Spring 1973

A Seasonal Distributional Survey of the Epiphytic Diatoms of the Outer Banks Region of North Carolina with Respect to Temperature and Salinity

Charles Welles Belin Jr.
Old Dominion University

136

A SEASONAL DISTRIBUTIONAL SURVEY
OF THE EPIPHYTIC DIATOMS OF THE
OUTER BANKS REGION OF NORTH
CAROLINA WITH RESPECT TO TEMPERATURE
AND SALINITY

by

Charles Welles Belin, Jr.

A THESIS

submitted to the Institute of Oceanography,
Old Dominion University, Norfolk, Virginia,
in partial fulfillment of the requirements
for the degree of

Master of Science

April, 1973

Committee

Chairman
Dr. Jacques S. Zaneveld

Dr. Harold G. Marshall

Dr. John C. Ludwick

ACKNOWLEDGEMENT

I would like to acknowledge and gratefully thank for their careful reading, and thoughtful help in the preparation of this thesis the following persons: Dr. Charles W. Reimer, Academy of Natural Sciences, Philadelphia, for his assistance in the identification of some species; Dr. Harold G. Marshall, for his kind criticism and the use of equipment of the Biology Department, Old Dominion University; Dr. John C. Ludwick, Director of the Institute of Oceanography, Old Dominion University, for his help with statistical expressions; and Dr. Jacques S. Zaneveld, Institute of Oceanography, Old Dominion University, for his continued direction and critical help at all phases of this thesis. Of particular assistance in the proofreading of this report have been Nancy I. Belin and Constance R. Belin. Additional thanks go to Susan P. Rasmussen for the typing of this report.

ABSTRACT

This study was initiated to investigate epiphytic diatoms at selected stations along the North Carolina coast and bay region. The relationships between these organisms and the season of the year and salinity and temperature of the water in which they were found are considered. The special affinity between the epiphytic diatoms and their host plants is also discussed. The specific data were analyzed statistically via linear correlation coefficients, graphs of least squares-best fit, and multiple linear regression analysis.

From the statistical tests, it was found that the mean number of epiphytic diatoms shows a significant multiple linear regression with respect to the pooled effects of salinity, temperature and season of the year. There appears to be a direct relationship between the presence or absence of *Synedra pulchella* and *Cocconeis scutellum*.

There is indicated a direct relationship between the number of diatoms present on any particular host and the surface configuration of that host.

In the limited area studied there is a pronounced gradient of epiphytic diatoms that appears to follow not only latitudinal but also seasonal and grazing pressures.

TABLE OF CONTENTS

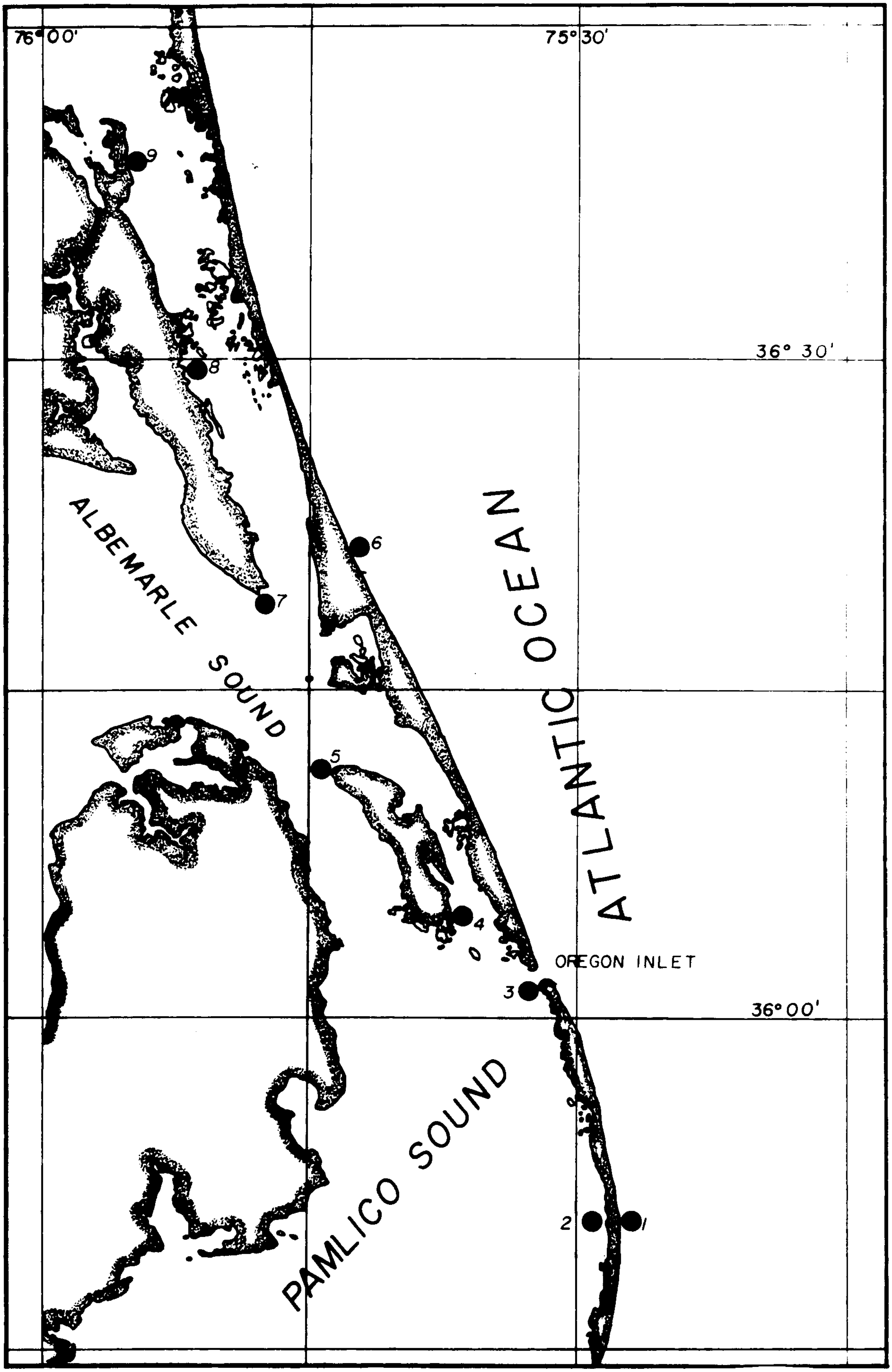
TABLE OF CONTENTS (CONT')

LIST OF TABLES

LIST OF FIGURES

LIST OF FIGURES (CONT'D)

Figure 1: Thesis study area: Stations are numbered arbitrarily.



76°00'
75°30'
36° 30'
36°00'
ALBEMARLE SOUND
ATLANTIC OCEAN
OREGON INLET
PAMLICO SOUND
1
2
3
4
5
6
7
8
9

I. PROLOGUE.

A 13 month study of the epiphytic diatoms occurring in the upper eulittoral zone from Rodanthe to Waterlily, North Carolina, was undertaken from June, 1971 to July, 1972. During this period, samples were recovered from each station during September, 1971, January, March, April, and June, 1972. These were preserved in buffered formaldehyde and treated according to the Swift (1967) method, as modified by Belin and Zaneveld (1972), to strip the available diatoms from the host plant parts. These diatoms were examined, identified, and photographed. Some physical data of the water at each station were taken and many meterological conditions were noted.

Epiphytic diatoms are common inhabitants of the marine coastal and estuarine habitats. Extensive algal and diatomaceous studies have been reported from New Jersey northward and from Beaufort, North Carolina, southward (Hoyt, 1920; Williams, 1948; Hustedt, 1955; Patrick and Reimer, 1966). The epiphytic diatoms of the outer banks region have been studied little (see page 17). Possibly they represent a biological index to changing environmental conditions.

The particular region of the North Carolinian Outer Banks as a study area was selected for three reasons. First, the area of a barred estuary provided a variety of environmental niches in which the occurrence of diatoms could be correlated with regard to their substrate, the host plants. Hence, two stations were established on sandy, neritic beaches; one was an ocean inlet to a sound; three were estuarine; and three were estuarine-lacustrine. Second, the location of these stations was accessible by means of the available research facilities to permit facile and frequent collections. Lastly, and perhaps most important, these stations represent an area that has not been widely studied in the past. Nonetheless, the Outer Banks region is an algal transitional zone comprising, during portions of the year, salinity and temperature characteristics of cooler North Atlantic regions due to the Labrador Current, of subtropical regions provided by the Gulf Stream influence, and combinations of these due to their mixing (Humm, 1968).

II. IDENTIFICATION OF MACROSCOPIC ALGAE AND DIATOMS.

The identification of the various macroscopic benthic algal and marine hosts that were collected during the period was based on Taylor's (1964) standard text. Phanerogamic hosts were identified using the text of Gleason and Cronquist (1963). Preliminary identifications of diatoms were made using Peragallo (1897) and Hustedt (1930). Subsequently, the species were compared with the standard and type collections of the Limnology Department, Academy of Natural Sciences, Philadelphia.

III. DESCRIPTION OF COLLECTION AREAS.

The Pamlico-Albemarle-Currituck Sound complex along with the associated waters occupies a "drowned" portion of the North Carolina Coastal Plain region (Pickett and Ingram, 1969; National Estuary Study, 1970). Numerous rivers and creeks feed into this shallow basin where fresh and salt water mix. This barred estuary comprises the second largest estuarine complex on the east coast of the United States (the Chesapeake Bay, an open estuary, being the first). It extends for a distance of 272 km and has a maximum width of 48 km.

Ecologically, the area is primarily controlled by the freshwater inflow from the river systems of the region, which mix with the salt water entering via Oregon, Hatteras, and Ocracoke Inlets. In this region are a variety of bottom types, wind and oscillating tidal currents, and unidirectional freshwater flow bringing nutrients from the uplands. These factors contribute to the biological productivity of the area (National Estuary Survey, 1970).

The survey area is characterized by shallow water which ranges from a maximum depth of 2 meters in Currituck Sound and 6 meters in Pamlico Sound to only a few centimeters on the myriad of shoals. The substrate near the mouths of rivers and at the head of Currituck Sound are composed primarily of mud and organic debris. The deeper regions of Albemarle and Currituck Sounds are characterized by sand and mud bottoms. The ocean beaches are exclusively quartz sands. The shoal areas are comprised of mud and sand flats (Pickett and Ingram, 1969).

Numerous authors (Pickett and Ingram, 1969; Roelofs and Bumpus, 1953) have observed there to be little or no lunar tides in the sounds except in and near the inlets. Currents diminish quickly as one proceeds into the sounds. Tidal heights vary from a few centimeters in the more restricted regions to a maximum of one meter near the inlets.

The lunar flood tides found near the inlets transport into the estuarine regions the salt water of the coastal areas. Vertical mixing with river water produces low salinity gradients that range from freshwater in the backwater reaches of Currituck Sound to coastal saltwater in the inlets (Hobbie, 1969).

The region has a temperate climate. January has the lowest mean air temperature of 4.4°C while August has the highest, 29.4°C. These air temperatures are similar to the surface water temperatures recorded on the ebb and the flood tides in the inlets (National Estuary Survey, 1970). The average annual rainfall is between 114.3 and 127.0 centimeters.

The diversity of species and high productivity of the area make it important economically. First, there are numerous commercially productive oyster beds in the shallow waters near the mainland from Albemarle Sound south to Pamlico Sound. Salinity ranges from 10 to 30 °/oo, and there is a firm substrate. Besides oysters, some clams and bay scallops are in the region.

Second, the numerous waterfowl nesting areas (Pea Island and Knotts Island) within this region draw tourists and hunters and serve as economic potential.

Third, in this region is a shrimping and crabbing industry based at Wanchese that has developed over a million dollars revenue during the period 1959 to 1963.

Lastly, the commercial and sport fishing industry of this region provides a large source of income, coming from the sale of the fish caught, and from the resulting tourist trade (National Estuary Study, 1970).

Thus, this area is economically important. Utilization of the area by the public will increase in the future. If it is to retain its productive nature, it is imperative that studies be conducted on those organisms which are readily affected by adverse conditions, which contribute to the primary productivity of the area, and on which larger organisms feed.

To this primary productivity, the epiphytic diatoms are likely to contribute significantly (Raymont, 1963; Wood, 1965). Although estimates, both quantitative and qualitative, of the utilization of the epiphytes by larger organisms have been made, no consideration has been made for zooplankton grazing nor for those epiphytes that have been mechanically removed from their host (Wood, 1965). Thus, in an effort to study the development of the primary production of phytoplankton, it is wise to learn as much as possible about the members of the epiphytic plants, including the epiphytic diatoms. Knowledge of their distribution with respect to temperature and salinity over a year's cycle should provide additions to our understanding of the conditions of this region.

I. STUDIES OF EPIPHYTIC DIATOMS.

One of the earliest studies of epiphytic diatoms began with the work of Cleve (1894). This work was a successful examination of one class, the Raphideae. Cleve's identifications are based on morphological characteristics which occur in the greatest number of species, among them frustule shape, striae number, and symmetry. He discounted the prior identification criteria that are obvious on examination but which may be found only in a limited number of species.

Cleve stated that it is not correct to distinguish species that are strictly epiphytic and others that are strictly free-floating. Such a distinction, he feels, is based on the fact that "many of the attached or enclosed forms also live in a free state; and that there are frequently very slight differences between species, which live attached or enclosed, and others which may never occur in such a state." Therefore, he regards "as a characteristic of very little importance the mode of occurrence in free or attached state" (Cleve, 1894).

Patrick's (1948) study of diatoms was an extremely useful compilation of the work of numerous authors since Cleve's (1894) publication on the naviculoid diatoms. A general discussion of the important ecological factors affecting diatoms is presented, followed by separate, limited portions dealing with such parameters as salinity, temperature, viscosity, light, depth, and nutrients. At the time of publication, it would appear that Patrick had produced the best publication on the factors controlling the distribution of diatoms. Her work places emphasis on "all phases of the ecology of living diatoms from all environments"

(Lohman, 1957).

Patrick (1948) makes specific reference to epiphytic diatoms in her report on the distribution of these organisms. She indicates that the epiphytic mode of life must be considered and, in her report, a specific category is created for those "which attach themselves by a secretion of jelly to the substratum". Some of these organisms may use this mucous as a cushion, as in the genera Cocconeis and Achnanthes; others employ a stalked structure at the end of which are found the diatoms, as in the genera Cymbella and Licmophora. She indicates that the most common sites for attachments are rocks, algae, marine fungi, Sphagnum, and many of the higher aquatic plants.

Castenholz (1960a, 1960b, and 1963) refers to a number of epiphytic diatoms. He describes the denuding of numerous concrete blocks and their subsequent recolonization by species of Rhodophycophyta, Phaeophycophyta, and Bacillariophycophyta. This recolonization study is interesting from the aspect of adaptability of these organisms. There is recognized a specificity of some epiphytic diatoms for particular plants, especially for algae (see also Aleem, 1950). However, when forced by the denudation process, these supposed epiphytes readily attach to other nonfloral surfaces, such as the concrete blocks (Castenholz 1960b).

Castenholz (1963) also indicates that the intensity of solar radiation is the major factor in determining the upper limits of the settling of diatom epiphytes.

Carpenter (1970) studied the epiphytic diatoms from oceanic samples of Sargassum natans and S. fluitans. His investigations found a rich community of eight species of *Mastogloia*, and one species each of *Cyclotella*, *Cocconeis*, *Nitzschia*, *Synedra*, and *Navicula* (see Figures 4 and 18). Carpenter offers a possible explanation for this rich community where nutrients of the aquatic environment are relatively low (i.e., the Sargasso Sea). He found that the nutrient concentration in very close proximity to the *Sargassum* plant is two to three times higher than in the adjacent waters, and he indicates that "it is possible that the *Sargassum* acts to provide a favorable environment for epiphytes".

An extensive study of the seasonal variation in the littoral epiphytic flora of the Ouse estuary in England was completed by Hopkins (1964). He concluded that the single major factor in the death of intertidal epiphytic diatoms was "the (high) air temperature in conjunction with desiccation". He indicated that although the heat may not be deadly to the organisms, the desiccation produced and accelerated by the higher temperature was tantamount to death.

In view of some of the chemical and physical measurements taken from this estuary, it would appear that the environmental conditions of the Ouse are comparable to many areas of the Outer Banks region, under examination in this paper. During the late winter, 1954, the salinity range in the Ouse varied between 20 °/oo and 29°/oo, while during the summer, after heavy rainfall, this value fell to 9 °/oo. The air temperature values ranged between -9.0°C in the winter and 33.0°C during the summer. Sea surface temperatures varied between the winter low of 5.0°C to a high of 17.7°C in August of 1954. The substrate examined were concrete blocks, chalk banks, wood, and species of macroscopic benthic algae.

Although Castenholz (1963) noted that a preliminary study of diatoms epiphytic on large algae showed no pattern of interest, Hopkins (1964) indicated that the occurrence on the host plant shows a definite pattern of succession and distribution with respect to exposure. In particular, a listing of abundant and lesser found species attached to Fucus spiralis, Fucus vesiculosus, Fucus serratus, Dictyota dichotoma, Enteromorpha spp., Ulva lactuca, Cladophora rupestris, and Ceramium rubrum is presented.

One of the dominant presence and absence factors offered by Hopkins (1964) is the average air temperature which influences a widespread decline in the frequency of a number of littoral epiphytic diatom species. He found that, while attached to algae, the following temperatures produced declines in the numbers of epiphytic diatoms:

DIATOM SPECIES	°C
Fragilaria striatella	18 - 20
Melosira moniliformis	18 - 20
Melosira nummuloides	10 - 15
Grammatophora marina	20 - 22
Achnanthes brevipes	Summer Maximum
Licmophora (5 spp.)	Summer Maximum
Synedra (4 spp.)	20 - 22

Table 1. Temperatures producing declines in numbers of some diatom species.

Among the diatoms epiphytic on algae, the most common were Melosira moniliformis, Grammatophora marina, numerous species of Licmophora, four species of Synedra, and Striatella unipunctata (Hopkins, 1964).

Hopkins divides the epiphytes into three categories based on their habit. The first group includes the filamentous forms such as Fragilaria and Melosira that may attach directly to the substratum or may be found in association with the algae or the substratum. Second, there are numerous solitary forms such as Cocconeis and some species of Synedra attached to the host plant. Lastly, one may distinguish the free-solitary diatoms of the mud that are thought to be carried to the region of recovery through the efforts of the tide. It is felt by Hopkins that these forms are to be considered true epiphytes due to the accumulation of silt and mucilage by which they attach themselves, and also to their recovery from algal thalli.

II. FRUSTULE CLEANING METHODS.

In order to facilitate the identification of diatoms, the encrusting growth must be removed. Numerous pieces of detritus as well as fungi and members of the Cyanophycophyta may obscure the detailed structure (Patrick and Reimer, 1966). The method must be carefully controlled to prevent the destruction of the less silicified species found in tropical waters.

Many processes are explained by Van Heurck (1896), and Boyer (1916), ranging from boiling in nitric acid, sulfuric acid, and hydrochloric acid (Van Heurck, 1896), to boiling in a water bath with brown soap (Boyer, 1916) and digestion with pancreatin (Swift, 1967).

In spite of these procedures, a new mechanism for cleaning the frustules and destroying the organic material on them has been developed. Armstrong, Williams, and Strickland (1966) found that after twelve hours of irradiation from an ultraviolet lamp, the encrusting material had been oxidized.

This procedure of oxidation via ultraviolet irradiation was further modified by Swift (1967) in the preparation of frustules for electron microscopy. Swift added five drops of 30% H_2O_2 per fifty milliliters of diatom solution. He placed this new solution "six centimeters from an air-cooled 1200 Watt mercury arc lamp of thirty centimeters length. The temperature in the tubes did not exceed 61.0°C during irradiation". It was found that only two hours of irradiation was needed to clean thoroughly the frustules. It is unfortunate that Swift makes no mention of the wave-lengths from his lamp.

Carpenter (1970) in his examination of the epiphytes on *Sargassum natans* and *S. fluitans* used Swift's method to strip these diatoms from the algae. He found that this procedure worked well and that "after oxidation, *Sargassum* was visibly eroded and attached diatom frustules were released into the water".

Belin and Zaneveld (1972) also examined the attached diatoms found on *Sargassum*. This was performed on a quantitative as well as qualitative level. It was found that the Swift method provided an efficient, fast, and simple means for recovering nearly all diatoms. An average of 96% of the diatoms were stripped from the plant parts irradiated. Belin (1972) also comments that "this procedure is easily moderated to accomodate the more fragile species of algae. Merely by increasing or decreasing the time of irradiation or the hydrogen peroxide concentration, the fragile and the hardy algae can be treated equally."

The Swift method, as modified by Belin and Zaneveld (1972), and as explained below, was used for the preparation of material in this thesis.

III. INVESTIGATORS OF THERMAL ADAPTABILITY OF DIATOMS.

Numerous authors have studied temperature reactions of organisms and have reported its effect on their distribution (Setchell, 1920a; Zaneveld, 1937; Chapman, 1962; Patrick and Reimer, 1966; Zaneveld, 1969).

Setchell (1903, 1920a, 1920b) examined the effects of temperature on algae. One of his important concepts described algal species being confined via temperature to specific isotheres (the mean maxima for the hottest month) of 0°, 10°, 15°, 20°, and 25°C. He also felt that consideration should be given to isocrymes (lines of equal mean minima) of 0°, 10°, 15°, 20°, and 25°C, as demarcation points in marine flora.

Both Setchell (1920b) and Patrick (1948) indicate that a species will be found in a particular temperature zone which is not only most compatible with reproduction but also has its greatest effect in the germination of spores.

Bernatowicz (1958) comments on how Hopkins expanded this temperature relationship with reproduction to include four combinations in which temperatures limit aquatic floral distribution. Patrick (1948) indicated that the higher the latitude, the more species and individuals abound in the plankton, although many exceptions reveal that higher latitudes are no more productive annually than similar areas in lower latitudes.

Pratt (1959) concludes that the seasonal dominance of many species, especially Skeletonema costatum, can be correlated with temperature, indicating thermal preferences, both eurythermal and stenothermal. It would seem that Setchell (1920b) might take exception to the abundance of eurythermal organisms.

Pratt further states that the flowering of diatoms found in Narragansett Bay must necessarily precede the establishment of vertical mixing in the water column.

Castenholz's (1963) study of the vertical distribution of littoral marine diatoms indicated that their limits were controlled by the consequences of exposure to insolation - namely desiccation, high temperature, high light intensity, and ultraviolet radiation. Further, the degree of exposure to direct solar radiation during low tide was the most critical factor in determining this upper limit as well as the density of the diatom cover and the enumeration of species.

According to Hopkins (1964) the numbers of epiphytic diatoms were not affected by low seawater temperatures (4.4°C) provided there was no death of the host. However, when the temperature in the spring rose above 6.0°C there was a pronounced increase in the number of these epiphytes.

Hopkins indicated that for *Fragilaria*, *Melosira*, and *Grammatophora serpentina*, the maximum air temperature was the direct controlling factor with respect to distribution. Temperatures above 20.0°C produced a dramatic decline in the frequency of *Licmophora* and *Synedra* species.

Zaneveld and Barnes (1965) stated, "The appearance and disappearance of (these) algae are closely related to the changes in temperature of the surface waters." They imply that air temperature does play an important role in the maintenance of algal growth.

Chapman (1962) revealed that temperature is a major factor in biogeographical distribution of macroscopic algae. But his conclusion is that this operates in an "all or nothing" manner in that the correct temperature range permits reproduction and growth of the organism. If this temperature range is not present, there is no reproduction. Chapman does indicate though that certain species may be found in areas incompatible to reproduction. This would appear to have a moderating effect in that temperature may allow certain subtropical species, for instance, to flourish in temperate or subtropical regions (Chapman, 1962; Zaneveld, 1966). This may be exemplified as either a vertical migration along the shore face or an extension or reduction of geographical range during the seasons.

According to Humm (1969), the temperature of the surface waters is the primary factor controlling the distribution of macroscopic algae along the Atlantic coast of North America. His studies show that two major geographical floral units are the suppliers of the inshore waters of this area. One of these units is found in the tropics, bound on its northern side by Cape Kennedy, Florida, and fed northward by the Gulf Stream. The second unit is concentrated in the colder temperate waters of the North Atlantic and has its southern boundary in the waters off Cape Cod. It is fed southward by the Labrador Current. Humm states that the macroscopic algal flora found between these regions is representative of the temperature of the water in which they are found, be it predominantly of a northern origin or of a southern origin.

Mulford and Norcross (1971) found that temperature played a significant role in controlling the distribution of unicellular neritic algae in Virginia coastal waters. The cycle of temperature, expressed as seasons, contributes towards the production of very high cell counts per liter in the spring and early summer months - i.e., May, June and July.

Thus, temperature is considered by most investigators a factor of prime importance in controlling the distribution of algal species. However, there exists disparity as to whether air or water temperature exerts the greater effect on intertidal organisms.

