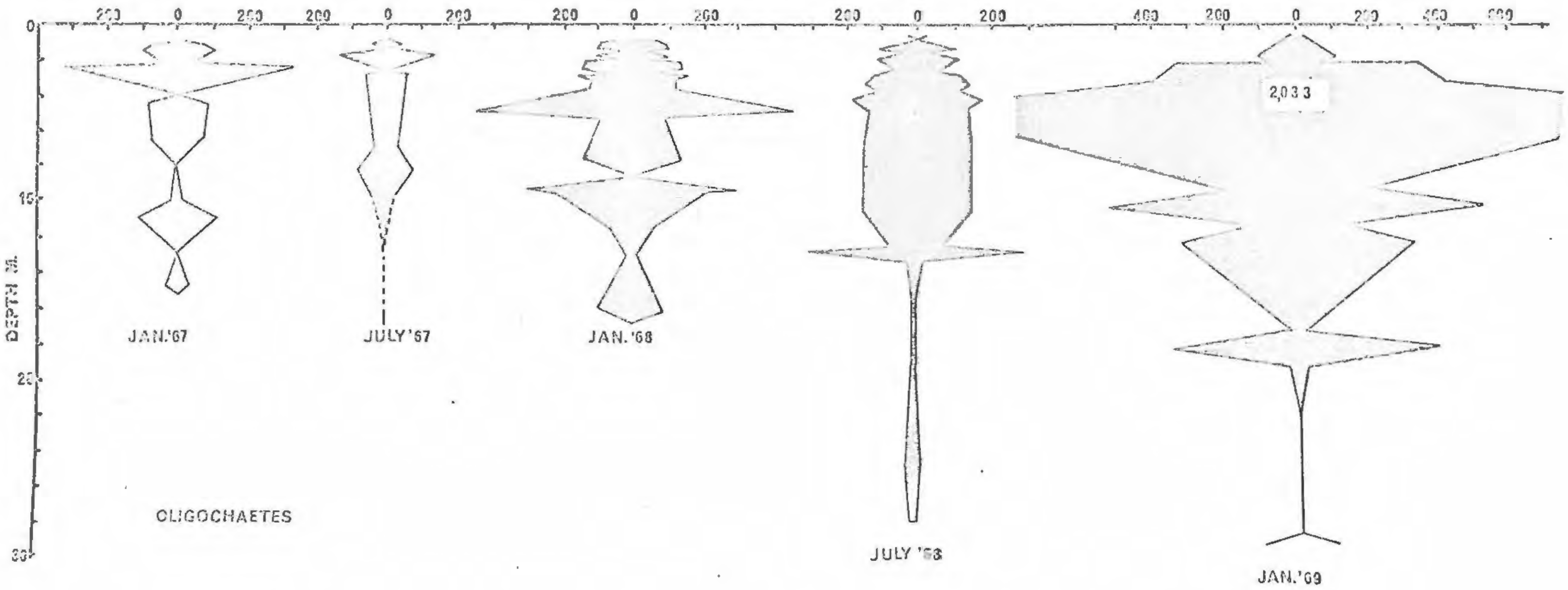


Fig. 12 Kite diagram of oligochaetes taken from grab samples from Lake Nhlange.
The different species were not identified and it is probable that a zonation of different species may have occured with depth.
Numbers of animals per grab sample is indeicated above each kite.
The number in the kite for Jan.'69 indicates a particularly rich sample that has not been plotted.

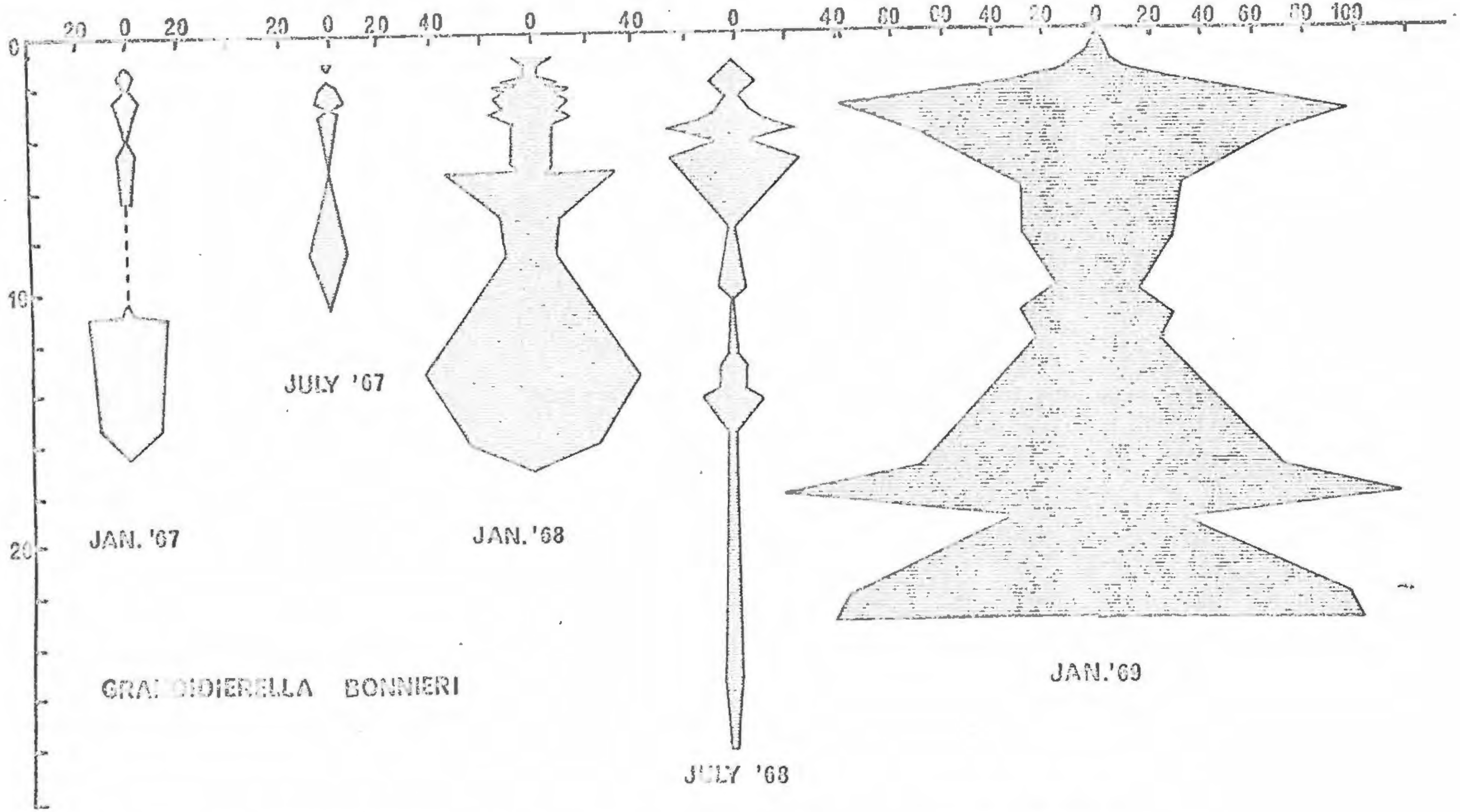


Fig. 13 Kite diagrams for G. lignorum from the benthos of Lake Nhlange. The number of animals per grab is indicated above each kite. For explanation see text.

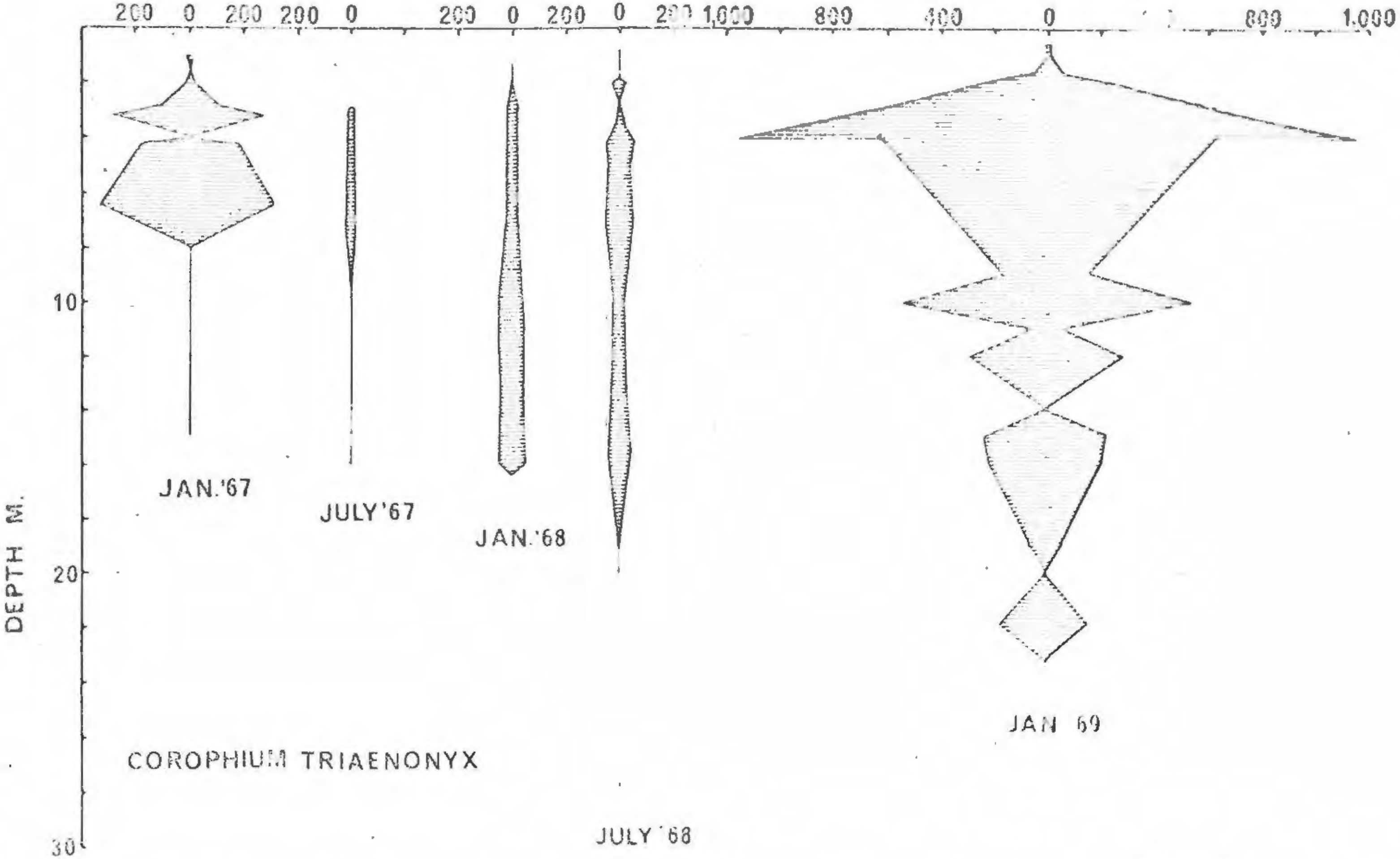


Fig. 14 Kite diagrams of the distribution of *C. triaenonyx* in the benthos of Lake Nhlange. The number of animals per grab is indicated above each diagram. For explanation see text.

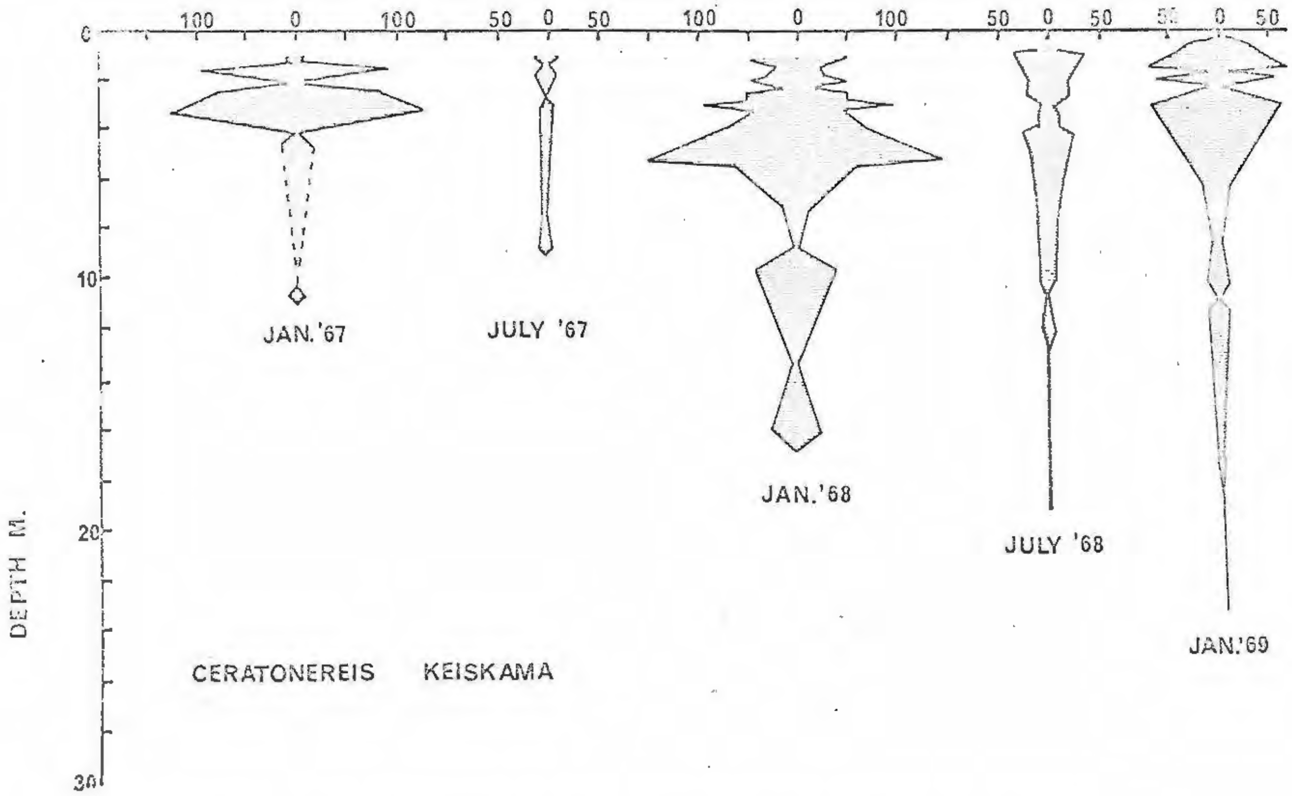


Fig. 15. Kite diagrams of the distribution of C. keiskama with respect to depth from the benthos of Lake Nhlange. The number of animals per grab is indicated above each diagram. For explanation see text.

the chironomids in January 1969, one sample in 29 metres of water yielded a high count of oligochaetes but no other groups of animals were found with them.

A general increase in the size of the populations in the grab samples was shown in the January 1969 sampling, a feature held in common with the amphipods *Corophium traenonyx*, *Grandidierella bonnieri*, and to a lesser degree with the bivalve *Lamya capensis*.

The pattern of numbers with respect to depth for the amphipods *G. bonnieri* and *C. triaenonyx* is given in Figs. 13 and 14. In both cases the population appeared to reach deeper depths with regard to time up until July 1968. In January 1969 they only penetrated to about 23 metres and 25 metres respectively. A curious fluctuation in numbers of animals per grab has been recorded for *C. triaenonyx*. In January 1967 five samples taken between 2 metres and 7 metres had populations between 20 and 627 animals per grab. Only one had 20, the rest had more than 211. In July 1967, and January and July 1968, only one sample had as many as 119 animals. In January 1969, of 13 samples one had 10 amphipods, one had 59, and the remainder ranged between 140 and 2,354 animals per grab. Six samples had greater than a thousand. The reasons for this dramatic increase in the populations of the amphipod in the lake are not known.

The polychaete *Ceratonereis keiskama* showed steady increases in the depth to which it has penetrated (Fig. 15). Together with the bivalve *Lamya capensis*, and the oligochaetes, the groups did not show the decreased penetration characteristic of the other forms reported on, in January 1969. It is interesting to note that in Lake Nhlange, *C. keiskama* reached a depth of 23 metres deeper than was found for samples from Lake Sibayi (Boltt

1969, p. 260). Further sampling would be interesting since it is felt that this polychaete was depth limited in Lake Sibayi at about 20 metres.

Having dealt with the results of the grab samples from Lake Nhlange purely from the point of view of depth, some attention must be paid to the fauna of different areas of the lake. The kite diagrams do not show, for example, that there is a considerable area of the lake at about 15 metres which was practically void of animals in the majority of samples. Thus, the kite diagrams deal mainly with samples showing the presence of animals with respect to depth, and ignore samples lacking animals.

Broadly, areas of the substrate of the lake may be categorised as follows:

a) Gently undulating plains of sandy substrates in water up to 15 metres deep. Grab samples from this area were invariably rich in faunal types and population densities. The kite diagrams reflect this area of the lake well, and since there were no obvious anomalies, the area will not be dealt with further.

b) The "15 metre plain". This area may be roughly defined by the area enclosed by the 15 metre contour, but excluding the area around the deep depression enclosed by the 25 metre contour (Figs. 3 and 7).

Although in general the substrate from the "15 metre plain" consists mainly of pure silt, depending on the presence of steeply sloping profiles, sandy mud and muddy sand have also been obtained. The infauna was generally low in numbers of animals per grab, and very often void. Because of the organic nature of the silt, this area of the lake bottom has a high oxygen demand. The presence or absence of infauna in samples from this area, therefore, will indicate in some degree conditions which obtained at any sampling date. A study of these samples at different dates of sampling has, therefore, been attempted in some detail.

Only four samples were recovered with animals in them before July 1968 from the "15 metre plain" (Table 8).

Table 8.

Details of samples from the "15 metre plain" with animals in them taken previous to July 1968 from Lake Nhlange.

Station No. and date		Depth	Substrate Type	% subsieve	Remarks
NHL 22	Jan. 1967	15.0m	Sand	5%	23 *G. bonnieri*, 9 *C. triaenonyx* Taken from a slope
NHL 30	July 1967	16.9m	Mud	71%	2 *C. triaenonyx* 19 Chironomid larvae
NHL 35	July 1967	17.7m	Sand	6%	378 Chironomid larvae 2 Oligochaetes Taken from slope
NHL 48	Jan. 1968	16.0m	Muddy sand	-	58 *G. bonnieri* 119 *C. triaenonyx* 144 Chironomid larvae 12 *Lamya capensis* 129 Oligochaetes 48 *C. keiskama* Taken from a slope

Three of the samples were taken from a slope at the sides of the "15 metre plain". The one sample taken from a muddy rather than a sandy area, NHL 30 was from July 1967. In all cases pure silt samples were void of animals. NHL 30 is important, since together with one sample from the north east depression containing *Chironomus* sp., we conclude that although, in the main, the conditions of the "15 metre plain" were anaerobic, the balance between supply to and the oxygen demand of the muddy substrates might have been near a point when demand just outweighed supply. This is supported by the fact that the oxygen in the water column over the deepest water varied little from 8 ppm from the surface to 30 metres. This is in contrast to the demand in the previous summer (January 1967), when greater oxygen deficits were demonstrated well above the substrate in the water column.

The position changed dramatically in July 1968, when eight samples were taken from the "15 metre plain". Two muddy sand samples contained animals, and of the remaining six mud samples, three were void and three contained Chironomus sp. larvae. One of the three also contained eight amphipods and chironomini larvae. An analysis of the chironomid fauna is shown in Figs. 10 and 11. Chironomids of the genus Chironomus are principally from the deep waters of the lake. They are noted for their tolerance of low oxygen conditions. Their presence in the pure silt substrates of the "15 metre plain" has been interpreted as evidence that by July 1968 the oxygen demand of the organic material in the mud had waned from previous levels obtaining just after the flood. The oxygen demand was still high enough, however, to create generally poor oxygen conditions in the mud substrata.

An extensive sampling programme in January 1969 on the "15 metre plain" showed that all sampling sites with sandy substrates had a full faunal complement of animals. All the muddy samples were void without exception. This feature has been interpreted as reflecting a raised oxygen demand by the organic matter of the mud, under the influence of raised summer temperatures. Measurements have shown that throughout the free water column above the mud, quantities of oxygen were present, but within a small distance of the mud, drastic reduction of oxygen concentration existed. The fact that Lake Nhlange was not at this time stratified in the classical sense of temperate lakes, is emphasised by the fact that in January 1969 all the sandy samples from around the edge of the "15 metre plain", and also from deeper water than some of the void mud samples, contained a full complement of faunal groups as well as reasonably dense numbers of animals. Over the sandy substrate, slight but not severe oxygen deficits were measured.

It may be concluded that from the point of view of the events in the benthos, there is some evidence that excessive loading of the substrate with allochtonous material consequent on the flood, may have caused abnormally severe deficits in oxygen in the years following the flood. It seems to be reasonably clearly established from the July 1968 and January 1969 sampling programme that the equilibrium between oxygen demand by the organic muds, and the rate of oxygen supply to the muds, balanced out in winter so as to allow limited colonisation of the muds of the "15 metre plain". In summer (January 1969) the supply of oxygen had fallen short of the demand, and rendered the muds sterile.

It would appear, therefore, that there was a cycle of serious oxygen deficits in deep water in the bottom of the lake leading to anaerobiosis in the deepest sediments in summer, alternating with increased oxygen availability to the substrata during the winter. The lowered temperatures of winter (27°C summer, 18.5-19°C winter, Allanson and van Wyk 1969) may have been responsible for reduced metabolic activity in the detritus of the bottom and allowed concentrations of oxygen in the bottom sediments to rise. The apparent deepening of water deficient in oxygen from January 1966 and January 1967 especially, may indicate oxidation of detrital material faster than it is being recruited from the inflowing streams. It is clear that the great quantity of humic water brought in by the floods was an unusual event in the history of the lake, and the evidence suggests that unusual amounts of organic material were added to the lake at that period of time.

A comparison between Lakes Nhlange and Sibayi

a) The character of the substrates.

The gross physical characteristics of the substrates of the lakes are not very different. The basins of both lakes were cut in tertiary sands, and therefore the sand particle size is similar. An average median phi value of 3.20 was found for Lake Sibayi, while for Lake Nhlange it was 3.21. However, the characteristics of the silt in the lakes is very different. Using the term silt in the sense only of particle sizes less than 0.04 mm, in both lakes this particle makes up the mud substrates and also forms a film of fine material overlying the substrate. The origin of this silt comes from two sources in both lakes, from allocthonous material brought in by the inflowing streams, and dust and plankton remains which rain down on the substrate from the open waters. This second component appears not to be very important, since in Lake Sibayi flat regions of sand have not been smothered in thick layers of silt and are only covered in a thin layer of fine material.

However, the organic character of mud from the two lakes appears to be different as judged from colour and consistency. That of Lake Sibayi is oozy grey (Boltt 1969, page 255) and of very uniform consistency. The mud substrates of Lake Nhlange are black and porridge-like. Aggregation into soft conglomerates was common enough to be noted while the mud was being analysed by sieving. Quite characteristic was the small but distinctive

Table 9

A comparison of the presence or absence of common animals of the benthos of Lakes Sibayi and Nhlange. + indicates presence, - absence.

	Lake Nhlange	Lake Sibayi
CRUSTACEA		
Amphipoda		
Grandidierella bonnieri	+	-
Grandidierella lignorum	-	+
Corophium triaenonyx	+	+
Melita zeylanica (Marginal vegetation)	+	-
Tanaidacea		
Apseudes digitalis	-	+
Isopoda		
Cyathura carinata	-	+
Pseudosphearoma barnardi	-	+
Cirolana fluviatilis	+	-
Decapoda		
Hymenosoma orbiculare	+	+
Rhynchoplax bovis	+	-
Cumacea		
Cumacea sp.	+	-
MOLLUSCA		
Lamya capensis	+	-
Bellamya capillatus	-	+
Melanoides tuberculata	-	+
Bulinus natalensis	-	+
ANNELIDA		
Polychaeta		
Ceratonereis keiskama	+	+
Annelid oligochaetes	+	+
COELENTERATA		
Hydroidea		
Ostroumovia inkermanika	+	-
Hydra sp.		
INSECTA		
Chironomid larvae		
Tanypus	-	+
Chironomus sp.	-	+
Chironomini	+	+
Clinotanypus	+	-
Cryptochironomus	-	+
NEMATODA		
Nematodes	+	+

volume of plant fragments and chironomid exuvae.

Over sandy substrates, a film of fine material consisting of silt particle sizes is found in both lakes. In the shallow water of Nhlange (0 to ± 5 m) the fine material resembles that of fine material over the whole of Lake Sibayi sands (Boltt 1969, page 253). Pale brown in colour, the fine material is rich in living organisms, algal cells, protozoa, very small crustacea and nematode worms. In Lake Nhlange in waters of intermediate depth (about 5 - 15 metres) the fine material overlying the sand tended to be dark grey to black, and showed fragments of decaying plant material in it. Although still showing a complement of algal cells and small fauna, it differed from that of Lake Sibayi, or of the shallow water of Lake Nhlange.

b) The animal life of the benthos of the two lakes compared.

Whilst fewer species may have been expected for Lake Sibayi, as compared with the more saline Nhlange, the presence of certain species in the freshwater Lake Sibayi, which are plainly of estuarine origin, and which are lacking in Nhlange, is remarkable. Four species, *G. lignorum*, *A. digitalis*, *C. carinata* and *P. barnardi*, are found in Lake Sibayi, but absent in Lake Nhlange. Perhaps even more surprising was the discovery of two different species of the same genus of amphipod, *G. lignorum* from Lake Sibayi, and *G. bonnieri* from Lake Nhlange. There can be no doubt that these are good, easily distinguishable, species. They occur in the estuaries around the Eastern Cape Province of South Africa, and the males can always be distinguished without difficulty (Barnard 1935 and 1940).

In seeking means for these differences, the following points should be borne in mind.

The salt concentrations of the waters from the two lakes is markedly different. Lake Sibayi has a sodium ion concentration of just over 80 mg/l, while Nhlange has one of just over 1800 mg/l (Allanson and van Wyk 1969). Neither lake shows ion ratios comparable with sea water, and thus cannot be considered simply dilute sea water. Nevertheless, differences in general chemistry of the lakes such as salt concentration, whilst being able to account for the absence of cumaceans, the amphipod *Melita zeylanica* and *Lamya capensis* from Lake Sibayi, cannot account for the estuarine species present in Lake Sibayi but absence from Lake Nhlange. These differences must be attributed to other factors which have not yet been identified.

These ideas are reinforced by noting that while the absence of *G. bonnieri* from Lake Sibayi may be due to differences in sodium ion regulation mechanism as compared with *G. lignorum*, much in the way suggested by Sutcliffe (1968) for various *Gammarus* species from Britain, the absence of *G. lignorum* in Lake Nhlange cannot be explained away by this sort of physiological difference.

It seems unlikely that the particle size of itself is important in determining the species of one lake as compared with the other. As is reported in the laboratory investigations on the behaviour of *G. lignorum*, this amphipod which is so characteristic of sandy substrates from Lake Sibayi, builds its tubes of silt particles if sand is unavailable or even lacking in food particles. Whether this is true of the other species must await further study.

Although the particle size of the fine material is similar in the two lakes, the chemistry of the water at the substrate/water interface is modified by the nature of the silt. The inflow of rich organic silt into Lake Nhlange may be responsible for even more profound differences in substrate/water interface chemistry than that of deoxygenation of the water; that is, the addition of substances to the water towards which animals may behave in such a way as to exclude them from the substrate of one lake and not in another. It has been found, for example, that the responses of *Apseudes digitalis* and *G. lignorum* towards rates of carbon dioxide concentration increase are very different (vide p. 105, Fig. 29). Similarly, the presence of typically estuarine forms in Lake Sibayi and their absence from Lake Nhlange may depend on subtle differences in the chemistry of the water at the substrate/water interface of the two lakes. That they may be complex is suggested by the response of *G. lignorum* to lowered oxygen conditions (vide page 122, Fig. 37). This amphipod does not react to lowered oxygen tensions until less than 2 mg/l O_2. The absence of *G. lignorum*, at least from the well oxygenated shallow water of Lake Nhlange, cannot therefore be attributed to this cause.

These findings direct our attention to the necessity for defining the conditions at the substrate/water interface, and also to an examination of the responses of different species of animals of the benthos towards such conditions. These studies are probably best started in the controlled conditions of the laboratory and then confirmed, if possible, in the real situation in the bottom of the lake.

The benthic fauna of Lake Sibayi was clearly divided into epiphytic fauna on the weeds, epifauna on the sand and an infauna. The poorly developed growth of weed found in Lake Nhlange except in very shallow water is obviously correlated with the rapid drop in light intensity in the water (Allanson and van Wyk 1969). The lack of epifauna on the sand may be due to one or more causes.

The epifauna of Lake Sibayi is principally composed of freshwater mollusca, and the estuarine crab *H. orbiculare*. The freshwater molluscs, as might have been expected, are absent in Lake Nhlange. On the other hand, *H. orbiculare* is much less common on the benthos as compared with Lake Sibayi. The burrowing goby, *C. mossambica*, is the only large form which is distributed similarly in both lakes.

The bivalve, *Lamya capensis*, is the most prominent epifaunal element in the substrate of Nhlange. It is absent in Lake Sibayi and may be so because of the low salt content of the water. This is really an epiphytic form as evidenced by the larger animals to be found on the one station (NHL 17) in which large plants were found. It is, however, numerically very abundant, though small, on the sands and provides food for fish. The fish, *Gerres oyena*, utilizes this bivalve extensively for food and may in part be responsible for the saucer shaped depressions in the sand and the ploughed up appearance of the sand. Much larger fish from the sea are to be found in Lake Nhlange than in the isolated Lake Sibayi. Their activities in the lake may well inhibit the production of a rich epifauna in the lake.

The next surprising differences occur in the infaunal elements of the two lakes, some more easily understood than others. The chironomid population of Lake Nhlange is on the average nearly a hundred times greater per unit area than that for Lake Sibayi. Although the population may have been artificially enhanced due to unusual enrichment of the bottom sediments by the flood waters, their commoness as judged by qualitative anchor dredgings in July 1965 tends to indicate that they have always been much more common in Lake Nhlange consequent on the eutrophic nature of the lake.

It is clear that factors governing the distribution of benthic fauna in these two lakes are complex and one cannot hope to disentangle their effects by simple surveys of the abundance and distribution of the animals in the benthos of the lake. Biological investigations on these animals may help to more clearly understand how two such superficially similar lakes have such widely different types of fauna in the benthos.

THE BENTHOS OF SOME SOUTHERN AFRICAN LAKES

IV. The Benthos of Lake Shengesa and Lake Sifungwe

The spatial relationships of these two lakes with Lake Nhlange is shown in Fig. 2. Lake Sifungwe is connected to Nhlange on the seaward side by means of a narrow winding channel. Lake Shengesa is, on the other hand, totally isolated at the present time from the Kosi estuary/lake system. The water chemistry of the two lakes is different, and in terms of salinity lies on either side of the conditions in Lake Nhlange. Lake Shengesa is freshwater, with a salinity of less than 0.5°/oo and its water is stained deep brown from humic material, and Lake Sifungwe has a salinity of 4.26°/oo in the epilimnion above a halocline rising to 14°/oo below the halocline.

Lake Shengesa

It was felt that an examination of the benthos of Lake Shengesa would be interesting since the water chemistry is much like that of Lake Amanzimnyama, which is connected to Lake Nhlange, but which could not be examined because of hippopotami which interfered with attempts to sample the lake. The isopod *Dies monodi*, characteristically an estuarine isopod, was found in a small collection of marginal vegetation fauna of Lake Shengeza. The fishermen on the lake take quantities of the small goby *Gobius giurus* and *Silhouetta* sp.

