UNIVERSITY OF SOUTHAMPTON

TAXONOMIC AND ECOLOGICAL STUDIES ON ANTARCTIC OCTOCORALS OF THE GENUS *Thouarella* (OCTOCORALLIA: PRIMNOIDAE)

by

Tânia Aparecida Silva Brito

A dissertation submitted in candidature
for the degree of Doctor of Philosophy
at the University of Southampton

December 1993

UNIVERSITY OF SOUTHAMPTON

ABSTRACT

FACULTY OF SCIENCE

OCEANOGRAPHY

Doctor of Philosophy

TAXONOMIC AND ECOLOGICAL STUDIES ON ANTARCTIC OCTOCORALS OF THE GENUS *Thouarella* (OCTOCORALLIA: PRIMNOIDAE)

by Tânia Aparecida Silva Brito

Amongst the primnoids collected by the Brazilian Antarctic Expeditions, the genus *Thouarella* is the most abundant and *Thouarella variabilis*, the dominant species. *Thouarella variabilis* shows a notable range of variation of characters. Some of the characters which have been applied in diagnosing this species are inconsistent and should be used with caution. An evaluation of the validity of these characters as reliable descriptors for this species is presented.

An investigation of the colony morphology; structure, formation and function of sclerites; growth and estimates of age; internal morphology and reproductive biology of *Th. variabilis* was undertaken.

The results of the studies on reproductive biology, showed that colonies of *Th. variabilis* are gonochoric brooders. The polyp fecundity is low with only one larva being released per female polyp at a time. The presence of oocytes in different stages of development within the same polyp suggests a two year cycle of oogenesis. The developing oocyte grows to a maximum size of 660 μm. A non-feeding, non-pelagic lecithotrophic planula larva is retained within the polyp until full development is attained. The larva reaches a length of 860 μm and occupies on average 80 % of the polyp volume.

The study of the extrinsic and intrinsic aspects which could cause variations in the features of the colonies show that: the colony form; branching pattern and density of the colony; arrangement, distribution and number of polyps; and the appearance of the sclerites varied as a function of current intensity and direction, water temperature, availability of food and essential compounds, and the presence of commensals. Intrinsic aspects such as growth and reproduction affect the shape of the polyp, the arrangement of the sclerites, the density of the branches and as a consequence, the whole appearance of the colony. It is suggested that these variations might have contributed to erroneous interpretations in the past about the taxonomic classification of the species of the genus *Thouarella*.

Thouarella variabilis is diagnosed and redescribed. The most consistent characters shown by this species and the range of variation of the inconsistent characters are defined. Through the work undertaken on this species the taxonomic position of the other species of the genus *Thouarella*, which present similar ranges of variation of characters, have also been evaluated and are discussed.

ACKNOWLEDGEMENTS

This research work was made possible through support provided by the CAPES (Coordenaçao de Aperfeiçoamento de Pessoal de Nivel superior) and SeCirm (Secretaria da Comissao Interministerial para os Recursos do Mar).

I wish to express my appreciation for the patience and support given by my supervisor Dr. Paul A. Tyler. I thank Dr. Andrew Clarke, British Antarctic Survey, Cambridge, for his constant interest and incentive during the elaboration of this research work. Particular thanks are extended to Dr. Manfred Grasshoff, Senckenberg Museum, Frankfurt, Germany, for his dedication and effort of teaching me the basic knowledge of the study of octocorals. I also thank Dr. Stephen Weinberg, Luxemburg, for his valuable help on the elaboration of part of my field work. I thank Dr. D. Kühlmann and Mrs. Karin Kühlmann, Museum für Naturkunde, Berlin, Germany, for their assistence with the type material. I am indeed grateful to Dr. Paul Cornelius, Natural History Museum, London, for the constructive criticism of the outlines of this research and to Dr. Elaine Robson, University of Reading, for helping me to see my work through different eyes.

I gratefully acknowledge my diving team: Monica Petti, Paulo Paiva and Uwe Walter for their help, friendship and above all for their patience with my bad mood after a bad dive in Antarctica. I specially thank Dr. Johann-Wolfgang Wägele for his assistance in diving and Antarctic matters. I also thank Dr. Martin Rauchert for providing additional samples for this study.

I am deeply indebted to Dr. Martin Althaus, Regina Asariotis and Sigrid Barkmann for their help with the exhaustive German translations. Thanks are also to the efficient and friendly team of staff of the Department of Oceanography, especially to Kate Saul, Sharon Rolfe, Lauri Wall, Jenny Mallinson, John Gibbs, Bob Stringer and Mike Crowfoot (if only for the nice smile that made many of my mornings). My gratitude is also expressed to Carol Chedzey and Pat Maughan of the reprographics unit at HR Wallingford.