IV. INVESTIGATIONS OF SALINITY ADAPTABILITY OF DIATOMS.

Frequently, the growth effects of temperature on algae cannot be separated from those of salinity. Salinity acts in many cases as a bio-barrier to the zone-invasion of many species (Williams, 1948). As indicated by Hoyt (1920) and later by Williams, salinity is one of the major variables causing seasonal variations in the aquatic flora of some particular regions of the sea. The area under investigation experiences quite stable salinity ranges with respect to season changes.

Patrick's (1948) report of the conditions contributing to the spatial distribution of diatoms deals with salinity in great detail. She indicates that it is recognized that diatom species are specific with respect to their salt requirements. In fact, an early classification of these organisms was made on the basis of this requirement. Patrick discusses how Gran and Hjort pointed out that the fluctuations of diatoms noted in the Lemfjord region feeding into the North Sea was predominantly due to the salinity gradient between these two regions. Patrick also recognized that ocean currents, having different salinities from surrounding waters, often contain separate and distinct flora communities which may retain their identity for thousands of kilometers.

Thus, a predominance in March of Skeletonema costatum and Chaetoceros socialis is succeeded in July by Rhizosolenia alata. As indicated, this is most probably due to particular requirements of each species for particular salinity, nutrient, and light conditions (Patrick, 1948).

Patrick (1948) discusses Kolbe's system of grouping diatoms according to their salt needs. These were divided into polyhalobiens, euhalobiens, mesohalobiens, and oligohalobiens. Although this system was successful, it became apparent that individual species of one genus responded to different salt tolerances. Navicula longirostris is a polyhalobien, needing salinities higher than sea water (i.e. 45-48 °/oo), while Navicula cincta has its optimum condition in water with a very low salt concentration (i.e. 1-3°/oo).

Whitford's (1956) report of algae indicated that in the springs and spring streams of Florida, salinity is one parameter controlling the distribution of diatoms. However, Whitford appears to be of the opinion that the salinity of the water operates in either a presence of absence manner, allowing or preventing diatom development.

Pratt (1959) studied the phytoplankton of Narragansett Bay and alluded to a dependence of the diatoms, as well as other phytoplankton, on salinity ranges. He noted that there is a stratification of relatively higher salinity water along the bottom of the Bay's three channels while the fresher waters remained at the surface. This, he thought, has a profound effect on the flora of the area.

An extensive floristic study by Castenholz (1960a and 1960b) was undertaken in several saline and fresh-water lakes in the northwestern United States. The relative salinity of these lakes was found to be an extremely important contributing factor in the distribution of diatoms (Castenholz, 1960b).

The study of the Ouse Estuary by Hopkins (1964) considered the seasonal variation of epiphytic diatoms. Again, the implication is that the salinity of the water has an "all or nothing" effect on the diatoms allowing their growth at some concentrations and preventing it at others. While Hopkins indicates this, he also feels that the effect of temperature ranges, nutrient concentrations, and duration of insolation modify the fundamental control by salinity.

The contributions of Zaneveld (1937, 1964, and 1969) are noteworthy in the area of delimitation of macroscopic littoral benthic algae. He states:

> "The upper limits of algal zones in the eulittoral region are primarily determined by the resistance of the species to loss of water during midday exposure, by the effects of desiccation upon metabolism, by the ability to adjust their osmoregulatory processes during the periods of exposure to the rapid changes of salinity, and by the degree of tolerance to diurnal and seasonal changes of air temperature." (Zaneveld, 1969).

Although in certain restricted regions, other parameters such as substrate composition, meterological conditions, and pollutants may control the distribution of diatoms, diurnal and seasonal temperature ranges, as well as the influence of salinity changes, control their distribution over a broad area, such as the east central coastal region of the North American continent (Zaneveld, 1969).

Pickett and Ingram (1969) indicate that the salinity characteristics of the Pamlico, Albemarle, and Currituck Sounds are extremely stable over a yearly cycle, except for those inlet areas, expecially Oregon Inlet. This would allow greater proliferation of indigenous species due to more constant water conditions (Wood, 1965).

Carpenter (1970) studied the phytoplankton of the Cape Fear River Estuary, North Carolina. Although temperatures and salinities of the region were taken for one year, there is little correlation with the phytoplankton counts. Carpenter does indicate that the numbers of diatom individuals increased from upriver regions to downriver areas. One could assume that the salinity also increased downriver, although this fact is not presented.

The salinity values taken by Mulford and Norcross (1971) with phytoplankton samples of the shelf waters east (35 km to 130 km) of Chesapeake Bay show seasonal fluctuations due to the variable land freshwater runoff. Salinity maxima occur in winter months, which typically have more diatoms than the spring or summer months when salinity values are at a minimum.

The studies by Marshall (1969a, 1969b, and 1971) concerning phytoplankton distributions are accompanied by salinity and temperature values of the shelf waters off North Carolina. Marshall (1971) has listed a seasonal expression of the phytoplankton that are found in these waters, which includes an indepth examination of the phytoplankton successional patterns as seen in the central Atlantic states' shelf waters. The importance of salinity is evident in various distributional matters. One of the important factors present in Marshall's study is its inclusion of five years of data and collections. The seasonal vicissitudes were kept at a minimum.

V. CLASSIFICATION OF DIATOMS.

Numerous systems have been devised to classify diatoms (W. Smith, 1853; Ralfs, 1861; Schutt, 1896; Mereschowsky, 1901; Hustedt, 1930;

Silva, 1962; Hendey, 1964; Patrick and Reimer, 1966). All but the latter three are based on artificial characteristics. Although the systems of Silva, Hendey, and Patrick and Reimer cannot be considered completely natural systems, these are far superior to those of the past.

Patrick and Reimer (1966) have used a combination of the classification of Silva as well as that of Hendey in which the diatoms were considered a single class of the Chrysophyta. Their classification is as follows:

Division - Bacillariophyta

Class - Bacillariophyceae

Order - Fragilariales

Order - Eunotiales

Order - Naviculales

Order - Bacillariales

Order - Surirellales

Order - Eupodiscales

Order - Rhizosoleniales

Order - Biddulphiales

In this paper the author will use the Patrick and Reimer system of classification.

I. RECOVERY OF HOST ALGAE.

Five trips were taken to the Outer Banks region of North Carolina where samples of aquatic plants were obtained. Four samples of plants from each of nine stations were taken in September 1971, January, March, April, and June 1972.

These host plant samples were rinsed gently in distilled water in the field to remove any pelagic or neritic diatoms that may have been present. They were then placed in fifty milliliter plastic pharmaceutical vials to which were added three milliliters of buffered formaldehyde and forty milliliters of distilled water to give a final concentration of 6.8%. The vials were then inverted numerous times to thoroughly mix their contents.

After the collection of samples, the physical and meterological data at the station were recorded. Included were the salinity of the water, the water and air temperature, the time of the collection, the distance of the host plant from the high water mark, the percent of cloud cover, the wind direction and speed, and the type of substrate of the host. The time of high tide for the general area (Oregon Inlet) was read prior to the collection in the current tide tables of the U.S. Coast Guard.

The samples were then transported to the laboratory.

II. IRRADIATION OF THE SAMPLES.

In the laboratory, the formalin solution was replaced with distilled water. To this was added eight drops of thirty percent hydrogen peroxide (H_2O_2). The vials were filled with distilled water to cover the plants. They were then placed approximately twenty-five centimeters from a Raytech Ultraviolet Lamp (Model LS-7, 60 cycles, 115 volts, 0.35 amperes) and were exposed to both long and short ultraviolet irradiation for a period of twenty-four hours. The simultaneous wavelengths emitted were 404 nm, 388 nm - 338 nm, 312 nm - 310 nm, and 252 nm. During this time, the vials were periodically capped and inverted several times to mix thoroughly the contents and to insure that all the host plant surfaces received equal irradiation.

At the end of the twenty-four hour period, the host plants were carefully removed from the vials and rinsed in distilled water. The rinse water was allowed to flow back into the irradiation beakers. The treated host plants were placed in tared Petri dish halves and were air dried for seven days. The gram weights of the dried plants were determined to the fourth decimal place on a Mettler Analytical Balance. These weights were then recorded for use in the computation of the diatom density per species of host plant.

The rinsed diatom solution was strained through a 1.5 mm mesh sieve in order to rid the solution of any minute pieces of algal thalli. The diatom solution was allowed to settle in the irradiation beakers for seven days to insure that the preserved diatoms sank to the bottom. At the end of this time, all of the solution except for five milliliters was suctioned from the vials, using a General Electric, one-sixth horsepower vacuum pump with a negative pressure of six inches of mercury.

The concentrated diatom liquor was then placed in separate ten milliliter vials for storage.

III. SWIFT METHOD OF CLEANING DIATOM FRUSTULES.

Since the characteristics of the frustule play an important role in the identification of diatoms, a variety of procedures have been used in the past to clean them in order to facilitate their identification (Belin, 1972).

Swift (1967) found that if irradiation with ultraviolet light was combined with a strong oxidizing agent, such as hydrogen peroxide, the frustules were cleaned effectively. Varying degrees and duration of oxidation have been proposed for various amounts of growth on the frustules. Quantitative methods of cleaning must be achieved so that the more fragile species which tend to have less growth can be treated less vigorously than the more encrusted ones.

The present author obtained superior results with _Sargassum fluitans_ recovered from the western North Atlantic, which was cleaned by irradiation for a period of twenty-four hours with eight drops of hydrogen peroxide per fifty milliliters of solution (Belin, 1972). Another result of this quantitative and qualitative study was that the degree of oxidation of the algae thalli could be controlled merely by increasing or decreasing either the time or irradiation or the concentration of hydrogen peroxide.

The Swift method, as modified by Belin and Zaneveld (1972) was the procedure employed for the preparation of material in this thesis.

IV. PREPARATION OF PERMANENT MICROSCOPE SLIDES.

From the stirred concentrated diatom solution, two drops were placed on a cleaned coverslip according to the procedure outlined by Patrick and Reimer (1966, page 95). This coverslip was allowed to air dry. Clean glass microscope slides were prepared and labeled. On each of these were placed two drops of Kleermount (toluene-based) Mounting Medium (see Figure 2, page 39). These slides were examined and diatom species were identified and counted.

V. PREPARATION OF HERBARIUM SAMPLES.

Samples of the plants that were used in the irradiation procedure were removed prior to this treatment and were used for voucher specimens. While one of the four samples taken from each station was used for these specimens, one was used for irradiation and two were kept as replacements should accidents happen.

A complete set of voucher specimens is present in the herbarium of the Institute of Oceanography, Old Dominion University, Norfolk, Virginia.

VI. IDENTIFICATION OF THE DIATOMS.

A binocular Bausch and Lomb phase contrast compound microscope was provided by the Biology Department of Old Dominion University for the examination of the diatoms. This microscope was equipped with an Ortho-Illuminator which allowed variable intensity of light as well as sunlight, red, and green filtered light. Both light and dark phase contrast microscopy were used.

The permanent slides were examined under low power (50X) phase contrast to determine the density of the organisms present. Following this procedure, oil immersion phase power (1000X) was used to identify the species. In addition to using the standard texts and atlases of diatom collections for identification (Peragallo, 1897; Boyer, 1916;

Hustedt, 1930; Cleve-Euler, 1951; Patrick and Reimer, 1966), comparison of the specimens was made with the standard and type collections at the Academy of Natural Science, Philadelphia, through the courtesy of Dr. Charles W. Reimer.

Using high-dry, phase magnification (450X) and a Whipple disc, diatom counts were made in ten randomly selected fields on each prepared slide. A movable stage was used to manipulate these slides under the microscope. Averages of counts were made to determine species densities. Only whole frustules were counted, since positive identification could rarely be made of the bits and pieces seen. The compound microscope was equipped with an integrated thirty-five millimeter camera. Following identification using the oil immersion phase power, the species were photographed using this camera. Since presentation and preservation of these slides were paramount, these photographs were made on negative, color film producing slides. Camera lucida sketches were also made (see Appendix A).

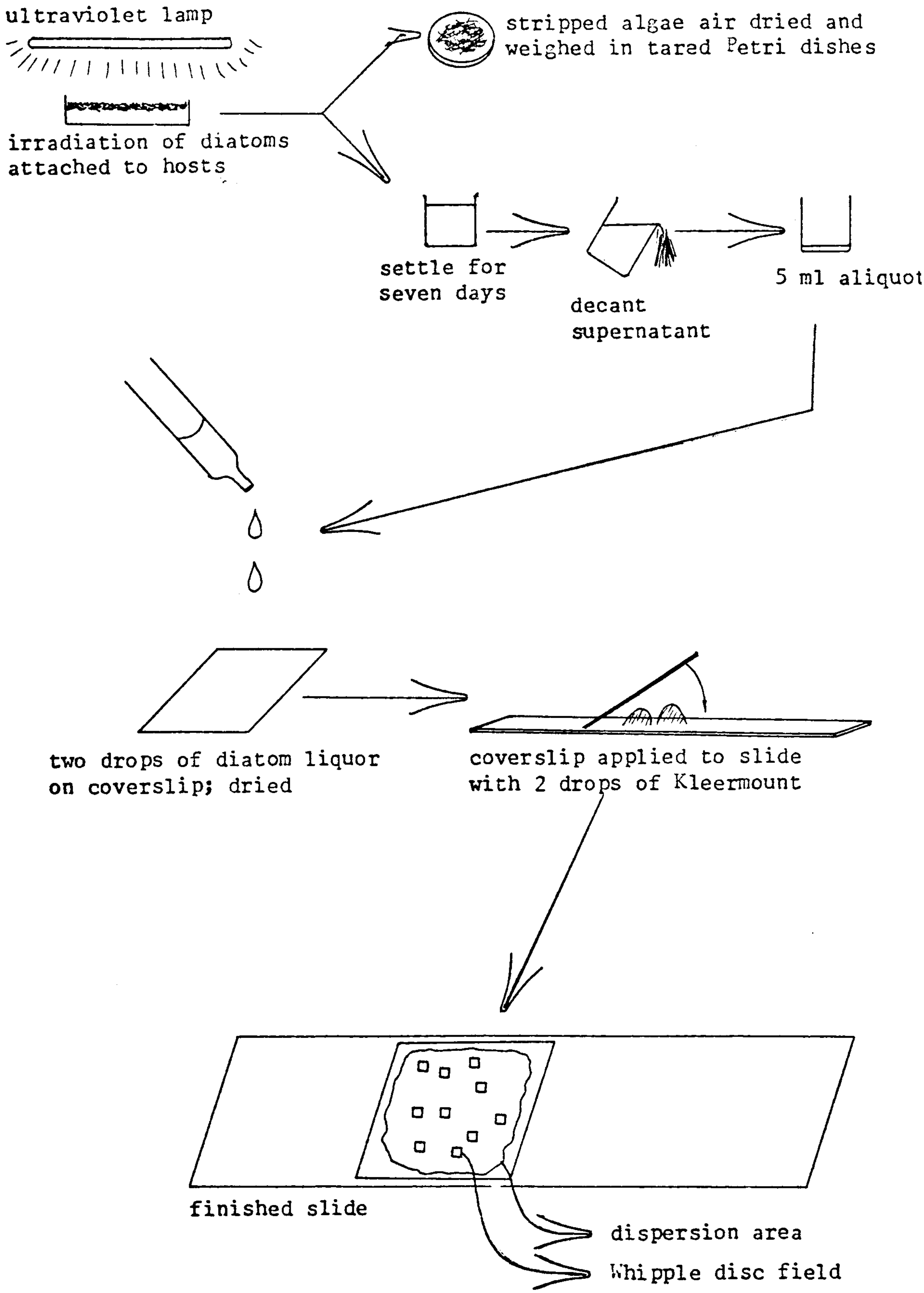
VII. COMBINING STATIONS.

The temperature and salinity ranges of the nine stations, as well as other meterological data, were examined at the end of the collection (see Figure 1). It was determined that because of the similar ranges of these parameters, the original nine stations could be combined to four types. These were: (a) the Ocean Type, comprised of Stations 2 and 6; (b) the Sound Type, incorporating Stations 1, 4, 5, and 7; (c) the Inlet Type, Station 3; and (d) the Quasi-Freshwater Type, including Stations 8 and 9.

VIII. RESTRICTION OF COLLECTIONS.

At each of these stations, efforts were made to collect the host algae belonging to the genus *Enteromorpha*, so that the correlation of the specificity of diatoms for a particular host could be discounted. This was not always possible - in fact at times there were no members of the order Ulotrichales. In this case, and where a choice was possible, the branched, cylindrical algae were chosen. In areas where no algae could be recovered, aquatic phanerogamic plants were chosen, i.e., *Zostera marina* and *Myriophyllum pinnatum*.

Figure 2: Diagram of Preparation of Microscope Slides:



ultraviolet lamp
irradiation of diatoms attached to hosts
stripped algae air dried and weighed in tared Petri dishes
settle for seven days
decant supernatant
5 ml aliquot
two drops of diatom liquor on coverslip; dried
coverslip applied to slide with 2 drops of Kleermount
finished slide
dispersion area
Whipple disc field

X. CALCULATION OF DATA.

Using the definitions and calculations described below an estimate was made of: (1) the number of diatom individuals per Whipple disc field, (2) the total number of individuals on a slide, and (3) the number of individuals per unit dry weight of host plant.

Let x_i indicate the number of individuals of a particular species in any Whipple disc field of a prepared slide. One Whipple disc field is 0.0036 mm^2 at a magnification of 450X. The index, i, is a field designator, and since ten Whipple disc fields were examined in each prepared slide, i ranges from one to ten. Thus,

$$\bar{x} = 1/10 \sum_{i=1}^{i=10} x_i \qquad \text{Eq. 1}$$

where $\bar{x}$ is an estimate of the number of individuals of a particular species in an average field on a given slide. Thus, for each species identified on a given slide, a separate value of $\bar{x}$ is obtained. Now if the values of $\bar{x}$ for a given slide are summed, an estimate of the number of diatoms in an average Whipple disc field is obtained for that slide. Let $\sum \bar{x}$ denote the estimate of diatoms/Whipple disc field.

By experimentation, it was found that when two drops of diatom liquor were placed on a coverslip, the area to which the drops spread was a function of the height from which they were dropped. However, by fixing the height of the drop delivery tube at two centimeters, it was determined that an average dispersion area was 0.529 mm^2. There are 147 Whipple disc fields in one dispersion area. Hence, this constant multipled times $\sum \bar{x}$ is an estimate of the number of individual diatoms in the dispersion area of a given slide. Since there are 120 drops of fluid in 5 ml of fluid,

$$\Omega = (\Sigma \bar{x}) 147 \cdot 120 \qquad \text{Eq. 2}$$

where Ω is an estimate of the number of diatoms per 5 ml aliquot from irradiation.

To arrive at an estimate of the number of diatoms per unit weight of a particular host plant, Ω is divided by the dry weight of the host. This new value is denoted by the symbol t.

XI. LINEAR CORRELATION OF DATA.

In an effort to determine whether there are correlations among salinity, water temperature, season, number of species, and number of individuals, linear correlation coefficients and graphs of least squares-best fit were prepared. These graphs compared the following pairs:

Station # (Latitude) <u>vs</u>. Number of Species
$\Sigma \bar{x}$ <u>vs</u>. Salinity
$\Sigma \bar{x}$ <u>vs</u>. Temperature
Number of Species <u>vs</u>. Salinity
Number of Species <u>vs</u>. Temperature
$\Sigma \bar{x}$ <u>vs</u>. Collection Season

For each of these pairs, five graphs were produced, one for each of the seasons - fall, winter, early spring, late spring, and summer.

The data pairs were fed into a Monroe 1655 desk computer which had been programmed by the author to yield best fit slope, y-intercept, the correlation coefficient, r, and the Student T value.

The results of these tests proved non-significant. The correlation coefficients were low (between -0.40 and 0.40). The T values were also low. At the accepted confidence interval of 95 percent, the T value should have fallen either below -2.306 or above 2.306. In many of the cases, the T values were between 1.25 and -1.25.

In the next section, the data are examined further using the methods of multiple linear regression analysis.

A. First Selection

Independent variables - 1. Salinity - expressed as °/oo
2. Temperature - expressed as °C
3. Season - expressed as 1, 2, 3, 4 and 5 (see Table 2)

Dependent variable - 1. Number of Diatom Species

Independent Variable	Mean	Std. Dev.	Correlation x *vs.* y	Regress. Coeff.	Computed t value
1.	16.77	12.35	0.21	0.09	1.77
2.	17.66	4.47	0.12	0.13	0.95
3.	30,000.00	14,301.90	0.14	0.00005	1.31
Dependent Variable					
1.	9.91	4.16			

Intercept 4.36
Multiple Correlation 0.32
Std. Error of Estimate 4.08

Analysis of Variance for the Regression

Source of Variation	Deg. of Free.	Sum of Sq.	Mean Sq.	F value
Attributable to Regression	3	78.26	26.09	1.57
Deviation from Regression	41	683.38	16.67	
Total	44	761.64		

R^2 value 10.2%

B. Second Selection

Independent variables - 1. Salinity - expressed as °/oo
2. Temperature - expressed as °C
3. Season - expressed as 1, 2, 3, 4 and 5

Dependent variable - 1. $\bar{x}$

Independent Variable	Mean	Std. Dev.	Correlation x *vs.* y	Regress. Coeff.	Computed t value
1.	16.77	12.35	-0.07	-0.19	-1.18
2.	17.66	4.47	0.13	0.41	0.91
3.	30,000.00	14,301.94	-0.45	-0.0005	-3.55
Dependent Variable					
1.	14.63	14.75			

Intercept 25.98
Multiple Correlation 0.50
Std. Error of Estimate 13.23

Analysis of Variance for the Regression

Source of Variation	Deg. of Free.	Sum of Sq.	Mean Sq.	F value
Attributable to Regression	3	2398.69	799.56	4.57
Deviation from Regression	41	7174.60	174.99	
Total	44	9573.29		

R^2 value 25.5%

Species	Ocean Type FWSSSu	Sound Type FWSSSu	Inlet Type FWSSSu	Quasi-F/W Type FWSSSu
Navicula incerta Grun.	-----	-x---	-----	-----
N. maculata (J. W. Bail.) Cleve	-----	---x-	-----	-----
N. mutica K. var. *ventricosa* K.	-----	-----	-----	----x
N. nitescens (Greg.) Don.	-----	----x	-----	-----
N. notabilis Grev.	-----	----x	-----	-----
N. peregrina (Ehr.) K.	-----	-----	-----	---x-
N. pusilla W. Smith	-x---	-----	-----	-----
N. salinarium Grun.	-----	--xxx	-----	-----
N. scandinavica (Lagst.) Cleve	----x	-----	-----	-----
N. spicula (Hickie) Cleve	-----	-----	-x---	---x-
N. viridula K.	----x	--xxD	-x-xx	-xxxx
Nitzschia acuminata W. Smith	-x---	-xxMx	-----	--x-x
N. attenuatum K.	-----	--x--	-----	-----
N. closterium W. Smith	-----	-----	x----	x--x-
N. constricta (K.) Ralfs	-----	----x	-----	-----
N. dubia W. Smith	-----	--x--	-----	-----
N. insignes Greg.	-----	--x--	-----	-----
N. lanceolata W. Smith	--x-M	----x	-----	---x-
N. longissima (Breb.) Ralfs	----x	-x---	-----	x---x
N. lorenziana Grun.	-----	----x	-----	-----
N. mirabilis Grun.	-xx-x	--M-D	--M--	----x
N. paxillifer (Gmelin) Grun.	-----	-----	-Dxx-	-----
N. socialis Grun.	-----	--x--	-----	-----
N. spathulata Breb.	-x---	-----	-----	-----
N. valida Cleve and Grun.	-----	---x-	-----	-----
Orthoneis cribosa Grun.	-----	--x--	-----	-----
Pinnularia socialis (T.C. Palm) Hust.	-----	-----	--x--	----x
Pleurosigma normanii Ralfs	-----	-----	---x-	-----
Podosira montagnei K.	-----	--M--	-----	-----
Rhabdonema adriaticum K.	-D---	-----	-----	-----
Rhizosolenia styliformis Brightw.	--x--	-----	-----	-----
Rhaphoneis amphiceros Ehr.	-----	----x	--x--	-----
Rhoicosphenia curvata (K.) Grun.	---x-	-xxxx	---x-	--x-x
Rhopalodia gibba K.	----M	----x	-----	x----
R. musculum K.	-----	-----	-x---	-----
Skeletonema costatum (Grev.) Cleve	-----	-----	-M---	-----
Stauroneis anceps Ehr.	-----	-----	-----	M-x--
S. ignorata Hust.	-----	M----	-----	-----
Staurosira mutabilis Smith	--Mx-	--DDx	---x-	M-x--
Surirella guinardii H.P.	-x---	-----	-----	--x--
Synedra closteroides Grun.	-----	-----	---x-	-----
S. gaillonei Ehr.	-----	--xx-	--x--	-----
S. parva K.	-----	--M--	--D--	-----
S. pulchella K.	-xM--	-MDxM	--x--	-DMDD
Tabellaria fenestrata K.	-Dxx-	--xx-	xxxxM	-----
Thalassionema nitzschioides Grun.	-x--x	--xMx	xM---	-----
Thalassiothrix longissima Cleve and Grun.	-----	-----	---x-	-----

Species	Ocean Type FWSSSu	Sound Type FWSSSu	Inlet Type FWSSSu	Quasi-F/W Type FWSSSu
Triceratium sp.	-----	x----	-----	-----

F = Fall = 1
W = Winter = 2
S = Early Spring = 3
S = Late Spring = 4
Su = Summer = 5

- indicates none found
x indicates presence
M indicates many
D indicates either dominant or codominant

SEASON DATA SHEET

	FALL	WINTER	EARLY SPRING	LATE SPRING	SUMMER
Air Temperature Range °C	15.0-24.4	8.3-17.8	6.1-15.6	12.2-22.8	15.6-23.3
Rainfall (24 hours) cm.	0.0	0.25	0.0	0.0	0.10
Noon Barometric Press mm. Hg.	76.61	75.41	75.79	76.83	75.82
Noon Cloud Cover %	20	100	40	100	90
Maximum Wind Speed and Direction km/hr.	25.6-NE	46.4-SW	35.2-W	24.0-NE	35.2-NW
	*				**

* Remarks: The effects of Hurricane Ginger were felt many days prior to its arrival in the collection area. These may have caused an appreciable change in the distribution of the diatoms.