The sampling of the benthos was carried out using the modified van Veen grab as described for Lake Sibayi (Boltt 1969). Depths on the lake were measured partly using an echo sounder (Hill 1969) and partly using weighted lines on calibrated winches (Boltt 1969). The oozy nature of the substrate in most cases

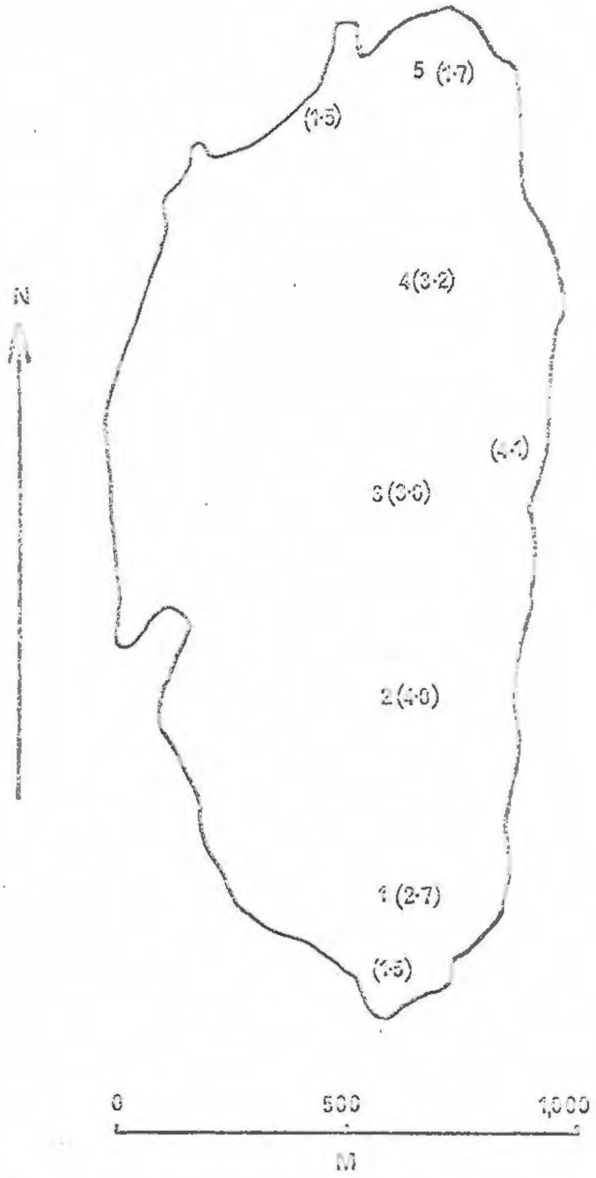


Fig. 16. Outline map of Lake Shengesa showing sampling stations and depths (bracketed).

gave a poor signal on the echo sounder, and hence a contour map of the bottom from transects could not be drawn. However, in general terms the bottom of the lake shelves generally from the west shore to a maximum depth of just over 4 metres very near the mid point of the east shore (Fig. 16). The substrate in waters of less than a metre was fine sand on the Wentworth scale (Morgans 1956). Consolidated lumps of algal bound sand were common on the sandy substrate. Chironomid larvae were common in the grab sample taken from this area, but no other animals were found when the samples were processed in the usual way (Boltt 1969). In the deeper water the bottom samples were composed entirely of thick lumpy porridge-like organic detritus. The presence of burnt grass fragments suggested that this material is largely derived from burnt grasslands around the lake. The samples were entirely barren of benthic fauna.

Lake Sifungwe

Lake Sifungwe has been described by Hill (1969) as a small lake with, relative to Lake Nhlange, very little shallow water. Hill's bathymetric chart, together with profiles calculated from his data is given in Figure 17. The lake is roughly divided into a shallow water plain above the 15 metre contour and a deep water plain below the 15 metre contour. Steeply sloping sides divide these areas between the depths of 5 metres and 15 metres. Only a few substrate analyses were done on Lake Sifungwe. As in Lake Nhlange, the shallow water plain consists of fine sand with a median phi value of 3.2. The substrate from the 15 metre plain was mud with a silt particle size with a median phi value of less than 6. Visual observations by SCUBA divers on the slopes leading down to the

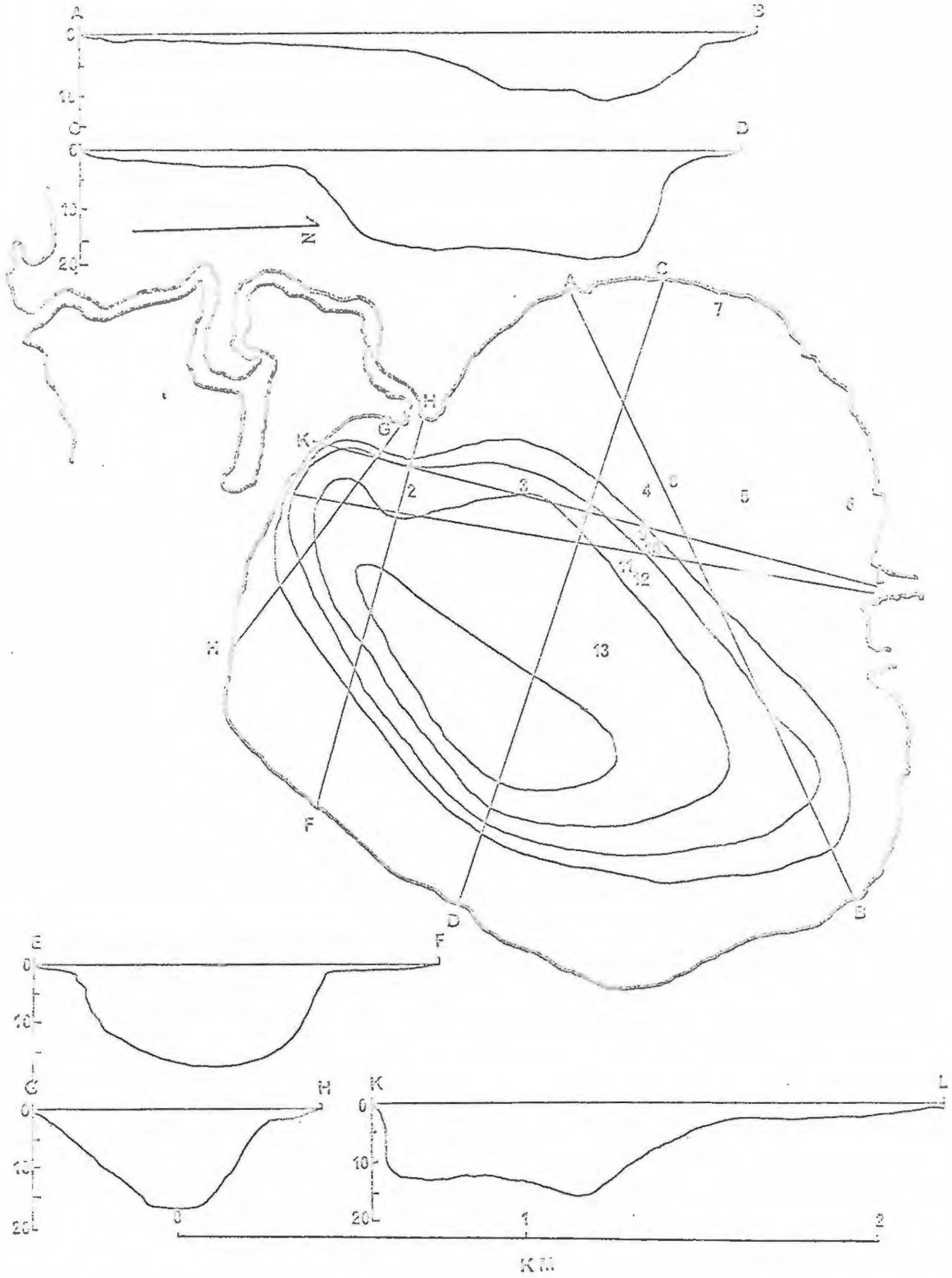
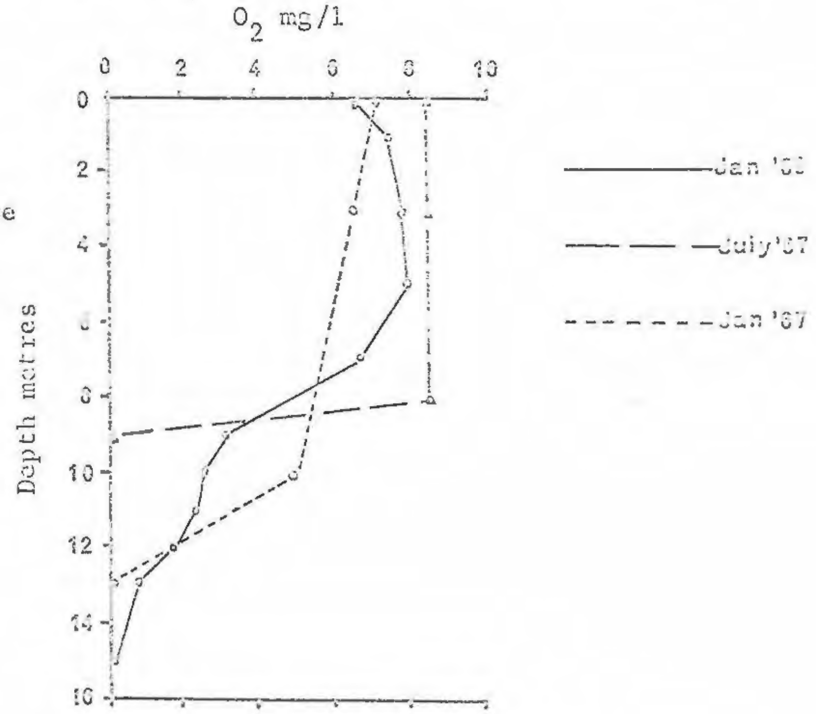


Fig. 17. Map of Lake Sifungwe showing sampling stations and profile transects. The contours in the lake are at 5 metre intervals. Depth on profiles in metres. Contour map from Hill 1969.

15 metre plain showed that the sand was generally [illegible] of fine particles until below 5 metres. An extremely thin film of brown material coated the sand from about 6 to 8 metres depth. The sand below this depth was coated with dark brown to black material tending to form a weak crust over the loose sand underneath. At about 10 metres, surprisingly the sand under the dark layer was white. Small heaps of broken bivalve shell were common on the sand. These were quite fresh, showing an uncorroded nacreous layer. This has been taken as evidence of feeding activity of fish, probably the spotted grunter <u>Pomadasys</u> <u>operculare</u>.

Figure 18.

Dissolved oxygen from Lake Sifungwe. The high surface temperature (30.5°C) depressed the amount of oxygen in the surface waters in January 1969. Data for the 1967 curves from Allanson & van Wyk (1969). Data for 1969 curve by kind permission of Professor B.R. Allanson.



Allanson & van Wyk (1969) have shown that the lake is permanently stratified at 9 metres into two distinct water masses by a halocline. The epilimnion has a salinity of 4.35°/oo and the hypolimnion 13.0°/oo. Associated with the halocline is a rapid decrease in the oxygen concentrations below 9 metres. In January 1967 and 1969, the decrease between 9 metres and 13 metres was very rapid (Fig. 18). In July 1967, dissolved oxygen fell

Table 11

Species list of the benthic fauna of Lake Sifungwe. Species from the marginal vegetation have also been included. (* denotes species reported by Broekhuysen and Taylor (1959)).

CRUSTACEA

Amphipoda

Grandidierella _bonnieri_ Stebb.

Corophium _triaenonyx_ Stebb.

*_Urothoë_ _serrulidactylus_ Brnrd.

*_Melita_ _zeylanica_ Stebb.

*_Afrochiltonia_ _capensis_ Brnrd.

*_Talorchestia_ _ancheidos_ Brnrd.

Isopoda

Cirolana _fluviatalis_ Brnrd.

*_Pontogeloides_ _latipes_ Brnrd.

*_Synidotea_ _variegata_ Cllge.

Tanaidacea

*_Tanais_ _philitraeus_ Stebb.

Decapoda

Hymenosoma _orbiculare_ Desm.

*_Callianassa_ _kraussi_ Stebb.

ANNELIDA

Polychaeta

Ceratonereis _keiskama_ Day

*_Dendronereis_ _arbifora_ Peters

Prionospio near _sexoculata_ (Personal communication Day)

Oligochaeta

(Not identified further)

MOLLUSCA

Pelycopoda

*_Lamya_ (_Modiolus_) _capensis_ Krss.

*_Psammobia_ ? _ornata_ (MPU 10)

?_Psammobia_ sp. (MPU 11)

INSECTA

Chironomid larvae.

from 8.4 mg/l at 9 metres to zero at 10 metres. The permanence of the halocline has been shown by the reverse gradient of bottom temperatures in winter when the hypolimnion was at 25.2°C and the surface was at 19.1°C (Allanson & van Wyk 1969).

Grab sampling stations are plotted in Figure 17. The results of the sampling programme are given in Table 10, and a list of the animals of the benthos in Table 11.

Broekhuysen and Taylor (1959) have reported on the marginal fauna of the lake, and from the present data their faunal list may be extended by some seven species.

There are more species in the benthos of Lake Sifungwe than there are in Lake Nhlange; Urothoë serrulidactylus, Callianassa kraussi, Prionospio sp., Dendronereis arbifora and the two Psamobiid bivalves not having been taken in Lake Nhlange. No species of estuarine animal found in Lake Nhlange was absent from Lake Sifungwe, although G. bonnieri was remarkably uncommon. It may be that the presence of more species in Lake Sifungwe is due to an increase of salinity of up to 1.2°/oo relative to Lake Nhlange. Although this is only a small difference, Sutcliffe and Shaw (1968) have shown that small differences in sodium concentration are sufficient to allow one race of a species to cope with dilute water and another not.

In general, the fauna extended downwards to the 10 metre contour. As the table shows, no animals were found below this depth. Urothoë serrulidactylus however, although common in shallow water up to 4 metres was not found below this depth. This amphipod does not build tubes, but burrows through the sand. At about 5 metres the character of the sand changed slightly. It would be interesting to know if there is any causal relationship between these findings.

Although the discontinuity layer has been reported to be at 9 metres by Allanson and van Wyk (1969), animals have been found up to a metre below this depth. In summer the oxygen only disappears at 13 metres, and hence it is not surprising to find animals deeper than the discontinuity layer in summer. However, in winter, as Figure 18 shows, dissolved oxygen had disappeared at 9 metres. An invasion of the substrate in deeper waters during summer had therefore taken place. This was confirmed by the observations of SCUBA divers, who noted that in the bottom there were the openings of burrows about 0.5 cm in diameter. Counts of burrow numbers for 0.25 metre square areas are given in Table 12.

Table 12

Large (0.5 cm) and small (2 mm) burrow openings in the substrate of Lake Sifungwe per $0.25 m^2$ counted by SCUBA divers. January 1969.

Depth	Large burrows	Small burrows
2m	4	35
2m	4	79
2m	4	70
4m	6	72
3.5m	2	79
5m	0	11
5m	6	56
6m	13	0
10m	5	0
10m	6	0

At 10 metres and just below, there were numbers of large burrow openings, but most were old and showed up in contrast to new burrows which had small piles of clean white sand around the entrance of the burrows. Close inspection of the old burrow openings showed that they tended to be uneven and crumbling and clearly unoccupied, whereas the new burrow openings were recently repaired.

The reasons for the sharp cut off of oxygen at the discontinuity layer in winter as compared with summer is not easy to explain. Since the lake has only been visited either in the height of summer or of winter, nothing is known of the changes that take place between these times. The effect of temperature differences on the density of epilimnetic water will be less severe than the density difference between the hypolimnetic water and the epilimnion caused by salinity differences. The lake probably cannot therefore turn over at any stage since at no time will the densities of the two water masses be the same. Some mixing must be taking place all the time, at the discontinuity layer, and this presumably under the influence of wind stresses on the surface. It may be that wind stresses are greater in summer than in winter, and these allow of better penetration of dissolved oxygen. However, no data on the wind forces operating on the lake are available.

Grab sampling only gave information about the smaller infauna of the substrate. Divers found evidence for the larger forms in the substrate. From diggings in the very shallow water of the margins of the lake, Callianassa kraussi and Dendronereis arbifora were found to construct permanent tubes in the sand. Two species of Psamobiid bivalves were also discovered.

The burrows of the worm and the prawn were reasonably easily identified. The prawn tends to build a rather large untidy mound at the mouth of the burrow, while the polychaete tends to build a much smaller but still recognisable mound which is much more symetrical around the lip of the burrow. One other burrowing animal was found in the lake, namely Croilia mossambica. This fish has practically no mound at the entrance to the burrow. To burrow the fish picks up a load of sand in the buccal cavity from the bottom of the burrow and swims a short distance away from the burrow to spit the sand out. In the lake they are not found more than about half a metre away from the burrow, and these usually could therefore be correlated with an animal. Using these criteria to identify burrows, it was found that burrows clearly belonging to *Callianassa* did not appear to occur below 1.5 to 2 metre depths. The burrows reported from deeper water seemed to belong either to *D. arbifora* or *C. mossambica*. The burrowing goby was much less common than *D. arbifora* and none of the quadrats counted actually incorporated any of their burrows.

The problem of inhabitants of small holes reported in Table 12 has not been resolved. These may belong to juvenile *D. arbifora*, but since no intermediate sizes were common, it was thought that they may belong to a different animal. Furthermore, small heaps of sand were not found around the entrance to the burrow.

The two psamobiid bivalve species were reasonably common in the substrate and were found by divers running their hands through the sand. No quantitative data was collected on the two short dives, but an impression was gained that the one *Psamobia* ? *ornata* may have been more common in slightly deeper water (6 or more metres).

General discussion on South African coastal lakes.

Clearly the main basins of the South African coastal lakes were cut in fine sands typical of the East and South coast. Sand particle sizes from the Zululand lakes, the Knysna district lakes and from the Kowie estuary lagoons all have a median phi value of about 3.1. The bathymetric surveys of the lakes indicate that they are the remains of old valley systems cut in the late pleistocene, the mouths of which have been partly or wholly blocked by sand from the seaward side. This is true for the smaller lakes as well. Groenvlei, for example, which at present has no connection with the sea, has a bottom up to 7 metres deeper than present sea level. This bottom is generally muddy in the deep water. Martin (1959) reported that the mud was uniform to a depth of at least 1.6 metres in the lake.

The original sandy basins of the lakes have had allocthonous and autocthonous material added to them which covers part or most of the sand as mud. The size of the catchment area and also the surrounding vegetation type have determined the nature and quantity of the allocthonous material added to the lakes. The basins of those lakes which have reasonably large rivers running into them, have been almost entirely filled with silt from inland erosion. Lake Umhlatuzi (Richards Bay) is 12 square miles in area, but in the greater part of the lake is only up to a metre deep (Millard and Harrison, 1952). The St Lucia lakes are 120 square miles in area, and the maximum depth recorded by Day, Millard and Broekhuysen (1952) is just over 2 metres. Bolangvlei, a small 5 metre deep lake in the Knysna district, which has a number of small streams coming in on the North shore, has very little sandy substrate and the whole of the lake shore is covered in a thick layer of mud.

Examples of lakes whose originally sandy basins have been mostly covered with organic detritus rather than silts from the land, are Lake Shengesa and Groenvlei. Windblown organic debris from grass fires has covered the substrate of Lake Shengesa but, as Martin (1959) has shown, autocthonous material is largely responsible in Groenvlei. The gyttja of the latter lake consists almost entirely of ostracod shells in some of our samplings. Addition of material of any kind has been least in the two Zululand lakes, Sibayi and Nhlange, largely because of their size relative to small lakes like Groenvlei, and the particular conditions surrounding them. The catchment area of Lake Sibayi is remarkably small, and the swamp forests around Lake Nhlange and lining the water courses leading into the lake act as silt traps. In any case, the catchment for both lakes drains from extremely sandy soil, in contrast to the catchment areas of the rivers of the St Lucia lake system.

Lake Sibayi and Lake Nhlange have of course not escaped the addition of allocthonous material entirely, and both lakes have muddy substrates in some areas. But the nature of the material contributing to the mud of each lake is very different. The muds of Lake Sibayi correspond closely to the description of gyttja given by Ruttner (1963), being largely autocthonous in origin and grey rather than black, at least in the main basin. This is a well degraded material which has been degraded in well oxygenated conditions. The catchment area of the lake is small and the mouths of most of the streams are blocked with Typha swamps which act as traps to silt. Further, the topography of the shoreline is such that any autocthonous material carried in by the streams must travel 2 or 3 kilometres from the stream mouths to the main basin of the lake along arms of the lake.

Lake Nhlange, on the other hand, is partly surrounded by swamp forest (Fig. 2), and undegraded humic material is dumped straight into the lake, especially in time of flood. We have no record of the frequency with which this might happen. The St Lucia lake system has been subjected to flooding on at least two well documented occasions (Day, Millard & Harrison 1952 and Millard and Broekhuysen 1964/65). Although flooding such as was experienced in January 1966 may be a less common event in the Kosi lake area, one would expect such an event to occur periodically.

Consequently, the substrates of the South African coastal lakes fall into the broad categories of either fine sands or muds, each of which may contribute more or less to the total substrate type.

Overlying the sandy substrates there is a thin layer of fine particle size material which may be likened to the "mulm" that gathers in aquaria. This feature is particularly important in Lake Sibayi and Lake Nhlange. Lake Sifungwe for example has little of this material overlying the sand, only having a skim of brown, probably diatomaceous, cells over the sand. The situation in the Knysna lakes is as yet poorly known. The origin of the material in Lake Sibayi seems to be almost all from autocthonous sources save perhaps for dust which may make up the clay fraction. In Lake Nhlange, plant fragments from the humic load from the swamps are common, and hence the "mulm" of this lake is derived from both autocthonous (the exuvae of insects and the ostracod remains) and allocthonous material. A discussion of its importance is best left until after the experimental studies.

Table 13

Lists of fauna from the highest stations of the estuaries reported by the Cape Town University Ecological Survey Group, and from the lakes of the coastal strip of South Africa.

Species	Klein River (Scott et al 1952)	Knysna (Day et al 1952)	St Lucia (Day et al 1953)	Mpungwini (Broekhuysen et al 1959)	Sifungwe	Nhlange	Rondevlei	Onderlangvlei	Bolangvlei	Groenvlei	Sibayi	Shengesa
CRUSTACEA												
Isopoda												
Pseudosphaeroma barnardi	+	+	-	-	-	-					+	+
Cirolana fluviatalis	+	+	+	+	+	+					-	-
C. luciae	-	-	+	-	-	-					-	-
Cyathura carinata	+	+	+	-	-	-					+	-
Dies monodi	-	-	+	-	-	-	-				-	-
Paramunna sp.	-	+	-	-	-	+					+	-
Corralana africana	-	+	-	-	-	-					-	-
Sphaeroma terebrans	-	+	-	-	-	-					-	-
Amphipoda												
Melita zeylanica	+	+	+	+	+	+	+	+	+	-	-	-
Parorchestia rectiplalma	+	+	+	-	-	-					-	-
Corophium triaenonyx	+	+	+	+	+	+	+	+	+	+	+	-
Grandidierella lignorum	-	+	-	-	-	-				+	+	-
G. bonnieri	-	-	+	+	+	+				-	-	-
Afrochiltonia capensis	-	+	+	+	+	+					+	-
Eriopsella chilkaensis	-	-	+	-	-	-					-	-
Talorchestia ancheidos				+	+	+	+	+	+	+	+	-
*Urothoë serrulidactylus	-	-	-	+	+	-	-	-	-	-	-	-
Tanaidacea												
*Apseudes digitalis	-	-	-	-	-	-	-	+	-	-	+	-
Leptochelia sp.	+	+	-	-	-	+	-	-	-	-	+	-
Decapoda												
Hymenosoma orbiculare	+	+	+	+	+	+					+	-
Rhynchoplax bovis	-	-	+	-	-	+					-	-
Cleistosoma algoensis	-	+	-	-	-	-					-	-
Callianassa kraussi	-	+	-	+	+	-	-	-	-	-	-	-
MOLLUSCA												
Pelycopoda												
Lamya capensis (Modiolus)	+	+	+	+	+	+	+	+	+	-	-	-
Psamobia sp.	-	-	-	+	+	-	-	-	-	-	-	-
Annelida												
Polychaeta												
Ceratonereis keiskama	-	+	-	+	+	+					+	-
Dendronereis arbifora	-	-	-	+	+	-	-	-	-	-	-	-
D. zululandica												
Prionospio sp.	-	-	-	-	-	-	+	+	+	-	-	-
Marphysa simplex	-	-	+	-	-	-	-	-	-	-	-	-

Notes: Klein River Canal region 0.36-7.9^{o}/oo salinity. Knysna, Charlesford Rapids mean sal. 3.9^{o}/oo. St Lucia, Hluhluwe River and Nyalazi River no salinity data given. Mpungwini 4.4-7.8^{o}/oo. Sifungwe 4.25^{o}/oo. Nhlange 3.3-4.0^{o}/oo. Rondevlei, Onderlangvlei and Bolangvlei 12-15^{o}/oo. Groenvlei 2.5^{o}/oo. Sibayi 135 ppm Cl^{-}. Shengesa 0.5^{o}/oo.

* denotes animals from Kowie estuary in 35^{o}/oo salinity.

In the lakes thus far studied intensively, the sandy substrates support the greater part of the biomass of the benthos. The muddy substrates support far less, and in some cases none at all. Initial surveys on the Knysna lakes confirms this point of view even for the smaller lakes.

The studies on the mud of the 15 metre plain of Nhlange show that at least for the lakes receiving some undegraded organic detritus, favourable conditions may exist for large chironomid populations, but that these are controlled by the oxygen demand of the mud. If the interpretation of events that have been observed in Lake Nhlange has not been amiss, it will be interesting to follow the further degradation of the mud in that lake together with population changes of chironomids.

The lack of such a chironomid population in Lake Sibayi in the muddy substrates, may mean that the mud no longer offers the food resources of less degraded material. Although McLachlan (1969) has shown that *Nilodorum brevibucca* (Chironomidae) will choose silt in preference to coarse or fine sand, he also draws attention to the necessity for sufficient food for the larvae to live off. Similarly, experiments reported later on *G. lignorum* indicate that these amphipods will build burrows in silt rather than sand, perhaps because the food resources of this material in the experiment were better than those offered by the sand.

Indeed, Meadows (1964) has already shown that with *Corophium volutator*, even on sandy substrates it is not so much the physical characteristics of the substrate which matter as the presence of the micro-fauna on the sand grains.

The species comprising the benthic fauna of the coastal lakes may be divided into those derived from the essentially marine environment of estuaries, and those derived from fresh water. The composition of the fauna of the benthos of the lakes varies from an almost totally estuarine fauna as in the St Lucia lake system, to the condition obtaining in Lake Sibayi, where the estuarine component only forms less than half of the important species of the benthos (Boltt 1969, Table 2, p. 253). But the estuarine fauna has never been found to be unimportant in contributing to the biomass of the benthos of any of these lakes, Lake Shengesa excepted, where benthic fauna is practically non-existent.

The status of species of the lake fauna is interesting in relation to Day's classification of South African estuarine animals (Day 1967). He has divided the estuarine fauna into five components, abolishing one from his previous lists (Day 1951, 1964 and 1967). They are a freshwater component, an estuarine component, a euryhaline marine component, a stenohaline marine component and a migratory component. Two of the categories are of special interest, the estuarine component including those species which are never found in the sea and which occur along the full length of the estuary. They are most important only at the head of the estuary. The euryhaline and marine component is found both in the sea and the estuary, and is the most important component of the estuary. The stenohaline marine component as defined by Day only occurs in the Knysna estuary (Day 1967, p. 400). The usefulness of these categories will be discussed after the experimental studies have been reported.

Considering the fauna of the least saline lake, with abundant benthic populations, the estuarine derived fauna is mainly of the estuarine component although two species, Cyathura carinata, and Hymenosoma orbiculare are euryhaline and marine.

Table 13 compares the fauna reported by the Cape Town

Ecological Survey group for the topmost stations of three estuaries they have studied, with the fauna from the lakes. With the exception of the Psammobiid bivalves, Apseudes digitalis and Urothoë serrulidactylus, the species found in lakes having salinities less than 5°/oo have all been taken from the topmost stations of at least one estuary or another. In all cases the species extend into the more saline lagoon regions of the estuaries as well.

The distribution patterns of animals in the lakes, and among the lakes, is of great interest because it clearly indicates that simple correlations between the gross physical and chemical characters is in many cases misleading, and does not afford an understanding into the reasons behind these phenomena.

For example, the range of salinities offered by the lakes varies from hypersaline conditions at times in False Bay, in the St Lucia system (Day et al 1952) to permanently freshwater conditions which have been reported from Lake Sibayi. While it is quite true that the number of species from the St Lucia lakes is much greater than for Lake Sibayi, and that certain forms such as for example Solen capensis (Day et al 1952) is present in St Lucia and absent in Lake Sibayi, probably for reasons of inability to cope with the salinity, or for that matter is absent from Nhlange for similar reasons, the lack of a great number of species in the more saline lakes which are present in Sibayi cannot be explained in this way. Thus, an attempt to guage the effect of salt concentration on the presence or absence of fauna from any particular lake or estuary meets with little success. Day (1951, 1964, 1967) clearly recognised that simple correlations between one factor and distribution were not enough to account for a distribution pattern. Several factors such as salinity, substrate type, current, wave action, etc. were important in determining the distribution of a species at least in an estuary. We may go one further and say that until the response of a particular species to a set of factors is understood, we cannot be sure that any correlations which have been observed by field survey methods are causal or incidental. For example, the problem of Urothoë serrulidactylus, which is present in many estuaries of high salinities (e.g. Kowie estuary at $35^{o}/oo$) is also present in Lake Sifungwe ($4.25^{o}/oo$) but absent from Lake Nhlange (3-$4^{o}/oo$). As has been suggested, this may be due to ineffective salt regulation below the salinity levels found in Lake Sifungwe. Equally, some as yet unknown factor in the substrate may be important. If one cannot place reliance on correlations from field data, what is to be done? Knowledge of the biology of a species in terms of physiological and behavioural responses towards certain conditions certainly increases the understanding of the reasons behind any particular distribution pattern as has been elegantly shown by numbers of workers in very different fields. Heeg, for example, (1967a & b 1969) studying the water economy and behavioural reactions in Thysanura (Insecta Apterygota) showed that although the lepismatids were sensitive to water loss in arid conditions, they have a wide distribution in apparently arid regions. He explains the observed wide distribution of the lepismatids in terms of their ability to take up water from undersaturated atmospheres, making them independent of the presence of water in its liquid state. Similarly, Sutcliffe (1968) has used biological data gained from laboratory experiments and field data to gain an understanding of the distribution of the various species of amphipods of the genus Gammarus in Britain.

However, not all the biology of an animal directly affects its pattern of distribution. The problem then is what responses should be investigated in order to gain insight into a particular distribution pattern? Perhaps the simplest way to start is by investigating fairly obvious factors from the environment and pursue them to a point where it becomes reasonably clear that further pursuit is not likely to help in explanation of a particular problem. One may, of course, have to return to apparently fruitless initial lines of investigation.

One further important point is that the laboratory study in itself is insufficient to establish the reasons behind a particular distribution pattern. Conditions shown to have an effect in the artificial laboratory experiment must be shown to exist in the real situation in the field.

Of the many problems posed by the field results from the South African coastal lakes, the problem of the distribution of G. lignorum in Lake Sibayi has been chosen. The second part of this work relates to laboratory and field work which has led to a partial understanding of the problem.

EXPERIMENTAL INVESTIGATIONS OF THE RESPONSES OF GRANDIDIERELLA LIGNORUM TO CERTAIN PHYSICAL AND CHEMICAL CHANGES UNDER LABORATORY CONDITIONS

Introduction

During the course of the field work, numerous problems of a purely ecological nature posed themselves. Why was it that the lakes Sibayi and Nhlange had such a different species composition? Why were the crustacea of Lake Sibayi living there in very low salinities regarded as freshwater, and lacking in water which seems to be suitable at the tops of the estuaries in the South African Eastern Cape Province? Considerations of the known chemical and physical environments from the field work of Allanson & van Wyk (1969) gave no clue as to answers to these problems. It was felt that too many varying factors are present at the same time in the field which lead to considerable difficulties of interpretation. Nevertheless, the field data gave an indication of the types of factors that might be influencing the distribution of animals among and in lakes. What was considered a reasonably defined problem was chosen, and some of the more obvious factors that could play an important part in the pattern of distribution were experimentally investigated in the laboratory. Thus, armed with better knowledge of the influence of these factors on a particular animal, the problem might once again be investigated in the field with a better insight into exactly what factors to measure.

The choice of the problem was governed by a number of considerations. The lakes are 800 miles from the laboratory and therefore only a limited amount of experimental material was readily available. However, *Grandidierella lignorum* was available in large numbers from the Kowie River estuary. Other species were also available, but in smaller numbers and, therefore, for the present work attention has been confined to *G. lignorum*. This animal was also chosen since it shows an interesting discontinuous distribution in Lake Sibayi. The tanaid Apseudes digitalis was used where it was thought that comparative results might help in elucidating the problem on G. lignorum.

The problem of the discontinuous distribution of G. lignorum in Lake Sibayi seemed to offer a reasonably well defined topic for a preliminary study of benthic fauna of at least one of the South African coastal lakes. A good deal of preliminary study on the limnology of the lake is known (Allanson & van Wyk 1969, Allanson 1969, Tinley 1958b and Hill 1969, Boltt, Hill and Forbes 1969, Boltt 1969, Forbes and Hill 1969). From this starting point therefore an enquiry into the phenomenon has begun.

Factors which might influence the distribution of G. lignorum with respect to depth which immediately spring to mind are:

a. Choice of substrate,

b. Pressure effects,

c. Light intensities,

d. Dissolved gaseous environments at the substrate-water interface, as being a reflection of conditions there.

It is inconceivable that there would be no interactions between these factors in the real situation of the lake. However, it is likely that some are more important than others in determining the distribution of the animals in the lake. The work has therefore been directed towards assessing the effect of these factors on G. lignorum. As soon as it became clear that the effect of the factor being studied was minimal, further work on that particular aspect was discontinued. It was felt that although fascinating in themselves, complete analyses would in all probability not really help towards solving the problem with relation to the lake; that of the reasons for the distribution of G. lignorum with respect to depth in the lake.

Initially it was important to establish if it was possible to use material from the Kowie River estuary as experimental material for the problem related to Lake Sibayi. If it could be demonstrated that the Kowie population was physiologically similar to the Sibayi population, Kowie material could be used to obtain some idea of the effects of the factors which might influence the distribution of animals in Lake Sibayi. The first section therefore reports the salinity tolerances and responses of animals from Lake Sibayi, the Knysna District lakes (Groenvlei, etc.) and Kowie estuary. It cannot be claimed that the responses to salinity changes prove that physiologically these three populations are identical. However, it does seem to argue that if there are any differences they are likely to be minimal, and therefore it is reasonable to suppose that the results of experiments carried out on a population normally living in the Kowie estuary at 35 $^{o}/oo$ salinity are applicable to the material from Lake Sibayi.

The later sections report the results of experiments on G. lignorum with respect to substrate, pressure, light, and gas concentrations. Some further data on conditions near the bottom from the field is included in a separate section.

Salinity tolerance of G. lignorum

Until the present study, save for one doubtful record, G. lignorum has been associated with estuarine conditions normally showing high salinities not much different to sea water. It has been recorded from the following localities by the University of Cape Town Survey: Isipingo, Umgababa, Umzimkulu, Umzimvubu, Umzimbosi, Bashee, and St Lucia. (Professor J.H. Day, personal communication). Barnard (1952) described the species from

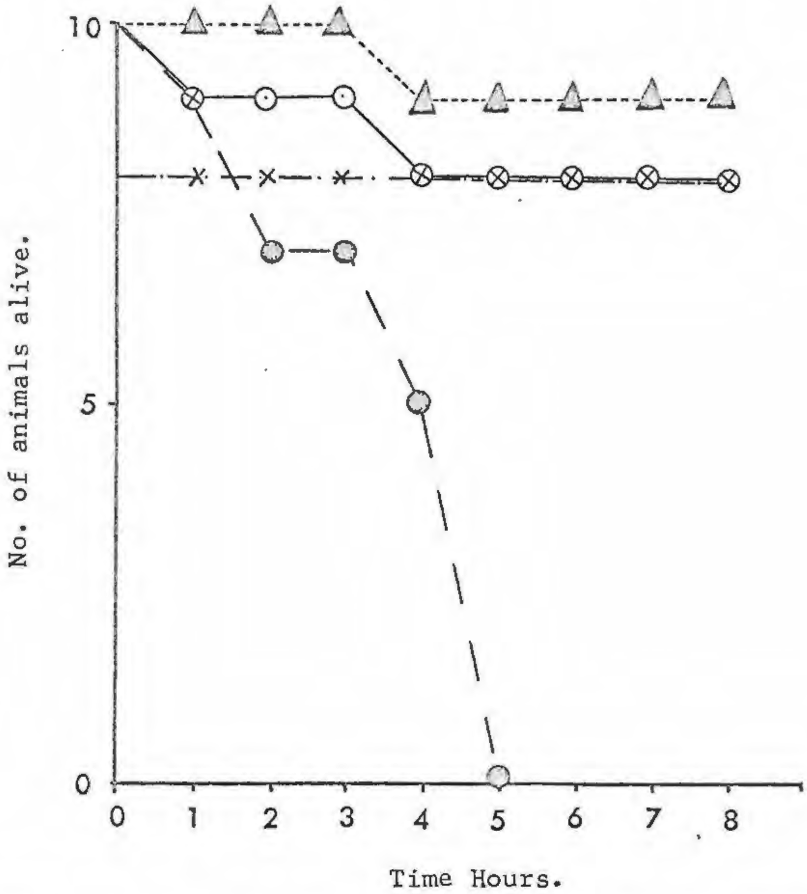


Fig. 19. The survival of G. lignorum in pond water after direct transfer from sea water (● closed circles) and after initial transfer to 50% sea water (▲ Triangles) and 25% sea water (X crosses). The control in 100% sea water is shown with open circles. Excellent survival is shown provided the animals are exposed to an initial dilution before being put into pond water.

Keurbooms River estuary and noted that two specimens from Zoetendals Vlei, Bredasdorp District, Cape, might doubtfully be referred to the species. Since then we have established its presence in further estuaries and also in isolated lakes. It has been taken from the East lagoon of the Kowie estuary and also at the head of the estuary some 16 km from the sea, from the Boknes lagoon and also from Kasouga estuary. G. lignorum has also been taken from Lake Sibayi and from Groenvlei, a small isolated lake in the Knysna district. It has been recorded from Bolangvlei, Rondevlei, and Onderlangvlei; also in the Knysna district. These are small lakes connected by channels to each other and the estuary. These localities represent a spectrum of salinities ranging from fresh water in Lake Sibayi to full sea water in the open estuaries like the Kowie estuary. Both stable and fluctuating salinity conditions are represented in the localities reported. Thus we have recorded 0.5^{o} /oo salinity some 16 km down stream from the top of the Kowie estuary immediately after heavy rains and flooding, when the estuary was temporarily flushed out. Airey (1968) has reported that salinities at the head of the estuary varied between 31^{o} /oo to 1.6^{o} /oo. Stable salinities are represented by the lakes.

1. Survival of G. lignorum when transferred from sea water to fresh water.

G. lignorum from the East Lagoon of the Kowie estuary were extracted from the substrate and either transferred directly to pond water or allowed 48 hours acclimation in 25% or 50% sea water and then transferred to pond water from the Botanical Garden, Grahamstown. Ten animals were maintained in plastic containers in 250 ml of test solution. The containers were fitted with tightly closing lids. The containers were inspected for deaths of animals every day; the results are shown in Fig. 19.

When animals were directly transferred from 100% sea water

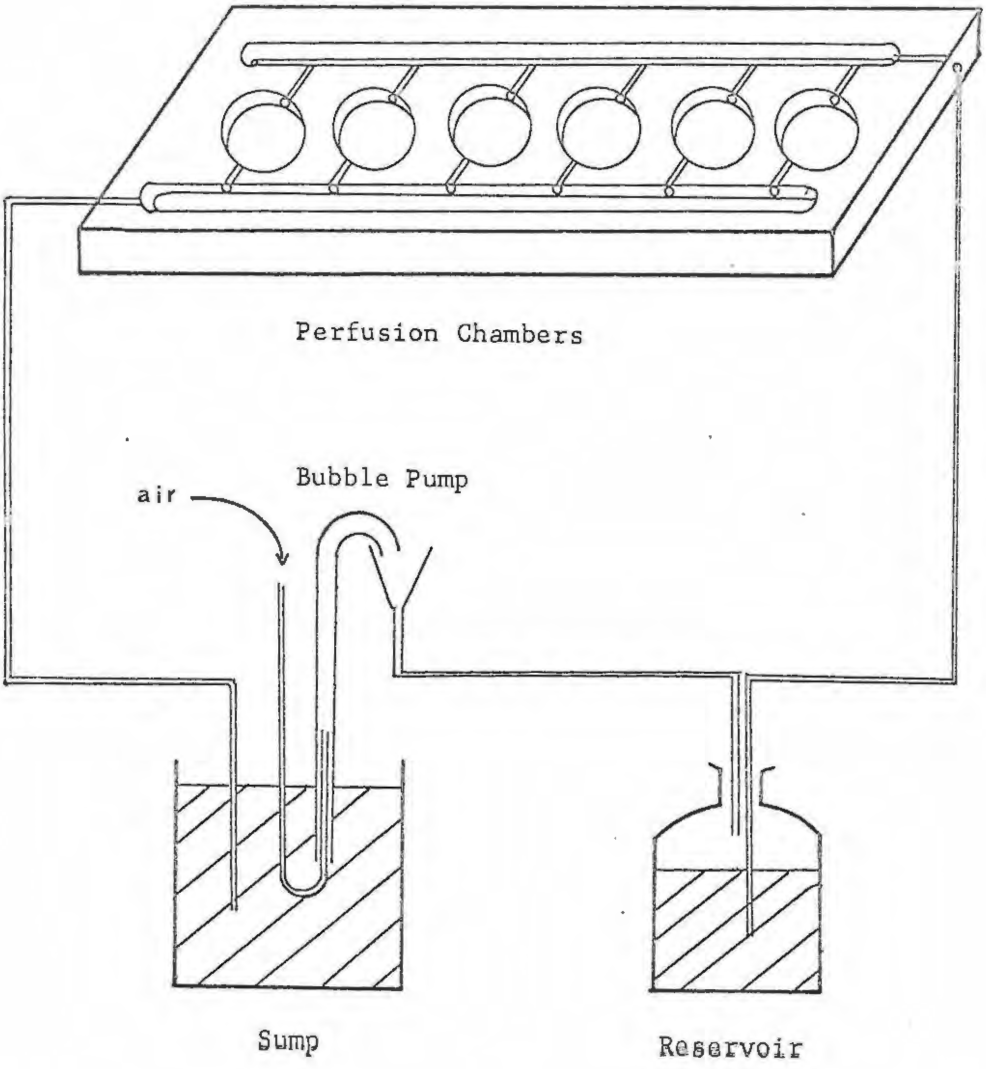


Fig. 20. Diagram of apparatus used to isolate individual *G. [illegible]norum* in moulting studies. The reservoir was at a slightly higher level than the perfusion chamber so that water siphoned through to the sump. The water was recirculated to the reservoir by the bubble pump.

to low dilutions of below 10% sea water or pond water, deaths occurred markedly sooner than if animals were first exposed to 50% or 25% dilutions of sea water. In the latter case few deaths had occurred after 8 days exposure and this was not different to the controls in 100% sea water. Considering the harsh environment offered of a substrateless box, this seems to be good evidence for the ability of G. lignorum from the lagoons to survive in fresh water. It did not prove possible to feed the animals, and the only food available was the corpses of the occasional animal which died in the boxes. After 36 days only one animal survived in 100% sea water, 3 in 25% and 3 in pond water.

2. The ability of G. lignorum to moult in freshwater.

The most sensitive time for a euryhaline crustacean exposed to water of low salinity is at the time of moulting when water is taken in by the animal to swell the new exoskeleton. Hill (1967) has shown that the mud prawn Upogebia africana puts off the moult in low salinities and eventually succumbs at moult. In order to study the ability of G. lignorum to moult in fresh water, individuals were isolated in separate chambers in a perspex block perfused with pond water (Fig.20). Only a single individual moulted in these conditions. The animal moulted successfully, but 24 hours later became stuck in the artificial burrow and died. This period is well after the time most critical in the moulting cycle. Upogebia africana died during the process of moulting and did not get out of the old exoskeleton (Hill 1967).

Moulting is a reasonably uncommon event in G. lignorum. During the survival experiments animals were inspected once a day on a total of 133 occasions. A total of only 9 moults was recorded in all salinities and only one occurred in pond water; this individual survived for at least six days after ecdysis. From this slight evidence it would appear that G. lignorum taken from full sea water in the Kowie estuary is able to moult successfully in fresh water.

A second critical period in crustacean life cycles is the hatching of eggs. In the crab *Hymenosoma orbiculare*, Forbes and Hill (1969) showed that the young larvae were the most sensitive stage to low salinity. In the survival experiments females of *G. lignorum* with eggs in the marsupial pouch were common. These eggs were in various stages of development. At no time was abortion of eggs in the pouch noted. The hatching of young amphipods from the marsupium was noted on five occasions. Two of these occurred in animals in pond water, indicating that the young stage of *G. lignorum* are also capable of surviving fresh water.

3. The control of the blood osmotic pressure by *G. lignorum* in different salinities.

Methods

G. lignorum from the Kowie estuary were exposed to various dilutions of sea water. For comparative purposes small numbers of animals from Groenvlei, Rondevlei, Onderlangvlei, Bolangvlei (lakes in the Knysna district) and from Lake Sibayi were brought to the laboratory for study in their natural waters. Animals from Groenvlei (salinity 2.5^{o}/oo) were also exposed to increased salinities.

Individual amphipods were blotted dry with tissue and immobilised by immersion in liquid paraffin oil, thus preventing desiccation during bleeding. The animals were bled by inserting fine silica glass capillaries filled with liquid paraffin in the region of the heart through the dorsal tergites. Care was taken not to break the lateral diverticulae of the gut and thus contaminate the blood. The freezing point depression of the blood sample was determined by the method of Ramsay and Brown (1955) using apparatus built by the workshops of the Council for Scientific and Industrial Research, Pretoria. Freezing point depressions were measured both

Table 15

Depression of freezing point of blood taken from *G. lignorum* from different localities having differing salinities.

Date	Place	Salinity °/oo e	Δi	Mean- Δi	st. dev.
27/5/66	Kowie	35°/oo	1.572		
			1.666		
			2.036		
			1.589	1.640	0.055
			1.528		
			1.497		
			1.597		
30/9/67	Kowie	35°/oo	1.583		
			1.504		
			1.480	1.487	0.064
			1.397		
			1.472		
6/4/67	Kowie	35°/oo	1.760		
			1.740		
			1.618		
			1.589		
			1.580	1.679	0.032
			1.838		
			1.827		
			1.709		
			1.600		
			1.532		
20/9/66	Rondevlei	12°/oo	1.294		
			1.089		
			0.981		
20/9/66	Onderlangvlei	6°/oo	0.905		
			0.974		
			0.981		
20/9/66	Bolangvlei	6°/oo	0.867		
			0.824		
			0.829		
19/9/66	Groenvlei	2°/oo	0.906		
			0.893		
			0.977		
19/3/67	Groenvlei	2.5°/oo	0.949		
			0.905		
			0.815		
			0.880		
			0.835		
			0.728	0.814	0.108
			0.876		
			0.885		
			0.675		
			0.595		

for internal and for external fluids by the same method. Measurements on the same sample were repeated three times until enough skill in controlling the rate of temperature rise of the apparatus was obtained so that small differences only appeared in the third decimal place of the reading. Thereafter single determinations were done on each sample.

During the taking of a sample from the animal under oil, small droplets of water could sometimes be seen on the hydrofuge cuticle of the animal. To check for the possible contamination of the samples from this source, duplicate samples were taken from each of two animals, and the depression freezing point checked. The results are shown in Table 14. The differences measured between the two samples from the same animal differed by small amounts in the third decimal place which can only be estimated on the Beckman differential thermometer used. Greater variation was experienced between animals, and therefore this source of error has thus been ignored.

Table 14

Results of the depression of freezing point of two samples of blood from the same animal repeated on two individuals of <u>G. lignorum</u>. Differences in the third decimal place on measurements on the same animal are less than differences between animals from 35^{o}/oo salinity.

Specimen No.	No. of Depression Freezing point determinations.			Mean	Difference
	1	2	3		
A	2.551	2.555	2.553	2.553	0.007
A	2.561	2.560	2.560	2.560	
B	2.625	2.627	2.622	2.625	0.005
B	2.620	2.619	2.620	2.620	

<u>Results</u>

Depression of freezing point measurements on animals from the same locality show considerable variation (Table 15).

<u>G. lignorum</u>, when exposed to dilutions, regulates rapidly

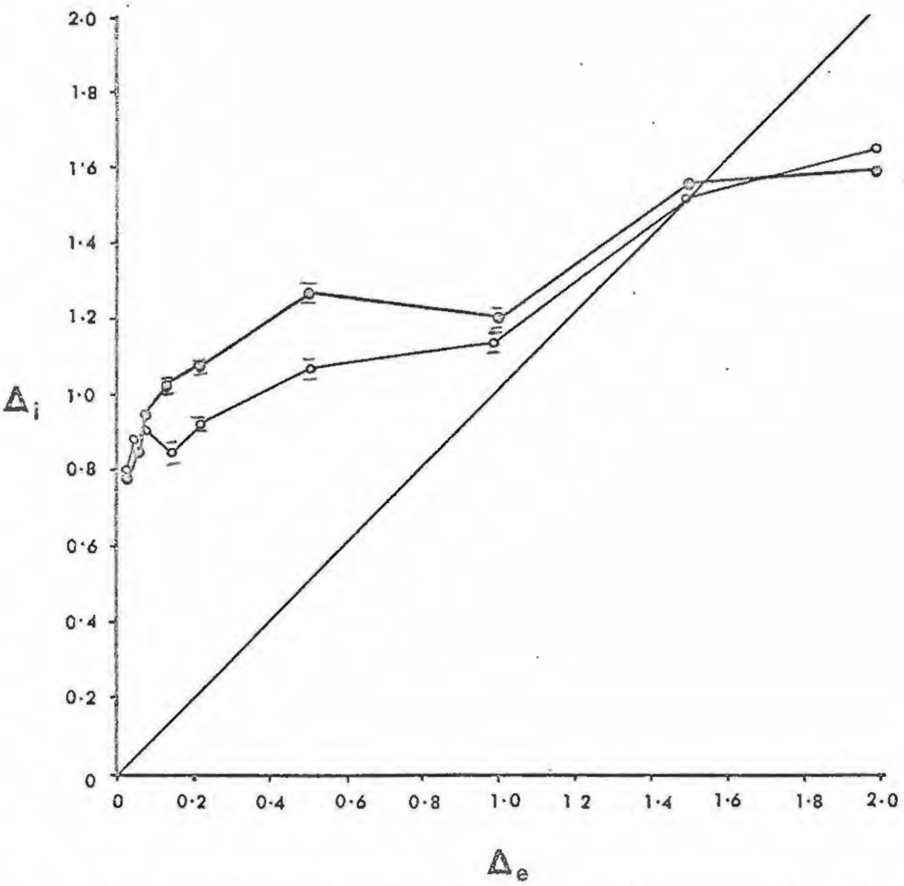


Fig. 22. $\Delta i / \Delta e$ for Grandidierella lignorum bled 5 hours (closed circles,) and 120 hours (open circles) after exposure to dilutions from 100% sea water. Bars on the points represent standard error of the means of 5 animals and 10 animals (closed and open circles respectively).

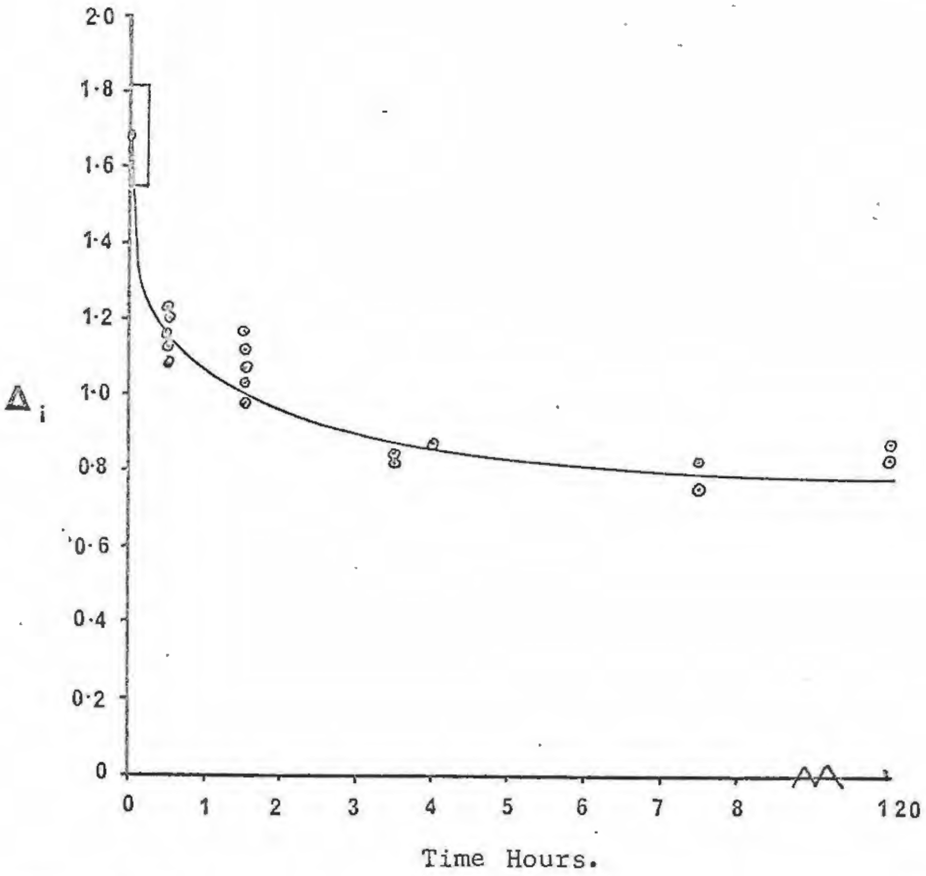


Fig. 21. The drop of Δ i in animals directly transferred from 35% salinity to pond water with respect to time. Equilibrium is complete by 5 hours exposure. The point between 1.8 and 1.6 represents the mean of 10 animals; the line and bars indicate the standard deviation of the mean.

and appears to have reached stable conditions after about 5 hours exposure to very low dilution (Fig. 21). However, in the middle ranges (Fig. 22) of 25% sea water, statistically significant differences were found between animals exposed for 5 hours, and 5 days (at 25% < 0.01 using Fisher's method for comparing small uncorrelated samples.) The measurements of depression of freezing point of the blood for G. lignorum exposed for 5 days has been used as a standard in the subsequent analysis.

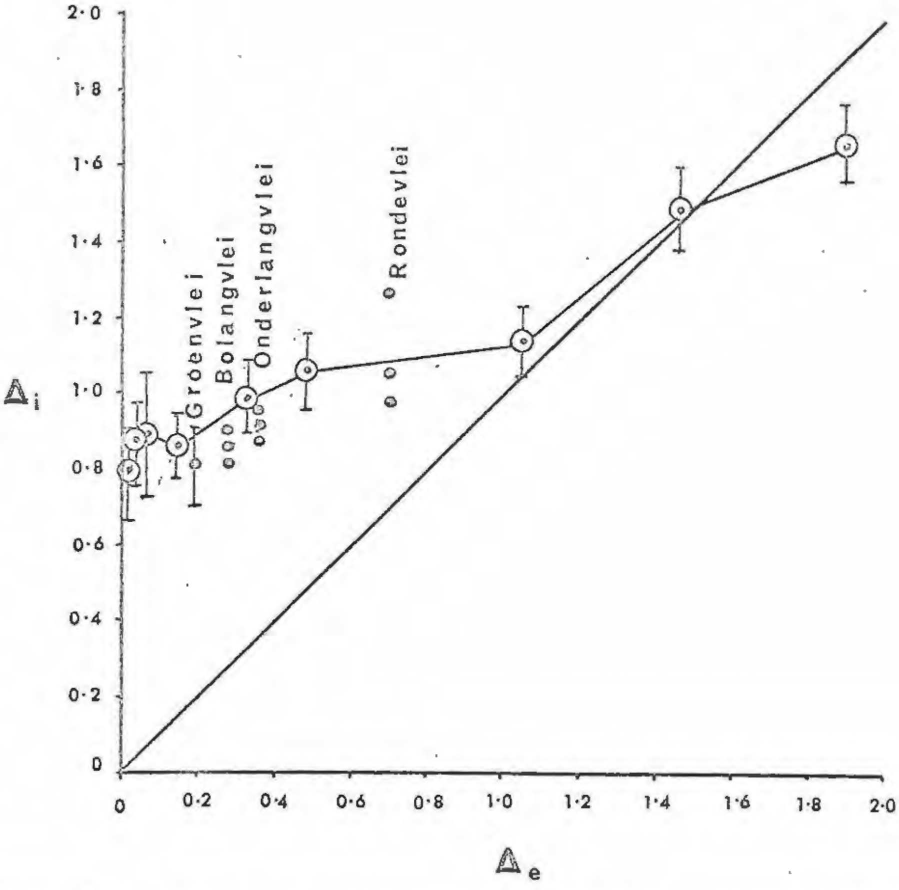


Fig. 23. $\Delta i / \Delta e$ determinations for G. lignorum from Kowie (open circles) in various dilutions as compared with animals from other localities in their natural waters (closed circles). Bars represent the standard deviation around the mean of determinations on 10 individuals.

The degree of regulation of the internal environment varies with respect to the external environment. At high salinities the blood is slightly hypotonic, whereas at low salinities it is hypertonic (Fig.23). Between 5% and 75% dilution of the external environment the blood is practically isosmotic. Below 50%, strong regulation takes place, and at very low dilutions the blood is lowered to about half the concentration found in animals from full sea water.

Figure 23 also shows that the blood concentrations of animals living in naturally low concentrations do not differ markedly from G. lignorum taken from 100% sea water in the Kowie estuary, and exposed to dilutions.

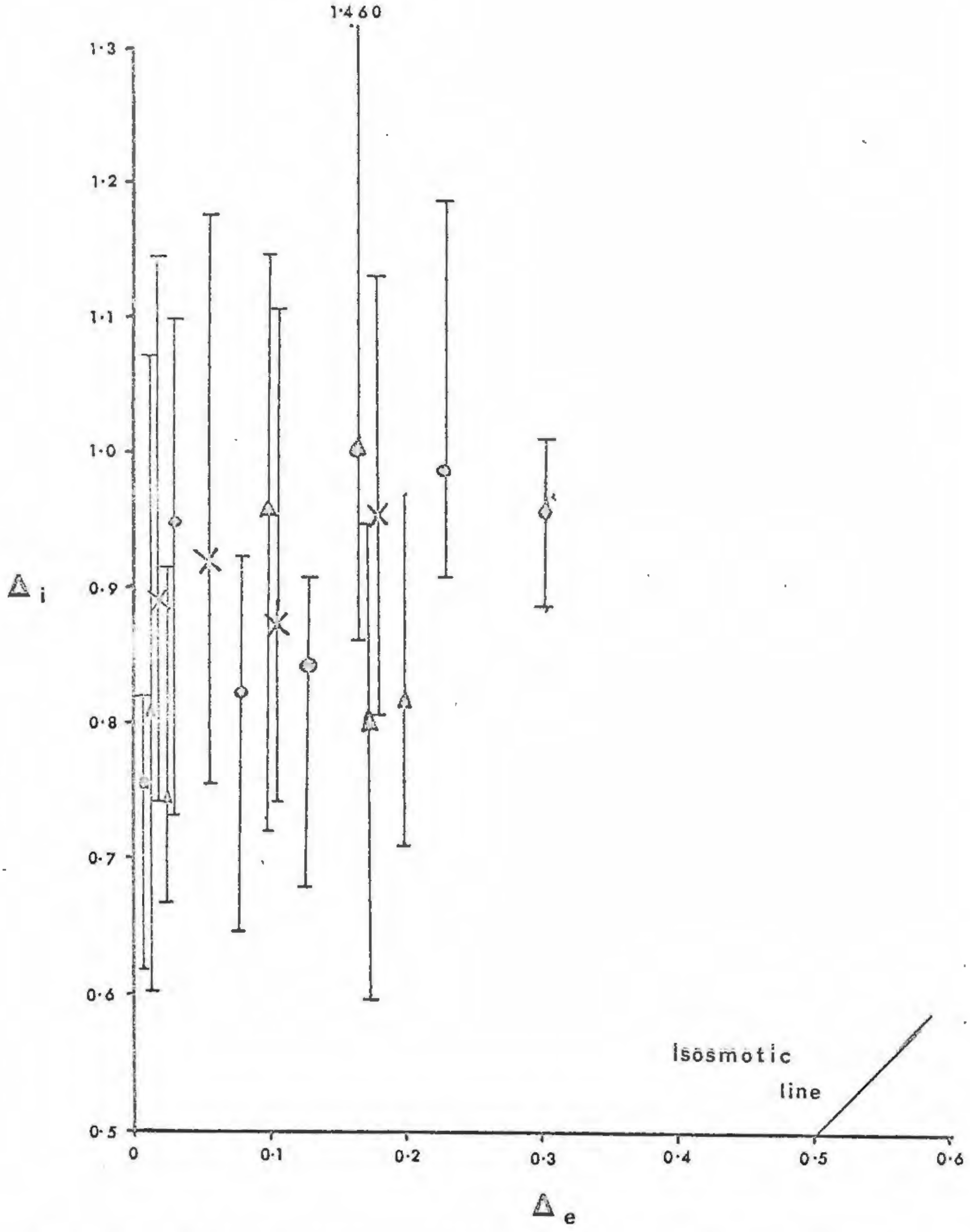


Fig. 25. Internal osmotic pressures of G. lignorum from various localities of different low salt concentrations.

- △ Groenvlei mean and range of 10 animals
- × Kowie mean and range of 10 animals
- ▨ Lake Sibayi mean and range of 5 animals
- ● Bolangvlei and Onderlangvlei mean and range of 5 animals.