** Remarks: The week prior to this collection date was remarkable in the amount of rain that fell in the collection area. On 6 June alone, 48.77 cm. of rainfall within a three hour period was recorded. This is indicative of a great deal of fresh-water that was added to the area via the rivers and their tributaries that feed the region.

TABLE 5: Parameter Data Sheet; (indicating salinity, surface temperature, $\Sigma \bar{x}$, Ω, t, the Host weight, and the host species at all nine stations during all collection seasons).

Season	STATION / DATA	1.	2.	3.	4.	5.	6.	7.	8.	9.
FALL	SALINITY (‰)	29.0	35.5	29.0	28.5	13.2	33.0	12.1	6.4	6.2
	SURFACE T (°C)	24.0	23.0	25.0	23.0	24.8	23.0	22.0	19.0	20.0
	$\Sigma\bar{x}$	20.7	43.3	10.6	30.6	42.4	1.5	12.6	36.0	60.7
	Ω ($\times 10^3$)	3.4	7.1	1.7	5.1	7.0	2.5	2.1	6.0	1.0
	t ($\times 10^3$)	2.8	4.7	1.4	4.3	5.9	1.7	1.8	4.7	6.2
	HOST WT. (gm.)	1.2	1.5	1.3	1.2	1.2	1.4	1.2	1.3	1.6
	HOST	i	f	f	j	j	d	f	n	m
WINTER	SALINITY (‰)	20.0	32.0	31.1	11.5	4.0	31.1	5.9	5.9	7.8
	SURFACE T (°C)	15.0	17.2	14.4	13.3	15.0	12.8	13.9	13.9	13.3
	$\Sigma\bar{x}$	5.6	8.9	2.1	8.2	5.2	1.1	10.3	13.5	51.2
	Ω ($\times 10^3$)	0.9	1.5	3.5	1.4	0.9	1.8	1.7	2.2	8.5
	t ($\times 10^3$)	2.8	2.1	1.0	4.0	2.7	0.5	22.5	22.3	58.1
	HOST WT. (gm.)	0.3	0.1	0.4	0.3	0.3	0.3	0.1	0.1	0.2
	HOST	d	m	g	k	g	d	e	g	g
SPRING	SALINITY (‰)	32.1	33.0	27.3	17.0	7.7	33.0	3.0	11.0	2.9
	SURFACE T (°C)	15.0	12.2	12.2	14.4	13.3	9.4	12.7	15.0	14.4
	$\Sigma\bar{x}$	11.7	11.4	38.0	26.2	8.4	0.7	33.1	19.9	3.7
	Ω ($\times 10^3$)	1.9	1.9	6.3	4.3	1.4	0.1	5.5	3.3	0.6
	t ($\times 10^3$)	11.1	11.0	297.8	15.4	56.1	0.8	5474.0	26.1	4.4
	HOST WT. (gm.)	0.2	0.2	0.1	0.3	0.1	0.1	0.01	0.1	0.1
	HOST	d	m	f	n	m	f	l	g	g
SPRING	SALINITY (‰)	17.0	29.0	17.8	12.3	14.8	31.2	0.9	0.9	0.8
	SURFACE T (°C)	17.0	12.0	17.0	19.0	18.0	12.5	17.1	17.0	15.7
	$\Sigma\bar{x}$	12.5	1.5	16.4	15.3	10.4	2.2	17.3	2.8	3.3
	Ω ($\times 10^3$)	2.1	0.3	2.7	2.5	1.7	0.4	2.9	0.5	0.5
	t ($\times 10^3$)	33.3	1.3	6.4	18.4	10.2	6.0	57.6	3.6	6.6
	HOST WT. (gm.)	0.1	0.2	0.4	0.1	0.2	0.1	0.1	0.1	0.1
	HOST	f	e	e	l	m	c	l	g	g
SUMMER	SALINITY (‰)	19.2	34.7	20.3	14.2	0.0	32.0	0.0	0.0	0.0
	SURFACE T (°C)	22.7	22.7	22.7	22.7	22.7	14.0	23.3	23.3	23.3
	$\Sigma\bar{x}$	2.4	10.9	21.3	8.6	1.4	2.2	1.3	2.0	91.1
	Ω ($\times 10^3$)	0.4	1.8	3.5	1.4	0.2	0.4	0.2	0.3	15.1
	t ($\times 10^3$)	2.4	9.6	10.9	2.9	1.0	1.5	0.5	1.3	54.5
	HOST WT. (gm.)	0.2	0.2	0.3	0.5	0.2	0.2	0.5	0.3	0.3
	HOST	a	m	d	a	g	d	g	g	g

REMARKS:

- a = Enteromorpha compressa
- b = E. confervoides
- c = E. intestinalis
- d = E. linza
- e = E. plumosa
- f = E. prolifera
- g = Myriophyllum pinnatum
- h = Nostoc sp.
- i = Polysiphonia nigrescens
- j = P. subtilissma
- k = Rhizoclonium tortuosum
- l = Ulothrix flacca
- m = Ulva lactuca
- n = Zostera marina

I. PRIMARY PRODUCTIVITY OF DIATOMS

In the effort to determine relative productive ability of diatoms as members of the photoautotrophic community, it is necessary first to establish their individual rate of net productivity. Most reports concerning the net productivity of phytoplankton have given total values for all phytoplankton have given total values for all phytoplankton and not for the separate members such as the Bacillariophycophyta.

Riley's (1944) studies of all phytoplankton divisions employed the oxygen method and produced a value of world ocean production of 1.55×10^{11} tons carbon per year.

Steemann-Nielson (1955), after receiving data from the Galathea Expedition, derived an average value of $1.2 - 1.5 \times 10^{10}$ tons carbon per year. This figure was determined using the radioactive carbon method. Ryther (1969) found a value of 2.0×10^{10} tons carbon per year. His figures take into account the efficiency factors of some of the ecological areas from which data are obtained.

Further, Koblentz-Mishke, Volkovinsky, and Kabanova (1969) have analyzed over 7000 separate productivity observations of recent world coverage. A new value of $1.5 - 1.8 \times 10^{10}$ tons carbon per year is offered for the world's oceans.

As indicated by Talling (1962), productivity, or the rate of photosynthesis, due to its dependence on solar energy, must be related to a unit area measure. Depending on differential depletion of nutrients in these unit areas, restrictions are placed on the specific photosynthetic rates of the autotrophic organisms. Thus, many measures of cellular productivity are carried out in the laboratory, where conditions can be controlled to emulate those found in nature. Talling (1962) cites 1 to 3 cell divisions per day and 4 to 12 mg CO_2/mg chlorophyll/hour as being a maximum value of growth rates for diatoms.

Round (1965) notes that intensive studies of the individual photosynthetic rates of particular species of planktonic algae have rarely been undertaken. He cites studies of Lund (1949, 1950, 1954) in which in vitro cultures of Asterionella formosa and Melosira italica subsp. subarctica were examined. It was found that A. formosa rapidly changes its growth rate with light intensity and temperature changes.

Although these data place in perspective the value of the productivity of the world's oceans, they do not indicate the individual worth of the members of this community. From the figure derived by the above authors, the average net productivity of diatoms cannot be determined because of great individual differences. Holm-Hansen (1968) has started to solve this problem on a cellular level. His results consider the amounts of DNA and organic carbon found in unicellular algae. He feels that DNA regulates cellular metabolism through the control of concentrations of free nucleotides by sequestration reactions (Holm-Hansen, 1968). Thus, the percentage of DNA as compared to organic carbon (pg/cell) is an indication of production. From his research, he has determined the DNA/carbon ratios for ten species of unicellular algae, among them Navicula pelliculosa, Skeletonema costatum, and Thalassiosira fluviatilis. These are listed as follows:

Species	@ DNA	@ Carbon
Navicula pelliculosa	0.09 pg/cell	10 pgC/cell = 0.009
Skeletonema costatum	0.70 pg/cell	28 pgC/cell = 0.003
Thalassiosira fluviatilis	8.00 pg/cell	110 pgC/cell = 0.073

As described by Holm-Hansen:

"The portionality between DNA and cell carbon can be used for biomass determinations in ecological studies. Some estimate of the total living crops of organisms, both autotrophic and heterotrophic forms, is often desired, and analyses for total organic carbon or protein are not feasible because of organic detrital material. The usefulness of DNA measurements for estimation of biomass may be limited to some extent by the rate of hydrolysis of cellular nucleic acids upon death of the cell. However, DNA has been used as a biomass indicator in ocean samples." (Holm-Hansen, 1968).

II. SEASONAL PRODUCTIVITY.

It is clear that the early spring is the season of greatest growth of epiphytic diatoms from the Outer Banks Coastal and Bay region. Not only do the numbers of species increase at a rapid rate, but also the number of individuals per species is larger. There were 29 species found in fall, 41 in winter, 46 in early spring, 39 in late spring, and 44 in summer.

If an increase in numbers of diatoms in early Spring is observed, it is often explained by the increasing length of time of solar energy, the general warming of the waters, and the prior winter accumulation of phytonutrients (Johnson, 1957). However, due to the corresponding and subsequent increase in grazing zooplankton in the late spring, the numbers of diatoms soon plateau and decline. They rise again in summer, perhaps due to toxic zooplanktonic metabolites. This, as well as mechanical detachment (Wood, 1965), is reflected in a relatively low number of epiphytic diatoms during the fall season as seen in this collection. During the fall, the numbers are far lower than in early spring due to zooplanktonic overgrazing.

The coming of colder weather in the late fall and early winter may produce a corresponding decrease in the numbers of zooplankton. Thus, the diatoms can have less grazing pressure applied to them. A slight rise in their numbers is often seen, but because of the shorter period of solar energy availability, the depletion in the water of the needed nutrients, and the lower water temperatures, their rise in numbers is often not pronounced.

III. ANALYSIS OF DATA.

From the data recovered there are four significant relationships that can be offered. These are: (1) there does not appear to be any linear correlation between the numbers of diatom species and the pooled effects of salinity, water temperature, and season of the year. (2) There is a marginally significant but stronger relationship between the average number of diatoms isolated, measured by $\sum \bar{x}$, and salinity, temperature, and season of recovery. (3) There appears to be a direct relationship between the presence of *Synedra pulchella* and *Cocconeis scutellum*. In each case, when one of these species is found in more than random numbers, the other is found in nearly the same numbers, regardless of the season, station, or species of host. (4) There seems to be a very strong relationship between the numbers of species and individuals of diatoms and the morphology of the host from which they are isolated. Perhaps this has to do with the morphological configuration of the host and the available surface area for attachment.

A) Diatom Species

Upon examination of the results of the multiple linear regression analysis, page 44, there is no significant relationship between the numbers of diatom species - the dependent variable - ant the three independent parameters: salinity, water temperature, and season. A number of statistics support this.

First, the F ratio value in this test is 1.57. This statistic is a ratio derived by dividing the between variance, S_b^2, by S_w^2, the within variance. If the null hypothesis is true and the data were not affected by the method of collection (i.e., destruction of less siliceous species), then S_b^2 will be approximately the same value as S_w^2 and the ratio will be equal to one. However, if the null hypothesis is false, these two variances will not be equal to each other. This can indicate outside factors causing this difference. The variability among the means is increased and S_b^2 is made larger than S_w^2. F values much larger than one indicate that the null hypothesis should be rejected. In order to determine just how much larger than one F should be, the F ratio is computed (i.e., 1.57), the number of degrees of freedom are determined (3 and 41), the confidence level is chosen, and upon inspection of the F value tables, the indicated F value is noted (3.56). In order for the computed F value to be accepted as significant at the indicated confidence level, it must be equal to or larger than the F value from the table (Sokal and Rohlf, 1969a and b).

In this case, the tabled F ratio value of 3.56 (95% confidence level) is the lowest value that would warrant rejection of the null hypothesis. Thus, because 1.57 is smaller than the F table value, one cannot consider the null hypothesis as false. This indicates that from the available data, I have not established a significant correlation between the number of species and the pooled effects of salinity, temperature and season.

A second factor indicating a lack of significance in these data is the relatively small regression coefficients. If applied to the formula:

$$\hat{Y} = a + b_1x_1 + b_2x_2 + b_3x_3 \qquad \text{Eq. 3}$$

where $\hat{Y}$ is the estimated dependent variable value, a is the y intercept, b_1, b_2, and b_3 are the respective regression coefficients and x_1, x_2, and x_3 are the independent variable, the equation would appear as:

$$\hat{Y} = 4.36 + 0.09\ (\text{Salinity}) + 0.13\ (\text{Temperature}) + 0.0005\ (\text{Season}) \qquad \text{Eq. 4}$$

However, upon inspection of the values of the Sum of the Squares attributable to Regression (78.26) and the total Sum of the Squares (761.64), it can be seen that the R^2 value, which indicates the relative amount of association that the measured parameters account for, is 10.2%. There must be other variables that are contributing the other 89.8% to cause the observed numbers of diatom species.

Third, with the computed T values of 1.76, 0.95, and 1.31 respectively, these independent variables are quite low and cannot be considered as significant. Only a Student T value of at least 2.56 would indicate the data was significant at the 95% level.

B) $\Sigma\bar{x}$ Relationship.

Upon examination of the second selection (page 44) in which the number of diatoms is compared with salinity, water temperature, and season, there is a greater degree of association.

First, the F value of this selection is 4.57. At the 95% confidence level, this is higher than the value of 3.56 needed to indicate that this association is not merely due to chance.

Second, the absolute value of the regression coefficients is larger in this test selection, indicating a greater individual credibility in the independent variables: salinity, water temperature, and season. In applying these coefficients to Equation 4, one finds:

$$\hat{Y} = 25.98 - 0.196 \text{ (Salinity)} + 0.406 \text{ (Temperature)} - 0.0005 \text{ (Season)} \qquad \text{Eq. 5}$$

The estimated mean number of diatoms from each host, $\hat{Y}$, is inversely proportional to the salinity and is directly proportional to the water temperature. They appear to be unaffected by the season during which they were recovered.

From the T values computed, it is evident that these regression coefficients are nearly numerically equal to those of the first selection, with one exception. The T value of -3.55 with respect to season represents a value that can be considered most reliable.

Lastly, the R^2 value, or percentage of these parameters contributing to the mean number of diatoms, $\Sigma \bar{x}$, is 25.5%. Although this value is greater than that found in the first selection, it indicates that there still are other parameters that account for nearly three times as much effect. It must be emphasized that the other 74.5% may be attributable to many variables, and thus, the effects of salinity, water temperature, and season may be the most important group controlling the mean number of diatoms.

C) Species Relationships: Cocconeis scutellum and Synedra pulchella.

It was found that in nearly every case when either Cocconeis scutellum or Synedra pulchella was found in the diatom recovery, the other species was there in nearly the same numbers (see Table 2). This relationship was prevalent at all four station types, and during all seasons with the exception of fall. When C. scutellum was found as the dominant form, S. pulchella was present in prolific numbers; when one of these species was found only in sparse numbers or not at all, the other was present with the same frequency. It would appear that there is a direct relationship between the presence and numbers of one of these species and the other due to their eurythermal and euryhaline tolerances (see Appendix B). S. pulchella is a member of the order Fragilariales. C. scutellum is classified in the order Achnanthales. The former has no raphes while the latter has one true raphe and one pseudoraphe.

D) Host Relationship.

There appears to be a notable relationship between the numbers of diatom species and individuals found on a particular host and the shape of that host at the site of attachment. This theory is held by Aleem (1950), Reimer (personal communication), Zaneveld (personal communication), and Marshall (personal communication).

It seems that host species with more convoluted surfaces and with numerous branches offer more acceptable surfaces to which the diatoms can adhere than species without this morphology. Broad, flat surfaces, or those without branching, do not appear to have the necessary interstices. Aleem (1950) indicates that the suitability of different hosts for diatoms crosses phylogenetic lines, there being numerous epiphytic diatoms on all divisions of algae. It is felt that the more foliose species of some genera with smooth surfaces, such as Ulva, Punctaria, and Porphyra, do not provide the necessary criteria for attachment. Branched, cylindrical species of some genera of algal host seem to be favored by the diatoms. Certainly this subject is an area on which further work might yield valuable information.

IV. DISTRIBUTION OF DIATOMS.

Of the 94 species of diatoms divided among 37 genera, there were only 25 that were found in abundant numbers. These are the species designated with either an "M" or a "D" on the Diatom Recovery Sheet, above (see Table 3).

Perhaps the most interesting environmental fact is that Cocconeis scutellum was found at all the station types during all seasons, with the exception of the fall recovery at the Inlet Station. This species is apparently euryhaline and eurythermal. Associated with this is the fact that this species was recovered from all of the host plant species. This included all of the three algal divisions - Cyanophycophyta, Chlorophycophyta, and Rhodophycophyta - as well as the higher plants, Myriophyllum pinnatum and Zostera marina. Of all of the species of diatoms recovered, C. scutellum possesses the greatest tolerance to wide ranges of temperature, salinity, and host conditions.

The genus that represented the greatest numbers of diatom species found during the collection period is Fragilaria. Although not found with as great frequency as C. scutellum, the combined numbers of F. capucina, F. gracillima, and F. virescens, represent the genus contributing the greatest number of epiphytic individuals throughout the year.

Grammatophora marina can be considered a neritic species that is generally found in the summer and fall. This species was recovered in dominant numbers at the Ocean Station (#3) and at the Sound Stations (#1, #4, #5), but it was only rarely seen at the Quasi-Freshwater regions. It should be considered as a more restricted species with stenothermal and stenohaline characteristics.

Although *Achnanthes brevipes* was recovered during all seasons of the collection period, it was found in greatest numbers only during the winter and early spring at the Sound Station and Inlet Station. This would indicate that this species prefers cooler water of mixo-mesohaline (Fredrich, 1969, p. 426) character.

Synedra pulchella was found in all seasons, except fall. Its numbers were prolific during winter, late spring, and summer at the Quasi-Freshwater Station type. Certainly, this species is intolerant of high saline conditions, but it is found in both warm and cool waters. *S. pulchella* was the second most abundant species.

Tabellaria fenestrata was found in great numbers during winter and summer at the Ocean and Inlet Station types, but was found infrequently, or not at all, during the other times of the year. This would indicate that this species is intolerant of great salinity fluctuations, but is relatively unaffected by temperature extremes; hence, it is stenohaline and eurythermal.

One species that was recovered at many of the station types during all seasons, except fall, in low numbers was *Navicula viridula*. The peak of numbers of this species occurred during summer at the Sound Station type. It would seem that this species favors warm, mixo-mesohaline conditions for growth, and that, because these conditions were found during only one of the collection periods, one can conclude that *N. viridula* occurs in low numbers during all other times.

The following listing compares the species and conditions under which they most often are found.

0-9 °/oo / 9-15°C

Cocconeis scutellum
Synedra pulchella

18-29 °/oo / 9-15°C

Cocconeis scutellum
Rhabdonema adriaticum
Tabellaria fenestrata

18-29 °/oo / 16-20°C

Cocconeis scutellum
Diatoma elongatum
Eunotia parallela
Grammatophora marina
Staurosira mutabilis

0-9 °/oo / 16-20°C

Cocconeis scutellum
Fragilaria capucina
Stauroneis anceps
Staurosira mutabilis

10-17 °/oo / 16-20°C

Cocconeis scutellum
Melosira moniliformis var. *octagona*
Nitzschia acuminata
Podosira montagnii
Thalassionema nitzschioides

18-29 °/oo / 21-26°C

Cocconeis scutellum
Coscinodiscus lineatus
Fragilaria gracillima
Nitzschia lanceolata
Rhopalodia gibba

0-9 °/oo / 21-26°C

Cocconeis scutellum
Synedra pulchella

10-17 °/oo / 21-26°C

Cocconeis scutellum
Navicula viridula
Nitzschia mirabilis
Synedra pulchella

10-17 °/oo / 9-15°C

Cocconeis scutellum
Synedra pulchella

An indication of the season of growth for diatoms from the collection area may be obtained by summing the $\Sigma\bar{x}$ values as they appear on the Parameter Data Sheet. However, this is misleading. Such as value will not indicate the dry weight of the host from which these organisms were stripped. Algae with a large biomass will necessarily yield greater numbers of diatoms than smaller samples of a host. A more appropriate indicator of diatom growth is the expression, t. By adding each t value derived from each station during one season, a total t value, or Σt is derived. Completing this for all five seasons, it is possible to observe increases and decreases in diatom production. The following values are obtained:

Season	Σt	Dominant Genera
Fall	3.35×10^6	*Grammatophora*, *Fragilaria*, *Cocconeis*
Winter	1.15×10^7	*Cocconeis*, *Rhabdonema*, *Synedra*
Early Spring	5.89×10^8	*Cocconeis*, *Synedra*, *Staurosira*
Late Spring	1.43×10^7	*Cocconeis*, *Fragilaria*, *Staurosira*
Summer	8.40×10^6	*Synedra*, *Nitzschia*, *Cocconeis*

Table 6: Seasonal Σt values.

Σt reflects the number of epiphytic diatoms per unit weight of host plant. It is felt that in future reports of this nature, when comparing colonization or distribution of epiphytic diatoms, or when examining these organisms with respect to salinity, temperature or season, the statistic Σt should always be used.

The growth cycle depicted in Table 6 follows predicted patterns (Talling, 1962; Round, 1965; Raymont, 1963). One expects an increase in numbers of diatoms in early spring when light energy is more available, when temperatures are rising, and when increasing river run-off provides additional nutrients to estuarine and coastal areas. There is a corresponding drop in numbers in the late spring and summer, not due to a decreased frustule division rate but rather due to the grazing pressure exerted by herbivorous zooplankton. In fall, when light energy decreases, water temperature falls, and the provided nutrients in the water are depleted, the numbers of diatoms decrease due to a drop in the division rate. Some species then may continue to survive and will exist throughout the winter. Others resort to a resting stage which then lies dormant until the favorable conditions of early spring are manifest, at which time the cycle is begun again.

In an effort to graphically display the temperature - salinity requirements of the species that appeared in dominant, codominant or many numbers in these samples, the graphs of Appendix B were prepared. It is felt that they indicate the requirements of these species as shown by their presence. Species that were found in only one, two or three samples were not depicted: it was felt that this number of samples would not show a significant trend with respect to temperature and salinity.

CONCLUSION

CONCLUSION:

There are five major points that should be made concerning the distribution of epiphytic diatoms of the Outer Banks region of North Carolina with respect to salinity and temperature. First, their mean numbers, $\Sigma \bar{x}$, show significant multiple linear regression with respect to the pooled effects of salinity, temperature, and season of the year. This is supported by equation 5, page 60, the F value of 4.57, the T value of 3.55, and the R^2 value of 25.5%.

Second, there is a direct relationship between the presence or absence of Cocconeis scutellum and Synedra pulchella. The Diatom Recovery Sheet, Table 3, indicates that in nearly every instance, when either of these species was recovered from a host plant, the other was also present in nearly the same numbers. Conversely, when one of these species was absent, the other was not found.

Third, the numbers of epiphytic diatoms present on any particular host may be a function of the surface configuration of that host. Convoluted, branched host plants nearly always had attached to them a greater number of diatoms than those hosts with broad, flat surfaces. This, however, warrants further investigation for confirmation.

Fourth, in this limited study area (see Figure 1) there is a pronounced gradient of epiphytic diatoms that appears to follow not only latitudinal but also seasonal and grazing pressures. This may be seen in the diagram on page viii.

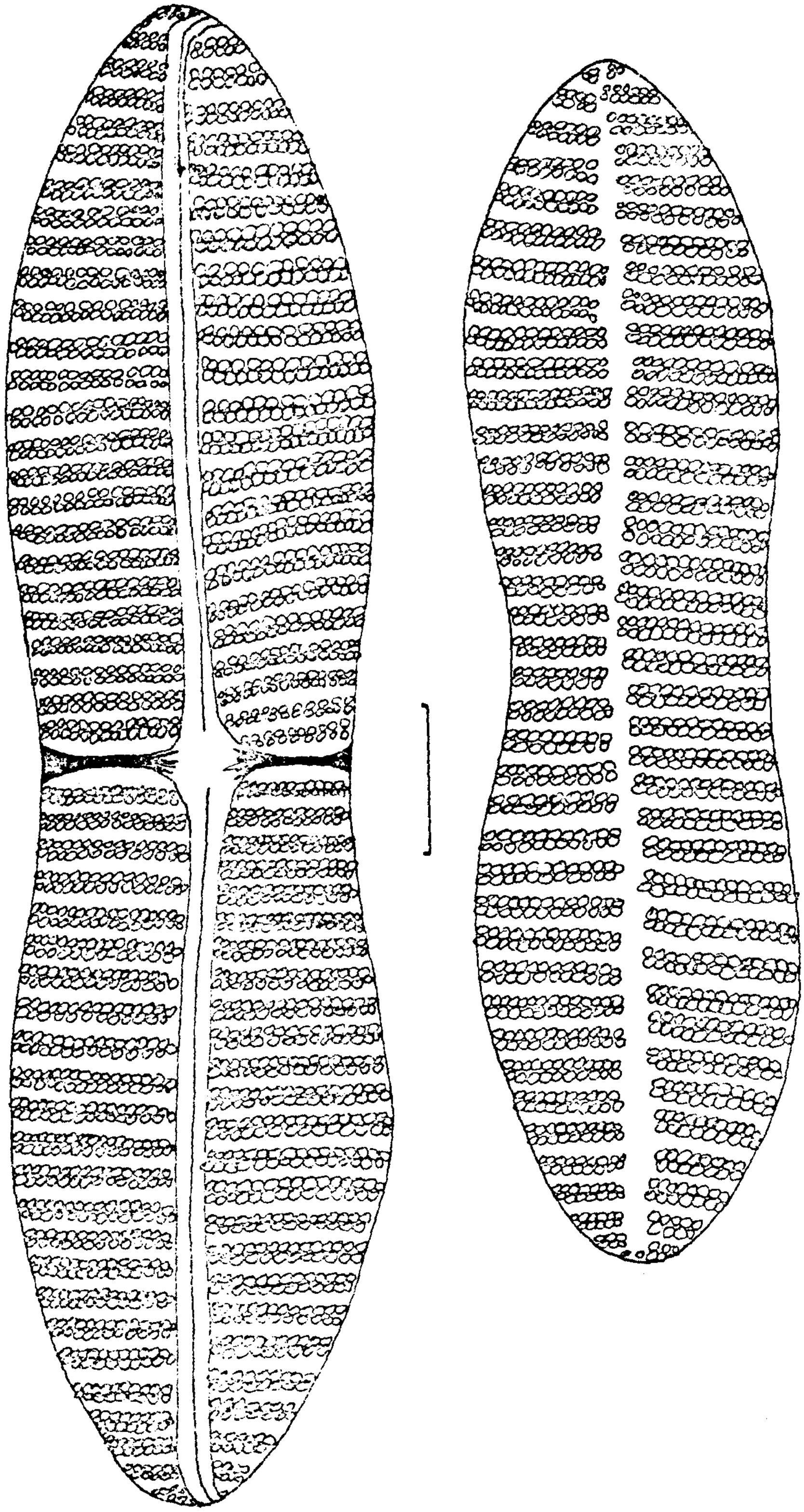
Fifth, the Swift method, as modified by Belin and Zaneveld (1972) is an extremely effective method both qualitatively and quantitatively for stripping epiphytic diatoms from their host plants.

Lastly, as the human population of this area increases, and with it the rise in amounts of sewage and industrial and commercial pollution, periodic studies of the epiphytic diatoms may indicate environmental tendencies.

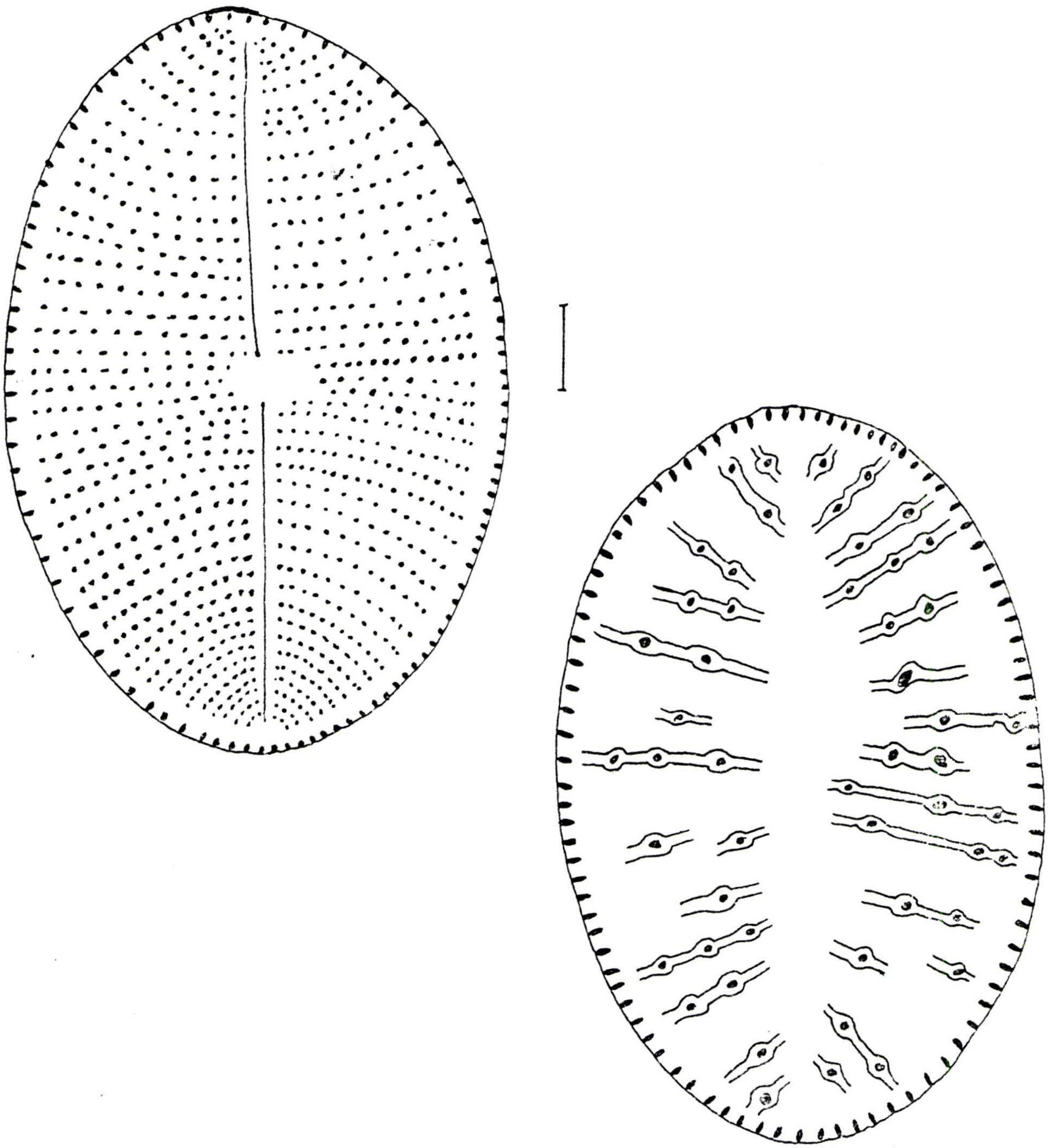
APPENDIX A

SKETCHES OF SELECTED DIATOM SPECIES

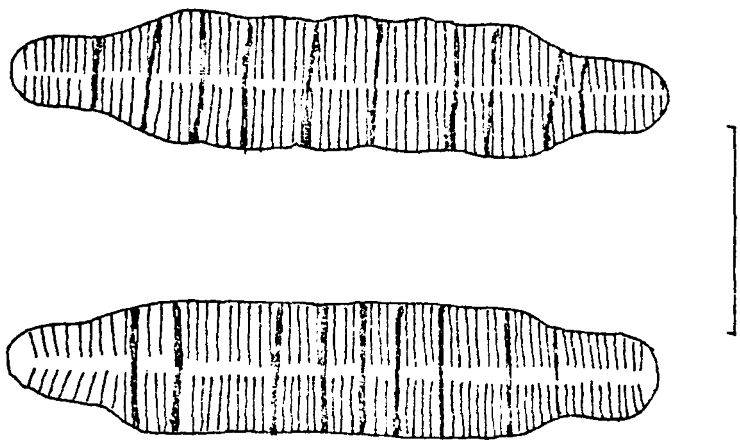
(Designator Lines Indicate 10μ.)