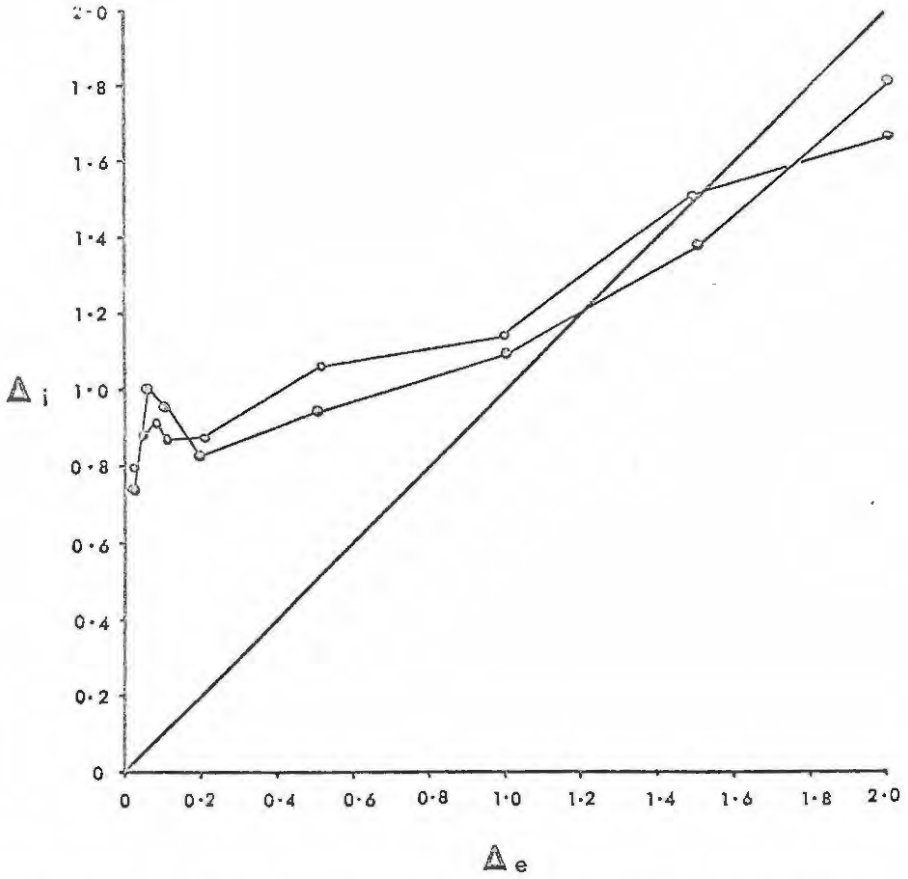


Fig. 24. Δi/ Δe curves for G. lignorum from Kowie estuary (open circles) in dilutions, and animals from Groenvlei in increased concentrations (closed circles). Both sets of data represent the mean of readings from 10 animals in each concentration.

The effect of increasing concentrations of salts on animals normally from low salinity environments was compared with the effects of dilution on the animals from Kowie estuary (Fig. 24). G. lignorum from Groenvlei, when placed in increased concentrations relative to their natural environment show blood concentrations slightly below the Kowie estuary material, when placed in dilutions from 10% to 75% sea water. Below 10% dilutions, there is much variation. This would seem to indicate that in middle ranges of dilution exact control of internal blood concentrations is of little importance, and only at lower concentrations there may be attempts to keep the internal concentration as high as possible.

Figure 25 shows the results of measurements of internal blood concentration of G. lignorum from a number of localities with varying natural salinities when placed in dilutions of water below 10% sea water. At these low concentrations, the means and the ranges of the readings have a large degree of variation and form no discernable pattern. These results suggest that in the populations of G. lignorum thus far studied, there is considerable variation in the control of internal concentrations and that the osmoregulatory mechanism is similar. Thus there is no evidence for physiological races.

Discussion of salinity responses.

The ability of G. lignorum to withstand transfer from high salinities to fresh water and vice versa for at least a week has been demonstrated. It has not been possible to demonstrate that these animals maintain themselves indefinitely and reproduce at low salinities under laboratory conditions. No method of reliably feeding the animals was discovered, and hence the experiments were performed on starving animals. The animals were maintained in clean boxes of water for as much as 40 days, and in one extreme case Kowie estuary material survived without substrate for as long as 120 days. The only source of food in the boxes was the corpses of dead animals which were quickly discovered and eaten, and the bacterial film which must have grown on the sides of the boxes. In the case of the Kowie material which survived for 120 days, of the original 20 animals, 3 animals remaining alive after 36 days were transferred to algal rich pond water. The cells of the algae were seen to pass through the gut of the animals, apparently unchanged. These three succumbed by the 120th day. The demands on the food reserves of G. lignorum were underlined by the observation that after a week in a substrateless environment, the white fat deposits which can clearly be seen beneath the exoskeleton had noticeably decreased. Thus starvation appears to be a severely limiting factor in this type of survival experiment.

The observation that *G. lignorum*, originally from a high salinity environment can moult in freshwater successfully, is important, especially since Hill (1968) has shown that the mud prawn *Upogebia africana* avoids moulting if possible in low dilutions by delaying the time to moult, and succumbs during moult if this is inevitable. This is to be expected since large volumes of water are taken in by crustacea during the moult, so as to expand the new exoskeleton. Thus the ability of *G. lignorum* to moult in freshwater indicates that these animals have the physiological capabilities of successfully coping with low salinities.

The existence of physiological races has been shown by Reigel (1959) working on the isopod *Gnorisphaeroma oregensis*. The two physiological races could be recognised with respect to their changes of Δi in response to exposure to different salinities. The freshwater form did not allow the Δi to drop below a concentration equivalent to 50% sea water. (Δi 1.0°C). An intermediate form from the estuary was also recognised, but this was more nearly like the freshwater form in its adjustment to internal concentrations on exposure to low dilutions. Menzies (according to Reigel 1959) had already subdivided the species into two subspecies and had shown that the survival of the sea water form was poor in freshwater.

Suttcliffe and Shaw (1968) have described two physiological races of *Gammarus duebeni* from England and Ireland. Survival of these two races in very dilute waters is different. The mechanism of the control of sodium uptake system in the Irish form allows for the balance of sodium uptake and sodium loss at minimum natural concentrations of 0.27 mM/l NaCl whilst the English form, which is found in streams with greater than 1 mM/l NaCl concentration, can only achieve sodium balance at between 0.17 mM/l and 0.21 mM/l NaCl for a short time. These differences are not reflected in the (internal) blood concentration levels which are the same for both races.

Details of the sodium transport mechanism and sodium losses from *Grandidierella lignorum* have not been carried out. The fact that the blood concentrations of *G. lignorum* from Lake Sibayi and from Kowie estuary, when placed in the same external concentrations have indistinguishable Δi values, and cannot be used as a criterion for checking the possibility of two physiological races. However, it has been established that Kowie estuary material can survive and moult in water of low dilutions, comparable to Sibayi water concentrations. There is thus no reason to believe that two physiological races do exist between Lake Sibayi material and Kowie estuary material.

Sutcliffe (1968) in his paper on the adaptations of *Gammarus* species to freshwater has analysed the mechanisms involved. In the series of animals from *Marinogammarus finmarchicus* able to regulate only down to 20% sea water for short periods, through brackwater species *Gammarus duebeni*, *G. tigrinus* and *G. zaddachi* to the freshwater species *G. pulex* and *G. lacustris*, three aspects to regulation were found: the control of the loss of Na^+ ions by increased impermeability, and reduction in loss from urine; the increased efficiency of the sodium transport mechanism; and finally the lowering of the internal concentration of the blood. The marine species have a high permeability and tend to regulate immediately when placed in dilutions, thus maintaining a high blood concentration. Loss of ions through the body wall and the urine is countered by the sodium transport system operating maximally with an efficiency such that it is only saturated at something of the order of 60 to 100 mM/l NaCl external concentration. Although *Marinogammarus* is able to maintain itself in 20% sea water, it probably cannot achieve this indefinitely (Sutcliffe 1968).

The brackwater species begin to regulate only at about

50% sea water dilutions (Beadle and Cragg 1940). The sodium loss is reduced by virtue of an increased impermeability of the body and, at very low concentrations, by the retention of sodium ions from the urine. The sodium uptake mechanism is only saturated at 10 mM/1 NaCl in G. zadachi and G. tigrinus and at 1 - 2 Mm/1 NaCl in G. duebeni. Thus there is a greater affinity for sodium ions and greater intake of sodium to compensate for the loss of ions (Sutcliffe 1968). However, the blood concentration is maintained at a high level of about 1.1 Δi °C (Beadle & Cragg 1940, Sutcliffe and Shaw 1968, Sutcliffe 1968).

The freshwater species in common with the brackwater species have high impermeability, control of sodium loss in the urine and efficient sodium transport mechanisms. They differ in having a low blood concentration, ± 0.5 Δi (Beadle & Cragg 1940), relative to the brackwater species. The concentration of ions required to maintain the low internal concentration is less, and therefore these animals can and do live in very dilute environments.

An essential difference between the brackwater Gammarus species and the freshwater species is the difference found in the blood concentration levels, since this determines the amount of ions required to be transported into the animals. The blood concentration of Grandidierella lignorum has a Δi °C of 0.75°C and lies between the values for the brackwater species (Δi about 1.1°C G. duebeni) and the freshwater species (Δi about 0.5°C G. pulex). Furthermore, the lowest concentration of water in which G. lignorum has been found (Lake Sibayi) has a NaCl concentration of about 3.7 mM/1. (Calculated from the mg concentrations for Na^+, and Cl^- for Lake Sibayi given by Allanson & van Wyk 1969). This would indicate that the amount of sodium required to maintain internal concentration balance in G. lignorum is less than for the brackwater Gammarus species, since the Δi is lower the sodium transport system may not be required to work at maximum capacity. Similarly, since the external concentration of sodium is much higher in Lake Sibayi compared with that to which the Irish G. duebeni are exposed, the transport system of G. lignorum is able to cope quite adequately with the intake of sodium ions needed to balance loss. The concentrations of salt in Lake Sibayi do not seem to require the special improvement of salt balance control mechanism required by Irish G. duebeni as compared with English G. duebeni in the very low salt concentrations of the Irish waters.

It is significant that G. lignorum has not been found in the truly freshwaters above the heads of the estuaries of the Eastern Cape Province. In such a situation sodium concentrations may easily drop to values where G. lignorum, like English G. duebeni, can survive for a period, but cannot maintain themselves since the sodium transport system may be strained to maximum capacity. This in itself may suggest that only a single physiological race of G. lignorum is established in the estuaries and the coastal lakes in South Africa.

It has therefore seemed a reasonable assumption that the animals from Lake Sibayi are physiologically identical to those from the Kowie estuary, and that other physiological responses shown by the Kowie population may be reasonably expected to occur in the Sibayi population.

Laboratory studies on G. lignorum to various conditions.

Having established that there are no obvious reasons to believe that Kowie estuary G. lignorum would behave abnormally as compared with amphipods from Lake Sibayi, investigations into the responses of G. lignorum from the Kowie estuary to a variety of factors which may operate in determining the distribution of the amphipods in Lake Sibayi were attempted. Each factor has been dealt with under separate headings.

1: Substrate choices in G. lignorum

The results of the field survey of substrates is reported previously (Lake Sibayi, Boltt 1969, p. 252, Lake Nhlange p. 253). The substrate of Lake Sibayi falls into two clear categories, sand and fine silt. Evidence was presented showing that fine silt substrates tended to be barren of animals. It was also shown that the percentage fine silt in the fine sand substrate did not appear to have an effect on the distribution of the animals in the substrate (Boltt 1969, p. 265). Nevertheless these points were checked in the laboratory, especially in an attempt to establish the importance or otherwise of silt in the substrate.

Materials and methods

Animals were obtained from the Kowie Estuary. Choice chambers were constructed from shallow plastic trays, 33 cm by 26 cm by 4 cm. The tray was partitioned in half by a high wall of plasticine and each side was then separated into 7 compartments by low ridges of plasticine about 1 cm high. Into each compartment a separate grade of sand particle was placed up to the edge of the plasticine wall. Sands of different grain size was initially obtained from the substrate in which the animals were found in the Kowie River East Lagoon. However, this sand is very deficient in grain sizes of both larger and small diameter. Sand from a local river was used to complete the size range for use in the choice chambers. The sands were aged for 24 hrs in water from the lagoon since Meadows (1964 and 1967) has shown the importance of fauna on the particles in choice experiments. The silt particle size came from the Kowie lagoon substrate.

Results

The results of the experiments are tabulated in Table 16

Table 16

Numbers of G. lignorum in substrates of different grain sizes. The animals overwhelmingly chose the fine silt substrate (51.7% p= < 0.02). There is no significant difference between the highest percentage and the lowest in the other grades of sand (cf C and D where p = < 0.2).

Trial No.	1	2	3	4	5	6	7	8	9	10	Total	%
Particle size mm												
A -210	3	4	5	6	0	0	0	1	5	7	31	6.5
B 0.5-2.0	0	2	1	0	1	12	4	9	3	7	39	8.2
C 0.25-0.5	1	1	5	4	14	4	2	8	9	2	50	10.6
D 0.125-0.25	1	1	7	1	3	2	2	2	1	2	22	4.7
E 0.074-0.125	1	3	4	3	14	2	8	1	1	2	39	8.2
F 0.040-0.725	2	3	5	10	1	14	4	4	1	3	47	9.9
G 0.040-	19	13	13	27	24	28	26	27	36	31	244	51.7
Totals	27	27	40	51	57	62	46	52	56	54	472	

Note: Trials 1-4 inclusive were arranged with particle sizes in sequence, whereas trials 5 to 10 were arranged with particle sizes randomised by drawing numbers from a hat.

From the table it is clear that the greatest choice was for the fine silt material. A simple 't' test between sand size G and G gave a probability greater than 0.02. A similar test between C and D, representing the greatest difference in choice between grain size other than the fine silt material, was not significant with a probability of less than 0.2 in Fisher's tables.

Discussion

These results are very interesting especially when regard is paid to the method in which G. lignorum usually builds its tubes in the field. The tubes in which the animals live are constructed from sand particles cemented together from secretions from glands housed in the pair of legs immediately behind the gnathopods. The burrow is then often lined with fine material. The behaviour as seen in the laboratory is as follows. The first pair of antennae are used to collect sand grains. These are passed to the mouthparts where they are worked over in a scrubbing motion much as has been described for Bathyporeia pilosa and B. sarsi (Nicolaisen &

Kanneworff 1969) although these latter animals do not construct a tube. The particles collected by G. lignorum are then fitted to the mouth of the tube and gummed into place by the secretions from the legs as they move back and forth as though the animal were "sewing" the grain into place.

G. lignorum may be induced to use artificial tubes. The animals were offered short lengths of glass capillary tubing of a suitable diameter. In order to get the animals to use glass tubes, animals are put into a container with no sand and having a selection of tubes of differing diameter and about 1 cm long. Within a short while the animals will have taken up residence within the tube. If a number of animals in glass tubes is returned to a sandy substrate, a small percentage of animals will retain the glass tube and continue building the burrow from the mouth of the glass tube. The glass tubing is not wholly acceptable since, being inflexible, unlike the sand tube, the animal may often have difficulty in turning around.

In glass burrows the behaviour for lining is easily observed. Apart from scrubbing of sand grains, G. lignorum also filter feeds by straining out the suspended material in the water. The antennae and gnathopods are held at the mouth of the tube so as to form a net of setae from these appendages. A current is set up through the tube with the pleopods and fine material is filtered from the water coming into the net. The material is transferred to the mouthparts and eaten. Passage of material through the gut is rapid. Carmine particles fed to the animal in this way appeared in the faeces 20 minutes after ingestion. At defaecation the animals turn the abdomen forward in the tube and collects the faeces in the gnathopods and mouthparts as it is extruded from the anus. The faecal pellet is then re-eaten. After a certain number of passages through the gut the animal then collects the faecal pellet and plasters it onto the wall of the tube. Some time

Fig. 26. Perspex pressure chamber used to subject *G. lignorum* to pressure.

later the faecal pellet may be removed from the wall of the tube and reprocessed through the gut.

Although the main fabric of the tubes is often built of sand grains rather than silt, the animals are perfectly capable of using silt. Loose material is bound together with sticky secretion and moulded into place. Given a single choice of either grains or silt, the animals construct the tube without difficulty from fine material. In all probability the choice of silt in the experiments cited is not so much a choice of particle size as of material on the particles for food. The evidence suggests that G. lignorum is not dependent on the particle size of the substrate for determination of distribution but rather on other factors in the bottom of the lake. This accords well with the results obtained for sandy type substrates where the percentage of silt in the substrate did not appear to influence the distribution of animals in the substrate (Boltt 1969, p. 265). It does not explain the absence of animals from the great majority of silt substrates (Boltt 1969, p. 261). The only interpretation permissable at this stage is that conditions of the water close to the substrate are important rather than the material of the substrate itself.

2: Pressure

It has already been suggested that pressure may conceivably form a barrier to the penetration of G. lignorum to depths of greater than 25 m in Lake Sibayi (see Boltt 1969, p. 266). In order to test this possibility a pressure chamber was constructed (Fig. 26). The main body of the chamber was turned out of laminated perspex. A removable lid was retained by six brass screws. Since O rings of a suitable diameter were not readily available, a channel was cut on the face of the main body of the chamber. This was then used as a trough in which liquid rubber was built up and allowed to dry. When the rubber ring so constructed was a few millimetres above the shoulder of the groove, it formed an excellent seal between the face plate and the body of the main chamber. Pressure was measured by a Smith oil pressure guage connected by standard fittings through a narrow bore copper tube to a narrow bore channel leading into the chamber. Pressure was introduced through a needle valve attached to a side arm on the chamber. The side arm formed a dead air space when the volume of displaced water was pushed into the guage. The chamber could thus be kept full of water at all times. The needle valve was attached to the laboratory compressed air system for pressurisation by means of rubber pressure tubing. Good seals were obtained between the brass fittings and perspex with Araldite epoxy cement.

Twenty animals subjected to pressure of 4.9 kg cm^2 did not come out of their tubes over a period of 6 hours. This pressure represents a depth of approximately 48 metres depth, a much greater depth than that encountered by the animals in the bottom of the lake.

There seems to be no a priori reason why the animals should react to pressure save that amphipods have been demonstrated to react to small changes of pressure which they use as a signal for migration with the tides (Enright 1966).

3: Light orientation

Grandidierella lignorum normally lives in tubes in the sand, and at least during the day does not venture far from the tube although it will stretch out to gather sand grains from around the tube to scrub with the mouthparts for food. Animals maintained in covered polythene containers in their natural substrate in the laboratory, had to be vigorously aerated. If this aeration broke down, and the container became stagnant the animals abandoned their burrows and collected on the side of the container closest to the light. Thus the animals showed a strong photopositive response to light when "unfavourable" conditions occurred. Four questions

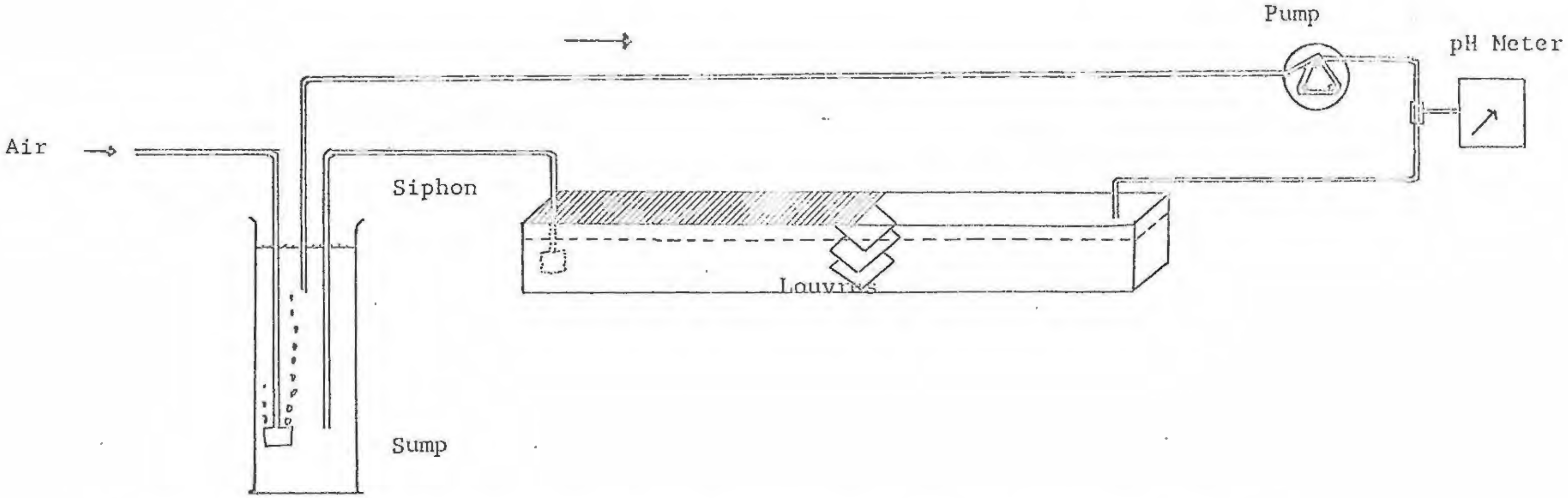


Fig. 27 Choice chamber for light and dark experiments on G. lignorum. The hatched portion of the chamber was covered. The louvres cut down light entering from the open region of the chamber.
The sump and aeration were added in later experiments (see text.).

are relevant: A. Are the animals normally phtonegative in response in good conditions? B. Will the animals migrate into dark conditions to burrow? C. Will the animals remain in dim or totally dark conditions in the burrow? D. What constitutes an unfavourable condition?

A consideration of D is left to later sections on the responses of G. lignorum to gas concentrations in the environmental water. The other questions were answered by some simple experiments with light in choice chambers.

i.) The response of G. lignorum to deteriorating conditions.

A long trough 90 cm by 10 cm was built of wood and lined with polythene. One half was painted black, and the other white. Substrate from Kowie estuary containing its natural fauna was placed along the length of the box to an even depth of approximately 0.5 cm. Water over the substrate was circulated by means of a peristaltic pump taking out water from one end, and returning it to the other. The black half of the box was covered with a board and light proof material. A light baffle or louvre was placed at the junction of the light and dark portions of the box. The louvre had the slats widely placed so as not to impede the movement of animals from one portion of the choice chamber to the other. A 100 watt light bulb was placed over the light half of the chamber to provide illumination. Measured values for the light in the light side were c. 300 ft ca. and in the dark half away from the louvre c. 0.35 ft ca. Just behind the louvre the light value was c. 15 ft ca. and half way along the covered part of the choice chamber c. 0.45 ft ca. The light values were measured with a light meter specially constructed for the purpose (see Appendix II).

In spite of the slowly circulating water, conditions deteriorated as evidenced by the peculiar "estuarine" odour of the water. It is believed that this comes from the bacterial populations on the sand grains. After 24 hours the animals were extracted from the sand in each half of the choice chamber and counted. The results are presented in Table 16.

Table 16

Numbers of amphipods recovered from substrate spread evenly in the dark/light trough in each of the four quarters. The light values for the four quarters were the same as in the previous experiment. The quarters in the box are numbered 1, 2, 3, 4. Quarters 1 and 2 were covered and are therefore "Dark".

	Dark		Light		
Animal species	1	2	3	4	Total
G. lignorum	18(3.4%)	72(13.4%)	318(61.2%)	111(21.3%)	519
C. triaenonyx	42(8.3%)	117(23.2%)	258(51.2%)	87(17.3%)	504
Urothoë serrulidactylus	25(4.2%)	77(12.8%)	304(50.6%)	195(32.4%)	601

It is clear from the results that the amphipods move away from the dark and into the light showing a photonegative response. Towards the last hours of the experiment many *G. lignorum* were seen swimming at the surface of the light half of the chamber and also on the walls of the chamber.

ii). The normal photonegative response of *G. lignorum*.

The apparatus previously described was set up. Temporary partions of plastic were inserted to give four isolated chambers along the length of the box and into each of these about 70 animals were introduced, but without substrate. The partitions were removed, the dark half covered and the experiment allowed to run for 3 hours. The conditions in the chamber did not deteriorate in this time. The results of counts of animals in the light half of the box and, by substraction from the total population, in the dark half of the box are given in Table 17. The light values in each side of the choice chamber were as in the previous experiment.

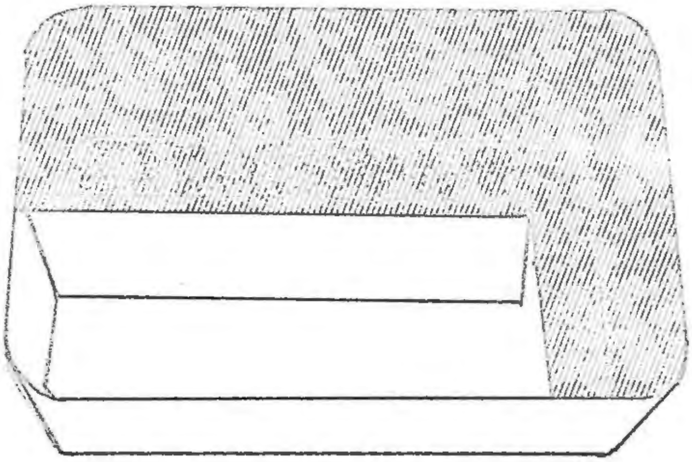


Fig. 28 Diagram of plastic box light/dark choice chamber. Cross hatched area covered with light proof material. Stippled area indicates substrate.

Table 17

Percentages of G. lignorum in dark and light after periods up to 3 hrs in light/dark choice chamber. ± 70 animals put into each of the four quarters of the length of the box initially. The animals were deprived of sand in which to burrow.

	Dark	Light
1 hr	85%(240)*	15%(40)
2 hr	89%(250)	11%(30)
3 hr	86%(241)	13%(39)

* (No. in brackets indicates number of animals in each half of box)

G. lignorum in a substrateless environment tend to move around constantly. If the animals were not behaving photonegatively one would expect them to be evenly distributed throughout the choice chamber. It is therefore clear from the results of this experiment that G. lignorum will move away from the bright light into dim light in a short time in good conditions, thus demonstrating a photonegative behaviour.

Not only will G. lignorum move into dim conditions, but these animals will also build tubes in substrates in practically dark conditions. In order to reduce the amount of light in one side of the choice chamber, an alternative apparatus was used. A 25 cm by 16 cm by 10 cm plastic container was divided by a perspex screen for 4/5 of its length (Fig. 28). A light tight cover was fitted over as shown in the diagram. Ghemically pure sand was placed in the dark half of the container. A temporary partition was placed in between the dark and light portions of the chamber and 30 G. lignorum were introduced into the light section. The partition was removed and six hours later 18 of the 30 animals (60%) had moved into the dim conditions with less than 0.01 ft.ca. and burrowed.

(ii.) G. lignorum remains in the substrate in dim or dark conditions.

The apparatus was once again set up as previously described. However, in the circulation path from the peristaltic pump a tall container was added. Water was siphoned into the container, and this was vigorously aerated. The aerated water was then pumped round to the other end of the choice chamber (Fig. 27). Thus optimal conditions were maintained throughout the duration of the experiment.

In this experiment substrate from the Kowie estuary with amphipods in it was placed in the covered half of the trough. The system was maintained for 36 hours. After 24 hours 19 animals appeared in the light half of the chamber and after 36 hours a further 9 animals appeared (Table 18). The light values for the four quarters of the chamber were as previously reported.

It is clear from this experiment that animals will remain in their tubes in very dim conditions of light.

Table 18

mbers of animals recovered from dark and light side of trough when conditions are not allowed to deteriorate.

	Dark side	Light side
Number	195	28
percentage	87.4%	12.6%

The experiment was repeated using the apparatus figured in Fig. 28. In this apparatus the light values were reduced to less than 0.01 ft Ca. Water was circulated and bubbled as in the vious experiment. The experiment was run for 72 hours. At the end of this time the numbers that had migrated to the light side of the box were 49 consisting mainly of small individuals. One hundred and twenty animals remained in the dark portion of the chamber. It is quite apparent that the animals will continue to remain in their burrows at extremely low light intensities. The appearance of a few animals in the light was due to a general tendency for the animals to spread over the available space. (Table 19).

Table 19

Numbers of animals recovered from dark and light side of choice chamber with very dark conditions, when conditions are not allowed to deteriorate. The animals were placed in the dark portion with the substrate from Kowie. Light values in the dark side of the chamber were less than 0.01 ft Ca., and ± 50 ft Ca. in the light section.

	Dark side	Light side
No. of animals	120	49
Percentage	71%	29%

Discussion

The presence of light by itself does not appear to affect the behaviour of *G. lignorum* in tubes in the substrate. The behaviour of the amphipods in artificial glass burrows appears to be perfectly normal compared with what can be seen of the animals in sand grain burrows. Further, unlike the mud prawn *Upogebia africana* which tends to close up "windows" in burrows built on the side of a glass aquarium, *G. lignorum* will build burrows using glass as portion of the burrow wall. Once in a burrow, the amphipods appear to be indifferent to light, or indeed to the absence of light. Only when the animals are out of burrows do they respond to light. Under optimal conditions they become photonegative. This mechanism will get them back into the substrate at the bottom of the lake. Under unfavourable conditions they become photopositive which will get them into shallower water which is more likely to be favourable. Thus the light responses of *G. lignorum* by themselves will not serve to affect the distribution of animals in the bottom of the lake. Only "unfavourable" conditions appear to get the animals out of their tubes in large numbers. The question of what constitutes an unfavourable condition is therefore of paramount importance. Together with the light responses, some understanding of the reasons for the pattern of distribution might be gained.

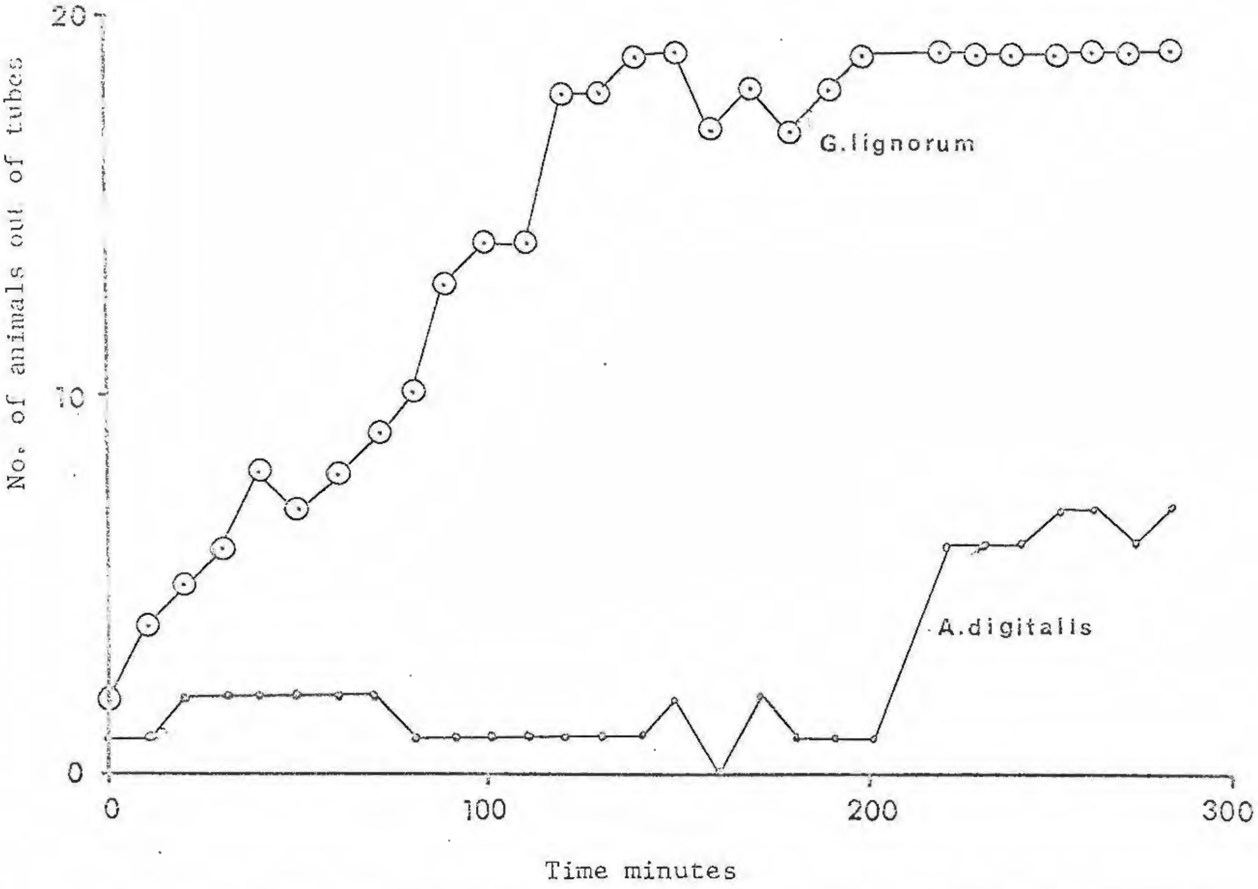


Fig. 29. 20 G. lignorum and 20 A. digitalis were allowed to burrow in acid treated sand in the experimental chamber. At time 0 in the experiment CO_2 was added at the rate of 40 mmHg/hr.