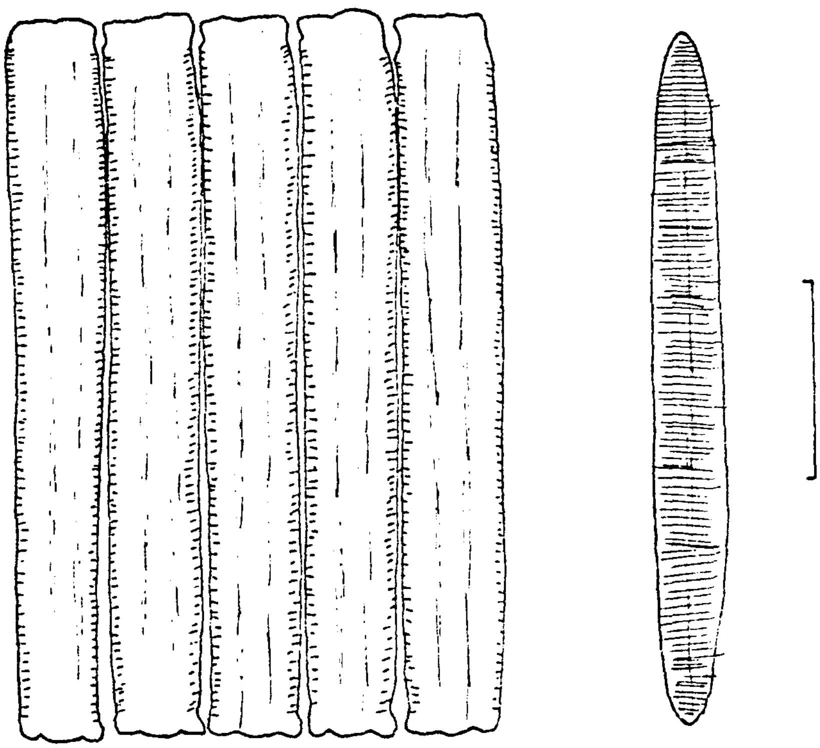
Achnanthes longipes



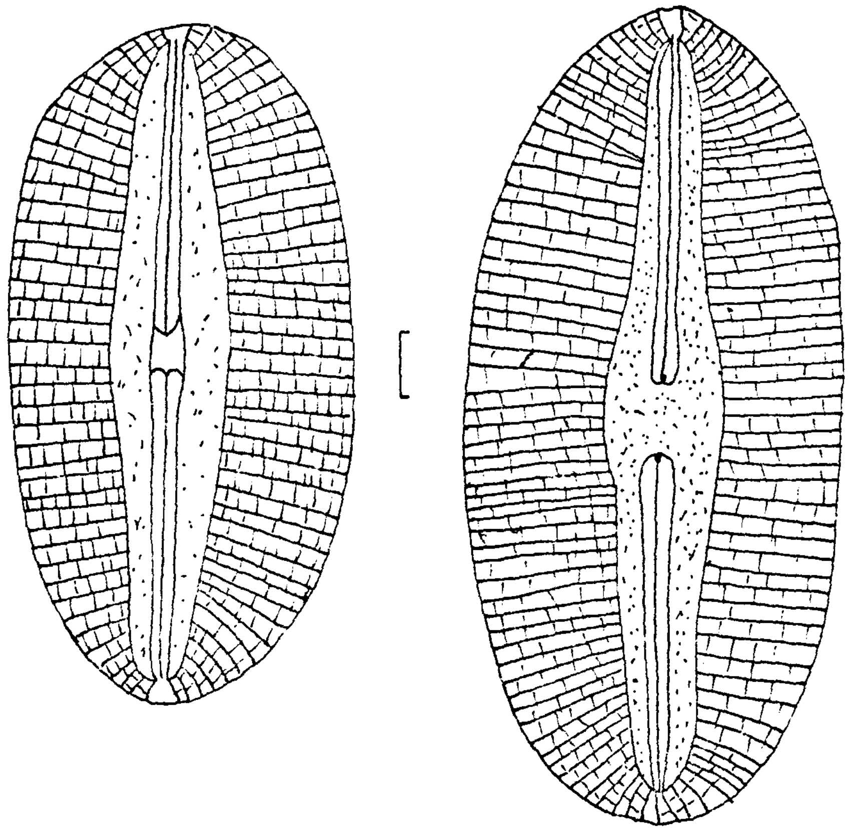
Cocconeis distans



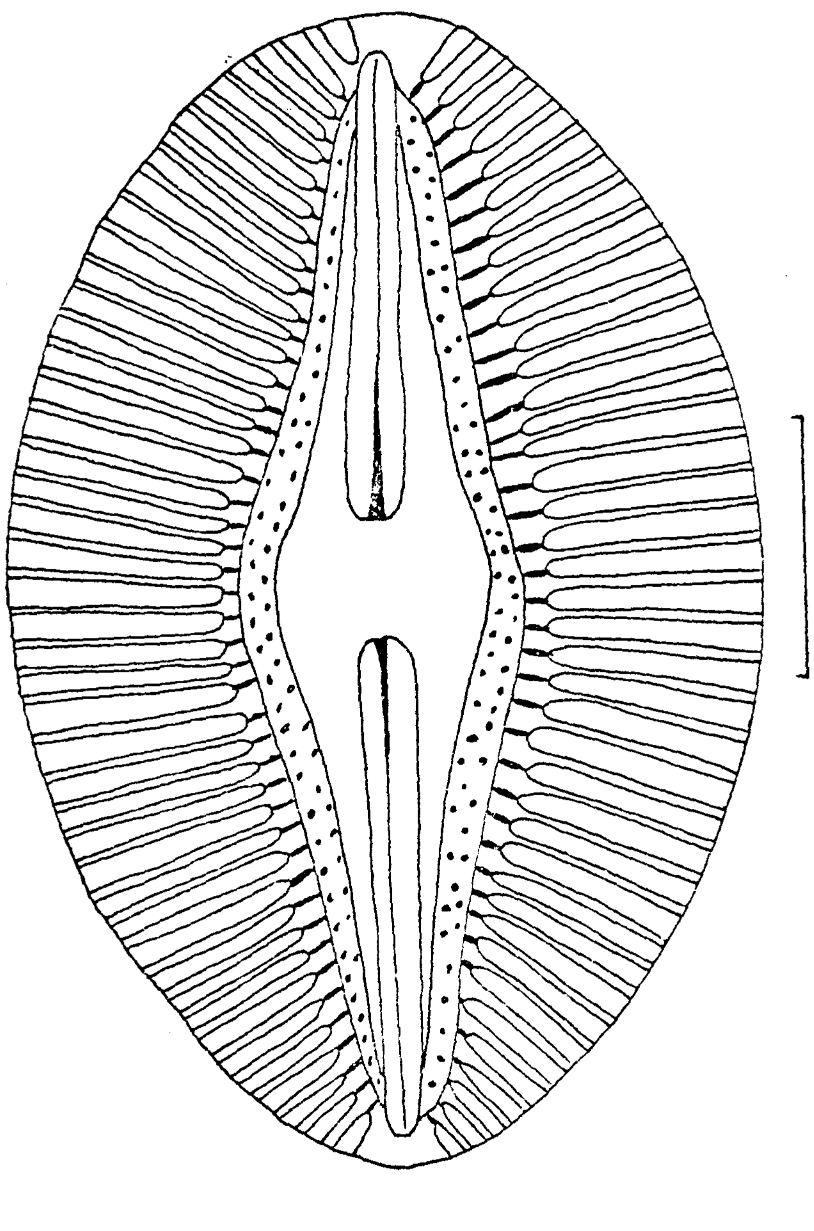
Diatoma anceps



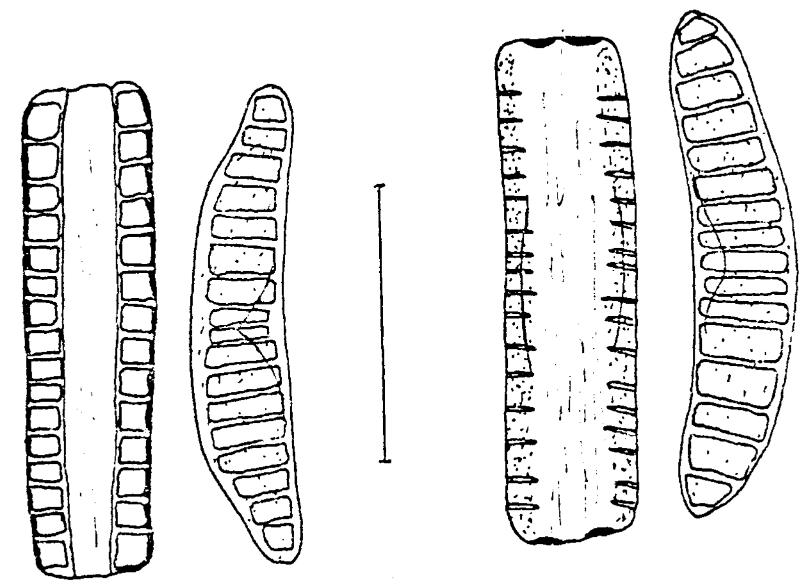
Dimerogramma dubium



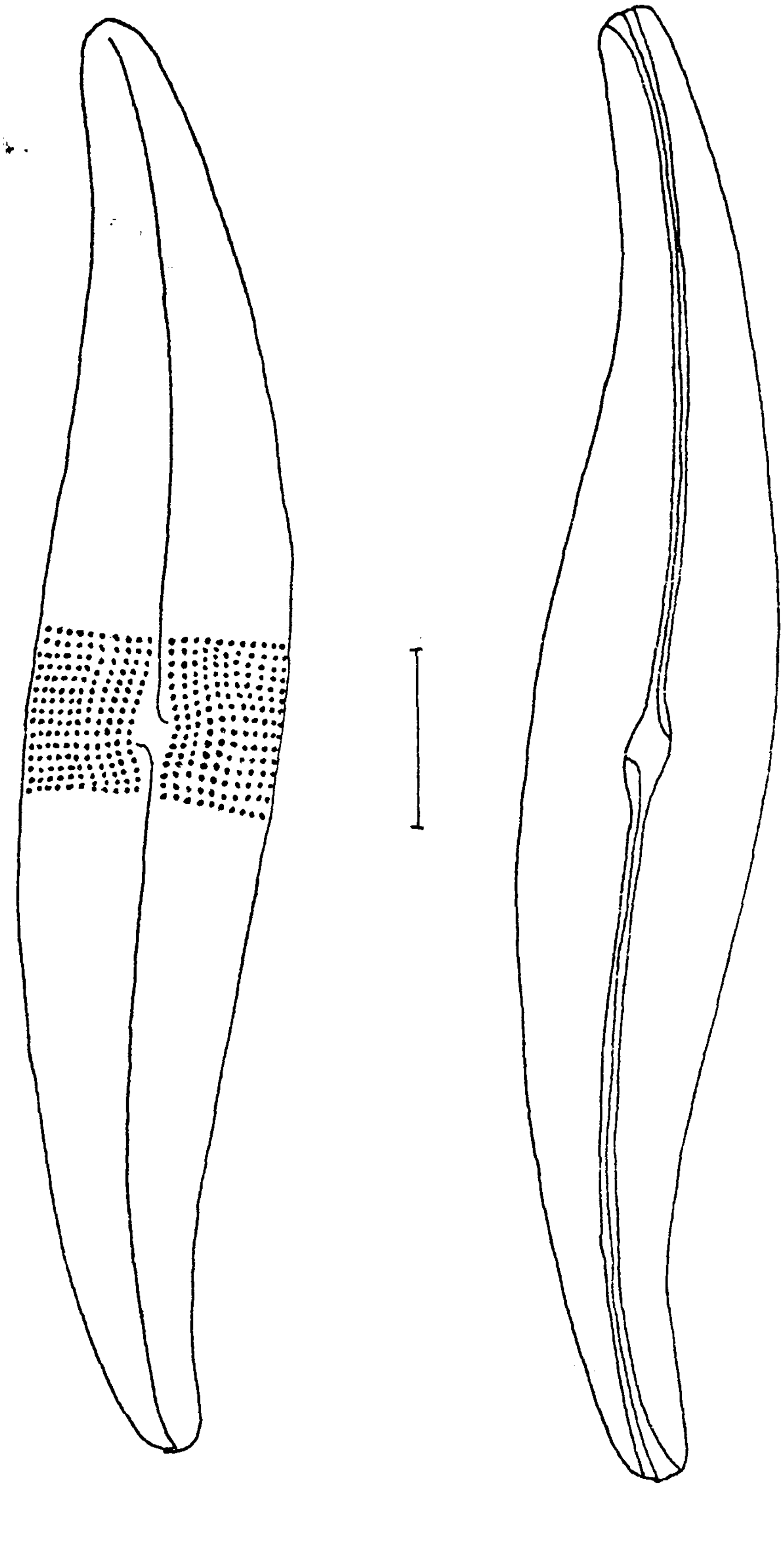
Diploneis fusca



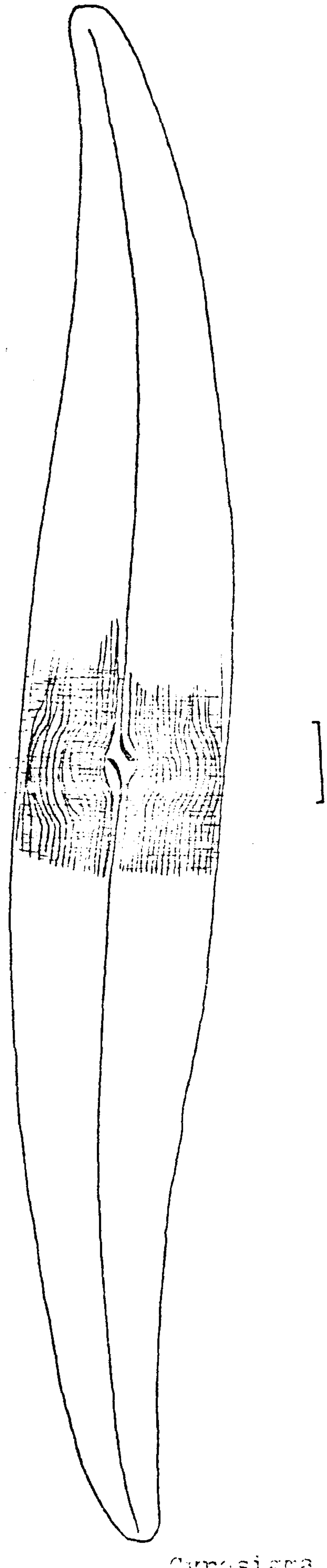
Diploneis smithii



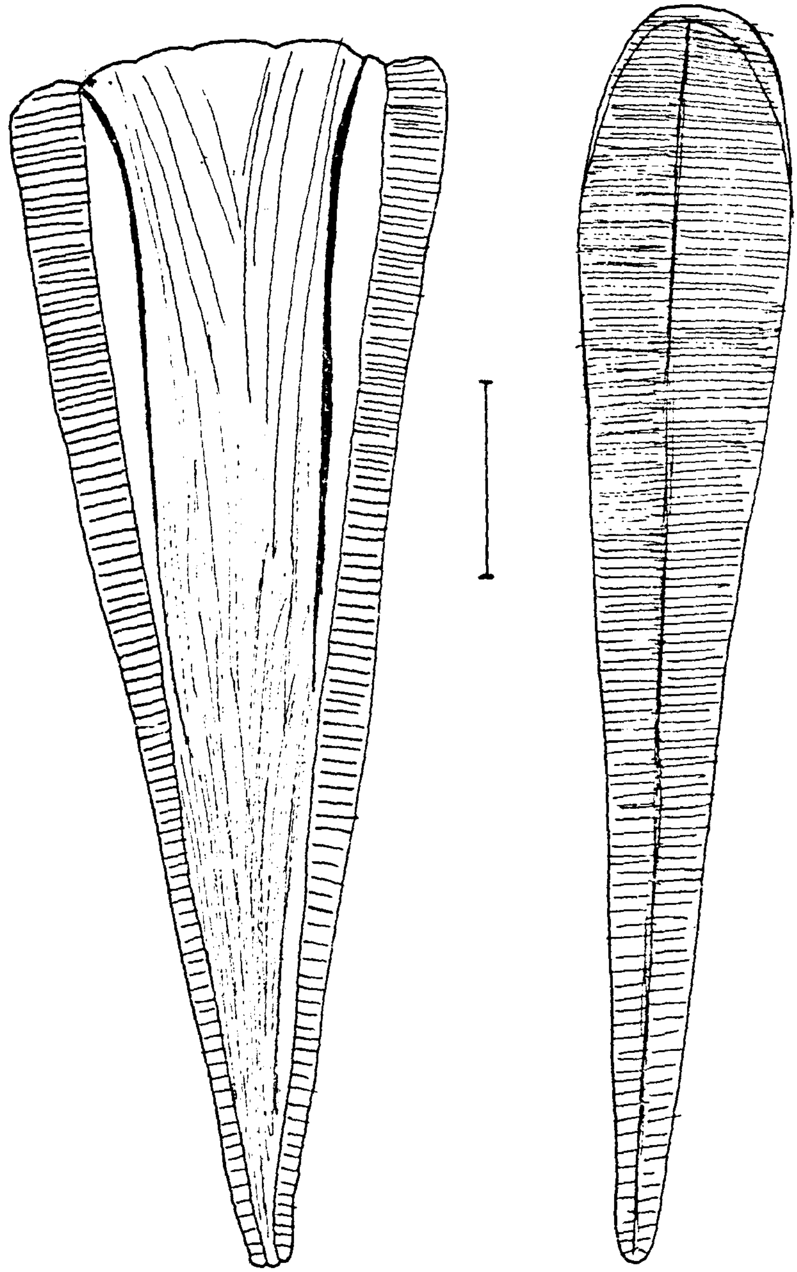
Epithemia zebra



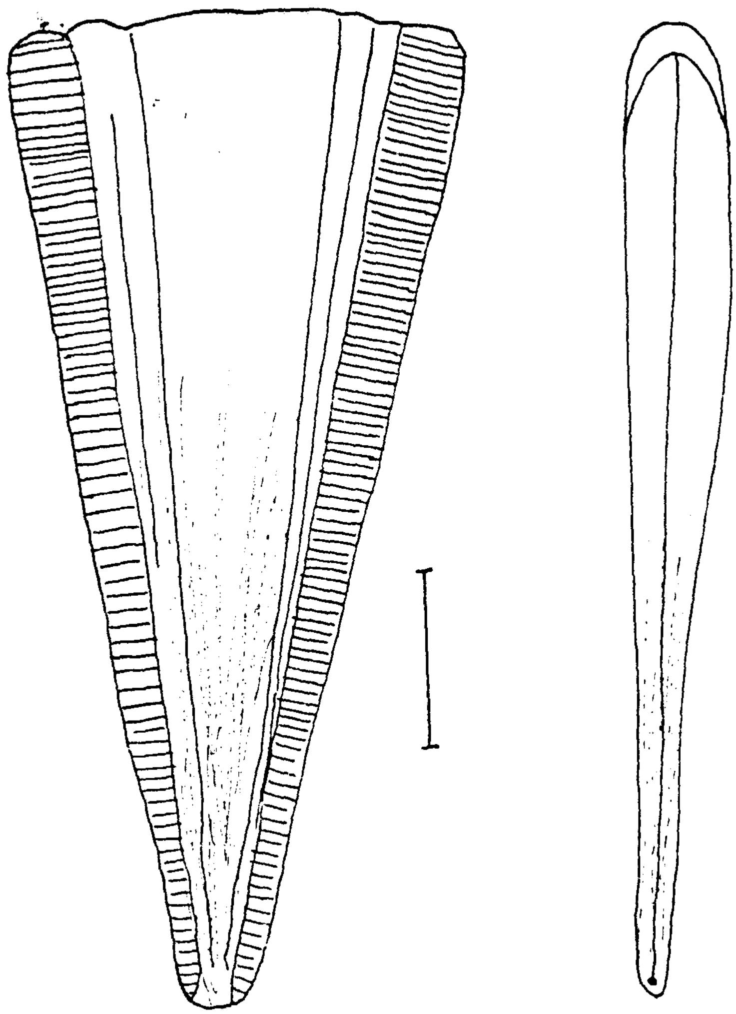
Gyrosigma acuminatum



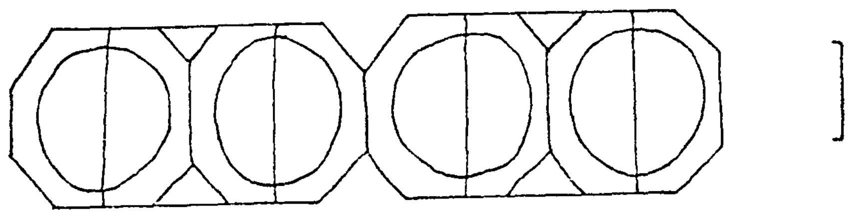
Gyrosigma attenuatum



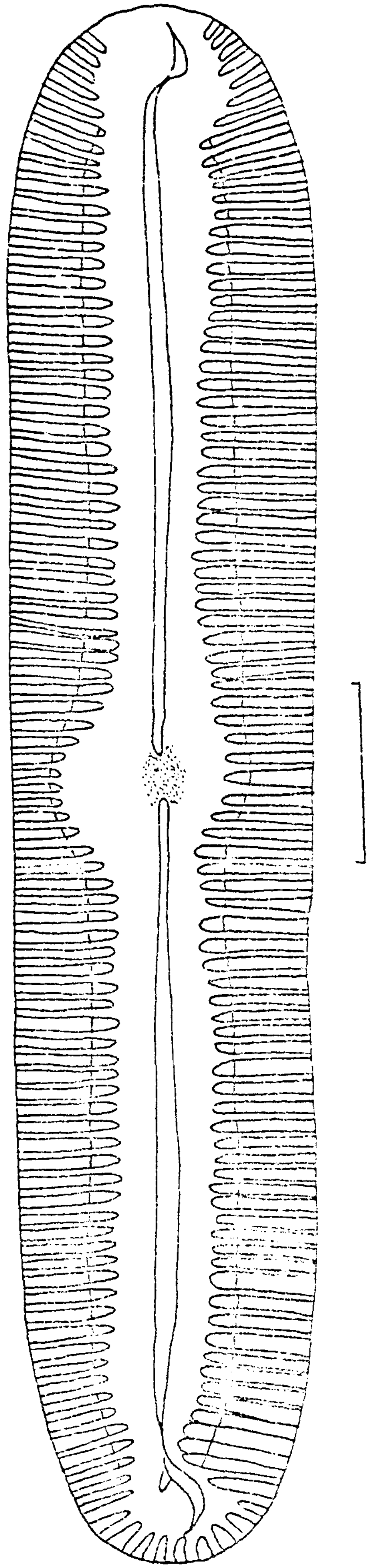
Licmorpha gracilis

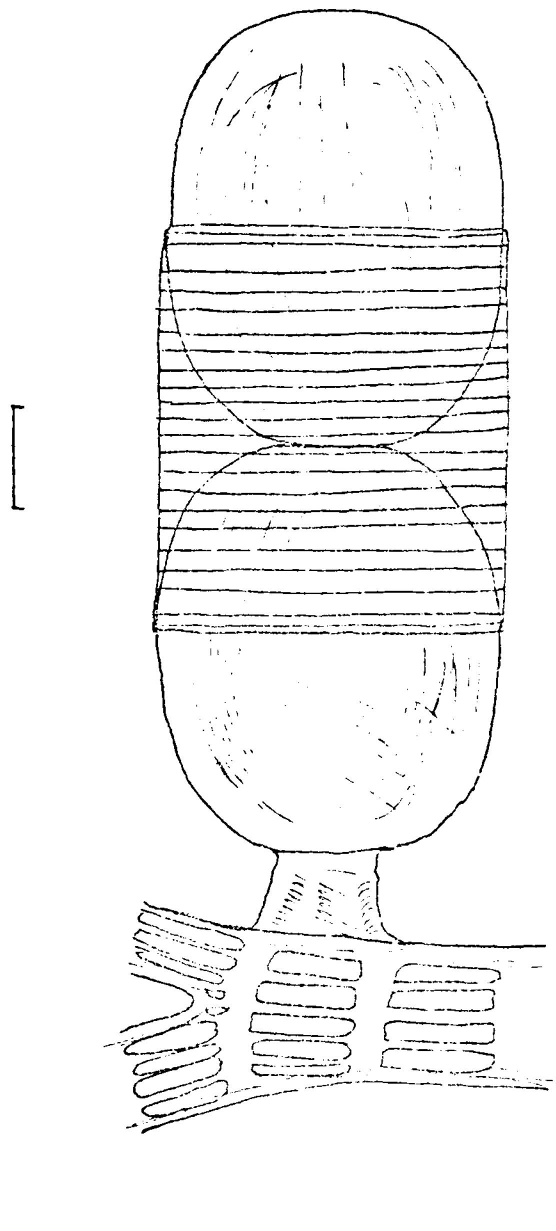


Licmorpha juergensii

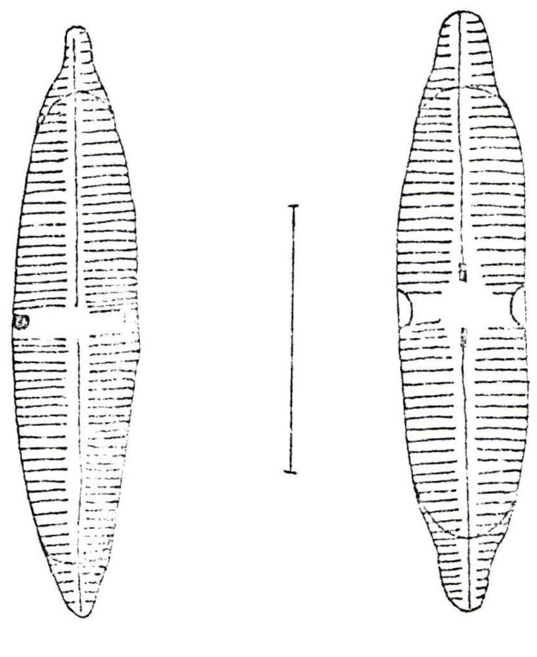


<u>Melosira moniliformis</u> var. [illegible]

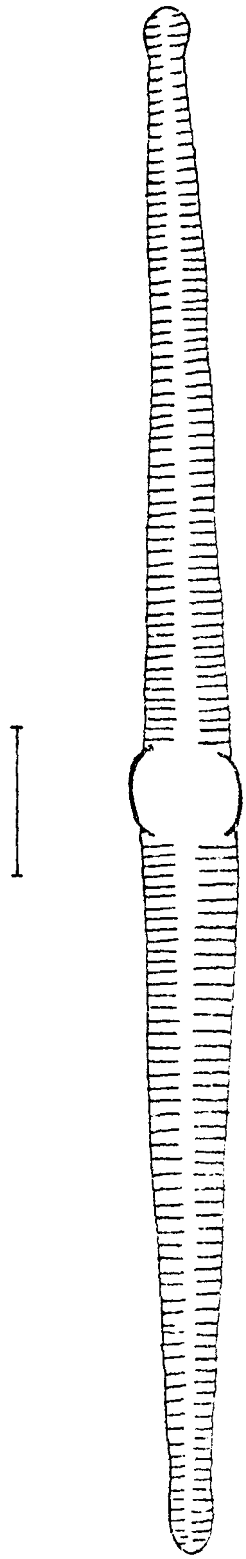




Podosira [illegible]
after Cleve-Euler, 1951



Stauroneis ignorata



Synedra pulchella

APPENDIX B

The species that appeared in dominant, codominant, and many numbers are included, separately, in Appendix B. Only those that appeared in more than three of the forty-five collections are depicted. The maximum and minimum temperatures and salinities are listed below:

	Minimum	Maximum
Temperature	9.4°C	25.0°C
Salinity	0.0°/oo	35.5°/oo

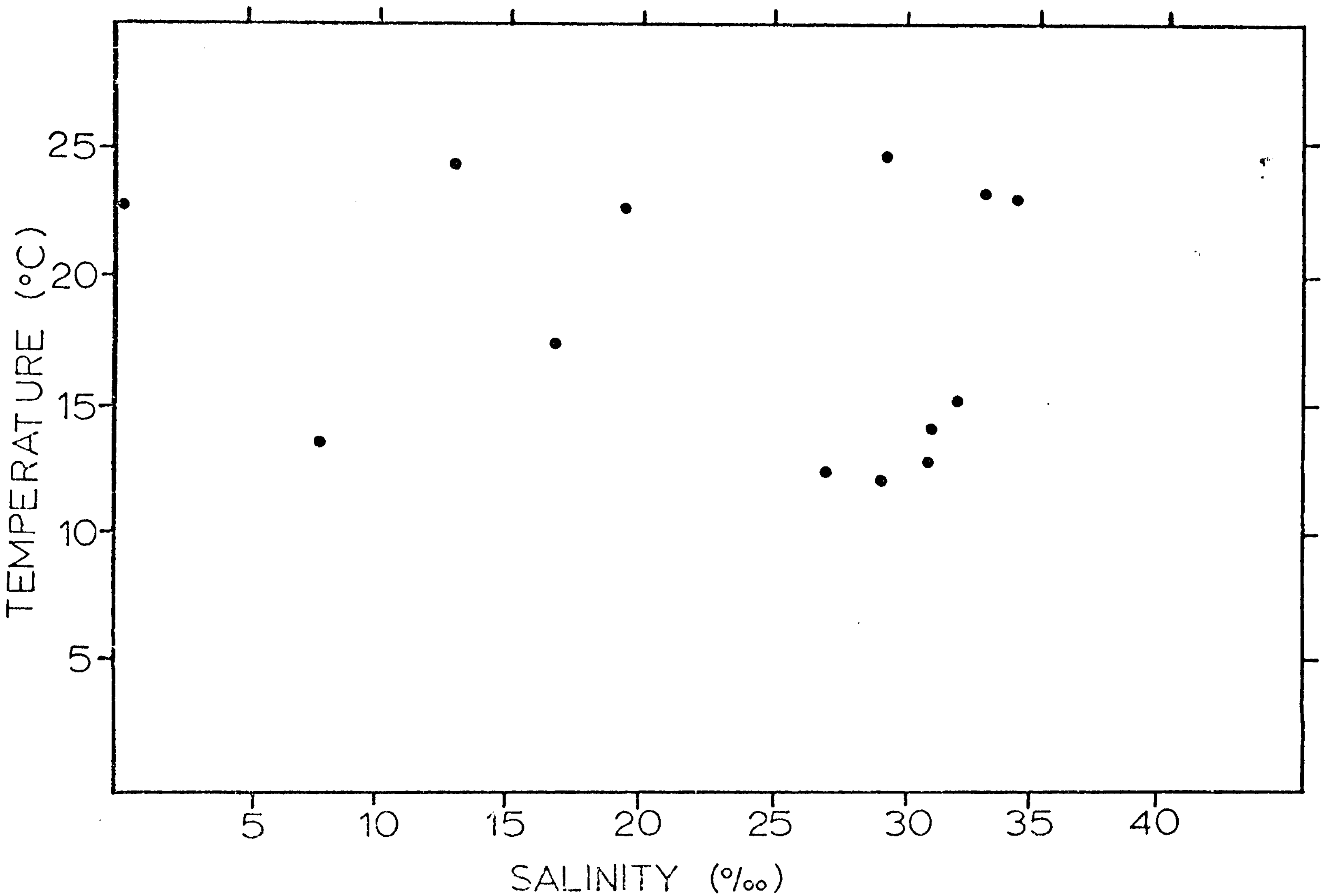
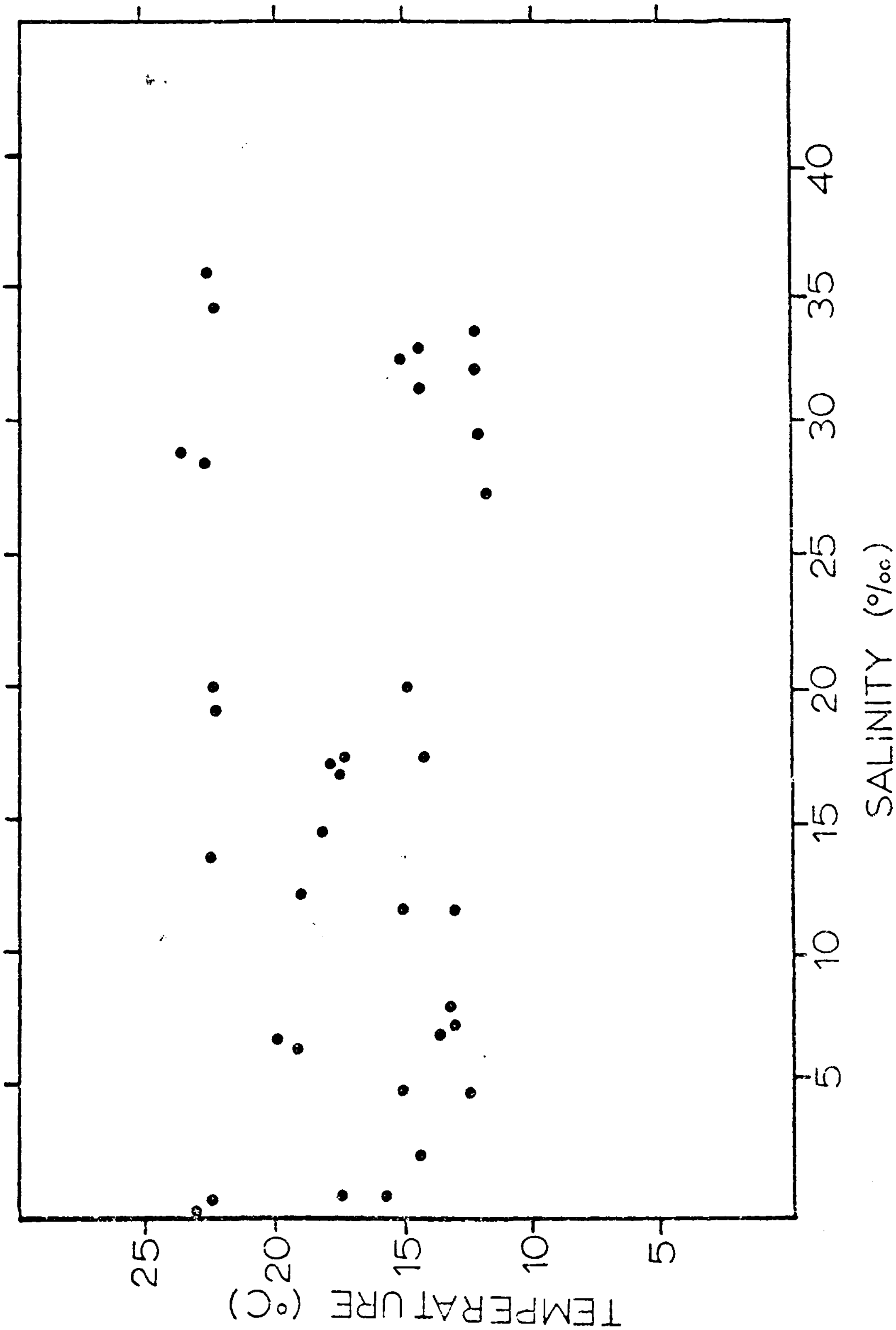


Figure 19
Achnanthes [illegible]



Cocconeis scutellum

Figure 20

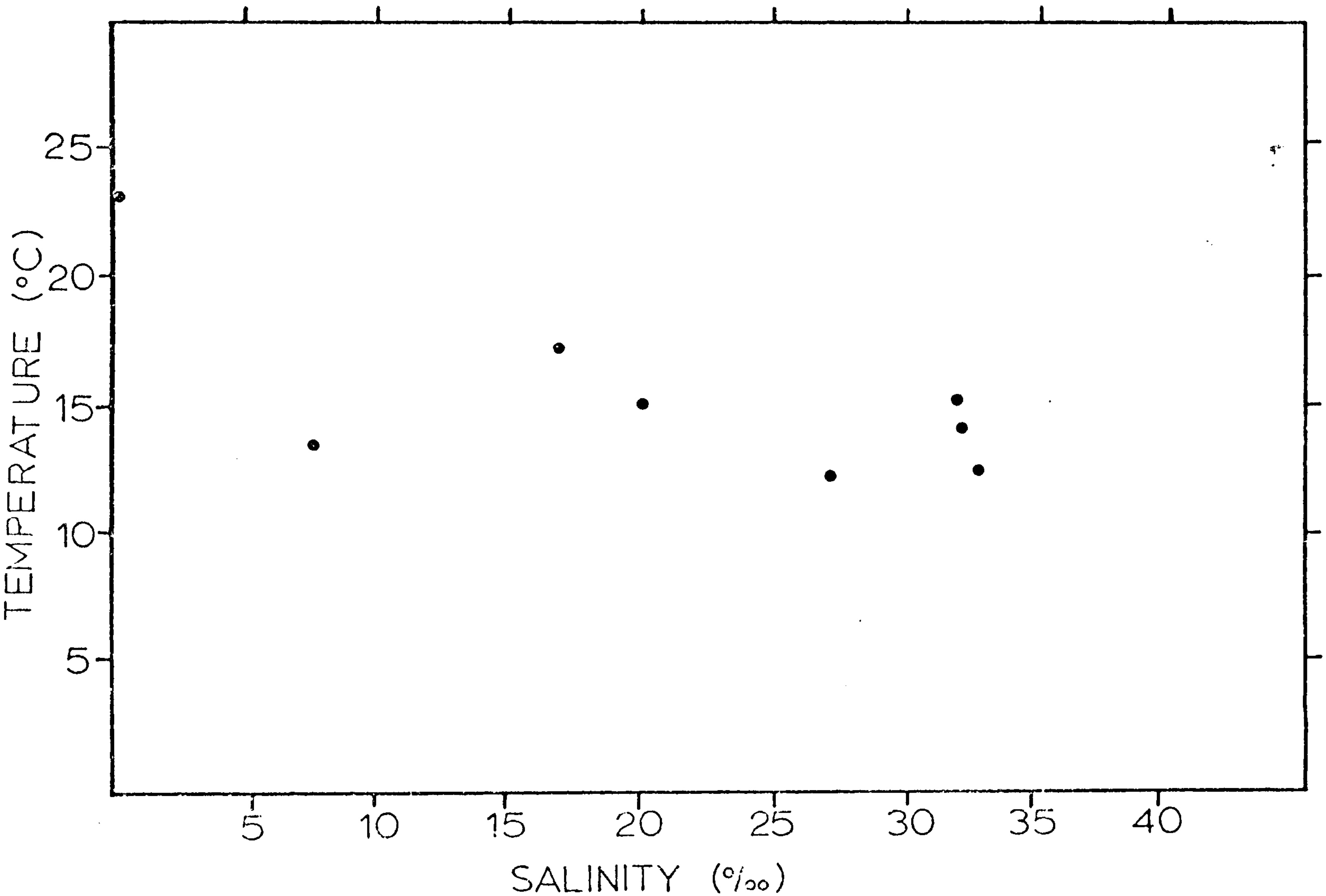
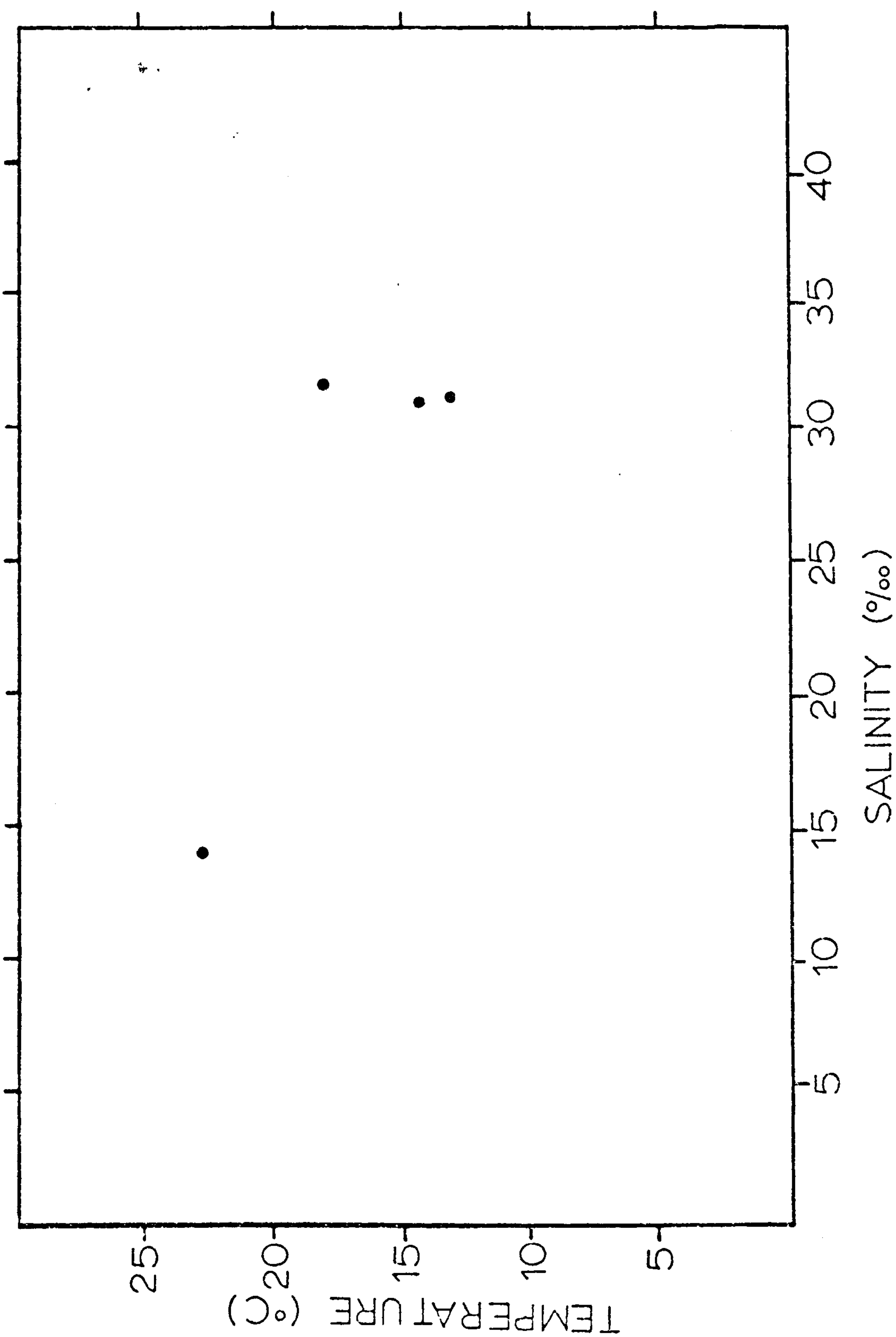


Figure 21

Coscinodiscus oculus iridis

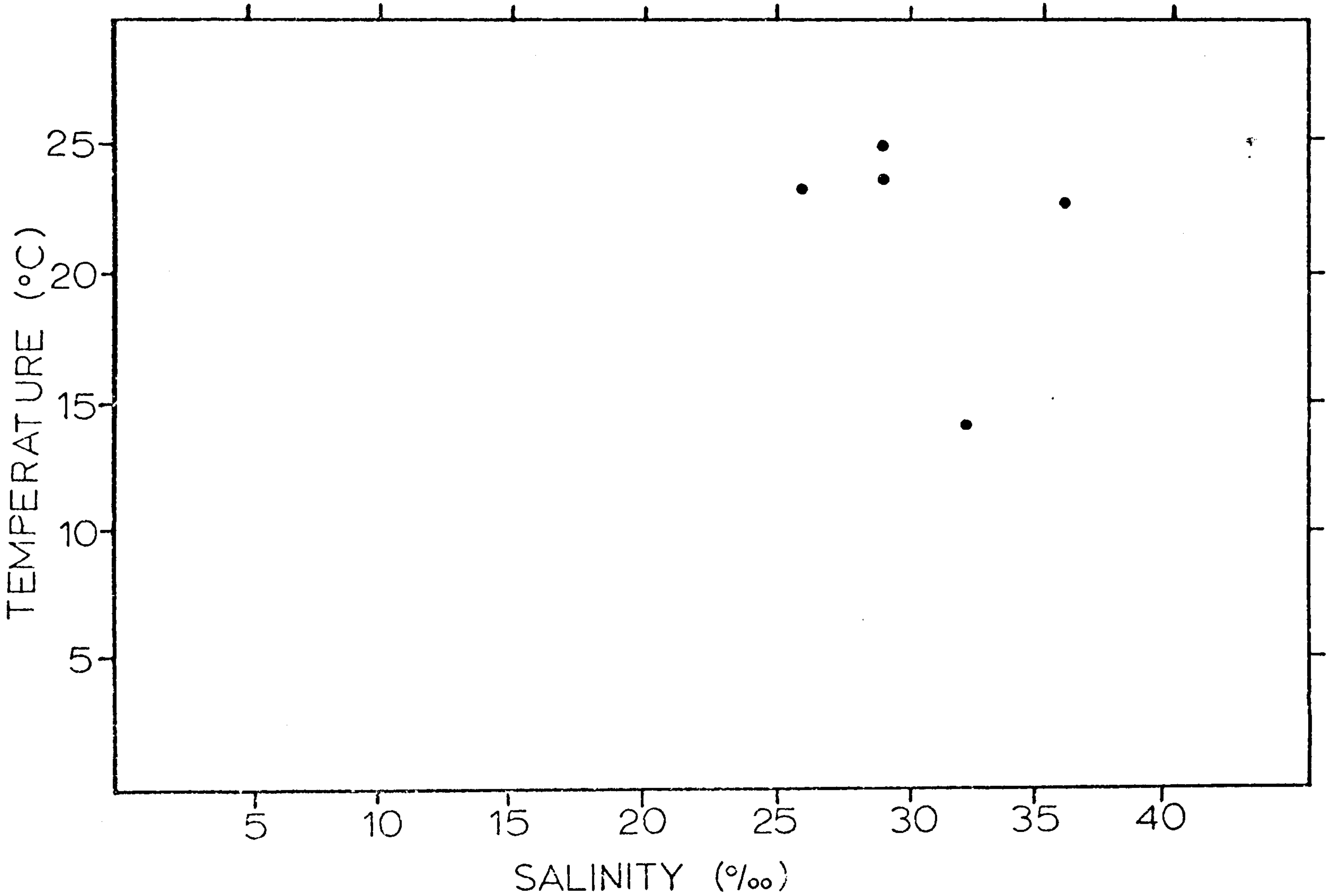


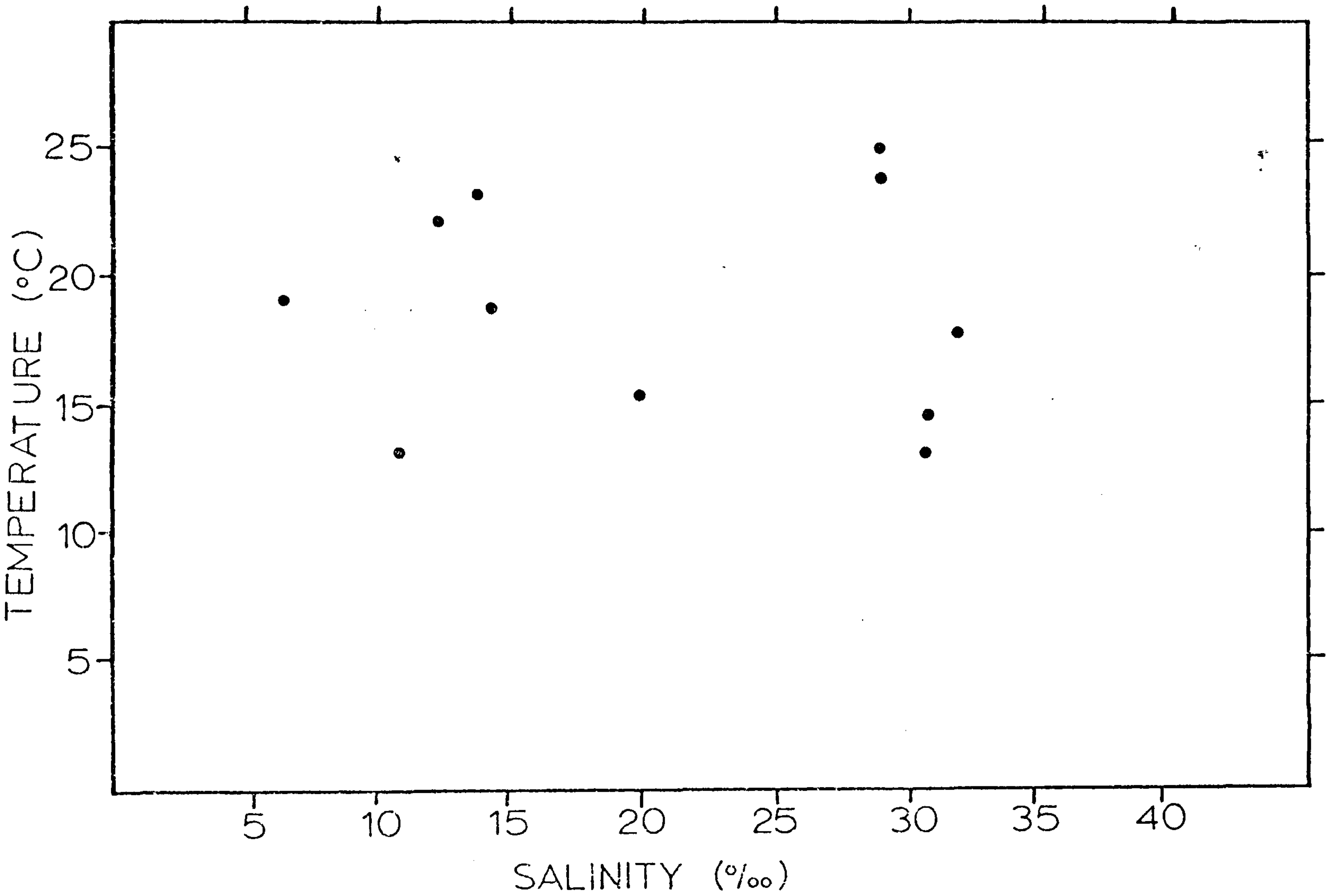
Coscinodiscus lineatus

Figure 22

Figure 23

Diatoma el[illegible]





Fragilaria capucina

Figure 24

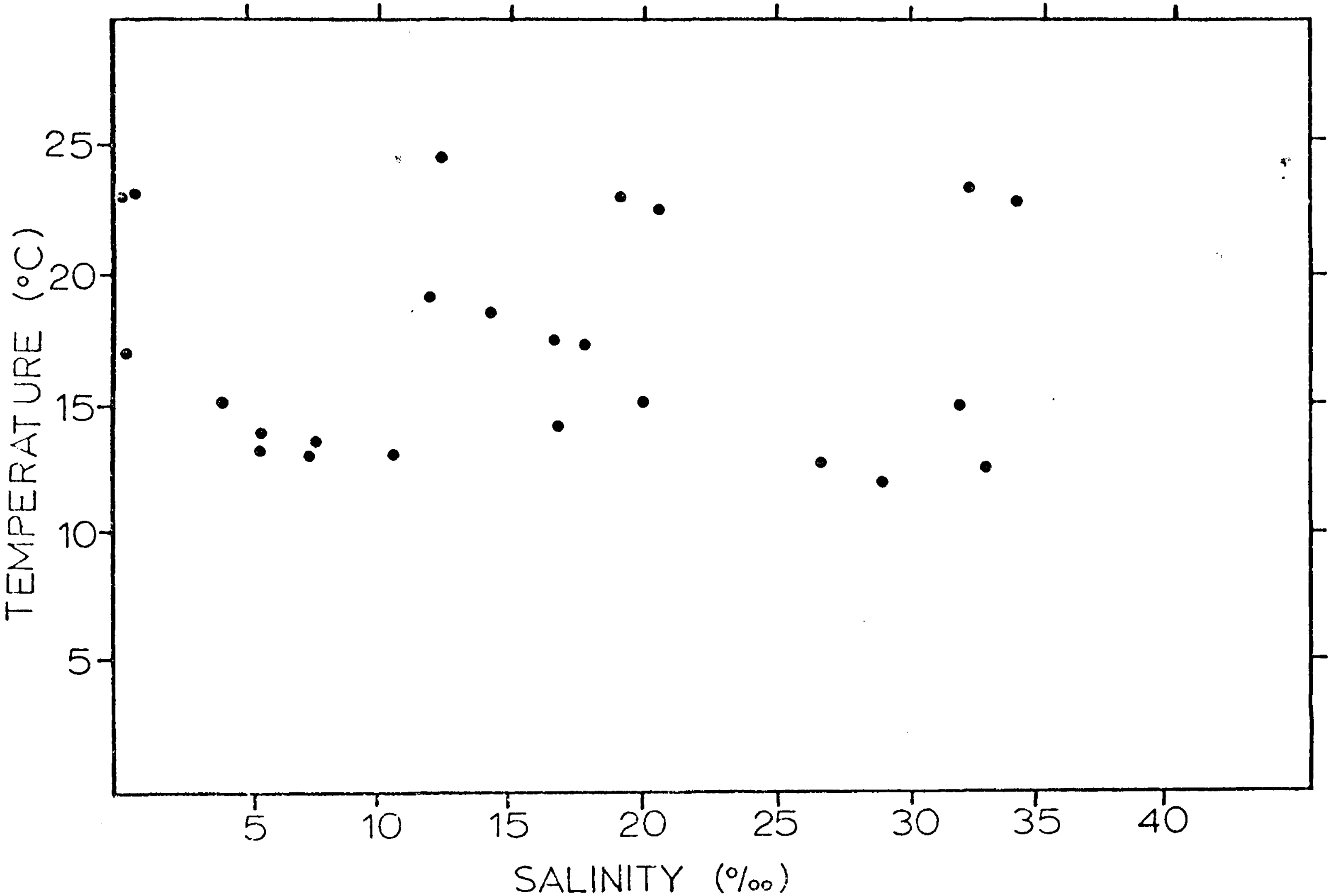


Figure 25

Fragilaria gracillima

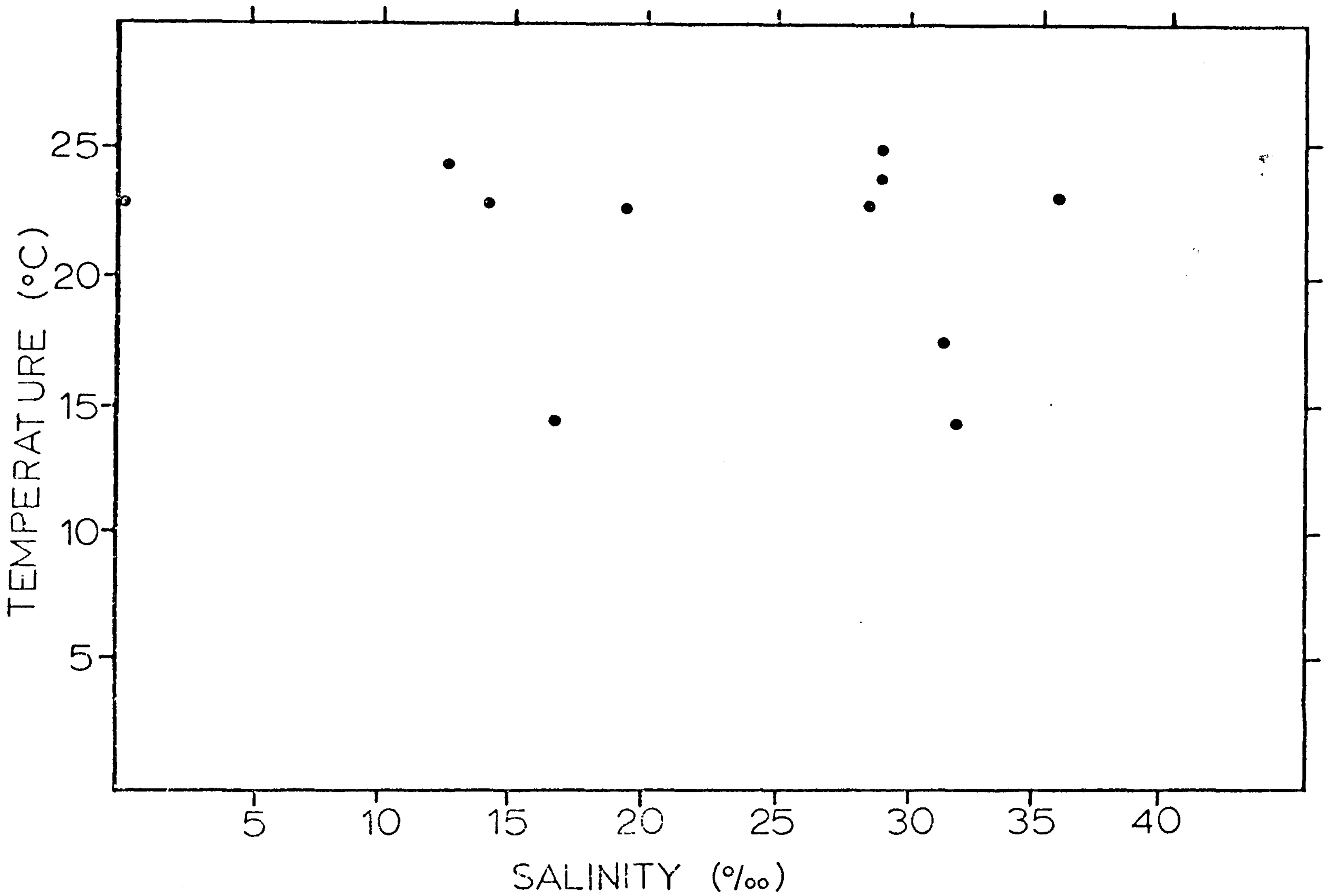


Figure 25

Grammatophora marina

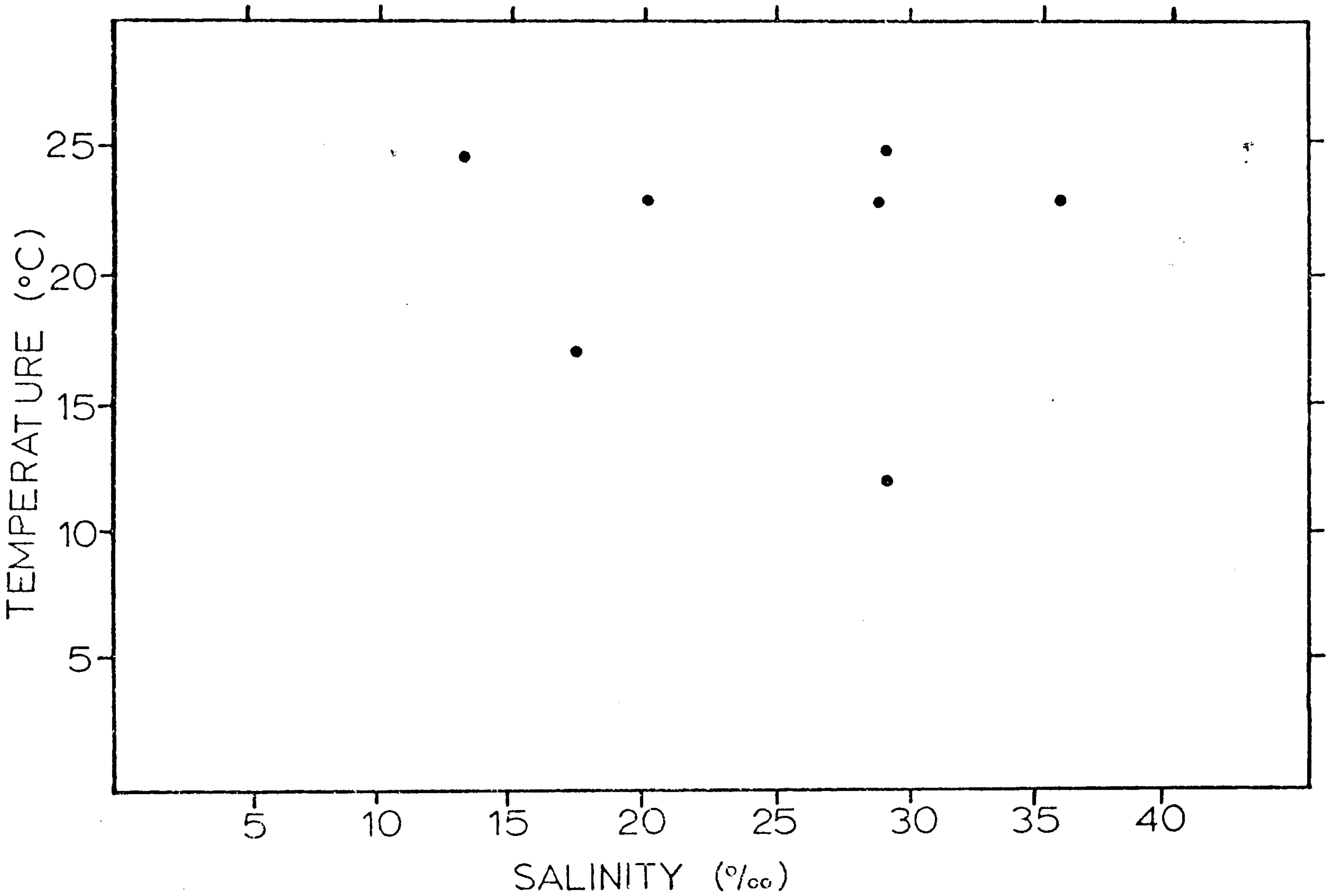
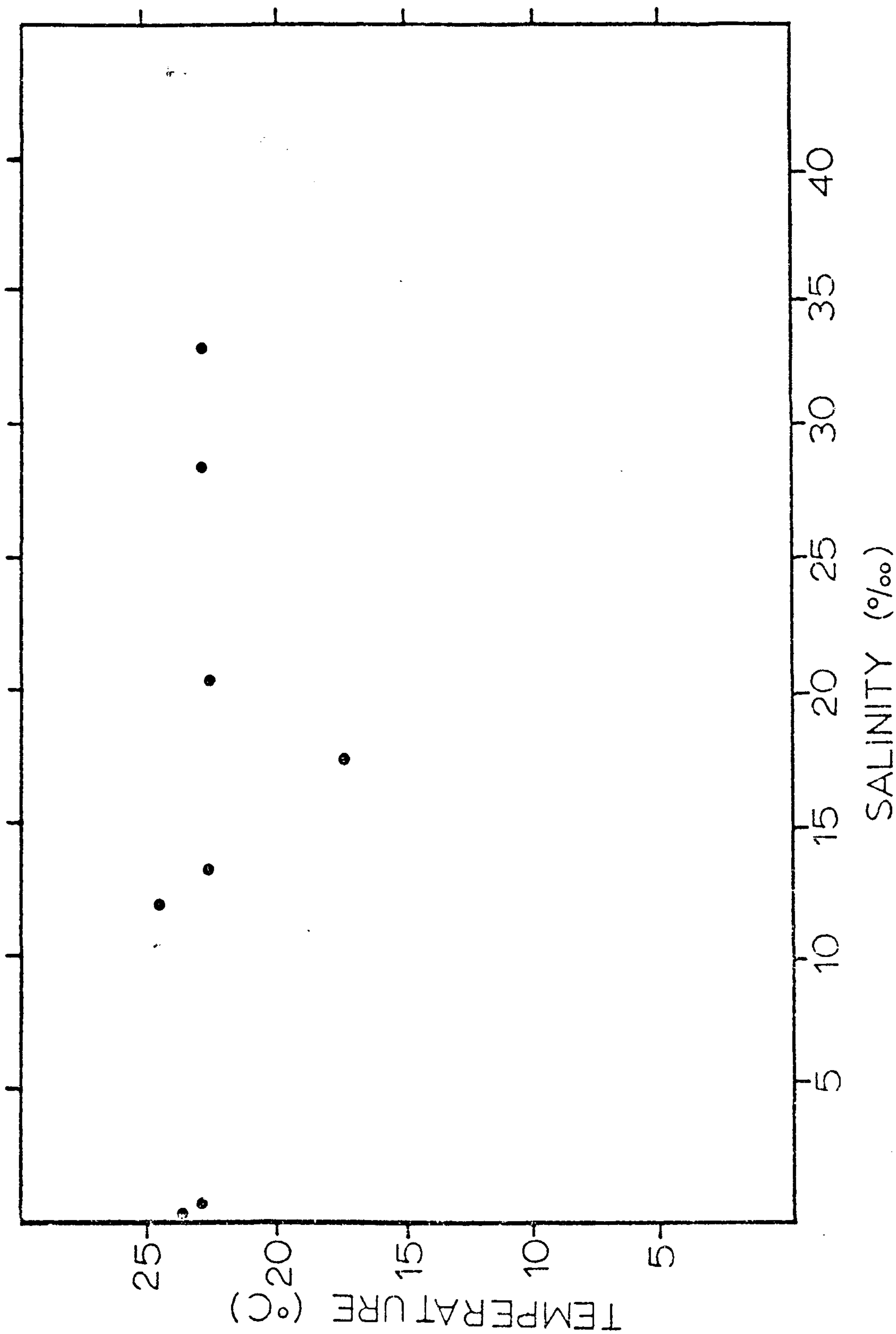