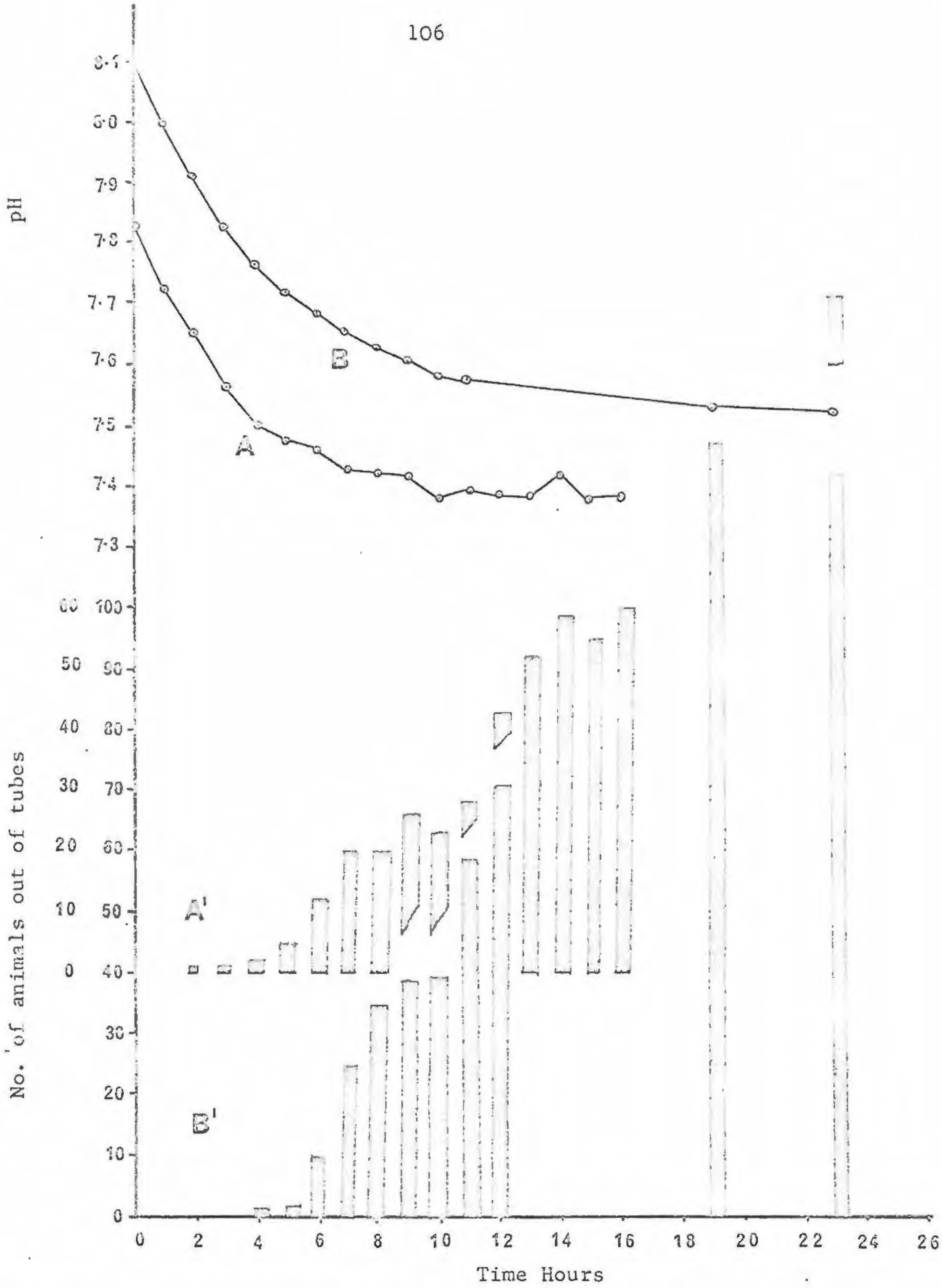


Fig. 30. G. lignorum were kept in a container which was open but not aerated. The drop in pH and the number of animals out of tubes is shown. A and A' Fresh substrate and animals. At the end of the A experiment the container was aerated for 12 hours. B and B' show the course of events when the aeration was turned off for a second time.

4: Behavioural responses to oxygen, carbon dioxide and hydrogen sulphide.

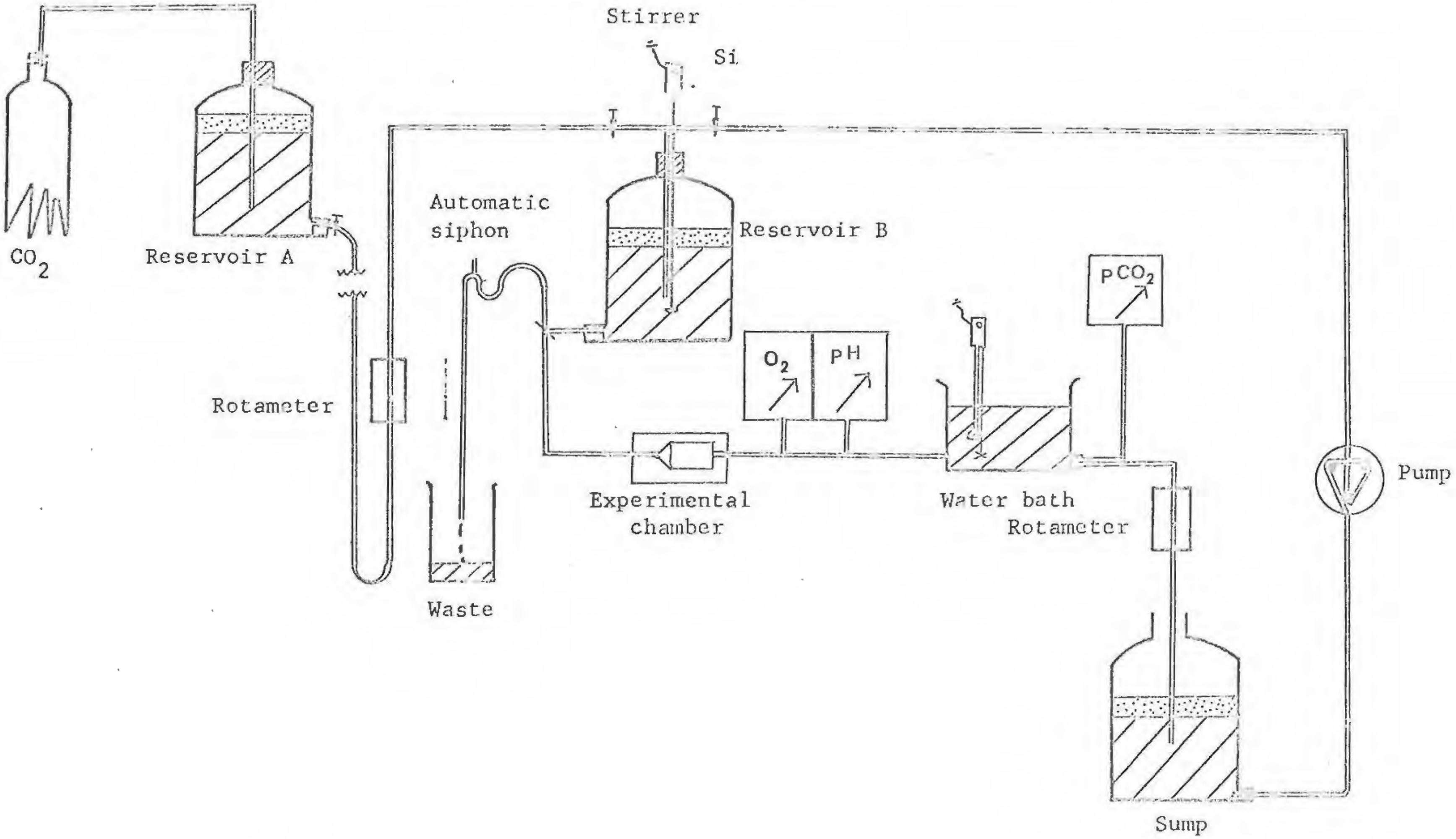
Introduction [illegible] gas problems.

It has been shown in experiments with light responses that certain "unfavourable conditions" reverse the normal photonegative response of *G. lignorum* to a photopositive response. This may well play an important part in the determination of the pattern of distribution of the animal in Lake Sibayi. However, some idea of what constitutes an "unfavourable condition" is necessary.

A priori the most obvious conditions which may affect the light responses are to be associated with metabolic waste products of the benthos and possibly the reduction of O_2 to critical levels for *G. lignorum* which may not affect the other fauna as much. The metabolic wastes of importance are CO_2 and possibly the products of deamination of protein for respiratory purposes. By chance it was discovered that CO_2 induced *G. lignorum* to abandon their tubes. When this effect was compared with the response of *A. digitalis* to a similar introduction of CO_2 to a system, it was found that the latter species does not abandon the substrate until a much higher value of CO_2 concentration was reached (Fig. 29).

The importance of stagnating conditions is shown by the response of the animals in the laboratory. Substrate from the Kowie estuary with its normal fauna, and kept in a container which is not vigorously aerated, stagnates, and after some hours *G. lignorum* abandons the substrate and comes to rest on the sides of the container. During this time the pH value of the water falls (Fig. 30), as does the oxygen concentration. In the experiment reported, after one period of stagnation the system was once again aerated, and then allowed to stagnate again. The difference in the two levels of pH for each period of stagnation in the experiment represents the effect of uptake of carbonate from the sediment.

Fig. 31. Diagram of apparatus used in flow system through the experimental chamber and sensing electrodes.



(This is a well known phenomenon occurring in the bottom of lakes cf. Mortimer 1941/42, Hutchinson 1957, Ruttner 1963). The alkalinity had risen from 2.62 m eq/l at the beginning of the experiment to 5.69 at the end of the experiment. In order to try and separate the [illegible] CO_2 concentration, O_2 lack, and other factors, apparatus was built in which controlled conditions could be obtained.

Methods

In order to allow for experiments lasting sometimes many hours, to ensure the adequate passage of water through the experimental chamber and across the electrodes (O_2 probe required at least 1L/hr), and to give reasonable economy in the quantity of sea water used, a recirculating system was designed. The general arrangement of the apparatus is shown in the diagram in figure 31.

Essentially the water in reservoir B was treated so as to allow for varying gas concentrations. The water was then siphoned through the experimental chamber, across measuring electrodes and flow meter to a sump. The effluent water was then recycled back into the reservoir B.

Great difficulty was initially experienced in controlling the rate of CO_2 added into the water of reservoir B. It was hoped that measured mixtures of gas bubbled through an air stone in the main reservoir would result in either stable quantities of dissolved gas or maintain constant rates of gas increase. This method used by Edwards and Learner (1960) on oxygen consumption experiments on Asellus, proved unreliable since although the quantitites of gas supplied to the air stone could be accurately controlled by rotameters, fluctuating pressures in the laboratory compressed air system, and from the pressure regulator of the CO_2 cylinder did not allow steady states to obtain for any length of time. It was found that better control could be effected by mixing gas saturated water at controlled rates of flow into a standard volume in the main reservoir.

Fig. 32. Regulating valve used to control the flow of CO_2 saturated water from reservoir A to reservoir B.

A detailed description of the various parts of the apparatus follows.

Reservoir A

A 6 litre bottle with a stopcock outlet acted as a reservoir for waters saturated in CO_2. It was discovered that slight pressure changes with saturated CO_2 water tended to evolve bubbles in the flow rate control system and upset it. It was found that the water in reservoir A should be bubbled freely with gas for some hours (at least 2) and then a layer of liquid paraffin was used as an oil seal. Liquid paraffin takes up quantities of gas but allows very slow diffusion of gas to the atmosphere once it has reached equilibrium with the concentration of gas in the water beneath the oil seal. After a week of standing with the bung open to the atmosphere the change in concentration of CO_2 in water under oil was hardly detectable. Consequently, during the run of an experiment, gas was not bubbled through reservoir A.

The water in reservoir A was metered into the main reservoir B through a regulating valve of simple construction. Initial trials with needle valves or a capillary drip feed proved unreliable. The construction of the regulating valve is shown in Fig. 32.

A micrometer screw was used because of the low angle of pitch of the screw allowing for fine control. The amount let through was metered through a Fisher & Porter Company precision bore Flowrator tube No. 02.F 1/8.20.5/36.

Reservoir B

Reservoir B was initially charged with 3 litres of sea water. The surface of this was covered with liquid paraffin to damp the rate of gas loss. The water was stirred with a long stemmed Gallenkamp mechanical stirrer to promote mixing of the return flow and water from reservoir A.

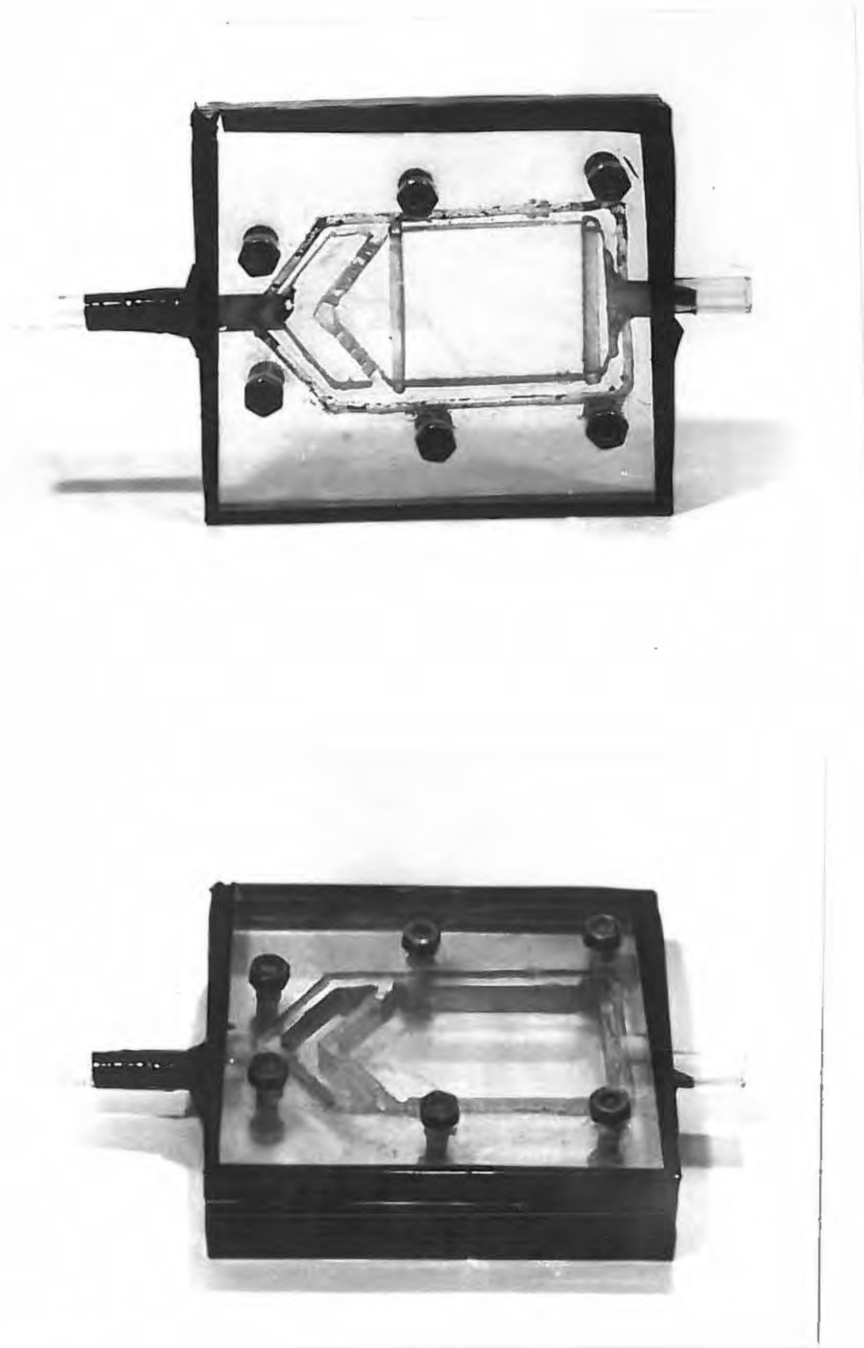


Fig. 33. Two views of experimental chamber. The screens preventing the escape of animals have been omitted in the lower photograph.

Water was led from reservoir B through a threeway stopcock. One arm was led to an automatic siphon which maintained a reasonably constant level in the reservoir. It was found that a large bore threeway stopcock was necessary to ensure that the automatic siphon was not unduly influenced by resistance to flow in the stopcock supplying both itself and the experimental chamber.

Experimental chamber.

An experimental chamber was constructed out of perspex. The inflow opened into an antichamber and thence through a partition with holes drilled at angles to facilitate mixing of water. Screens of nylon mesh were introduced to prevent animals escaping from the chamber. Tests with dyes showed that fluids advanced evenly through the chamber leaving no backwaters of stagnant water in the chamber.

pH Probe

A Phillips Harris model pH meter was connected to an Activon combined glass/reference electrode. The electrode was housed in a perspex chamber designed to allow free flow of water across the glass electrode. The details are seen in Fig. 34.

Oxygen probe.

A Beckmann oxygen analyser was used for estimations of O_2. The probe was housed in a perspex chamber built by Hill (1967).

Carbon Dioxide Probe.

Titration methods have been described for freshwaters by Mackereth (1963) and "Standard methods for the examination of water and water waste" (12th ed. 1965). Mortimer (1941) used Maucha's method (Maucha 1932) and Milburn & Beadle (1960) working in the specialised swamp conditions evolved a conductrimetric method. Titrametric methods have the serious disadvantage in that they are not easy to continuously monitor the changes in concentration of CO_2 in the water being examined. Alause (1968) has demonstrated that there

Fig. 34. The pH probe mounted in a chamber so that water flowed in the bottom around the glass bulb, and out at the top. Ducts from the top of the chamber ensured that bubbles were not trapped.

Fig. 35. pCO_2 probe in water bath with heater stirrer.

is a straight line relationship between dissolved free carbon dioxide and pH for any one fresh water. He has also shown that for waters of different salt concentrations (defférentes salinités) these straight lines representing the relation between the log concentration of CO_2 and pH are parallel for any one alkalinity concentration. Alause claims that the results are more reliable than back-titration methods.

An even more convenient method is available using a Severinhaus electrode (Severinghaus & Bradley 1958). This instrument is routinely used in the measurement of blood CO_2 in man. Gambino (1961) demonstrated that this instrument was very accurate for measuring blood pCO_2, but emphasised the need for temperature control. The applicability of this electrode to other systems is given by Severinghaus (1960) where he advocated the use of it in satellites. In this case he suggested a polythene membrane instead of teflon to stop water vapour loss from the electrode. Polythene membranes render the electrode slow in response. The best membrane is silicone rubber, but was not available. Severinghaus also noted that the electrode operated at low temperatures requires a relatively long time to come into equilibrium with the sample pCO_2.

In order to continuously monitor the pCO_2 concentration in the experiments to be reported, a Severinghaus electrode model 9987100 supplied by National Welding Equipment Company, Medical Division, U.S.A. was employed. The electrode was connected to a Beckmann S-22 Expandomatic pH meter, readable to within .003 pH units on an expanded scale. The sample passing the electrode was warmed to 35°C by passing it through 15 cm of narrow bore nylon tubing in a thermostatically controlled waterbath controlled to 0.1°C by a heater stirrer. The sample was cooled back to the temperature of the constant temperature room on its passage through pipes back to the reservoir. The water bath and electrode is shown in Figure 35.

Calibration.

Calibration of the response of the electrode is normally obtained by passing gas mixtures of accurately known concentrations across the sensitive membrane of the electrode. Gas mixtures of known concentration were not available. Attempts at mixing gas by taking up measured volumes in glass syringes gave very erratic results and were not repeatable in immediately consecutive trials. This method was therefore abandoned. CO_2 was bubbled through sea water for 12 hours so as to allow of saturation. Carefully measured quantities of saturated water were then diluted with CO_2 free water in a glass syringe and passed over the electrode. The saturated water was considered to be in equilibrium with air pressure and to have a partial pressure of 100% of 760 mm Hg CO_2. Neither of these assumptions is strictly true since the barometric pressure fluctuates daily, and slightly supersaturated conditions may have occurred due to increases of water pressure at the bottom of the container being bubbled relative to the free air space above the water. However, in the interests of being able to monitor the CO_2 continuously with the apparatus these inaccuracies were accepted for the present purposes. The pH readings obtained for various percentages of dilution of saturated waters were then plotted on semi log graph paper, thus giving a calibration curve. Calibrations were repeated with most of the experiments although the same calibration curve was used for immediately consecutive experiments on the same day.

Specimen calibration curves are presented in Fig. 36. A line was drawn through the points of the graph by eye. The various calibration curves had different slopes and positions on the graph, indicating drift in the sensitivity of the electrode and possibly the effects of dilution over a long time by water vapour

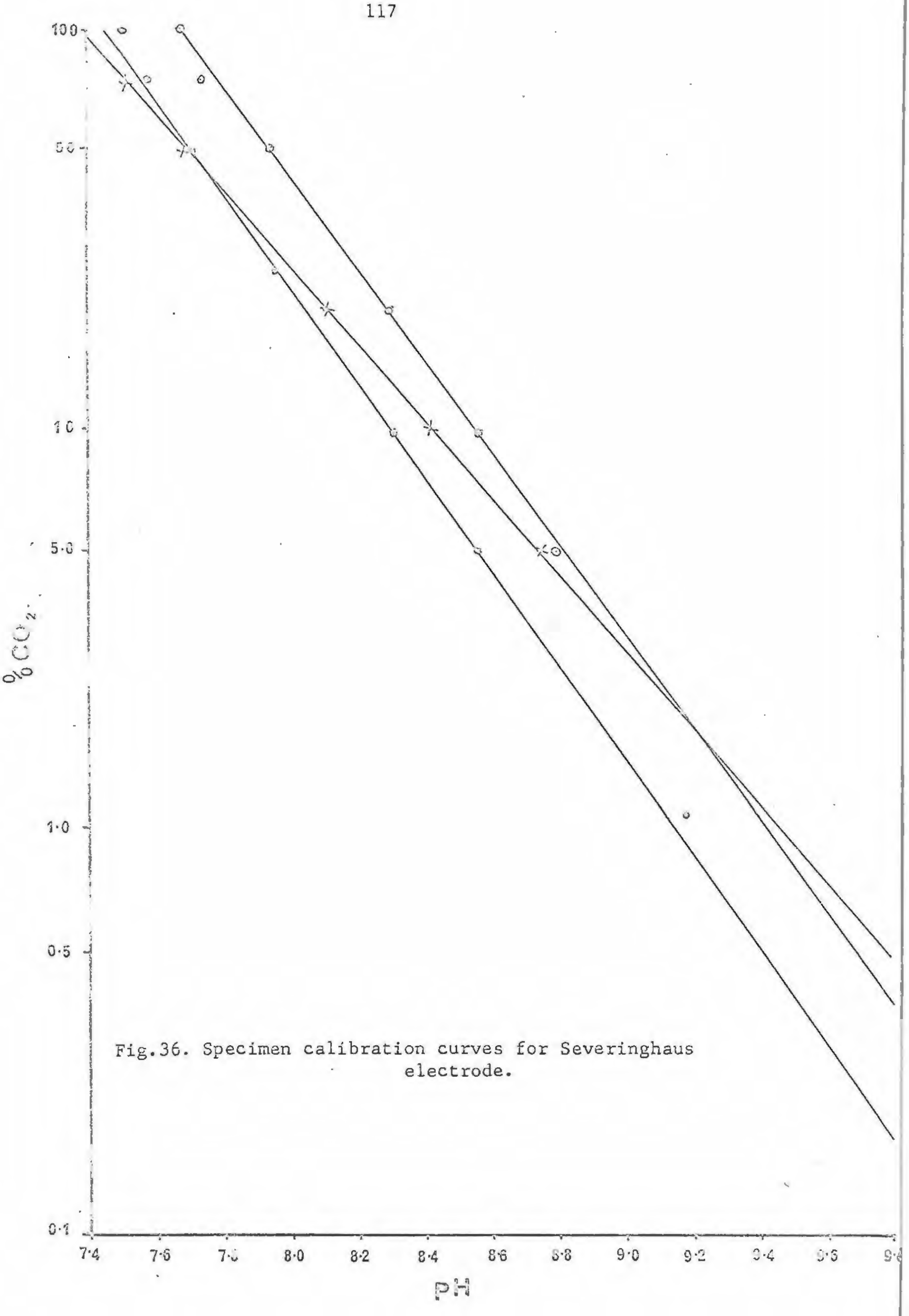


Fig.36. Specimen calibration curves for Severinghaus electrode.

either being added to or removed from the electrode. However, each calibration curve was self consistent in that the expected straight line relationship between the pH value and the log of pCO_2 percentage concentration was obtained. Errors in dilution of the saturated water were shown up by points off the main line of points on the curve and could safely be ignored. Enough points were in line so that the drawing of the curve by eye could only result in small changes of slope. The effect of this would be to shift the percentage CO_2 value by about 2% at the most. Any greater accuracy at this stage was not considered meaningful, since accurately calibrated glassware for dilution was not available. Attempts at mixing saturated and CO_2 free water, using burettes with the water covered with paraffin oil, gave greater scatter than using a syringe. The oil over the fluid in the mixing chamber quickly dissolved CO_2 from the water and upset the nominal concentrations of CO_2. The values of CO_2 therefore must be accepted with reserve, but do give a good indication of the order of magnitude of CO_2 concentration for any one experiment.

Effluent collection and return.

The effluent water from the pCO_2 sensor was piped through a rotameter to ensure that adequate flow of water was going through the system. This is important, especially for the O_2 probe which uses small amounts of oxygen and will record too low if less than about 1 Ltr./hr. passes the probe. The overflow was collected in the sump C (Fig. 31) and the effluent was sealed with a further layer of liquid paraffin. The effluent was then returned to reservoir B via a simple peristaltic pump.

Results.

Preliminary observations on behaviour of the animals used as criteria of distress and movement away from a particular environment.

In early trials for maintaining the animals in the laboratory it became clear that the *G. lignorum*, *C. triaenonyx* and *Urothoë serrulidactylus* swim up into the water away from the substrate as soon as conditions in the maintenance vessel became stagnant. As has been demonstrated, *G. lignorum* move towards the light under these conditions. It was also accidentally demonstrated that CO_2 bubbled through the water acted as an irritant on the animals, provoking a positive light response. The behaviour of the animals was then investigated in glass tubes. *G. lignorum* was placed in the experimental chamber and irrigated with sea water. Normal activities of the animals included periodic ventilation of the tube with the antennae and gnathopods held out to filter material from the inhalent current. The animals turn round in the tube, though infrequently in these conditions. The animals lean out of the entrance to the tube and attempt to gather sand grains which they scrub, as previously described, and then often build these into the fabric of the tube. Much time is spent on scraping the walls of the tube and manipulating the scrapings with the mouthparts. Collection of the faeces from the anus is often seen as is the plastering of exhausted faeces on the walls of the tube. In the glass tubes the animals are often seen moving the second and third legs rhythmically back and forth as though patching up the secretory binding of the walls in the tubes. In glass tubes in which the animals have been maintained for some time, the walls are often lightly coated [illegible] thin film of secretion contaminated with fine particles.

On the introduction of irritant CO_2 water through the experimental chamber the animals react by a rapid beating of the pleopods providing a fast current of water through the tube. If this is ineffectual the animals will turn round in the tube and beat in the opposite direction. This turning and beating is carried on for a short time and eventually the animals abandon the tube and swim towards the light.

Should conditions keep on deteriorating, the animals begin to become uncoordinated and fall to the bottom of the chamber making feeble uncoordinated movements. There are often vigorous cleaning movements of the gills on the bases of the legs in the following characteristic fashion. The abdomen is bent forward and the brushes of setae on the sides of the pleopods are pushed between the legs over the gills.

Cessation of movement begins with the gnathopods and pereiopods and then of the pleopods. At this stage the animals are anaesthetised and can be revived by placing them in fresh sea water. Presently the back of the animal arches and they can no longer be revived.

In the following experiments, abandonment of the tube is used as the criterion for the movement away from stressful conditions. Although G. lignorum uses the glass tubes freely as burrows, the tanaid Apseudes did not and experiments in which Apseudes digitalis were used, either substrate from Kowie East Lagoon was used or chemically pure laboratory sand was used. This latter proved quite satisfactory as a substrate for all the animals. In all other cases, G. lignorum was provided with glass tubes for burrows.

The tolerance of the animals to the chamber was good. There was always a small number of animals out of glass tubes at any one time. This is due to the fact that the tubes are "uncomfortable" unless of exactly the right diameter and a few animals were always looking for a "better fit". The results of a longterm experiment are shown in Table 21. In this experiment, open water surfaces of the apparatus were covered with paraffin oil, but there was no addition of saturated CO_2 water. The animals which were out of tubes were counted at 10 minute intervals during certain periods of the run of the experiment.

Table 21

Periods of time during which the control experiment on the tolerance of *G. lignorum* to the conditions in the experimental chamber was observed. During each noted period of time the animals out of tubes in the chamber was noted as were O_2 levels, pH, and $P.CO_2$. Observations were obtained inclusive for beginning and end times.

No. of Readings	Time in Minutes Beginning of observation period	End of observation period	Mean no. animals out	Max.-Min. Nos.	
11	0	100	2.5	4	2
6	280	330	4.5	6	3
14	530	670	2.3	3	1
3	1320	1340	1.4	3	1
14	1470	1600	1.0	1	1
5	1680	1720	0.8	2	0
15	1850	2000	2.9	4	1
4	3300	3330	2.0	3	1
72					

From the Table it is clear that small numbers of thirty test animals were always out of the tubes. The mean of all the samples counted was 2.3 with a standard deviation of 1.1. The mean plus two standard deviations gave a figure of 4.5, so that the occurrence of six animals out is a rare enough event to ignore and consistent readings of 5 or more have been taken as significant in the following experiments.

The sensitivity of the animals to oxygen lack.

The system figured in Fig. 37 was used for these experiments. The oxygen tension was lowered in reservoir B by bubbling nitrogen through the water. The level of oxygen was monitored by the oxygen probe, as were the pH and the CO levels in the water. In extremely low oxygen conditions of less than 0.5 mg/l in the flowing system, the animals abandoned their tubes progressively with time. 50% of the 30 animals used were out by the time the first animals had ceased to beat their pleopods. This occurred after the first hour of the experiment. The results are given in Fig. 37.