Figure 27
Licmophora communis



Licmophora gracilis

Figure 28

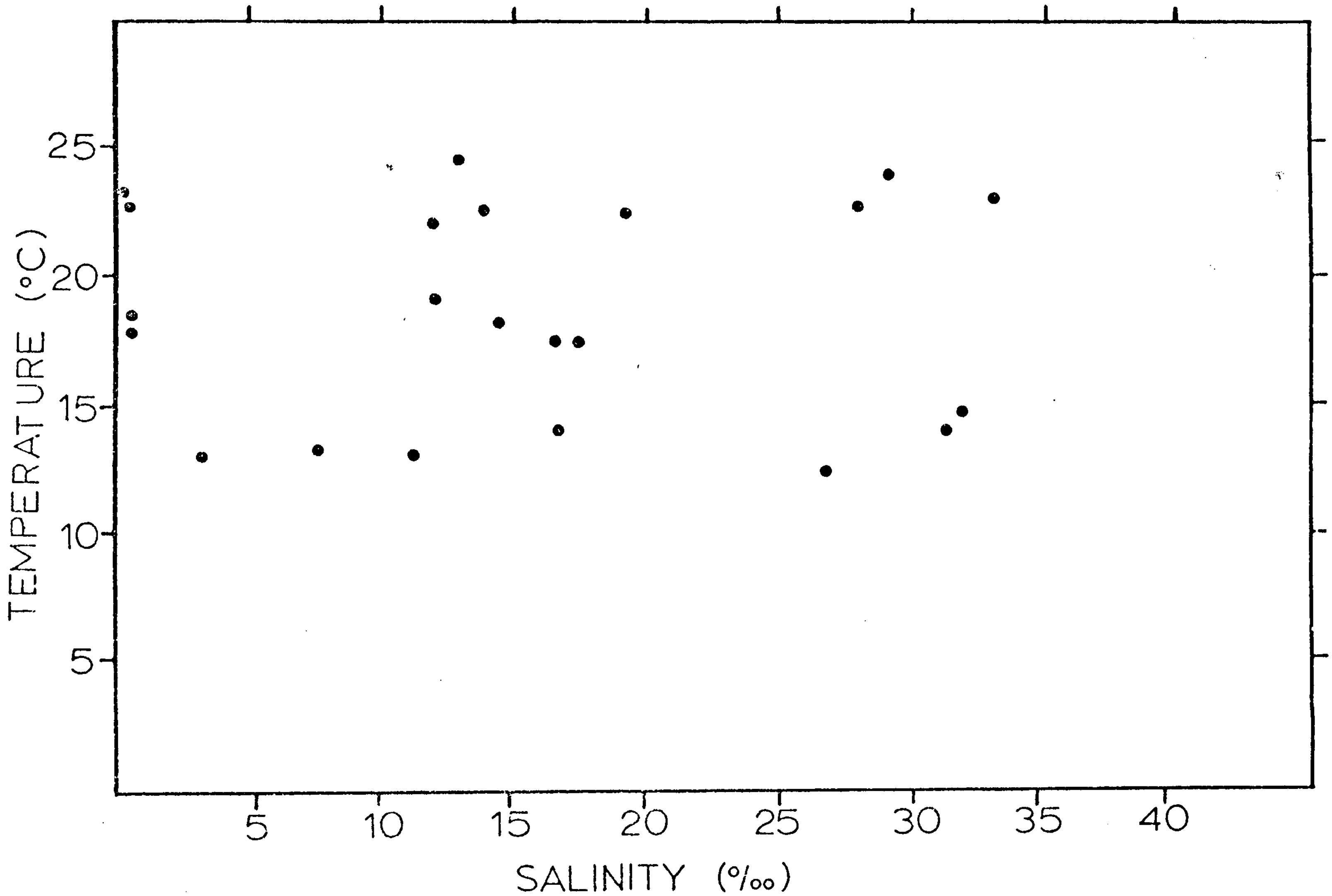


Figure 29 *Melosira moniliformis* var. *octagona*

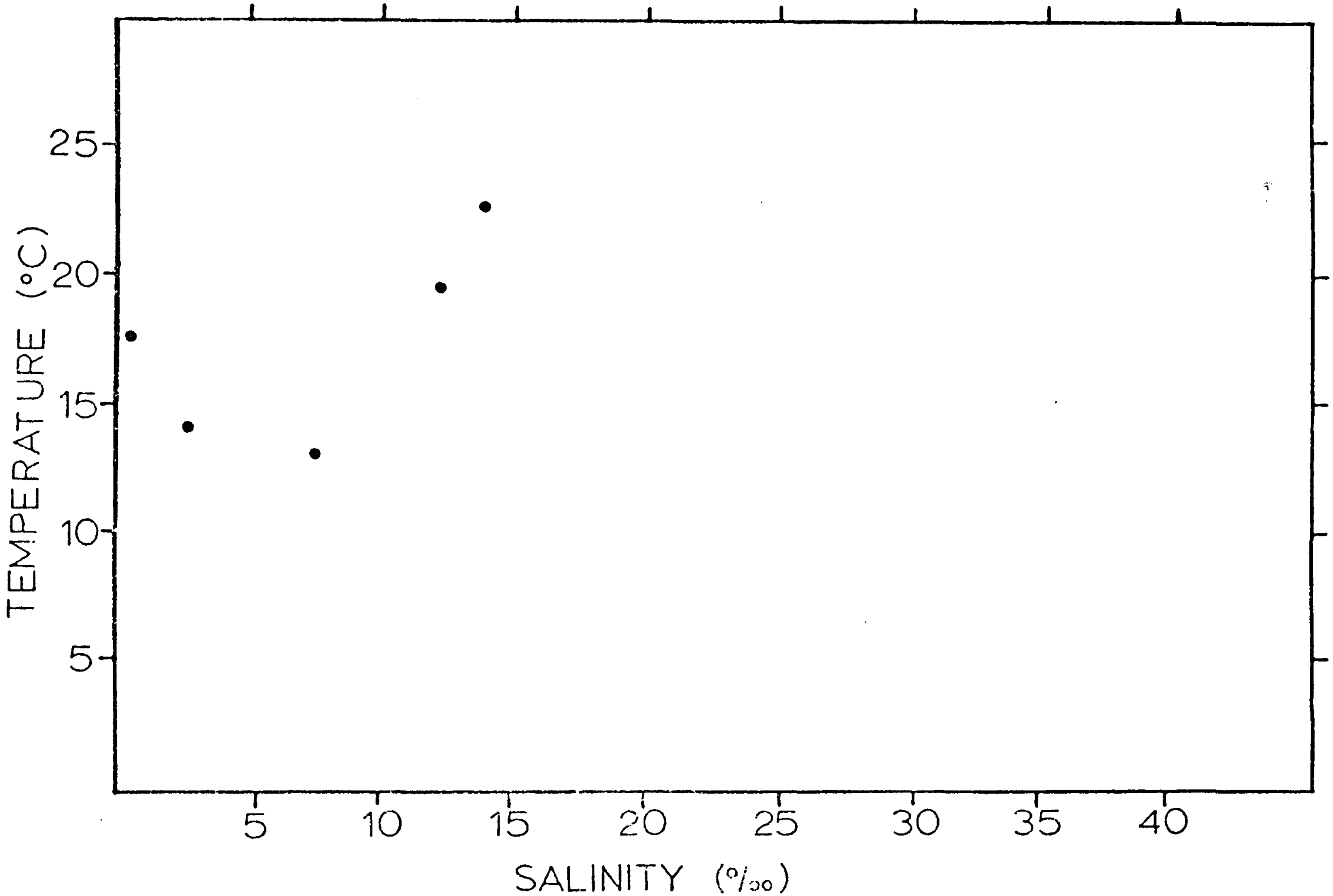


Figure 30 *Nitzschia acuminata*

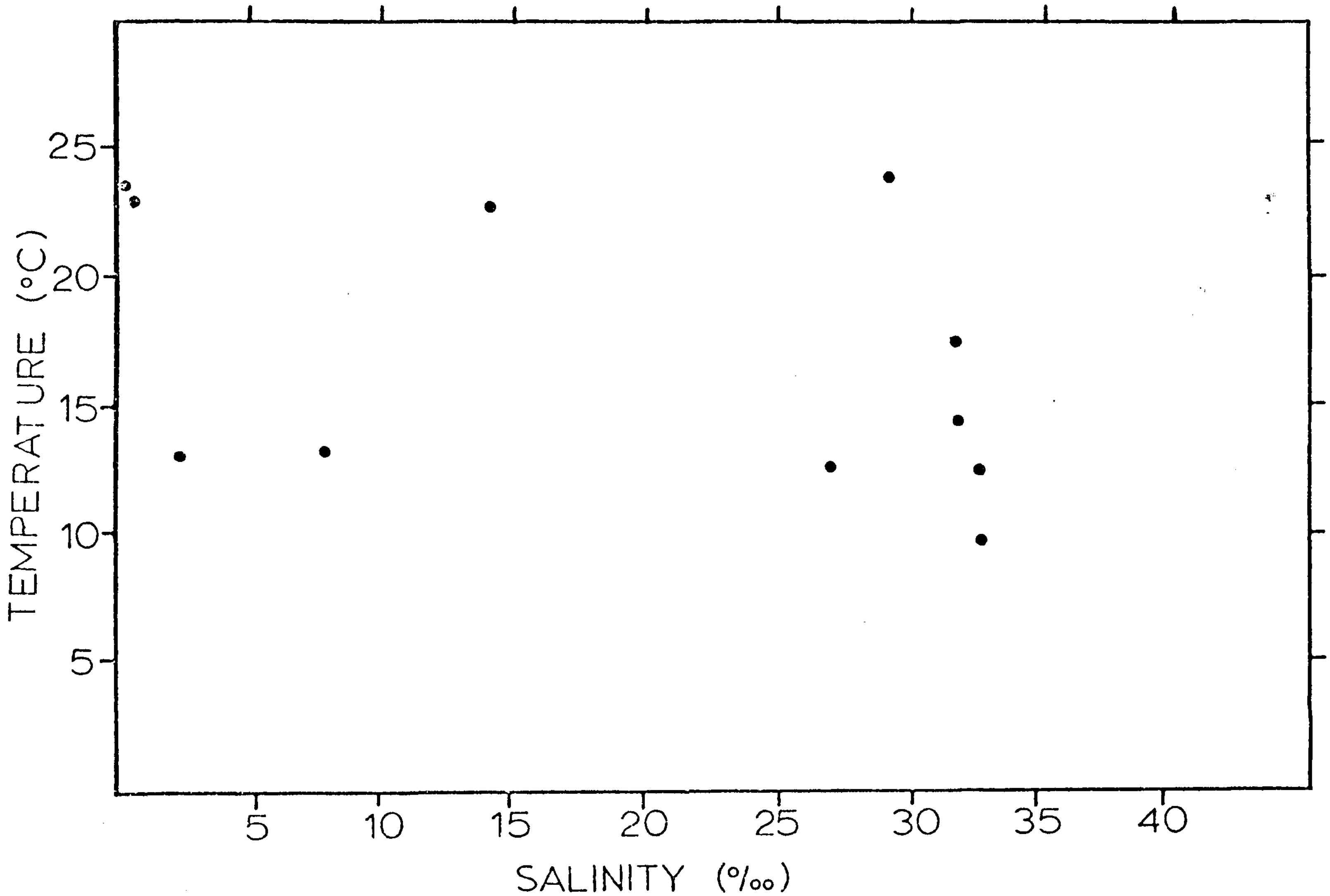
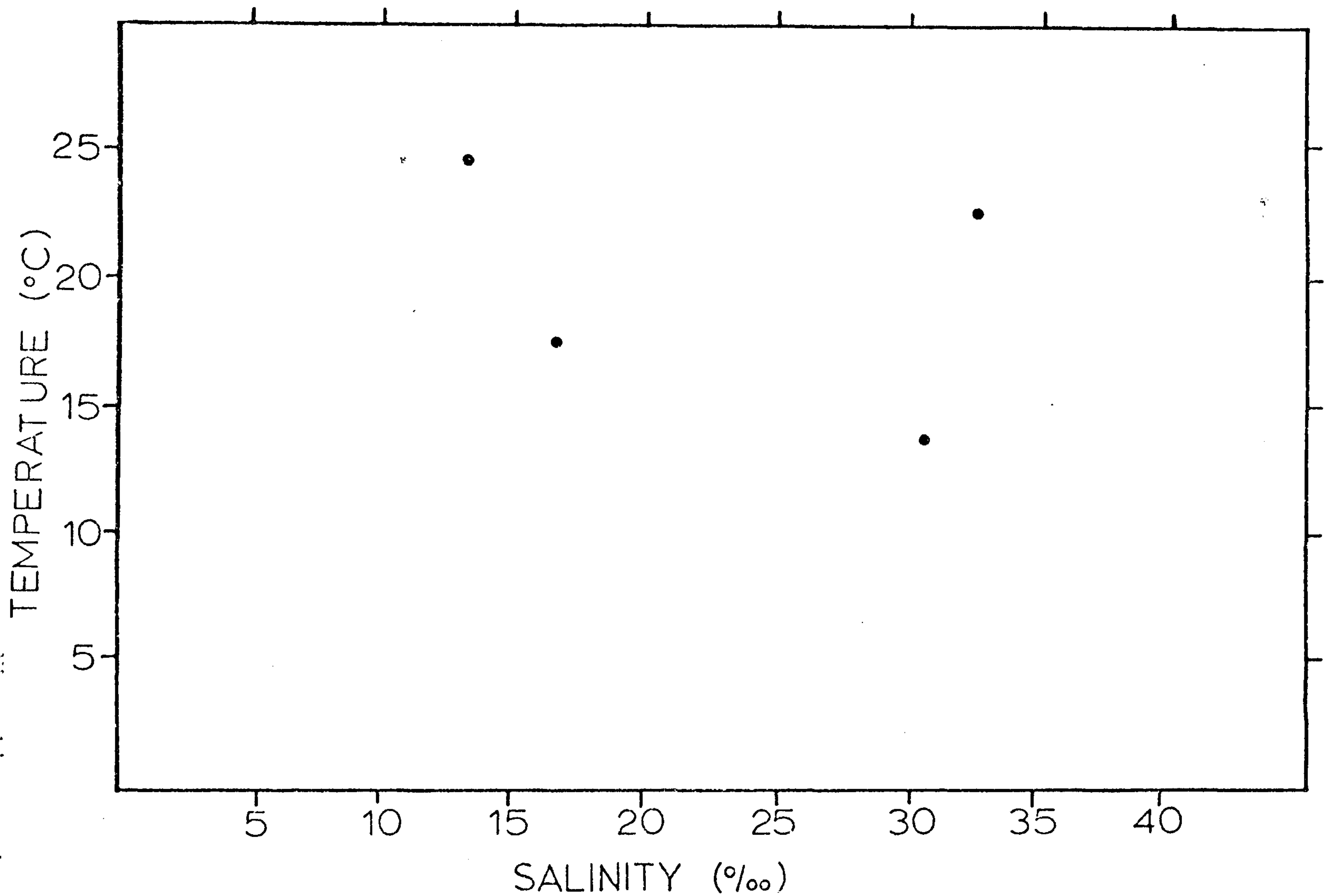


Figure 31
Nitzschia mirabilis



Nitzschia paradoxa
Figure 32

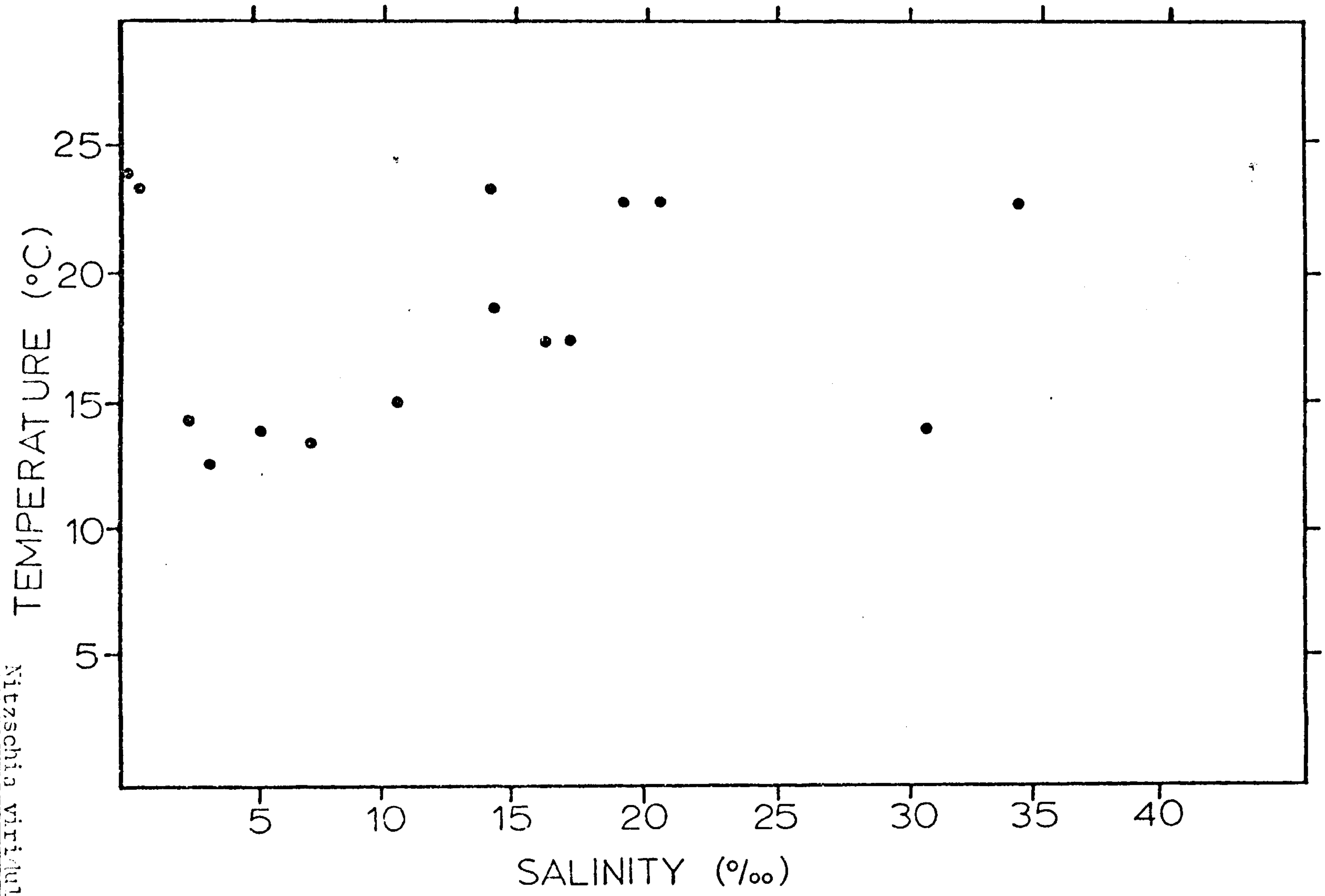


Figure 33

Nitzschia viridula

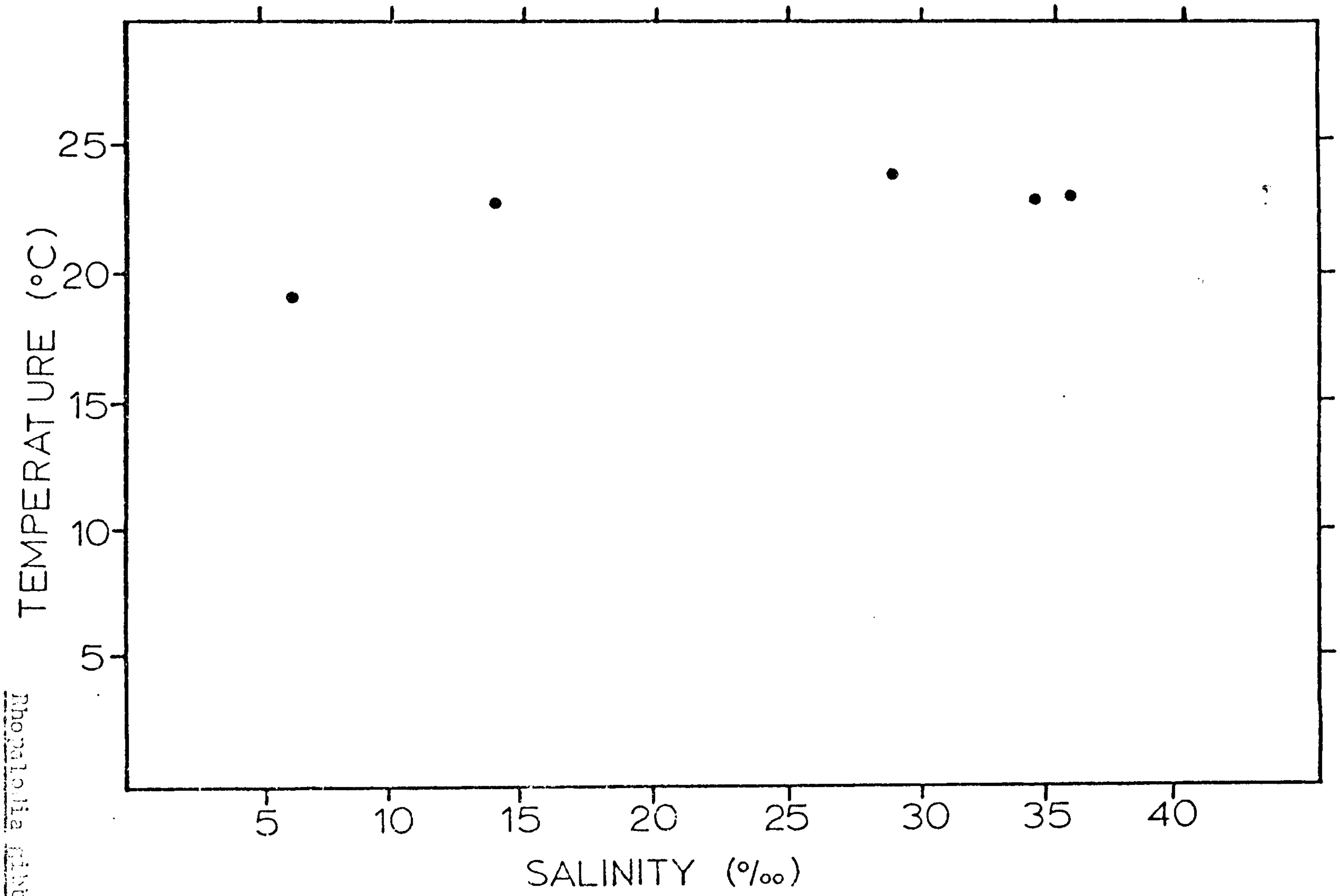


Figure 34
Rhopalodia gibba

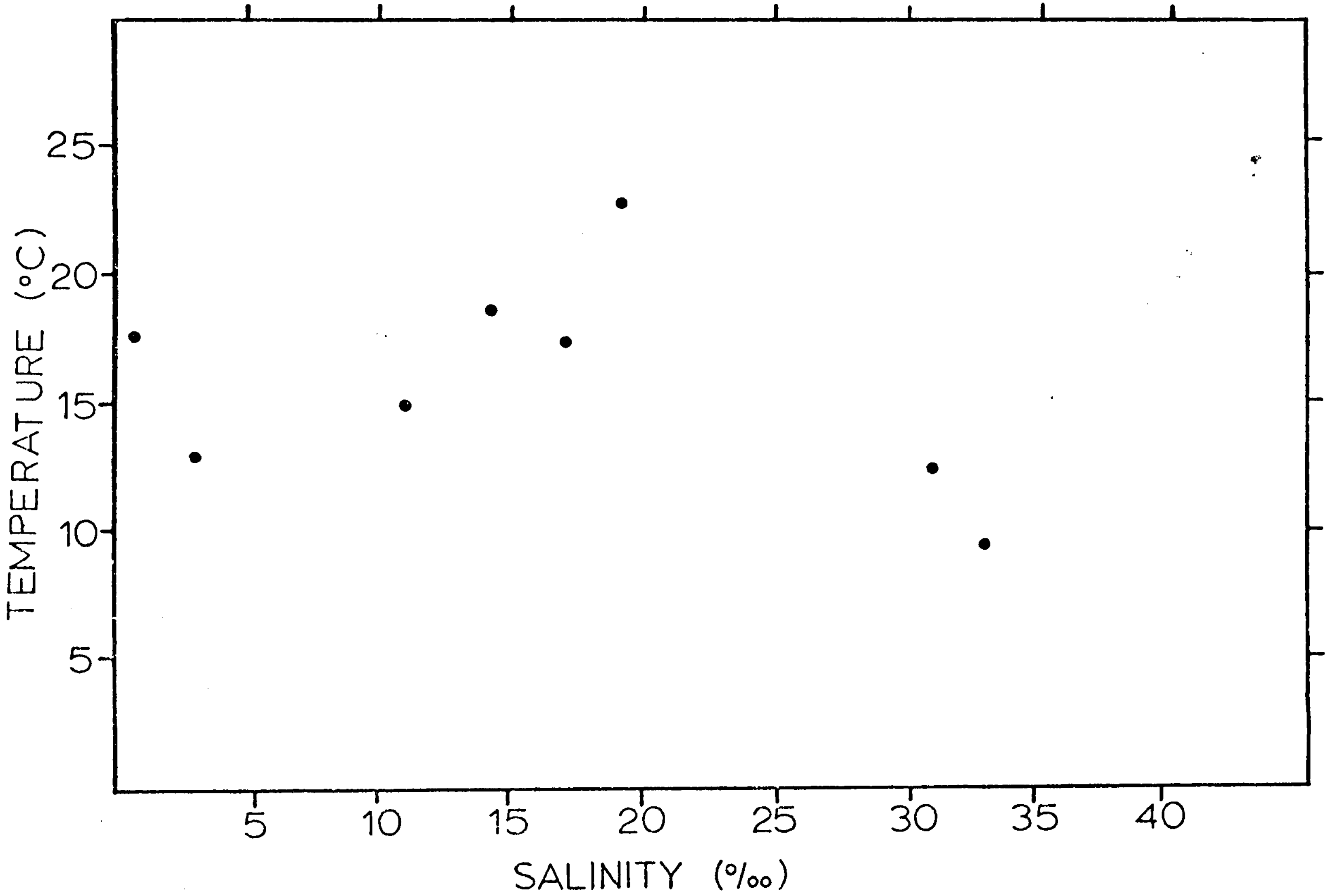


Figure 35

Staurosira mutabilis

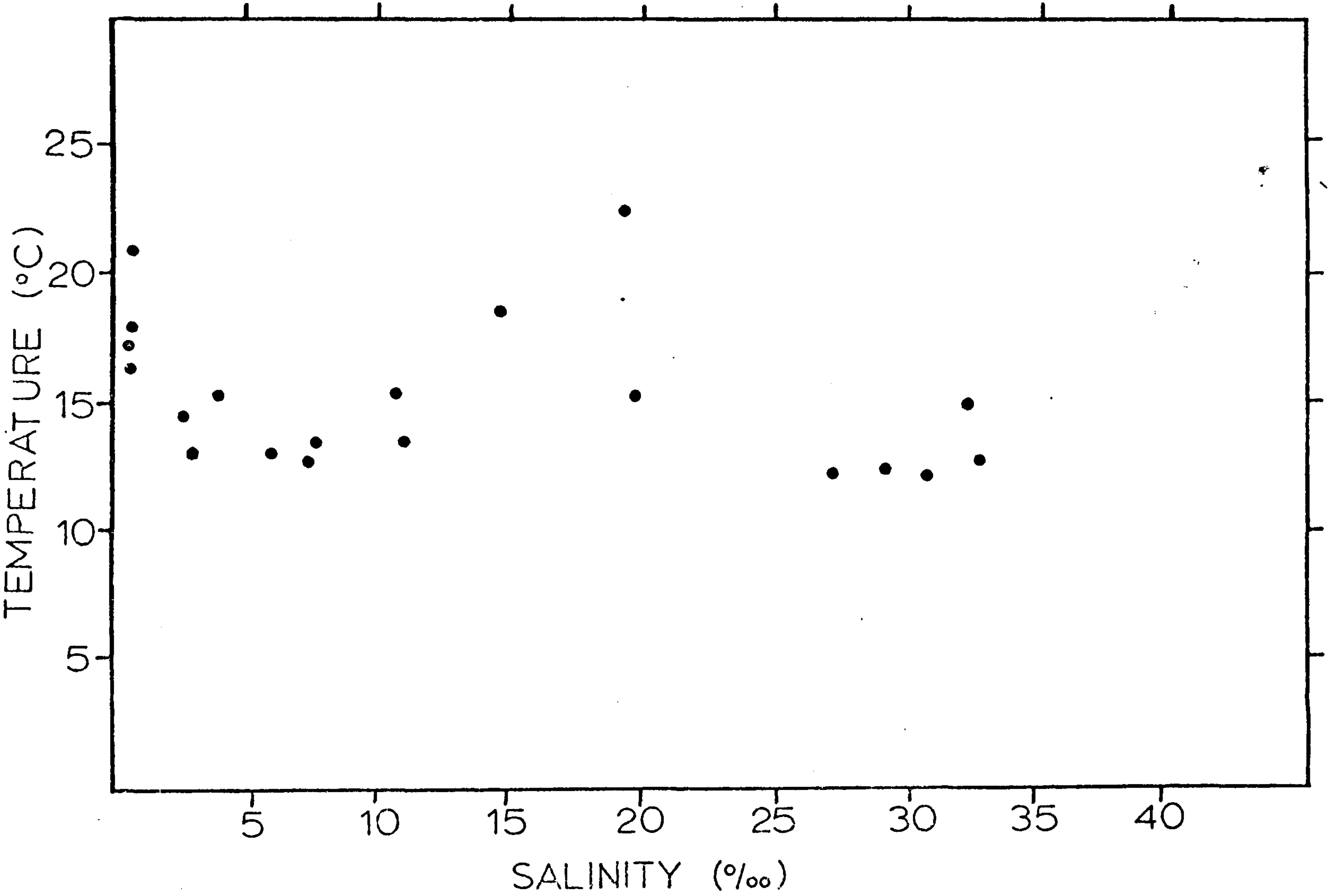


Figure 36
Synedra pulchella

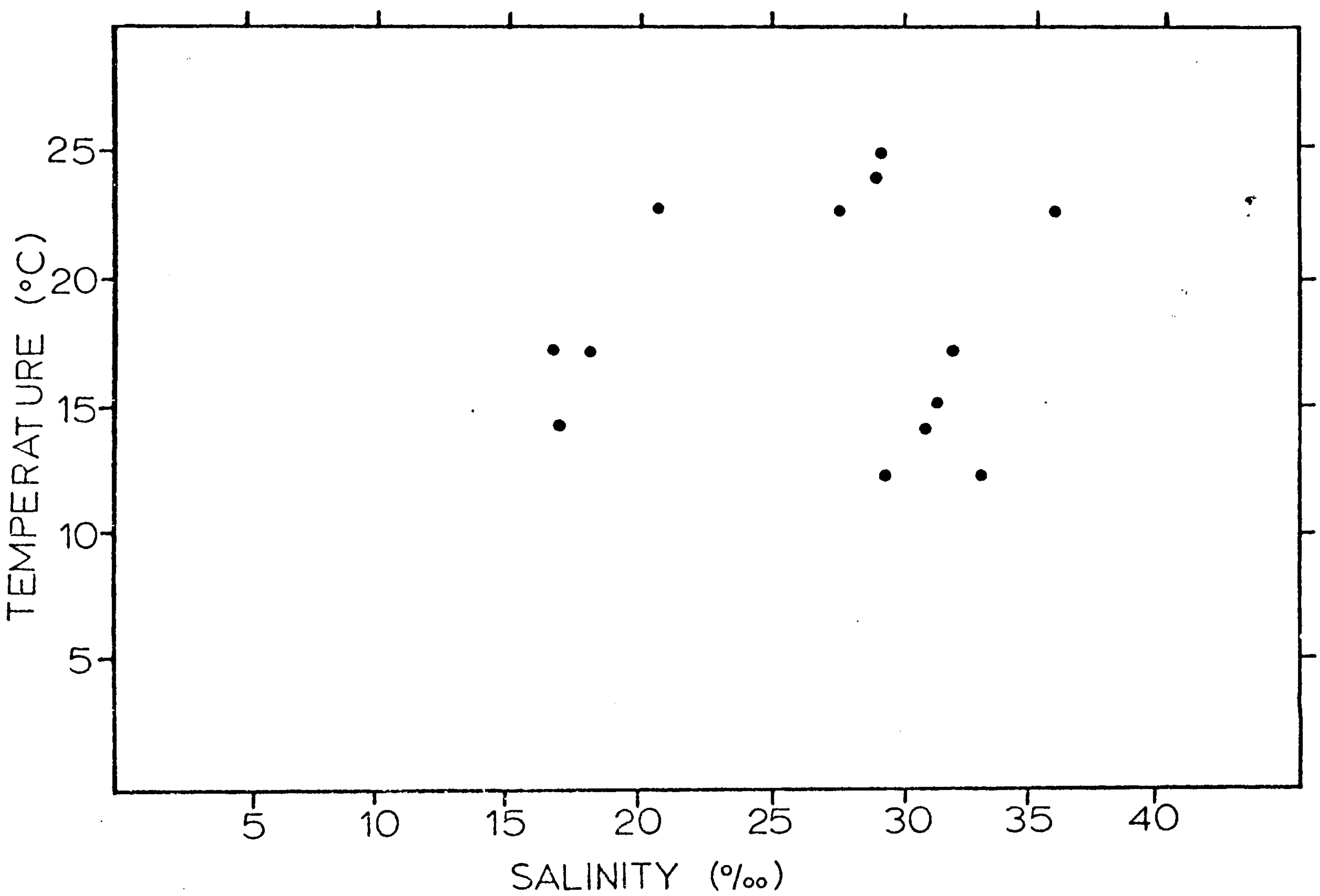


Figure 37

Tabellaria fenestrata

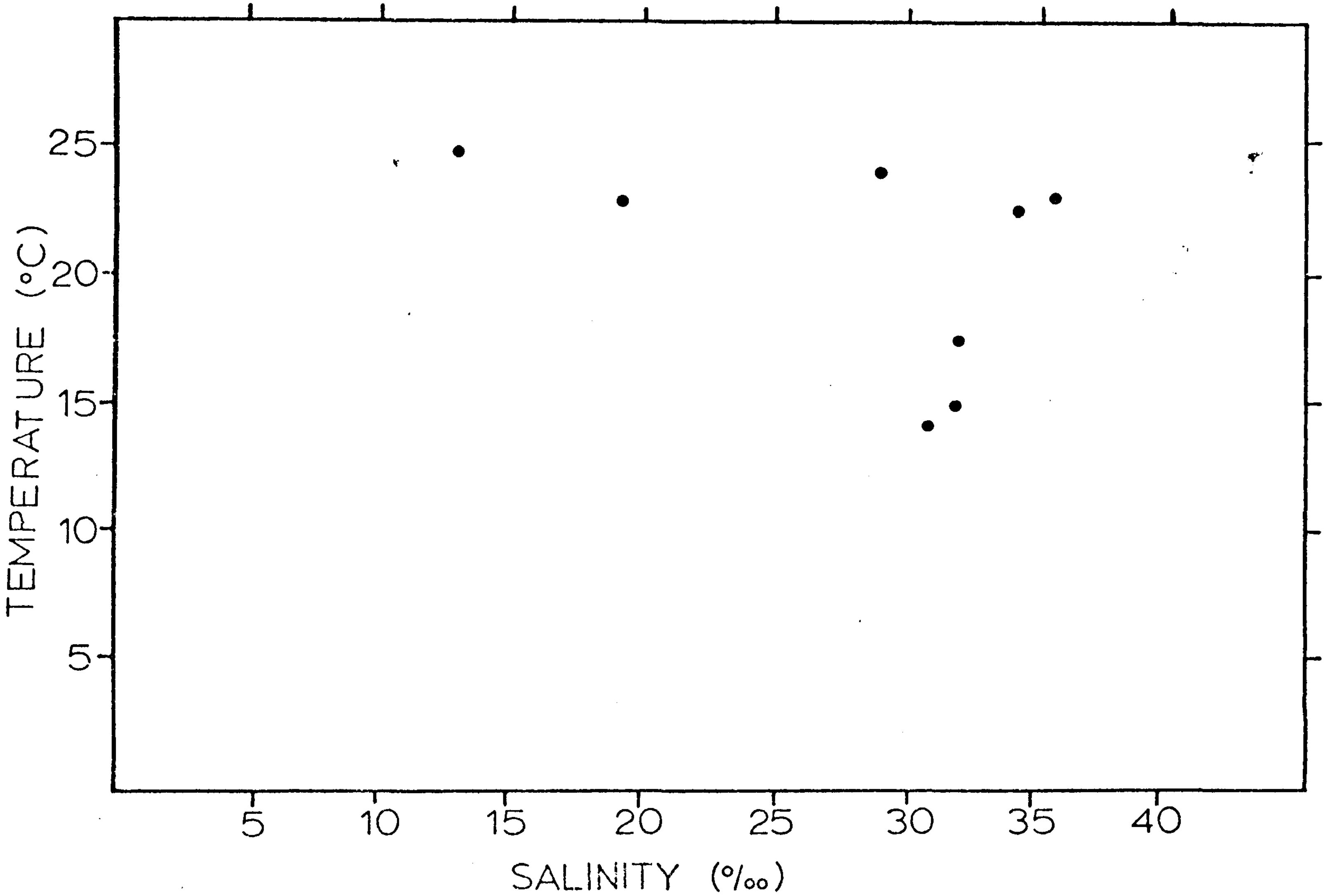


Figure 38

Thallassionema nitzschioides

REFERENCES CITED

Aleem, A. A. 1950. Distribution and ecology of British marine littoral diatoms. Journal of Ecology 38: 75-106.

Allen, W. E. 1936. Occurance of marine planktonic diatoms in a ten year series of daily catches in southern California. American Journal of Botany 23:#1: 60-63.

Armstrong, F. A. J., P. M. Williams, and J. D. Strickland. 1966. Photo-oxidation of organic matter in sea water by ultra-violet radiation, analytical and other applications. Nature 211: 481-483.

Belin, C. W. 1972. A quantitative study of the Swift method of cleaning diatom frustules, unpublished, 18 pages.

__________ and J. S. Zaneveld. 1972. An evaluation of the Swift method of cleaning diatom frustules. Virginia Academy of Science Journal, abstract only, p. 43.

Bernatowicz, A. J. 1958. Ecological isolation and independent speciation of the alternate generations of plants. Biol. Bull. 115:#2: 232.

Boyer, C. S. 1916. The Diatomaceae of Philadelphia and vicinity. J. B. Lippincott Co., Philadelphia, currently available through the University Microfilm Service, Ann Arbor, 143 pages, 40 plates.

__________. 1927. Synopsis of North American Diatomaceae. Proceedings of the Academy of Sciences, Philadelphia. 78: 1-228 and 79: 229-583.

Carpenter, E. J. 1970. Diatoms attached to floating Sargassum in the western Sargasso Sea. Phycologia 9(3/4): 269-274.

Castenholz, R. W. 1960. Seasonal changes in the attached algae of freshwater and saline lakes in the lower Grand Coolee, Washington. Limnology and Oceanography 5(1): 1-28.

__________. 1960. The algae of saline and freshwater lakes in the lower Grand Coolee, Washington. Limnology and Oceanography 5: 125-155.

__________. 1963. An experimental study of the vertical distribution of littoral marine diatoms. Limnology and Oceanography 4: 450-462.

__________. (abstr.) 1968. The vertical zonation of littoral marine diatoms. Bull. Ecol. Soc. Amer. 43(3): 98-99.

Chapman, V. J. 1962. The Algae. St. Martin's Press, Macmillan and Co., Ltd., Glasgow, 472 pages.

REFERENCES CITED (CONT'D)

Cleve, P. T. 1894. Synopsis of the Naviculoid Diatoms. Reprint, 1965, A. Asher and Co., Amsterdam. 220 pages.

Cleve-Euler, A. 1951. Die Diatommen von Schweden und Finnland, Vol. 1 and 2, Almquist and Wiksells Boktryckeri A. B., Stockholm. 639 pages.

Friedrich, H. 1969. Marine Biology. University of Washington Press, Seattle. 474 pages.

Fritsch, F. E. 1935. Structure and Reproduction of the Algae, Vol. I. Cambridge University Press, Cambridge. 643 pages.

Ghazzawi, F. M. 1933. The littoral diatoms of the Liverpool and Port Erin shores. J. Mar. Biol. Ass. U. K. 19: 165-183.

Gleason, H. A. and A. Cronquist. 1963. Manual of Vascular Plants of Northeastern United States and Adjacent Canada. D. Van Nostrand Co., Inc., Princeton, New Jersey. 810 pages.

Hendey, N. I. 1964. An introductory account of the smaller algae of British coastal waters, Part V. Bacillariophycophyta. Her Majesty's Stationery Office, London. 317 pages, 45 plates.

Hobbie, J. E. 1970. Hydrography of the Pamlico River Estuary, N. C. North Carolina State University Report, Project No. B-004-NC; 69 pages.

Holm-Hansen, O. 1969. Algae: Amounts of DNA and Organic Carbon in Single Cells. Science 163: 87-88.

Hopkins, J. T. 1964. A study of the diatoms of the Ouse estuary, Sussex. III. The seasonal variation in the littoral epiphyte flora and the shore plankton. J. Mar. Biol. Ass. U. K. 44: 613-644.

Hoyt, W. D. 1920. Marine algae of Beaufort, N. C., and adjacent regions. Bull. (U. S.) Bur. Fish. 36: 367-556.

Humm, H. 1968. Distribution of marine algae along the Atlantic Coast of North America. Phycologia 6(3/4): 37-51.

Hustedt, F. 1930. Die Kieselalgan Deutschlands, Osterreichs und der Schweiz, Band VII of Rabenhorst, J., Kryptogamen-Flora Deutschlands, Osterreich, und der Schweiz, Teil 1, 920 pages, 542 text figs., Teil 2, 576 pages, 465 text figs.

Johnson, M. J. 1957. Plankton. In, Hedgpeth, J. W. et al., Treatise on Marine Ecology and Paleoecology, Vol. 1, Geol. Soc. America, 67: 443-460.

Koblentz-Mishke, O. J., V. Volkovinsky, and J. G. Kabanova. 1971. Plankton primary production of the world ocean. In, Scientific Exploration of the South Pacific, W. Wooster, ed. (National) Acad. Sci., Wash. 183-191.

REFERENCES CITED (CONT'D)

Lohman, K. E. 1957. Marine diatoms. In, Hedgpeth, J. W. et al., Treatise on Marine Ecology and Paleoecology, Vol. 1. Geol. Soc. America, Memoir #67: 1059-1068.

Lund, J. W. G. 1949. Studies on *Asterionella formosa*. I. The origin of the cells producing seasonal maxima. J. Ecology 37: 389-419.

__________. 1950. Studies on *Asterionella formosa*. II. Nutrient depletion and the spring maximum. J. Ecology 38(1): 1-35.

__________. 1954. The seasonal cycle of the plankton diatom, *Melosira italica* (Ehr.) Kutz. subsp. *subartica* O. Mull. J. Ecology 42: 151-179.

Marshall, H. G. 1967. Phytoplankton in the James River Estuary, Virginia. I. Phytoplankton in Willoughby Bay and Hampton Roads, Chesapeake Bay Sci., v. 8, #2: 90-101.

__________. 1969a. Phytoplankton distribution off the North Carolina coast. Am. Midl. Nat. 81: 241-257.

__________. 1969b. Observations on the spatial concentrations of phytoplankton. Castanea 34: 217-222.

Merseschlowsky, C. 1901. Etudes sur l'endochrome des diatomees. Mem. Acad. Imp. St. Petersburg, ser. 8, vol. 11.

Mulford, R. A. and J. J. Norcross. 1971. Species composition and abundance of net phytoplankton in Virginia coastal waters, 1963-1964. Chesapeake Science 12: 142-155.

- - - - - . 1970. National Estuary Study, Volume 3, House Document #91-286, Part II, United States Department of the Interior, Fish and Wildlife Service, Bureau of Sport Fisheries and Wildlife, Government Printing Office, Washington, 326 pages.

Patrick, Ruth. 1943. The diatoms of Linsley Pond, Connecticut. Proc. Acad. Nat. Sci. Phil. 95: 53-110.

__________. 1948. Factors affecting the distribution of diatoms. Bot. Rev. 14: 473-524.

__________, M. H. Hohn, and J. H. Wallace. 1954. A new method for determining the pattern of diatom flora. Notulae Naturae, No. 259, 12 pages.

__________, and C. W. Reimer. 1966. The Diatoms of the United States. Monograph of the Acad. Nat. Sci., Phila. #13, 688 pages.

Patten, B., R. Mulford, and J. Warinner. 1967. An annual phytoplankton cycle in the lower Chesapeake Bay. Chesapeake Sci. 4: 1-20.

REFERENCES CITED (CONT'D)

Pickett, T. E., and R. L. Ingram. 1969. The modern sediments of Pamlico Sound, North Carolina. Southeastern Geol. 11(2): 53.83.

Pratt, D. M. 1959. The phytoplankton of Narragansett Bay. Limnol. and Oceanogr. 4(4): 26-59.

________. 1965. The Winter-Spring diatom flowering in Narragansett Bay. Limnol. and Oceanogr. 10(2): 173.

Ralfs, J. 1843. On the diatomaceae. Ann. Nat. Hist., xi, p. 447-457, and xii, p. 104-111.

Raymont, J. E. G. 1963. Plankton and productivity in the oceans. Pergamon Press, London, 660 pages.

Riley, G. A. 1957. Phytoplankton of the North Central Sargasso Sea. Limnol. and Oceanogr. 2(3): 133-151.

Roelofs, E. W., and D. F. Bumpus. 1953. The hydrography of Pamlico Sound. Bull. Mar. Sci. Gulf and Carib. 3: 181-205.

Rohlf, F. J. and R. P. Sokal. 1969. Biometry. W. H. Freeman and Co., San Francisco. 541 pages.

________ and R. P. Sokal. 1969. Statistical Tables. W. H. Freeman and Co., San Francisco. 253 pages.

Round, F. E. 1965. The Biology of the Algae. Edward Arnold (Publishers) Ltd. London. 269 pages.

Ryther, J. H. 1969. Photosynthesis and fish production in the sea. Science 166: 72-76.

Setchell, W. A. 1903. The upper temperature limits of life. Science Vol. XVII, 441: 934-937.

________. 1920a. Stenothermy and zone invasion. The American Naturalist, vol. LIV: 385-397.

________. 1920b. The temperature interval in the geographic distribution of marine algae. Science, Vol. LII, 1339: 187-190.

Silva, P. C. 1962. Classification of Algae. In, R. A. Lewin, ed., Physiology and Biochemistry of Algae. Academic Press, New York, Appendix A.

Smith, G. M., et al. Manual of Phycology. Waltham, Mass., Chron. Bot. Co., 375 pages.

Steemann-Nielsen, E. 1955. Production of organic matter in the oceans. J. Mar. Res. 14(4): 375-386.

REFERENCES CITED (CONT'D)

Swift, E. 1967. Cleaning diatom frustules with ultraviolet radiation and peroxide. Phycologia 6(2/3): 161-163.

Talling, J. F. 1962. Freshwater algae. In, Lewin, R. A., Physiology and Biochemistry of Algae. Academic Press, New York, p. 744-757.

Taylor, W. R. 1964. Marine Algae of the Northeastern Coast of North America, Vol. 1 and 2. University of Mich. Press, Ann Arbor, 870 pages.

Van Heurck, H. 1896. A Treatise on the Diatomaceae. Baxter, London, 558 pages, 35 plates.

Whitford, L. A. 1956. Communities of algae in springs and spring streams of Florida. Ecology 37: 433-442.

Williams, L. G. 1948. Seasonal alterations of marine floras at Cape Lookout, North Carolina. Amer. J. Bot. 35: 682-695.

Wood, E. J. F. 1965. Marine Microbial Ecology. Chapman and Hall, Reinhold, and Co., New York. 243 pages.

Zaneveld, J. S. 1937. The littoral zonation of some Fucaceae in relation to desiccation. J. of Ecol. 25: 431-468.

__________. 1964. The benthic algal vegetation of Antartica. I. The Ross Sea. Bull. U. S. Antarctic Proj. Off. 5: 66-69.

__________. 1966. Vertical zonation of Antarctic and sub-Antarctic benthic algae. Antarctic J., U. S. A. 1: 211-213.

__________. 1969. Facotrs controlling the delimitation of littoral benthic marine algae. Amer. Zool. 9: 367-391.

__________, and W. D. Barnes. 1965. Reproductive periodicities of some benthic algae in Lower Chesapeake Bay. Chesapeake Sci. 6(1): 17-32.

Collections Observed:

Diatoms; edited by Cleve, P. T., and Moller. 1877. Upsala. Reviewed by Grunow. At the Academy of Natural Sciences, Philadelphia.

Original specimens of Diatomacearum, Species Typical, of H. L. Smith. 1869. At the Academy of Natural Sciences, Philadelphia.

Diatom Herbarium, accession of the William Smith Collection. 1853. At the Academy of Natural Sciences, Philadelphia.

Types du Synopsis des Diatomees du Belgique. Dr. H. Van Heurck. At the Academy of Natural Sciences, Philadelphia.