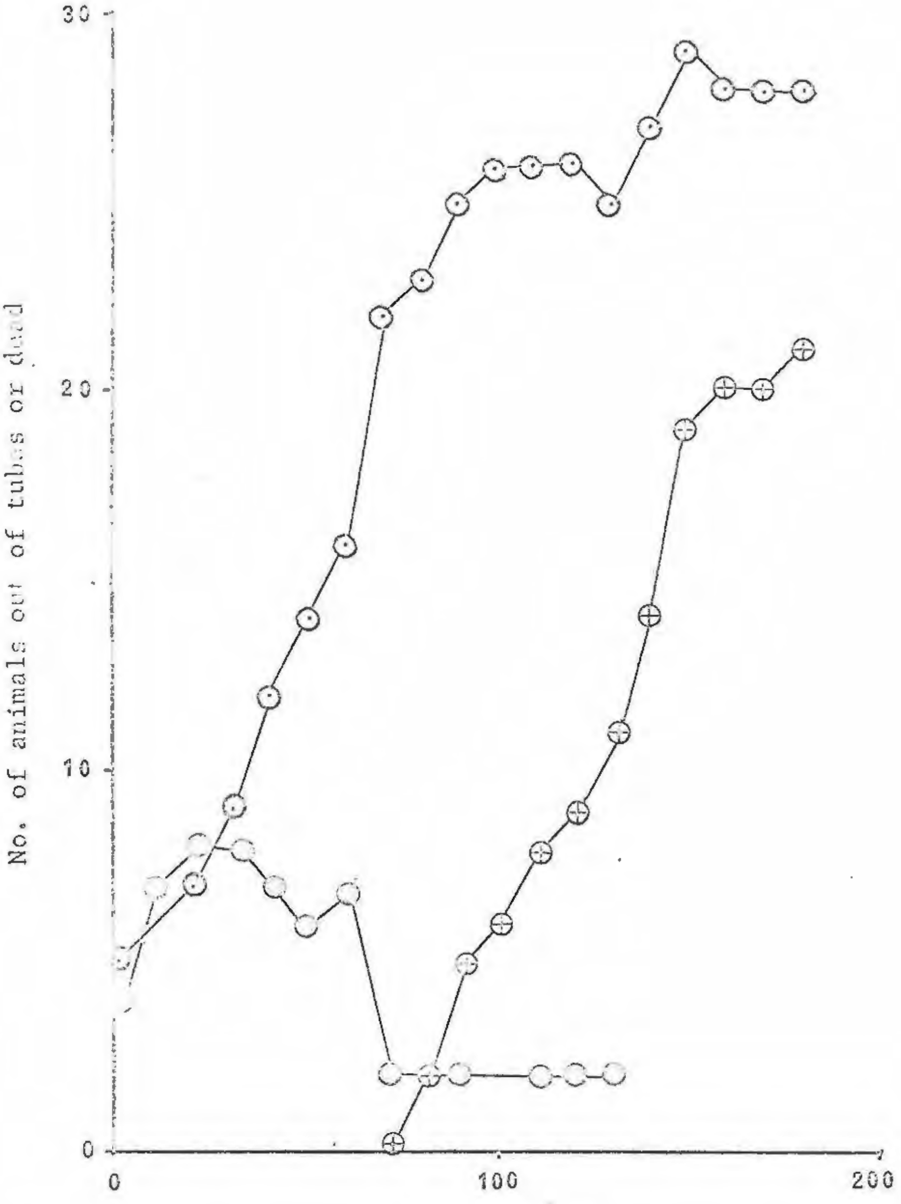


Fig. 37. The effect of two low O_2 tensions (⊙ 0.5 ppm and ○ 1.6-2.3 ppm on the behaviour of *G. lignorum*. Animals which stopped all movement in the 0.5 O_2 ppm concentrations shown as ⊕. No CO_2 was added to the system.

When the oxygen levels were raised to between 1.6 and 2.3 mg/l the animals were unaffected.

Carbon Dioxide.

It has been shown that the addition of CO_2 to the water will cause C. lignorum to abandon its tubes in the substrate. The effect of the addition of CO_2 is to change the pH of the water when aggressive CO_2 is present. The question of whether the hydrogen ion was important, rather than the CO_2, raises itself. In order to study the effects of hydrogen ion without the effects of CO_2, a container was set up with animals established in Kowie estuary substrate. The container was stirred with a mechanical stirrer to allow for mixing, and dilute HCl was dripped in at a slow speed, thus dropping the pH of the water. No animals appeared from their tubes throughout the whole pH drop from 8.3 to 4.5. This is taken as evidence that the animals are reacting to dissolved CO_2 rather than the hydrogen ion.

Initially it was thought that specific static concentrations of CO_2 would cause the animals to abandon their tubes. However, this is not the case since the animals will return to tubes, after having been driven out by an initial introduction of CO_2 into the system. An extreme case of this is illustrated by the results presented in Table 22.

Much more important in stimulating the behaviour of C. lignorum is the rate of change of increasing CO_2 concentrations. This was especially evident when an attempt was made to increase the concentration of CO_2 only very slowly in order that the animals would not be irritated out of their tubes prematurely.

In Figure 38a and b, the course of such an experiment is shown. In this early experiment where the control over the addition

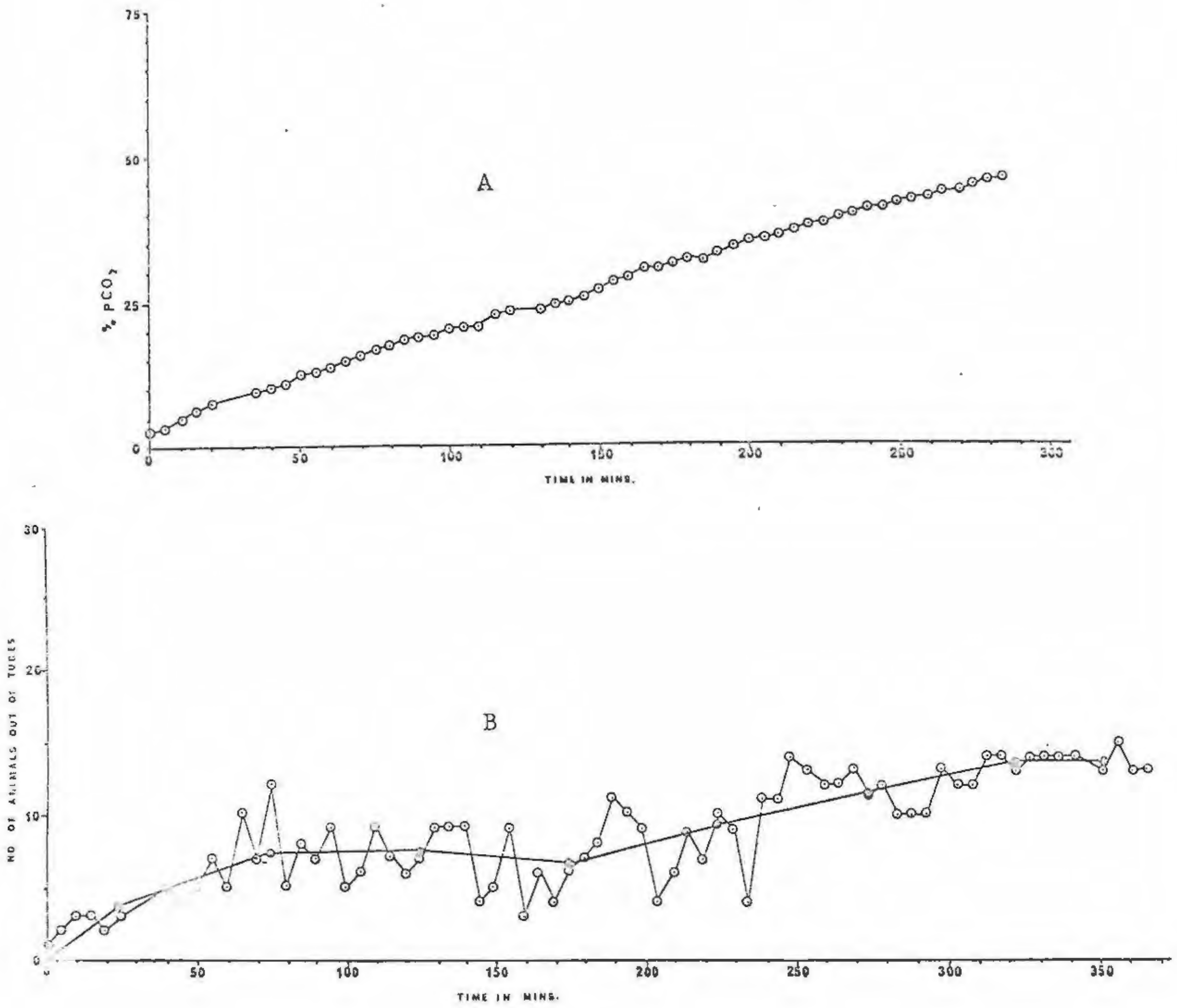


Fig 38 30 *G. lignorum* were placed in the experimental chamber and exposed to very slowly rising pCO$_2$ concentration. A; is a record of the pCO$_2$ concentration. B; is a record of the number of animals out of tubes. (Open circles; numbers of amphipods out at each 5 minute interval. Closed dircles; Numbers of animals out averaged for each 50 minuite period.)

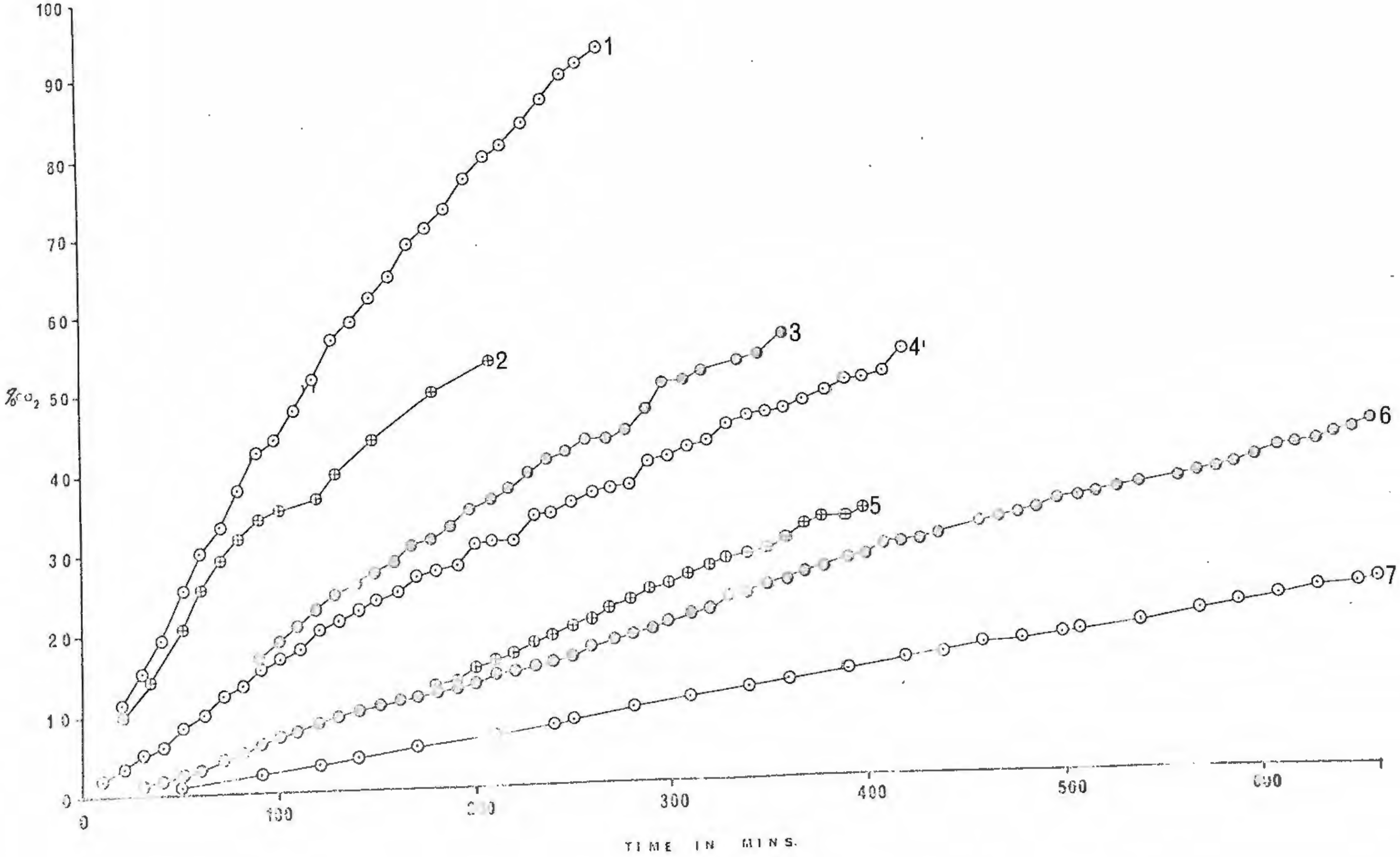
Table 22

The response of G. lignorum when exposed to a high concentration of CO_2 which is kept static, with respect to time. The O_2 tension was 4.8 mg/l and the ° water was at 20°C. The water was loaded with a pCO_2 of 59.8 mm Hg, which dropped the pH of the water to pH 5.75. After 310 minutes the pCO_2 was raised to give a pH in the water of 5.59.

Time mins.	pH Water	Out of tubes	In tubes	Remarks
0	5.75	0	20	
5		20	0	
7		20	0	Some animals show uncoordinated swimming
15		18	2	3 moving only feebly
40		12	8	2 " " "
50		9	11	3 " " "
65		6	14	All recovered.
240		3	17	
310		0	20	
		Increase pCO_2		
314	5.59	20	0	Many showing uncoordination
374		16	4	The same
510		14	6	7 no movement. The rest not moving much.

of CO_2 was very imperfect because of the drip type of feed in the apparatus, great variations in the numbers of animals in and out of tubes during the course of the experiment was recorded. In some cases the numbers of animals out at a particular time could be matched with a transient rapid rate of CO_2 rise. Thus, between the 110th and 120th minute of the experiment, the animals out of tubes dropped from 9 to 6. At this time the pCO_2 rose only 0.5%. Between the 120th and 130th minutes the animals out of tubes rose again to 9 and the pCO_2 rose 2.5%. Between the 130th and 140th minute the animals out of tubes remained at 9, and the pCO_2 concentration rose up 1.5% and returned to its original value again. These data must be interpreted in the light of the finding

Fig. 39 Records of the rates of pCO_2 increase plotted as a percentage of 100% saturation of CO_2 at a pressure of 1 atmosphere at 20 °C. Note occaisional irregularities in the curves, and the curvilinear shape. Where overlap of lines occurs one has been left out for clarity.



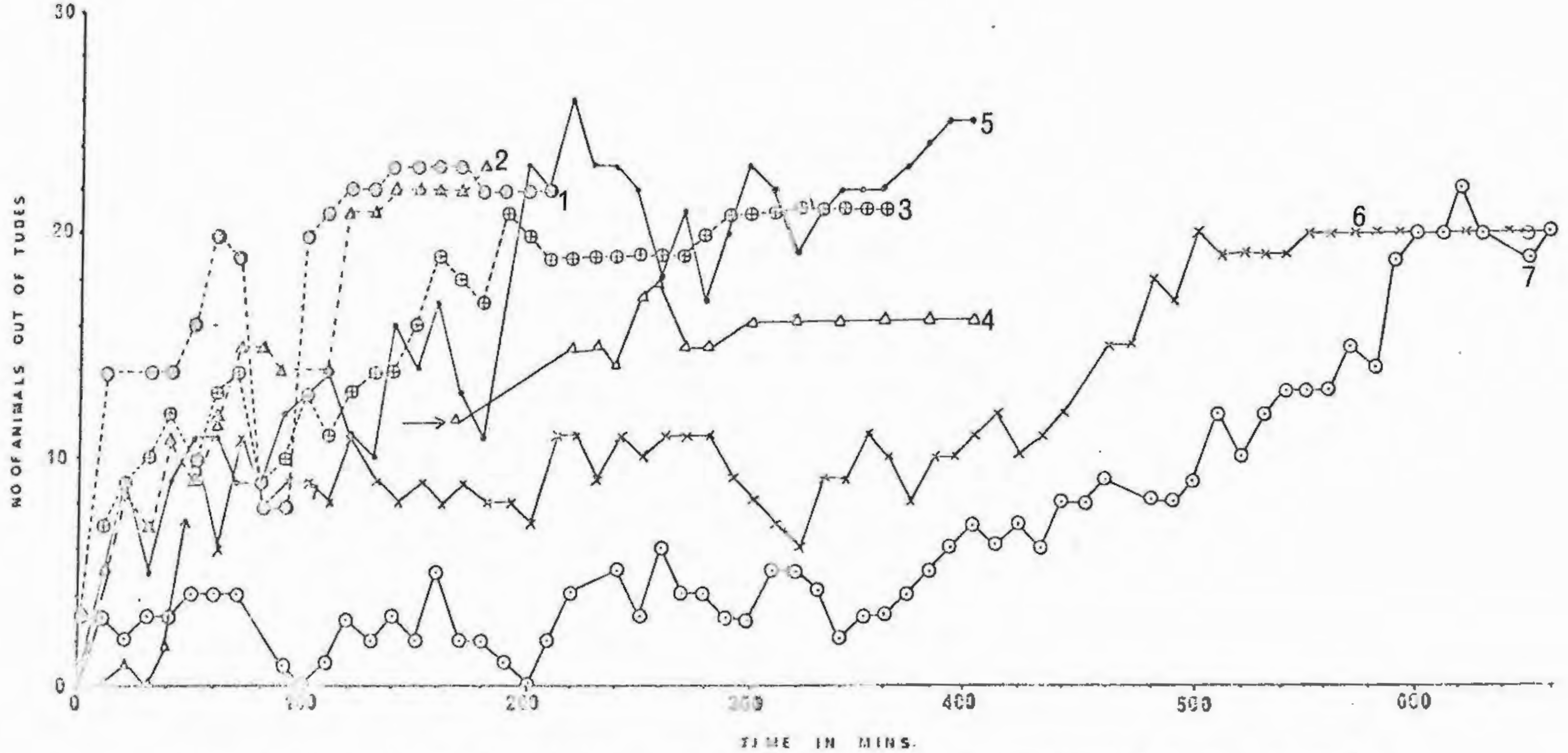


Fig. 40 Numbers of animals out of tubes in the experimental chamber, under the influence of different rates of pCO_2 rise. Each experiment started with 30 animals in the chamber.

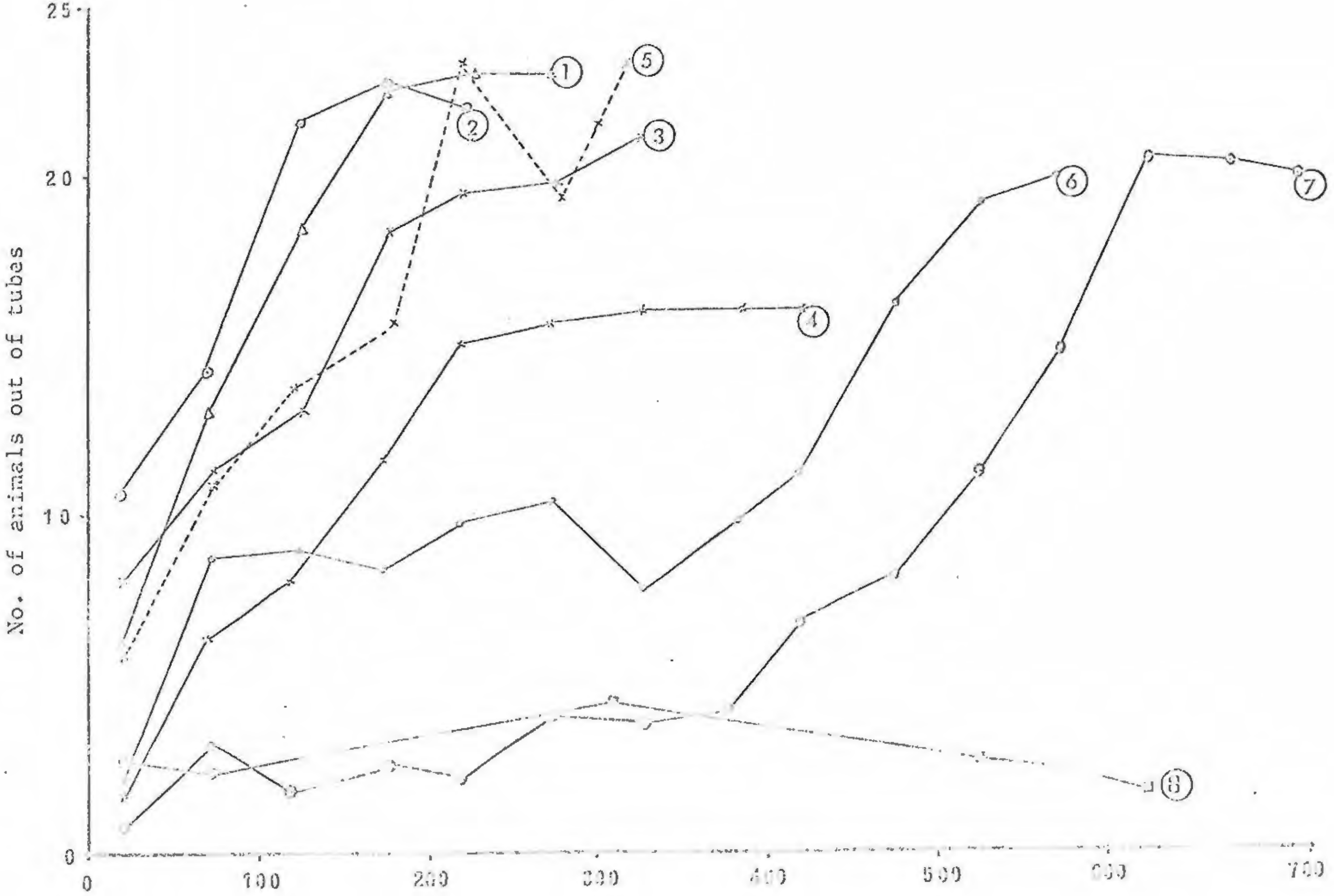
that even at relatively high concentrations of pCO_2, G. lignorum will return to burrows providing the pCO_2 remains constant, drops or rises at a very small rate.

In spite of the imperfect control over the rate of pCO_2 rise in this experiment, and the variations in the numbers of animals out of tubes at any one time, just over half of the animals died in their tubes. The experiment thus indicates that at very slow rates of pCO_2 rise there is no particular concentration which will drive the animals permanently from their tubes.

In order to obtain an estimate of the minimal rate of rise of pCO_2 necessary to move G. lignorum out of burrows, experiments were performed using the micrometer screw regulating valve for the addition of CO_2 saturated water to the main reservoir of the apparatus. Even with this modification, completely smooth control was not obtained, although it was much better. Records of the rates of pCO_2 rise are given in Figure 39. The numbers of animals out of tubes with respect to time are given in Figure 40. For simplicity, the average number of animals out of tubes during 50 minute periods was calculated, and is plotted in Figure 41.

The trends in the data are clear. At more rapid rates of pCO_2 increase, more animals are driven out of tubes in a shorter time (Graphs 1 to 5, Fig. 41). The minimum rate of rise used which drove animals out of their tubes within 50 minutes, was about 28.9 mm Hg CO_2/hr. At the rate of about 14 mm Hg CO_2/hr, 400 minutes elapsed before more than 5 animals were consisently out of their tubes. From my note book record, this was the first time that animals showed typical distress symptoms by clustering at the lighted end

Fig. 41. 30 G. lignorum in glass tubes were subjected to different rates of increasing concentration of CO_2. The chamber was inspected every 10 minutes. Readings for every 50 minutes are combined and a simple arithmetic average has been plotted.



of the experimental chamber. Severe anaesthetic effects appeared at 450 minutes when some individuals stopped beating their pleopods and ceased all movement. Thus, although significant numbers of animals appear out of tubes at low rates of pCO_2 increase, this effect appears at a time when severe physiological disturbances are taking place, in contrast to the active abandonment of the tube well before obvious physiological distress at higher rates of pCO_2 increase.

The nature of the physiological distress may be respiratory at high pCO_2 concentrations, even though oxygen levels in the water are well above those required to move the animals out of the tubes without CO_2. There is an exaggerated cleaning of the gills at the base of the legs in much the same way that Upogebia africana (Hill 1968) cleans its gills when exposed to low oxygen conditions, or blockage of the gills is caused by large amounts of suspended material in the water.

Having reasonably shown that the rate of pCO_2 rise must be between 14 and 28 mm Hg CO_2/hr, to drive G. lignorum from their tubes, it is also important to try and establish the absolute concentration at which animals move out of tubes for any particular rate of rise of pCO_2. High concentrations are not likely to occur in the bottom of Lake Sibayi, although transient sharp rises in pCO_2 may occur over low CO_2 concentration ranges. Unless the behavioural responses of the amphipod are shown at low concentrations, no matter what the transient sharp rates of pCO_2 rise may be, they will not be effective in determining the distribution of the animal in the bottom of the lake. Furthermore, any attempt to match the laboratory findings on the behaviour of G. lignorum in response to pCO_2 rates of rise with field results must rest for the moment on indirect measurements of CO_2 concentrations from the bottom of the lake. The problem of estimating rates of change in the real situation in the lake has not yet been solved.

What data are available on the minimum quantities of CO_2 require to move *G. lignorum* from its tubes are assembled in Table 23.

Table 23

Minimum pCO_2 apparently required to move 5 or more *G. lignorum* from tubes at different rates of pCO_2 increase. See text also.

Rate of pCO_2 rise mm Hg/hr	159	122	120	73	63	38	31	23
pCO_2 when first 5 or more animals	13	77	18	13	21	0.6	65	22

Since the control experiment indicated that 5 or more animals out of tubes could be taken as a significant response, the partial pressure of CO_2 at the time this occurred in the experiments has been recorded in the Table, and was taken as a measure of the minimum pCO_2 required to move the amphipods out of tubes for any particular rate of rise of pCO_2. The apparent concentration required varied between 0.6 and 77 mm Hg CO_2. No systematic arrangement of concentrations of CO_2 is seen when the rates of pCO_2 rates of rise are arranged in order.

These data tend to indicate that the minimum concentration required at any rate of pCO_2 rise is not likely to be more than 77 mm Hg CO_2 or less than 0.6 mm Hg CO_2. More probably it may be between 10 mm Hg and 20 mm Hg CO_2. The higher figures may be in error because of the method of taking readings at 10 minute intervals which only gives information at points of time. Should 5 or more animals have emerged early in the 10 minute interval between readings, the concentration recorded would be too high at the next reading. The low reading of 0.6 mm Hg may be in error because of the curvilinear relationship of the relationship between electrode response and CO_2 concentrations below 2% CO_2 (17 mm Hg CO_2 when 100% is 760 mm Hg).

More sophisticated experiments than those on which these figures are based are really required to get a more reliable measure of exactly the concentrations required. It does seem, however, that we may be safe in assuming that it will not be larger than 70 mm Hg CO_2.

Hydrogen sulphide.

H_2S readily forms beneath the surface of substrates, to form the sulphuretum layer. Addition of small amounts of H_2S saturated water to the water flowing through the experimental chamber causes the animals to abandon their tubes. However, H_2S can only exist very transiently in the presence of oxygen in water, and hence is not likely to be an inhibiting factor in the distribution of animals which live in the first few mm of a lake substrate. It is interesting to note that H_2S was never smelt in the muddiest of samples from Lake Sibayi, although it was demonstrated from the black slush bottom sediments in Lake Nhlange.

Discussion.

Some indications of what might constitute "unfavourable" conditions causing G. lignorum to abandon its tubes has been gained. These need not necessarily be all, or indeed the most important factors which might contribute to this behaviour, for example nitrogenous excretory waste products, products of plankton decay or other chemicals may form part of a complex "unfavourable" condition. For the present, however, and faced with the lack of further information, carbon dioxide appears to be the most likely factor contributing to "unfavourable" conditions at the substrate/water interface. The presence of this gas is well established in lakes (Hutchinson 1958, Ruttner 1963) and is usually found in concentrations in excess of that found in air.

We are now in a position to erect an hypothesis which offers an explanation of the reasons for the distribution pattern of the amphipods with respect to depth in Lake Sibayi. Briefly this would be that rates of CO_2 concentration increase between 14 and 28 mm Hg CO_2/hr should serve to remove amphipods from the substrates in which these changes occur. It is suggested that these conditions occur in water deeper than 25 metres in Lake Sibayi.

The formulation of this hypothesis directs our attention to what should be examined in the real situation of the lake. What little data we have on this point is assembled in the next section, together with an examination of what might occur in the lake bottom water/substrate interface.

FIELD INVESTIGATIONS ON CONDITIONS AT THE SUBSTRATE/WATER INTERFACE

Introduction

The experimental results have suggested an hypothesis for the distribution of G. lignorum in Lake Sibayi. Further evidence, either in support of, or otherwise, can be gained from the field. Some indication of changes in the rates of pCO_2 rise at the substrate water interface is necessary from the point of view of the generation of "unfavourable" conditions. The results so far are disappointing, since problems in the technology of measuring dynamic changes in the substrate surface interface of the lake have not been solved. It would be desirable to sample water a few mm below the surface of the substrate, at the surface, and perhaps 1 cm above the surface in at least 35 metres of water, since the changes that are envisaged by the hypothesis must take place where the animals build their tubes. Further, Mortimer (1941/42) has shown that considerable change takes place over the first few mm of water interface.

Methods of water sampling

Apparatus for taking water samples very near the bottom has been described in the literature. Thus, Joeris (1964) describes a horizontal sampler for the collection of water near the bottom. But this type includes water from up to 7 cm away from the substrate since the barrel is 7.1 cm internal diameter. In order to try and get samples at smaller distances from the substrate, an apparatus was designed to take 20 cc samples of water at known distances from the bottom sediments. The design of the apparatus is given together with an analysis of the sampling action of the apparatus in Appendix I . From these considerations, it is clear that water is being sampled from an area around the needle points. The nominal heights of the needle tips, especially in the case closest to the substrate, gives a false impression of the height from which the water is being sampled. Although the apparatus will not function exactly as indicated by the model, the figures given by the analysis give a good indication of what the 20 cc water sample represents in terms of height of water sampled, relative to the substrate. These are set out in Table 24.

Table 24

Theoretical distances from the substrate of the water samples taken by the syringes of the apparatus for sampling water very near the bottom.

Height of the needle tip above the substrate. cm.	Minimum and maximum height of water sample from the substrate. cm.
0.5	0 -2.4
5.0	3.1-6.9
9.0	7.1-10.9

Although the apparatus obtains samples of water nearer the substrate than that described by Joeris (1964), it is not entirely satisfactory since the needle at 0.5 cm from the substrate mixes in water from as high as 2.4 cm from the substrate. Conductivity measurements and redox potential measurements reported by Mortimer (1941/42) for Lake Windemere, have indicated that over mud substrates extensive changes may occur at less than 0.5 cm, indeed a few mm above the substrate/water interface.

The concentration of CO_2 was not measured directly, since the volume of the sample, 20cc, is too small to handle without the possibility of serious changes on exposure to the atmosphere. However, since there is a linear relationship between alkalinity and CO_2 concentration (e.g. Alause 1968), differences in pH from adjacent water samples from the same water body will give an indication of the CO_2 concentration. Providing no great change in alkalinity occurs, the free CO_2 concentration can then be estimated for the nomograms in "Standard Methods" (12th ed. 1965) if the alkalinity of the water is known.

The pH of samples was measured with either a Metrome portable pH meter or a Beckman model G portable pH meter. pH determinations by portable pH meters with glass electrodes are notoriously suspect if they have not been carefully calibrated against reliable buffers shortly before use. Being well aware of this source of error, great care was taken in the determination of the pH results reported. The instrument was checked against buffers before the reading of each set of samples. This was particularly important since only small changes were to be expected. The pH was estimated to the second decimal place, so that complete confidence in the first decimal place could be established.

The concentrations of oxygen were estimated by means of Winkler method. Unfortunately, disposable plastic syringes were used on the first visit to the lake with the apparatus when time was available for an extensive sampling programme. Late in the sampling programme, it was discovered that significant quantities of oxygen could leak through the plastic walls of syringe barrels, at a rate of about 1 mg per hour. This meant that most of the determinations had to be discarded. In an attempt to overcome the difficulty, and since glass barrelled syringes could not be obtained during the visit to the lakes, the samples were acidified

Table 25

The results of pH measurements from water near the substrate from Lake Sibayi. A mean was calculated from up to four determinations of water from one syringe at one depth from one side of the water sampling apparatus together with determinations of pH from the duplicate sample on the other side of the apparatus at the same depth. Figure in brackets is the number of determinations from which mean pH and standard deviation at nominal sample depths was obtained.

Station	Date	Depth m	Time hrs.	Surface pH	9.0cm	5cm	0.5cm	Remarks
A	25/9/69	19.0	14.00	8.40(3)	8.43±0.06(6)	-	8.37±0.00(6)	21.5°C
B	31/10/69	19.0	05.30	8.36±0.074(4)	8.26±0.04(6)	8.21±0.04(7)	8.37±0.09(4)	Wind force 1 22°C
C	30/10/69	20	18.30	8.53 (3)	8.46±0.03(7)	8.44±0.05(8)	8.47±0.03(6)	Wind force 2 22°C
D	25/9/69	30.6	18.50	8.50 (3)	*8.05 (3)	*7.91±0.06(4)	*8.01±0.07(7)	21°C
E	23/10/69	31.5	18.05	8.38 (3)	*8.19±0.03(7)	*7.72±0.04(4)	*7.29±0.04(4)	Wind force 2 22°C
F	24/10/69	31.5	0.630	8.53 (3)	8.43±0.01(6)	8.35±0.04(7)	^*7.93±0.10(6)	Wind force 3 22°C
G	26/9/69	31.6	0.630	8.50 (3)	8.38±0.3(6)	8.40 (3)	8.32±0.07(5)	21°C

* Fine material found in samples.

^ Tanaids found in syringe.

to release iodine as soon as a clear space had developed above the precipitate of manganous hydroxide in the barrel of the syringe. This procedure was done in the boat on the sample station, and the samples were then carried back to the laboratory for titration. Some iodine was taken up by the plastic of the syringe barrels but this was only serious after very long periods (± 24 hours). Titrations were carried out within 3 hours of the collection of the sample. The data on oxygen concentrations therefore are not accurate, but indicate that not less than the figure obtained was present in the water sample. As in the microWinkler method (cf. Barnes 1959), the dead space in the syringe was filled with manganous chloride. The Winkler reagent was introduced by means of another syringe with a needle into the outlet of the main syringe.

RESULTS

pH measurements are only available for a limited number of samples from Lake Sibayi. Ideally the samples should be taken in different weather conditions on the lake. Day and night samples from different depths are also needed in order to assess the influence of wind action on the surface in the deep water of the lake. The data gathered thus far indicate that further efforts in this direction could well be rewarding.

The results of the few pH measurements taken simultaneously from surface waters and waters near the substrate, are reported in Table 25. The surface values obtained varied from day to day, and also from station to station. Thus, on the 25/9/1969, at two different stations over 19 metres and 30 metres (A and D, Table 25), the pH was 8.40 and 8.53 respectively. Similarly, on

23/10/1969 and 24/10/1969 over 31 metres (E and F, Table 25) values of pH 8.38 and 8.53 were obtained. Since there is at present no reason to believe that anything other than carbon dioxide is responsible for the pH differences, these values tend to suggest that even in open water in the main basin of Lake Sibayi, the carbon dioxide budget is in a state of flux. Using the bicarbonate alkalinity value for the hot season, reported by Allanson and van Wyk (1969) for Lake Sibayi, a maximum difference of 0.37 mg/l free CO_2 was estimated from the nomograms in "Standard Methods" (1966, 12th ed.).

The greatest pH difference between surface water and water from very near the substrate was recorded from 30 metres depth. On 23/10/69 (E, Table 25), a difference of just over 1 pH unit was measured. Other pH differences from 30 metre depths were 0.6 pH units (D, Table 25), 0.5 pH units (F, Table 25) and 0.18 (G, Table 25). In 19 metre to 20 metre depths, the greatest difference recorded was 0.06 pH units (A, B, C, Table 25).

Presuming no great change in the bicarbonate alkalinity between surface and bottom, the free CO_2 value was estimated from the nomograms in "Standard Methods" (1965, 12th ed.). This presumption is not strictly true since any aggressive carbon dioxide will take up carbonate from the substrate as bicarbonate. However, the differences are relatively small, for if the bicarbonate alkalinity increased to 200 mg/l from 150 mg/l at pH 8.0, the increase in the free CO_2 would be 1 mg/l. The differences of free CO_2 estimated from pH values were for Station E, 13 mg free CO_2/l, Station F 1.19 mg free CO_2/l, Station G 0.4 mg/l and Station D 1.6 mg free CO_2/l.

Other differences in pH estimates were found in

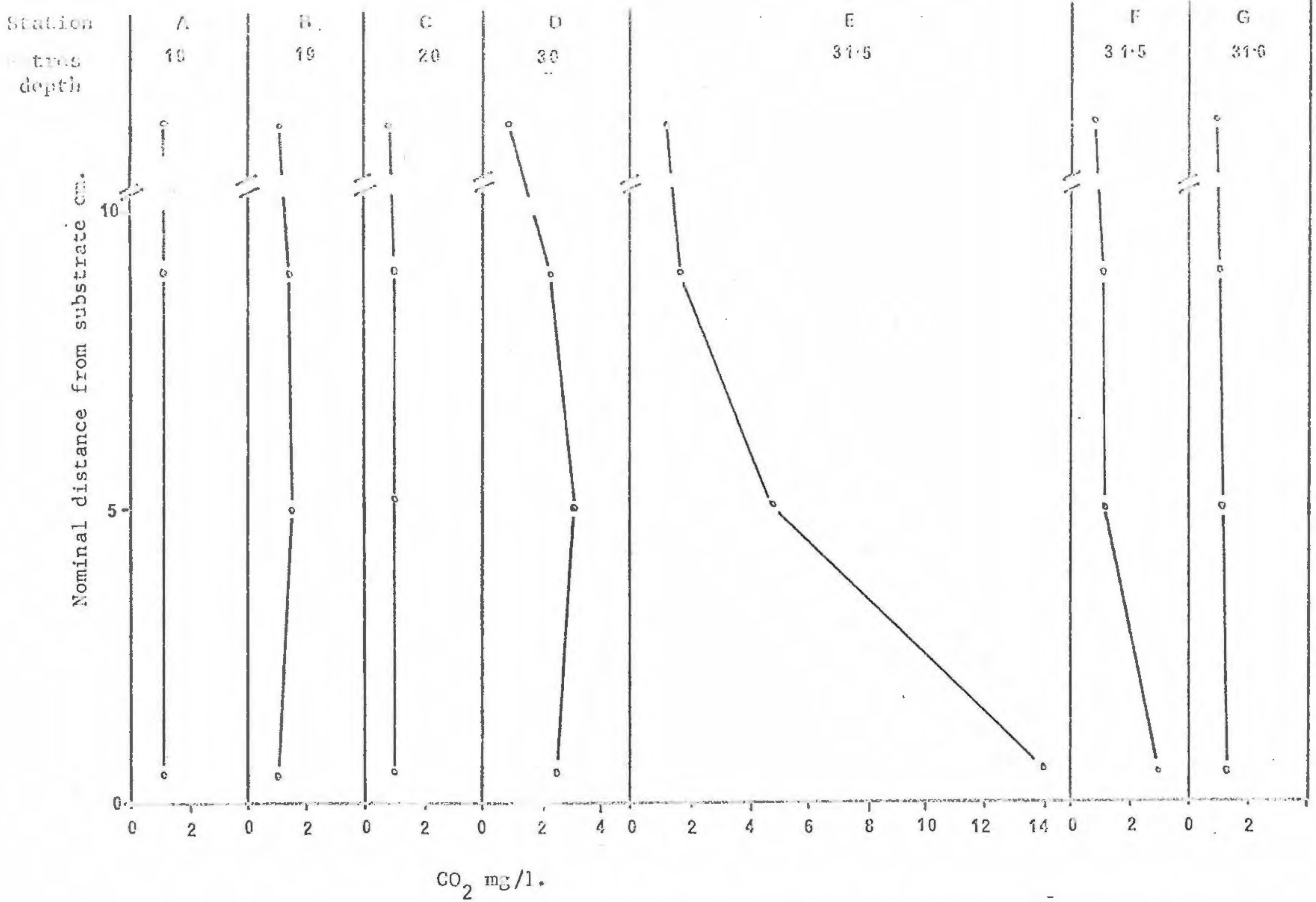


Fig. 42. Concentrations of CO_2 very near the substrate calculated from the nomograms of "Standard Methods" from the information in Table 25. Stations E and F show marked gradients of free CO_2 very near the substrate/water interface.

samples from surface and bottom waters in 19 metres (A, B and C, Table 25). The greatest difference in free CO_2 was 0.15 mg/l on 31/10/1969 (B, Table 25).

As far as these few samples taken allow, there seems to be evidence that in deeper waters, concentrations of CO_2 in the water near the substrate may very widely from surface values measured at the same time. Furthermore, the concentration gradient is steepest in water close to the substrate. The most marked gradient was from Station E where the three samples at 9 cm, 5 cm and 0.5 cm nominal depths (Table 25) had pH values of 8.19, 7.72, and 7.29 respectively. The free CO_2 concentrations estimated from the nomograms were 1.65 mg/l in the top sample, 4.7 mg/l in the middle and 14.1 mg/l in the bottom sample nearest the substrate. Stations D and F gave smaller free CO_2 differences and G practically none (Fig. 42).

<u>Oxygen</u>

Allanson and van Wyk (1969) published results which showed that in winter, oxygen was almost fully saturated down to near the bottom of Lake Sibayi, only dropping to 94% of saturation at 35 metres. In summer a slightly greater deficit was experienced at times. In January 1968 the percentage saturation at 38 metres was 68%. These authors used Friedinger sampling bottles which do not allow samples to be taken closer than one sampling bottle height away from the surface. Careful, as yet unpublished determinations, from one station at 29.6 metres gave a figure of 6.6 ppm O_2 one bottle height from the surface of the substrate. This represents a percentage of saturation of about 80% for the temperature at the time (Allanson, personal communication). Using the apparatus described for sampling near the bottom of the lake with disposable syringes, values of the same order were obtained in January 1969. The absolute values are unreliable because of leakage of oxygen into the water samples; nevertheless, taking this error into account, not much less than 6 ppm was present at the bottom in 38 metres. Since the same apparatus easily indicated much lower values in Lake Nhlange, it can be concluded that in the layer of free water closest to the bottom of Lake Sibayi, no serious oxygen deficits existed.

Discussion

The best published information on the conditions at the substrate water interface of a lake comes from the classical work of Mortimer on Lake Windemere (Mortimer 1941/42). Mortimer, however, was concerned with the reintroduction of nutrient and other chemicals from the mud during the stratified period, and furthermore he worked on the more common mud type of bottom rather than a sandy substrate. The substrate of Lake Sibayi contains both mud and sand type. The muddy bottom is poor in fauna, whereas the sandy type has quantities of fauna. It is on the sandy type of bottom that the discontinuous distribution is found. That there is little information on the conditions of a sandy substrate water interface, is not surprising, since the fine sand type of bottom of Lake Sibayi, or indeed of the South African coastal lakes, is an uncommon feature of lakes in general.

Not enough detailed work on the conditions at the sand water interface from Lake Sibayi has been done. Up to the present, the distance of the lake from the laboratory has dictated far more emphasis on other types of work on the lake. The whole of the problem requires a special study in itself. We can at this time only attempt an educated guess at what might obtain in this region of the lake, using whatever principles are available from previous work on substrate/water interface studies, and incorporating the flimsy field evidence from Lake Sibayi.

The sandy substrate of Lake Sibayi has been shown to have a layer of fine material over it in which is incorporated both plant and animal life. Doubtless the sand particles also have a micro-organism film. As in all lakes, the bottom of Lake Sibayi must take up oxygen and release CO_2 from normal metabolic processes. In winter in Lake Sibayi there is no lack of oxygen in the deepest waters of the lake. High oxygen tensions have been measured as near the bottom as normal apparatus would allow. In summer a small deficit of oxygen has been shown to obtain in the deeper waters of the lake. Ephemeral stratifications appear (Allanson, personal communication) from September through to January, but these do not successfully block off the deep water from the oxygen in the lake. Anaerobic conditions have never been detected in the surface of the sandy substrata in Lake Sibayi, and indeed cannot obtain since large populations of the Tanaid *Apseudes digitalis* occur in the sand of deep water. Therefore, there appears to be adequate circulation of water to all the substrates in the lake throughout the year.

Little is known of the pattern of circulation in Lake Sibayi. Scuba divers have measured currents of 0.6 metres per minute approximately, using the rate of travel of a dye through the water at 8 metres, with a force 3 wind blowing on the lake. Visual observations on the fine material overlying the sandy substrate show that this is swept away in shallow water (up to 2m.) and varies greatly in depth over the sand below about 5 metres. The material shows no ripple pattern, which might indicate disturbance by currents. However, it is most easily disturbed by the gentlest movements of divers.

The existence of slow currents in deep water has been shown in a number of both stratified and unstratified lakes. In Loch Garry, when there was a surface current of 28.0 cm/sec at 30 metres, it was 3.7 cm/sec and at 61 metres 3.1 cm/sec when the lake was unstratified (Wedderburn 1910 *vide* Hutchinson, 1957).

Mortimer (1941/ 1942) has shown that even within the hypolimnion, what he has termed turbulent diffusion carries away nutrient materials from the mud at rates in excess of those that can be accounted for by molecular diffusion in a static system. However, turbulent diffusion takes place in the free water and not in the water held in the substrate. In this case molecular diffusion is the only mechanism for ingress and egress of chemical substances from the substrate (Mortimer 1941/42, Bouildin 1968).

It seems reasonable to suppose that the undisturbed layer of fine material in Lake Sibayi traps water, and allows gas out only by molecular diffusion. Under the influence of a steady rate of gas transport away from the substrate/water interface by turbulent diffusion, a gradient of gas concentration will establish itself in the fine material overlying the sand of the substrate. Concentrations of gas at any particular level in the water phase of the fine material will then remain constant. Variations in the rates of transport of gas away from the substrate will alter the concentration of the gas at the substrate/ water interface and hence disturb the rates of molecular diffusion of gas out of the substrate. For example if the turbulent diffusion stops for any reason,gas will be transported away from the substrate/water interface more slowly, and hence lead to raised concentrations of gas there. The difference in gas concentration within the substrate and in the free water over the substrate will fall, and the rate of molecular diffusion of the gas from the substrate will also fall. The concentration of gas at any particular level within the substrate will then rise until new equilibrium conditions are set up.

Mortimer (1941/42) has pointed out that turbulent diffusion is an uneven process which varies considerably with the effects of wind forces on the surface of the lake. Variations in the rates of turbulent diffusion will seriously affect the rates of CO_2 build up in the water phase held in the substrate, and consequently influence the occupancy of the substrate by animals sensitive to changes, such as G. lignorum.

Further insight into the behaviour of gas exhange in water at the water/substrate interface is afforded by studies at Lake Nhlange by Allanson (personal communication). Bathythermograph recordings of temperature profiles showed temporary stratifications in summer lasting but a few hours. The presence of these discontinuities must help resist wind generated water movements. In Nhlange, water immediately above the mud surface over the 15 metre plain was shown to have only 0.3 mg/l of oxygen, while water taken at 5 cm away had 2.6 mg/l of oxygen. Thus, in spite of water movements clearly being observed by the depression of the temporary thermoclines in Lake Nhlange, enough stasis of water occurred in 14 metres to allow of deoxygenation by the rich bottom mud. For Lake Sibayi, at this point we can only postulate that stoppages of water can occur during calm periods on the lake, but this does not seem unreasonable in relation to the observations on Lake Nhlange. Certainly the evidence is not strong, but surely indicates that further effort on detailed measurements of dynamic events close to the sandy substrate/water interface may not be fruitless. *Grandidierella lignorum* will abandon its tubes in the substrate providing the rates of pCO_2 change are large enough.

The carbon dioxide equilibrium in the bottom sediments may also be disturbed by the effects of photosynthetic activities in the surface of the substrate by algal cells. Microscopic examination of the fine material from over the sand in even the deepest waters has revealed the presence of considerable numbers of diatoms and other algal cells. *Melosira granulatum* and *Synedra ?acus* are two of the most abundant species present, both in the freshwater and the bottom sediments. Recent evidence by Hart (unpublished) suggests that these algal cells circulate to all depths of the lake, and as they are abundant in the sediments no strictly benthic algal community has been found. This may well be a function of lake depths since Lake Sibayi has a mean depth of only 13 metres in spite of a large area below 25 metres.

Obviously the presence of these facultative algal species in the substrate must affect the carbon dioxide balance. Should there be sufficient light they may use CO_2 for photosynthetic processes, and in the dark they will add to the production of it.

It is difficult to judge the depth at which the plants will have an important effect in the abstraction of CO_2 from the water for their own metabolism under the influence of light. Levring and Fish (1956) have shown that in Lake Victoria, the compensation point for photosynthesis lay between 5 and 10 metres for planktonic algae, on a clear day, and was somewhat less on a cloudy day. Judging from their graph, the light had dropped to between 0.2% and 1.5% of the surface illumination. Light meter readings from Lake Sibayi reported in Allanson & van Wyk (1969) for

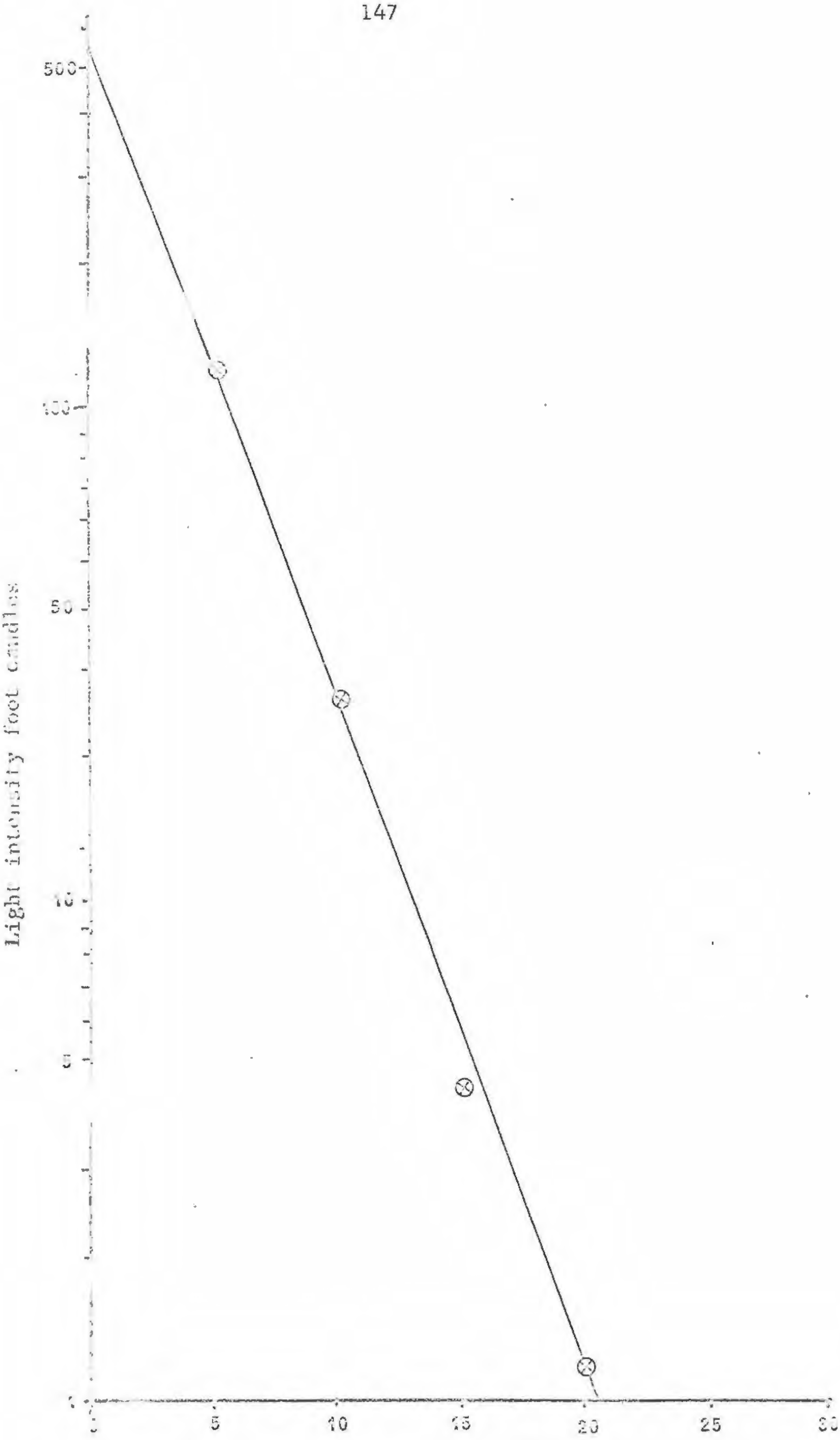


Fig. 43. Absolute light intensities measured in Lake Sibayi on 26/8/69 at 5 metre intervals, using the light meter described in Appendix II.

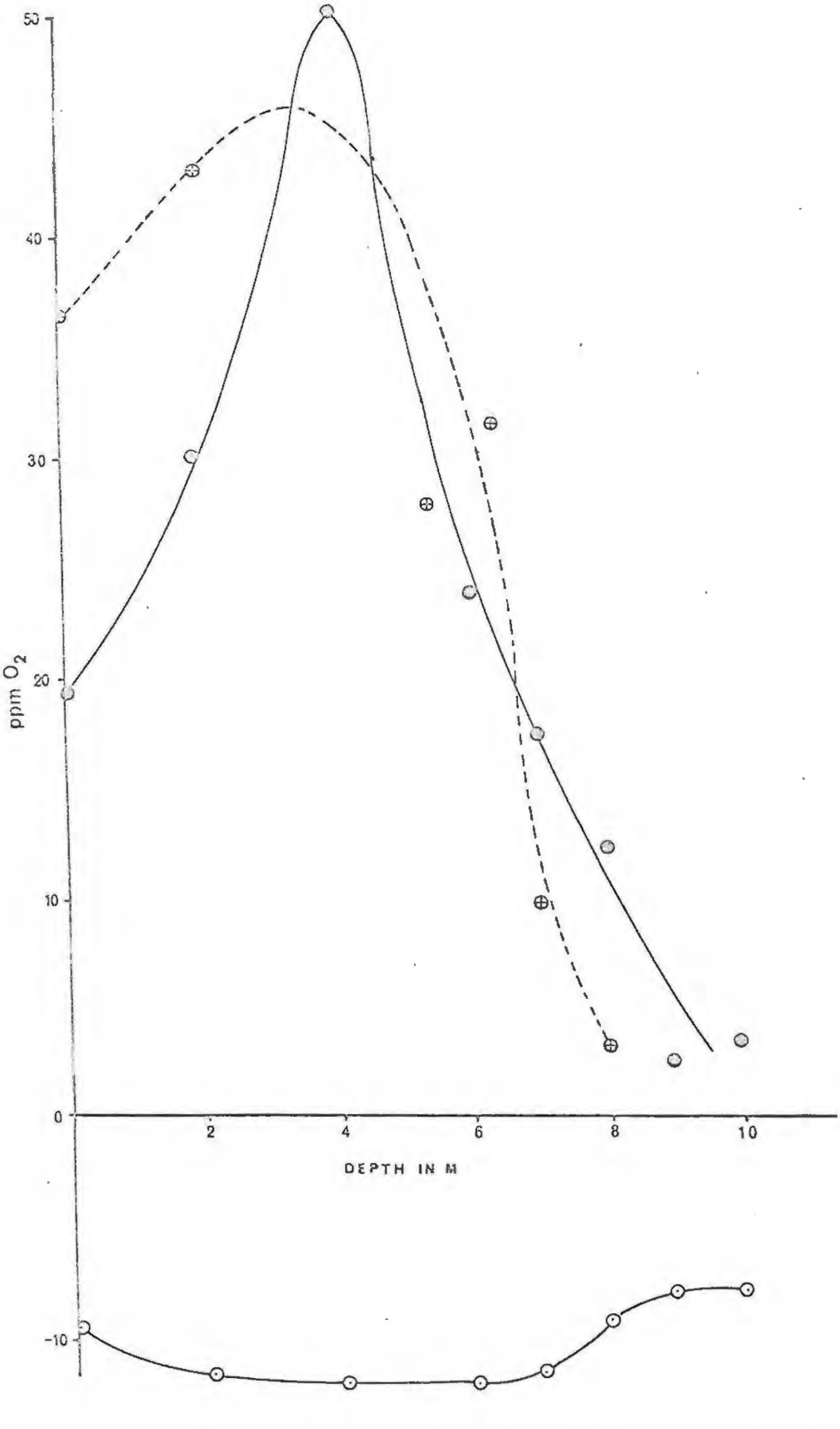


Fig. 44 Oxygen evolved or used by Myriophyllum spicatum in ppmO_2/gm wet weight of weed in dark and light bottles in Lake Sibayi according to the method used by Beadle (1932) in Lake Naivasha. Open circles ; Dark bottles. Open circles with crosses; 25/7/68 closed circles; 26/7/68. The compensation point lies between 7 and 8 metres.

Lake Sibayi indicate that 1% of total illumination was present at 13 metres depth. In Lake Sibayi, measurements of absolute values of light were taken on 26/8/69 at 5 metre intervals. These show that using the apparatus reported in Appendix II at 1500 hrs, one foot candle was present at just below 20 m (Fig. 43). It was difficult to judge the 1% extinction of incident light with the meter employed since at high light values the discrimination between hundreds of foot candles is poor. Nevertheless, the incident light value was not likely to have been less than 300 foot candles or more than 500 foot candles. This would tend to suggest that the 1% extinction occurs at between 15 and 16 m depth, which is slightly deeper than that reported by Allanson & van Wyk (1969). From this kind of information, one might guess that effective photosynthesis ceases at about 12 metres depth, for planktonic algae in Lake Sibayi. However, a compensation point for the photosynthesis of Myriophyllum, using Beadle's (1932) method from Lake Naivasha was found at between 7 and 8 metres (Fig. 44). This is the depth at which vascular plants disappear from the substrate of the lake (Boltt, Hill & Forbes 1969, page 244). But increments of oxygen were being generated in the light well below the compensation point at between 9 and 10 metres, although this was not enough to balance out the uptake of oxygen during the dark. It might, therefore, be argued that algal cells carried temporarily into the benthos below the compensation point, may deplete, or at least reduce, the rate of CO_2 production in the surface film during the light, and increase it at night.

A further important point is brought up by Talling (1966) working on the photosynthetic behaviour of planktonic diatom in English lakes. He found that previous light history could increase the photosynthetic ability of *Asterionella* in poor light when it became adapted to this condition, and thus could speak of sun and shade adapted plants, which must therefore have different compensation depths. Hutchinson (1957) has given a classification of four kinds of photosynthetic algae. "Low light" species which are divided into low and high temperature forms, and "High light species" also divided with respect to temperature. Without measurement of the actual conditions in Lake Sibayi, the effect of light on the production and uptake of CO_2 at the surface of the substrate cannot be dismissed as a factor that may help to regulate behaviour of *G. lignorum*.

Nocturnal migration of *G. lignorum* into the plankton

One other important observation of the biology of *G. lignorum* remains to be reported. This amphipod, in common with a number of other infauna of the benthos, is found in low numbers in plankton hauls at the surface even over the deepest water of Lake Sibayi. The animals identified from the plankton which are known from the substrate are given in Table 26.

Table 26

Benthic animals from night surface hauls of plankton from Lake Sibayi.

CRUSTACEA			
	Amphipoda	*Grandidierella lignorum*	
		Corophium triaenonyx	
	Tanaidacea	*Apseudes digitalis*	
	Isopoda	*Pseudosphaeroma barnardi*	(rare)
	Decapoda	*Hymenosoma orbiculare*	(small crabs apart from the normal planktonic zoea larvae).
INSECTA		*Cryptochironomus* larvae	

The same phenomenon was exhibited in experimental conditions by the population of amphipods from the Kowie estuary population.

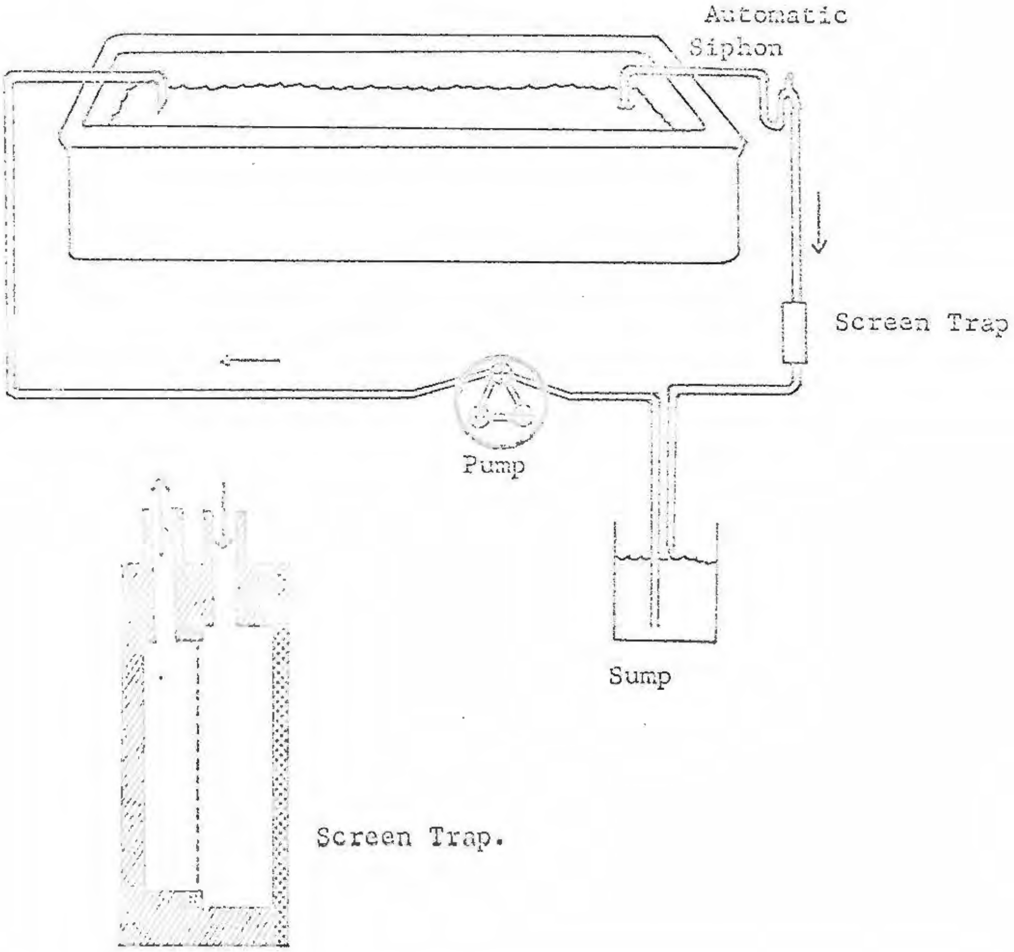


Fig. 45 Diagram of container with automatic siphon and screen trap to catch animals drawn in by the siphon. A sectional diagram of the screen trap is given. Arrows indicate the direction of flow.

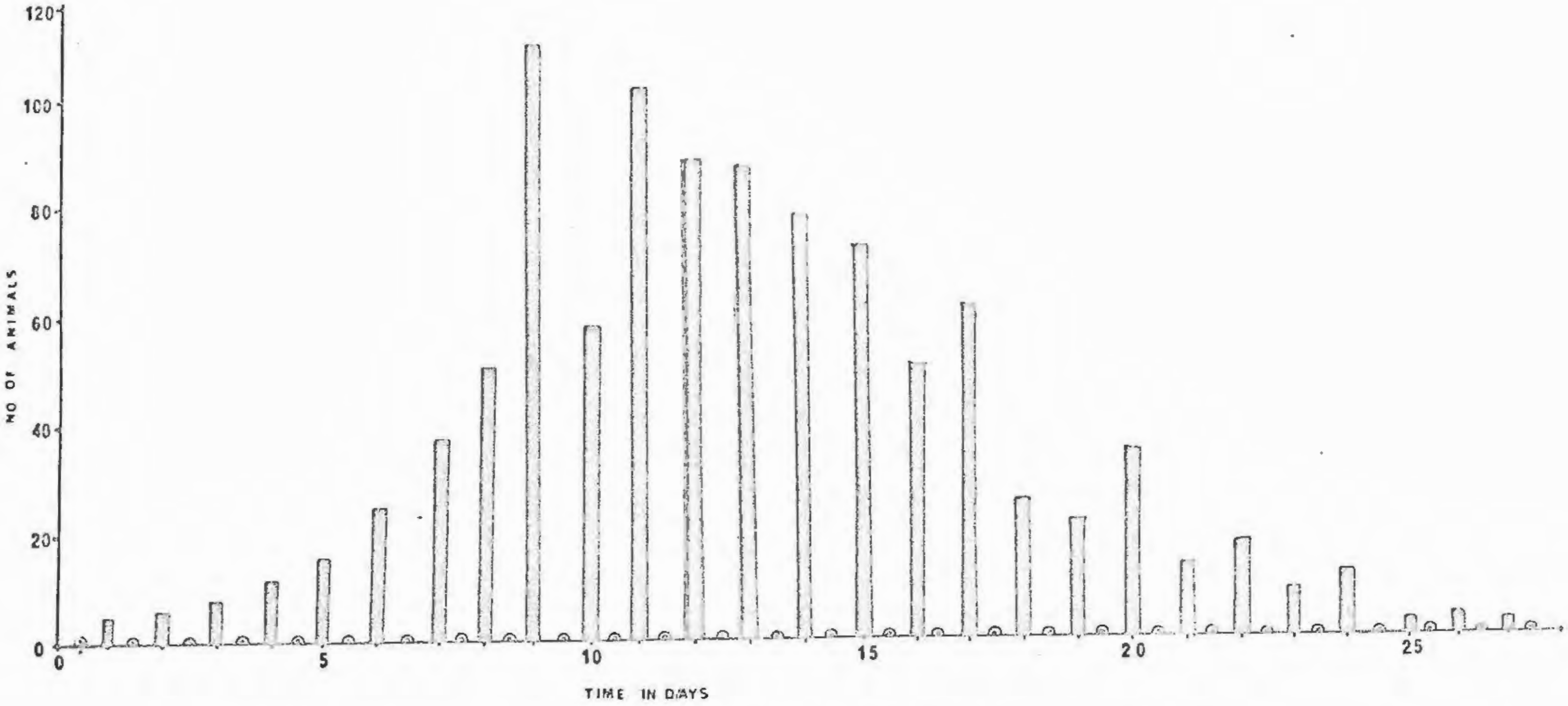


Fig.46 Numbers of *G. lignorum* taken in overflow screen trap in the apparatus shown in fig. 45. Collections from the night are shown as histograms, collections from the day as semicircles.

Substrate containing animals was brought from the Kowie estuary and placed in a porcelain trough 50 cm by 25 cm by 10 cm. This container was placed in a constant temperature room at 20°C with controlled light of 12 hrs darkness and 12 hrs light. The sea water was circulated by means of a pump (Fig. 45). An automatic siphon removed the excess head of water from the tank. In the line of the automatic siphon, a screen trap was installed which would allow the retention of any animals taken in the overflow. The screens were changed every 12 hours. The water flowed through to a sump, and this was then recirculated back into the main container.

The experiment was monitored for 28 days. During this time animals were trapped only from the night collections, and never from the light collections (Fig. 46). This result accords well with the analysis of samples from Lake Sibayi where amphipods were only collected from the night plankton hauls and never from the daytime plankton hauls. In the plankton hauls most of the animals were small juvenile forms. This is also the case for the experiment reported. During the course of the experiment, a total of 809 animals were recovered and classified as juveniles and adult. 84.8% were juvenile forms and 5.2% were adult.

The importance of this observation lies in the mechanism which such behaviour provides for the distribution of amphipods and other benthic fauna over the whole surface of the lake bottom. Animals over deep water at a time when they must return to the bottom could potentially colonize the deep substrates of the lake, but are presumably prevented from doing so by conditions in the substrate.

It is interesting to note that the amphipod *Pontoporeia affinis* has also been reported to migrate into the plankton (Segerstråle 1959). The males are good swimmers and copulation with the females takes place in the plankton. This is unlike *G. lignorum*, where few adults occur in the plankton. In common with *G. lignorum*, *P. affinis* only migrates into the plankton at night. Apparently photonegative behaviour is shown during the day and the amphipod is driven down to burrow in the substrate.

SUMMARY OF THE EXPERIMENTAL AND FIELD RESULTS

The physiology of salt balance in the populations from estuarine and lake conditions appears to be indistinguishable. From this it was concluded that it is reasonable to use the Kowie estuarine population as experimental material, and that the results could be applied to Lake Sibayi.

The reactions of *G. lignorum* to subtrates in the laboratory confirmed the view that substrates play no important role in determining the choice of substrate in the bottom of Lake Sibayi.

Similarly, exposure of animals to pressures greater than those found in the depths of Lake Sibayi illicited no response. It is concluded that this factor cannot play a great part in determining the distribution of the animals in the benthos.

A study of the reactions towards light, of *G. lignorum*, indicated that under normal conditions the animals are photonegative, and that they will enter regions of very low light intensities, burrow and remain in these conditions. However, under "unfavourable" conditions the animals become photopositive and will swim towards the light, away from the substrate.

Stagnation under laboratory conditions elicits the photopositive response and the abandonment of burrows in the sand.

In the laboratory, three factors may elicit this response; severe O_2 deficiency, rates of CO_2 increase above 14 mm Hg CO_2/hr, and the presence of H_2S. Carbon dioxide may be the more important factor since the tanaid *Apseudes digitalis* does not react to CO_2 increase in the same way as *G. lignorum*. Severe oxygen lack does not appear to exist in the bottom of Lake Sibayi. The presence of H_2S is precluded if oxygen is present. Field results support the view that carbon dioxide is held in the fine material over the substrate in greater concentration than in the free water. It is presumed that the greater concentration of CO_2 in the fine material is due mainly to molecular diffusion only allowing relatively slow egress of the gas into the water above.

It is supposed that equilibrium conditions which might obtain if constant rates of turbulent diffusion occurred, may be upset by transient stoppages of slow water movement at the bottom of Lake Sibayi. This would induce changes of carbon dioxide concentration at rates of change which would elicit the escape responses of *G. lignorum*, but would have no effect on *Apseudes digitalis*.

The nocturnal migration of *G. lignorum* into the plankton offer a mechanism whereby the amphipod can be distributed over the bottom of Lake Sibayi.

FINAL DISCUSSION

The two main areas that remain to be discussed are the problem of salinity tolerance of the fauna of lakes and estuaries, and the kind of research that is likely to yield the most understanding about this fauna.

The fauna of the South African coastal lakes has been shown to belong in the greater part to Day's (1967) estuarine component, but at least two species from Lake Sibayi belong to the euryhaline marine component (*Cyathura carinata* and *Hymenosoma orbiculare*). The assignment of the fauna to these categories in point of fact tells us little about the biology of the animals. It merely indicates that they are intolerant of wave action and are therefore "quiet water forms". Day has stressed that it also does not indicate much about the salinity tolerances of the species concerned.

Nevertheless, ultimately salinity must be of paramount importance in determining the distribution of the estuarine fauna.

A consideration of the faunal lists and the recorded salinity regimes of the South African estuaries and coastal lakes only indicates in a general way the salinity tolerances of the fauna. The reason for this may be twofold. Firstly, the recorded salinity data from the South African estuaries are sporadic and often do not include the complications introduced by the effects of seasonal floods, and secondly the salinity responses of the fauna are more complex than simple minimum salt concentration survival experiments indicate.

With regard to South African blind estuarine systems (Day 1951) and open estuarine systems, although there are many species common to both types, there is no generalisation which will adequately describe salinity patterns. Local conditions

Table 28

Records of salinities one day after the beginning of a flood in the Kowie River estuary. Salinities sampled on 29/5/67.

Distance from mouth	Depth	Salinity °/oo
1 km	0.0 m	4.69
	1.5 m	31.38
	3.0 m	2.35
2.5 km	0.0 m	1.11
	1.5 m	2.40
	3.5 m	31.45
	7.0 m	32.70
5 km	0.0 m	0.50
	1.5 m	0.51
	3.0 m	0.48

Note: Salinities were measured using the modified Moore titration and standarised against Copenhagen Eau de Mer Normale (Barnes 1959).

Table 29

The results of salinity samplings 5 days after the flood in the Kowie River estuary (taken 4/6/67). Samples were taken on the rising tide and also on the falling tide, starting at the mouth and following the tide upstream. The depths also indicate the depths of the estuary bed at the point of sampling.

Distance from the mouth	Depth m	Salinity °/oo Rising tide	Salinity °/oo Falling tide
1.1 km	0.0	10.05	18.25
	1.0	-	-
	2.0	12.09	-
	4.0	17.48	-
	5.0	-	35.5
2.5 km	0.0	7.04	-
	3.0	10.60	-
	6.0	28.32	-
	9.0	33.18	34.3
	11.3	33.70	28.83
3.5 km	0.0	5.93	-
	3.5	13.17	-
	7.0	34.40	-
	9.5	33.18	-
5 km	0.0	3.52	7.42
	1.5	4.33	-
	3.0	13.00	-
	4.0	-	24.90
9 km	0.0	1.13	2.98
	3.0	3.32	8.69
	6.0	28.22	28.30
11 km (Measured shortly before and after turn of tide)	0.0	0.72	-
	4.5	-	2.87
	8.2	0.88	-
	9.1		3.29

of each estuarine system vary such that open estuaries may show well developed salinity gradients most of the time (e.g. Knysna Estuary (Day, Millard & Harrison 1952) and Umlalazi Estuary (Hill 1966)) or, as in the case of estuaries like the 19 km long Kowie estuary salinities above 30^{o}/oo are shown for the greater part of the time up to the top of the estuary. Data collected by Airey (1968) are given in Table 27.

Table 27

Date	Salinity ^{o}/oo
28/2/68	30.44
5/3/68	32.03
5/5/68	29.59
26/5/68	31.44
11/8/68	1.65

Closed estuaries show varied salinity conditions. Brown (1953) showed that Kleinemond estuary ranged only from 21.06 to 35.68^{o}/oo during 8 months in 1953, although salinities must drop drastically during floods. Salinity records collected from a blind estuary near to Kleinemond showed a value of 15^{o}/oo (K.L. Pringle personal communication).

The salinities of the estuaries are also markedly affected by flooding. Data collected by Dr B.J. Hill and myself (Table 28 and 29) for the Kowie estuary following a severe but typical flood, shows a complex pattern of extreme dilution over the greater part of the estuary shortly after the flood and five days later.

Clearly the substrata of the estuary were exposed to considerable salinity fluctuation. The salinity regimes of the estuaries are therefore so complex that it is unlikely and probably impossible to discover meaningful correlations between salinity and faunal distribution from survey methods.

Complications in the interpretation of such data are also compounded by the physiological responses of euryhaline animals to salinity changes. Two levels of salinity tolerance must be clearly recognised, salinities at which animals may survive for sometimes extensive periods, but which are not suitable for completion of all the phases of the life cycle, and minimum salinities which must be present at some time for completion of the whole of the life cycle. Sutcliffe (1968) for example, has indicated that *Marinogammarus finmarchichus* may withstand temporary dilution of the medium to a very low value, but probably cannot cope with it indefinitely. Hill (1967) has shown that *Upogebia africana* requires at least 10^{o}/oo to live in indefinitely, but can withstand much greater dilutions of the external medium for long periods of time. At very low dilutions the moult is delayed and moulting occurs on return to higher salinities.

The fauna of the estuaries must therefore be adapted to wide fluctuations in salinity. On a long term basis, salinities below certain values will exclude certain animals, but many can survive extreme dilutions on a short term basis and, indeed, must do so in estuaries like those of South Africa which are periodically flooded.

It is likely that there is a complete spectrum of long term lower limit salinity tolerances among the various species found in an estuary, but the data, especially from the South African estuaries, are very limited. An orderly arrangement of species into categories must therefore await data from experimental

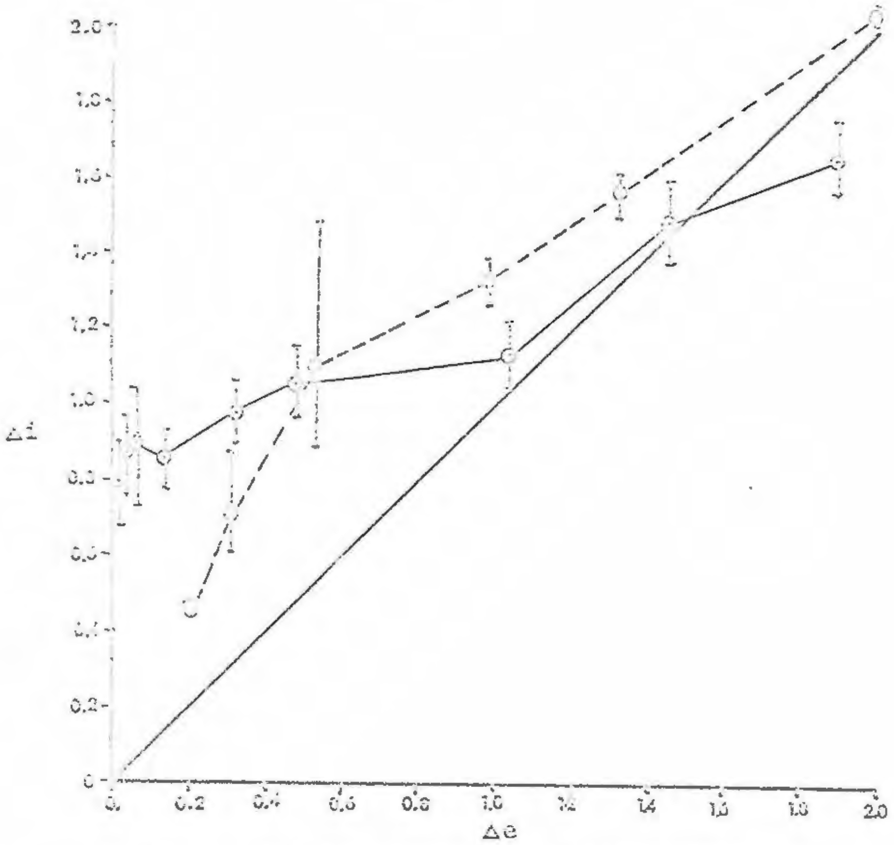


Fig. 47. Δi/Δe curves for *Melita zeylanica* (closed circles) and for *G. lignorum* (open circles). Both species were exposed for 5 days to dilutions of sea water. *M. zeylanica* did not survive below 5% sea water. Only 2 remained alive in 5% at the end of 5 days and they were almost moribund. Means of 10 *G. lignorum* with standard deviations are shown; means of 5 *M. zeylanica* with ranges shown, except at 5% dilution).

sources, rather than survey methods. Collection of this experimental data is important since it can help to explain certain distribution patterns of animals. A case in point is that for the amphipod *Melita zeylanica*. This is a ubiquitous species in both the lakes and the estuaries, except for Lake Sibayi and Lake Shengezi. Although a complete picture of its salinity tolerances is not available, laboratory findings show that it has poor control over its internal blood osmotic pressure in different external salinities compared with *G. lignorum* (Fig. 47), suggesting that it may be absent from the very low salinity lakes because it cannot live permanently in very low dilutions.

Although salinities are of prime importance in the ability of an animal to colonise any particular place, other factors must also be taken into account. A demonstration of the need for detailed knowledge of the responses of a particular species, so as to be able to understand reasons behind distribution patterns, comes from a consideration of the possible causes for the presence of G. lignorum in Lake Sibayi and its absence from Lake Nhlange.

Investigations of the distribution pattern of this amphipod in Lake Sibayi has indicated that characteristics of the chemistry of water very close to the substrate probably play a role of paramount importance in determining the distribution of the species in Lake Sibayi. The question as to whether G. lignorum is absent from Lake Nhlange for the same reason is difficult to answer. Superficially one might be inclined to say yes. It is true that the lake in general is much richer in partially degraded organic material than Lake Sibayi. The fine material over the sand in water of intermediate depths tends to show a greater oxygen demand and presumably therefore a greater carbon dioxide budget. It could therefore be argued that even in water as shallow as 5 metres, one might expect conditions to be such that the rates of carbon dioxide concentration change would exclude G. lignorum from the substrate. But this does not give any adequate reason for the lack of G. lignorum from water less than 5 metres deep. It may be that the profiles of the bottom of Lake Nhlange are important. It has been noted that the bottom of the lake shows a series of terraces set apart from each other by steeply sloping regions where the water depths change over a short distance, sometimes as much as 5 metres. Such rapid changes of depth are correlated with changes in the substrate. The sand of the shallow plains tends to be white, while the sand in 5 metres is grey and, when disturbed, stinks of hydrogen sulphide from the decaying plant material in the fine material over the sand. SCUBA divers noted that in the shallow waters up to 3.5 metres, ripple patterns were found on the sand. At 3.5 metres these tended to die away. The greater part of the substrate which has been investigated in detail above the 5 metre contour is less than 3.5 metres deep, and therefore directly affected by wave action and currents in the lake. Wave action has been suggested as a reason for the absence of G. lignorum on the shelf region of Lake Sibayi. Presumably the same would hold true for Lake Nhlange.

This leaves only a relatively small area of substrate between the 3 and the 5 metre contours where conditions must change from shifting sands, to sands covered in a skim of organic detritus. Such a small area may not be large enough to maintain a population of G. lignorum in Lake Nhlange.

From this line of reasoning it is clear that more detailed work is required at specific sites on the bottom of Lake Nhlange to establish the validity of the ideas which have evolved out of the present knowledge of the requirements of G. lignorum. A clear direction has been given as to what to measure where, but perhaps more importantly, it has shown that although the overall general physical and chemical properties of the lake are known in some considerable detail from the work of Allanson & van Wyk (1969), this has not been sufficient to answer the questions raised by even a superficial acquaintance of the biology and distribution of G. lignorum.

However, sight must not be lost of the importance of measurements of the overall physical and chemical properties of a lake. The microenvironment of the lake must depend to a large extent on the conditions in the macroenvironment. In any case, as the work on *G. lignorum* has shown, many important questions were answered by reference to the general physical and chemical characteristics of Lake Sibayi.

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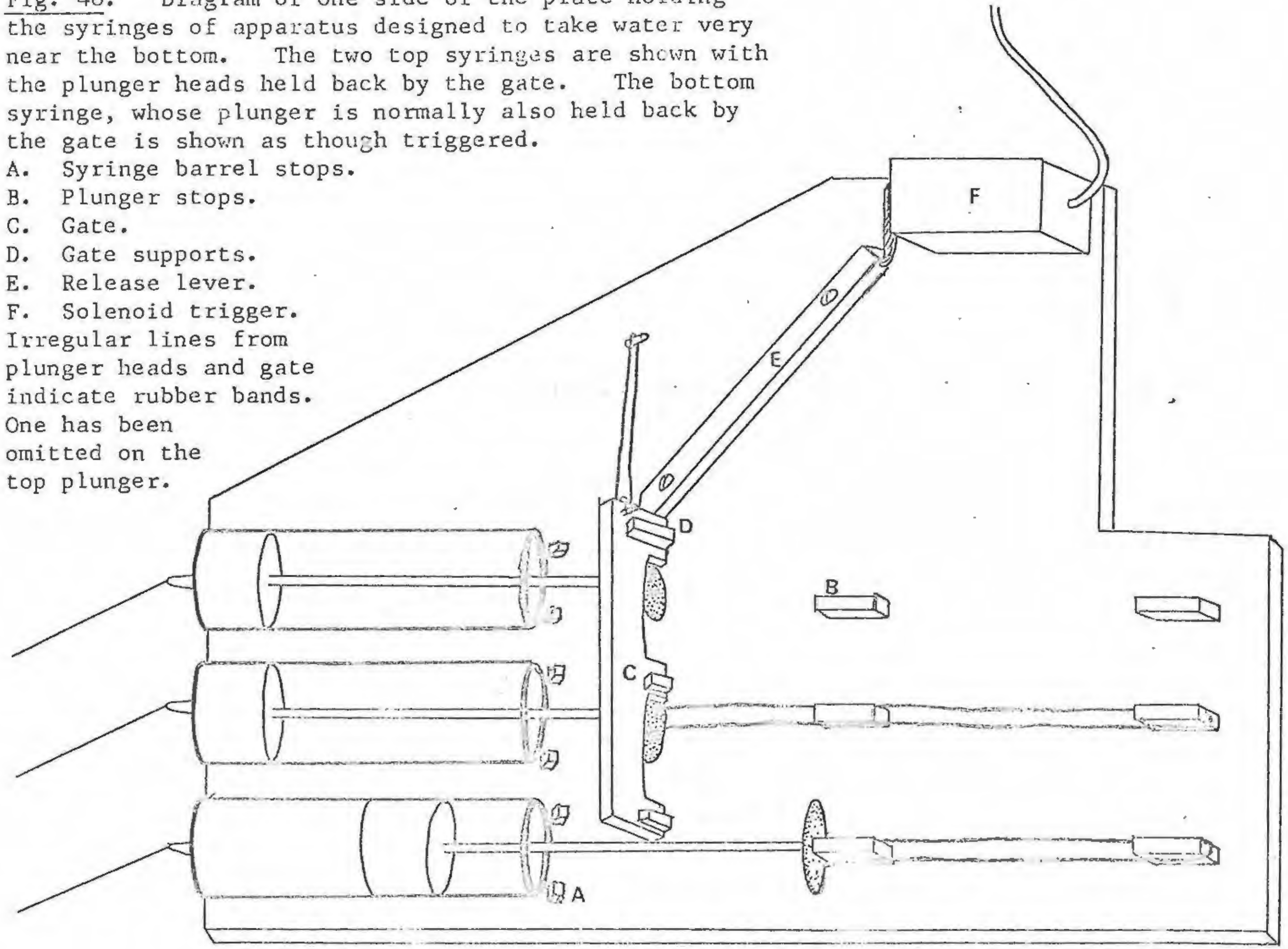
Fig. 48. Diagram of one side of the plate holding the syringes of apparatus designed to take water very near the bottom. The two top syringes are shown with the plunger heads held back by the gate. The bottom syringe, whose plunger is normally also held back by the gate is shown as though triggered.

A. Syringe barrel stops.
B. Plunger stops.
C. Gate.
D. Gate supports.
E. Release lever.
F. Solenoid trigger.

Irregular lines from plunger heads and gate indicate rubber bands. One has been omitted on the top plunger.



APPENDIX I.

APPARATUS FOR TAKING WATER SAMPLES CLOSE TO THE WATER/SUBSTRATE INTERFACE

The apparatus was constructed of perspex (Fig. 48). Three 20 cc syringes were held on to fixed positions determined by channels, by rubber bands. The shoulders of the barrels of the syringes towards the heads of the plungers were restrained by stops (A) (Fig. 48). The heads of the plungers were connected to rubber bands which withdrew the plunger head to the stop (B) allowing about 20 cc of sample to enter the syringe barrels. The heads of the plungers were restrained from moving before triggering by a gate (C) which could slide on gate supports (C). The gate was held down by a lever in turn held down by the arm on a solenoid (F). The solenoid was activated by a switch connected through an 18 volt dry battery and electric cable which at the same time acted as a tether for the instrument. The solenoid arm released the lever which in turn released the gate. The gate was pulled up from the plunger heads in such a way that these heads slipped through the arches in the gate (C). The plungers were withdrawn by the rubber bands and water samples were taken at different levels.

The upright plate holding the syringeswas fastened on to a triangular footplate arranged so that the syringes overhung one corner. The syringes were fitted with bent wide bore needles to place them as near to the substrate as possible.

The syringes and gate mechanism were duplicated on the obverse of the plate so that duplicate samples could be obtained at one time.

The apparatus was let down on to the substrate and allowed to settle for 10 minutes before being triggered. After triggering a further three minutes was allowed for retraction of the syringe plungers.

The positions of the needles were designed to be 0.5 cm, 5 cm and 9 cm from the surface of the substrate. However, the water sampled will in fact not come from as near as the nominal heights of the needles from the substrate, but will rather be recruited from all directions in the vicinity of a needle tip. In unobstructed water, the sample will be taken from an imaginary sphere equal to the volume of the sample with its centre congruent with the tip of the needle. For a sample of 20 cc, a sphere of radius 1.971 cm will be sampled. Presuming the 5 cm and 9 cm syringes to be sampling from water free to flow in from all directions, these samples will be taken from bands of water 3.1 to 6.9 cm, and 7.1 to 10.9 cm respectively.

The proximity of the substrate within the radius of the imaginary sphere of the needle 0.5 cm away from the bottom will interfere with the free flow of water into the syringe. Calculations based on the assumption that the water being sampled will come from a sphere with a segment removed of volume 20 cc, show that water will be recruited from up to 2.4 cm height away from the substrate, although the mean height of the water sample (when as much water comes from above as below that height) is about 0.9 cm from the substrate.

APPENDIX 11.

Light meter.

A lux meter was not available in the laboratory. An instrument capable of being used under water was constructed. The circuit of the light meter is given in Fig. 49. The light sensor was a Phillips light sensitive resistance. The circuit was driven by a Mallory nickel cadmium cell so as to provide a standard voltage through the circuit. The resistance of the light sensor varies as a non linear function with the amount of light falling on the sensor. The amount of current passing through the circuit therefore bears a relationship to the amount of light impinging on the sensor. The current passing through the circuit was measured by means of a μ ammeter, whose sensitivity was regulated for three different levels by using resistance shunts (R2 and R3), or for very low light sensitivities passing the current directly across the meter.

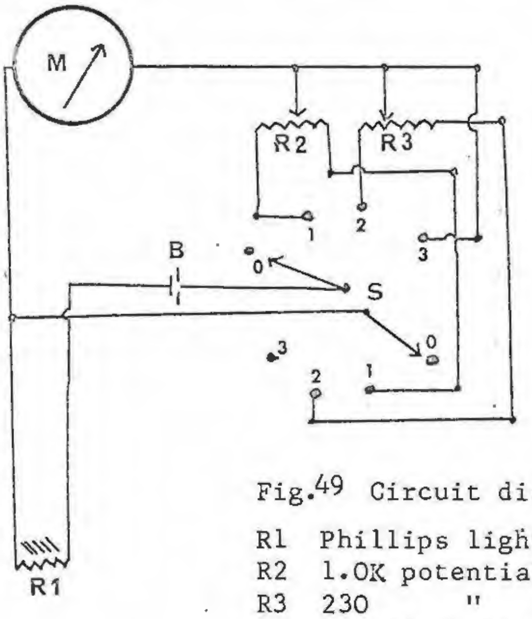


Fig. 49 Circuit diagram of light meter.

R1	Phillips light sensitive resistance.
R2	1.0K potential divider.
R3	230 " "
S	Ganged switch.
M	u ammeter.
B	Mallory battery Cadmium cell for 1.3 v.

The light sensitive resistance was calibrated against a "Lunimatt" light meter which when used as an incident light meter gave readings which could be converted to Ft candles or Lux. However, the readings were cited as only being approximate from the light meter, and the calibration therefore was not accurate.

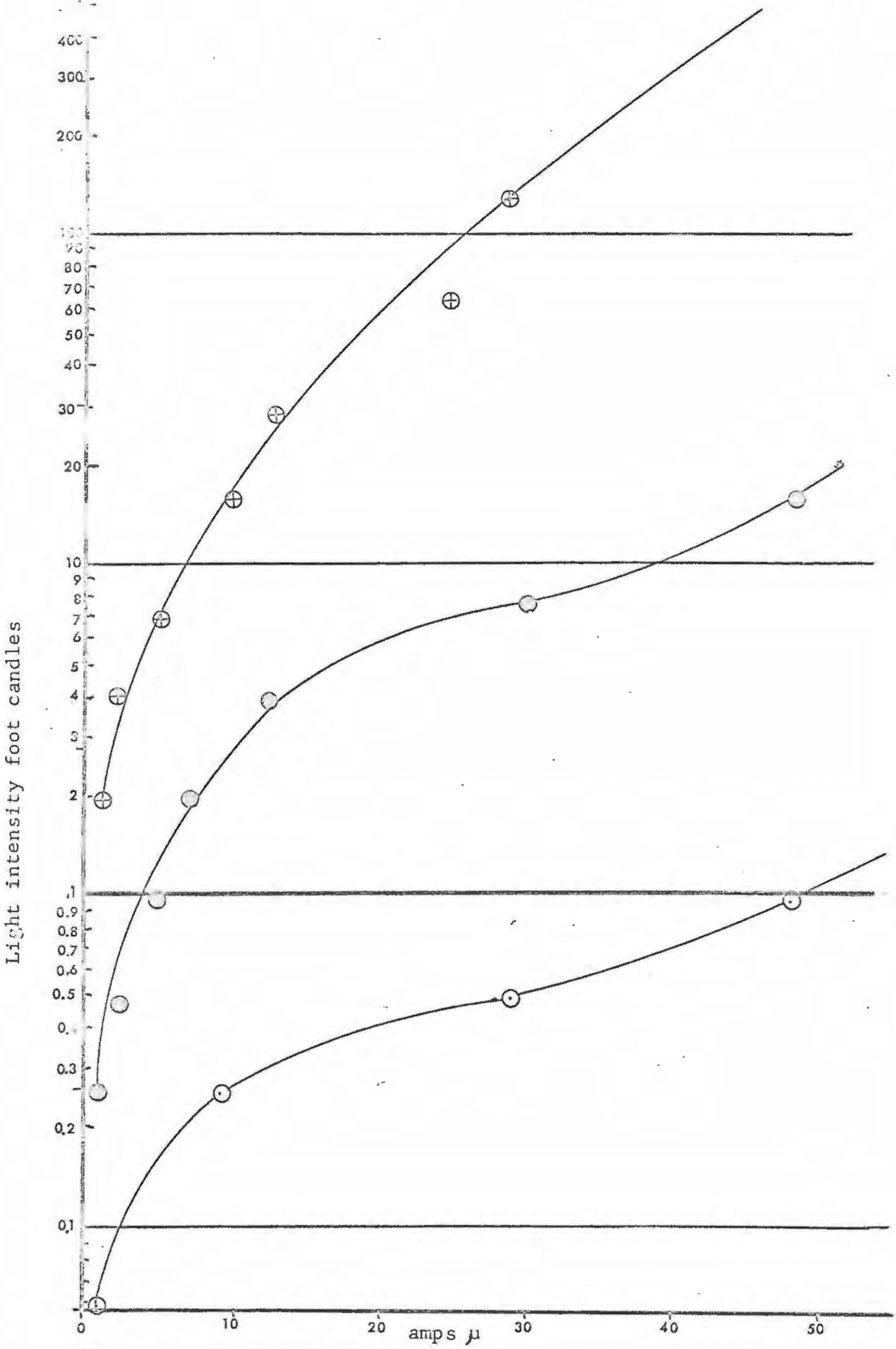


Fig. 50. Calibration curves for underwater light meter. The values are only approximate since a commercial photographic light meter was used as a standard.

However, some approximation was given, using this instrument.

The light sensitive resistance was embedded behind a window of perspex in epoxy resin, to render it water tight. Thus the sensor could measure light values under water.

The curvilinear nature of the response was very convenient, since very low light values could be estimated, indeed much lower than could be obtained using a selenium cell as a light sensitive current generator. Light values at very high intensities were much less conveniently measured, since large changes in light intensity was measured by small changes in current in the circuit. The calibration curve is given in Fig. 50.