Very special thanks go to Dr. Lucia Campos-Creasey, Dr. Virginia and Dr. Carlos Garcia for providing familiar support on my arrival in this country, which was kept by many other special friends, who have made pleasant my stay in England.

My most grateful acknowledgement is expressed to Prof. Edmundo F. Nonato, my master of always, with whom I have learned to like and respect science; to Dr. Michael Dearnaley, for sharing the burden and for his most valuable support, and to my parents, Aparecida and Celso Brito for their incentive, love and above all for their respect for the ways I have chosen.

Aos meus pais e ao Prof. Nonato

CONTENTS

CHAPTER 1
INTRODUCTION

Octocorals have a striking presence in benthic communities because of their beauty, diversity, abundance and inter-specific relationships. The subclass Octocorallia is represented by an enormous variety of forms, from encrusting, filiform or membranous to complex and elaborated branching architectures.

The wide geographical and bathymetric distribution of species of the Octocorallia is a result of their ability to adapt successfully to varied ecological situations, where they can also be well represented in density and biomass.

Despite their importance as conspicuous components of benthic communities, which would alone justify their study, gorgonians are also of anthropocentric interest. Their beauty has been exploited for economic interest in underwater tourism and exploitation as jewelry, mainly red coral (*Corallium rubrum*), has been for long a serious business. As this commercial exploitation could deplete the stock, intensive investigation into the ecology and life history of these precious corals has been supported (Bayer, 1981a).

Biochemical investigation of this group has shown these animals to be a potential source of chemicals, with important pharmaceutical applications, such as prostaglandins. These chemicals, previously only found in human semen and in the semen and vesicular glands of sheep, can be extracted in significant amounts from the gorgonian *Plexaurella homomalla*. The biochemistry of octocorals has become of particular interest to the pharmaceutical industry and harvesting or farming of gorgonians has been considered (Pike, 1974; Bayer, 1981b). Among the wide range of effects of prostaglandins on humans, the action on the female reproductive system is the most striking and has been widely studied. They can be used for therapeutic abortion and may become important agents in controlling population growth! (Pike, 1974). However, prostaglandins can now be synthesized in the laboratory.

The calcium carbonate skeletons of certain species of corals have been applied as a template for the production of microporous biomaterials which are used to replace bone, blood vessels, trachea, and other damaged organs and tissues (White & White, 1981).

This is possible because the aragonite skeletons, typical of reef-building corals, have a high degree of uniformity of pore diameter and high permeability since all the pores are connected to each other (Weber & White, 1973).

Despite the majority of octocorals being ahermatypic their importance as contributors to carbonate sediments should not be underestimated (Velimirov & Böhm, 1976; Konishi, 1981). Investigation of their chemical composition aids the understanding of the physical and chemical nature of the seawater in which these organisms live (Ohde & Kitano, 1981; Goldberg, 1991). The study of deep-sea octocorals allows time histories of conservative and transient tracers to be reconstructed (Druffel *et al.*, 1990). The periodicity of growth layers in octocorals can be used to interpret past variation in length of day, lunar month and year (Grigg, 1974). The orientation of the fan-shaped gorgonians perpendicular to the dominant current flow might be an useful indicator of the average direction of water motion (Grigg, 1972).

The applied use of octocorals has not been limited to the 20th century. Octocorals have been known from mythology when "goddess Minerva endowed the red coral with magical properties that protect travellers from danger" (Plinius Secundus, 1469, in: Bayer, 1981a). The ancients also valued them for their magical, medicinal, and decorative properties (Hickson, 1924, in: Bayer, 1956).

Above all, the pure study of octocoral systematics tell us much about phylogeny, evolution, zoogeography and animal diversity (Bayer, 1981a).

The importance of the study of Antarctic and deep-sea octocorals is primarily to contribute to the knowledge about the benthic communities inhabiting some of the most poorly known and supposedly undamaged areas of the Globe. Octocorals have been shown to be abundant in these areas and a relevant assemblage of unidentified specimens lies in existing collections in many of the natural history museums around the world.

The knowledge about Antarctic and deep-sea octocorals has been restricted to taxonomic studies resulting from the scientific expeditions realized mostly in the second half of the last century and the beginning of this century. Little is known about their biology.

The main problems faced when working on the taxonomy of Antarctic octocorals lie in the fact that the first descriptions of the species were based on a limited number of specimens, and sometimes on badly preserved material. The illustrations of the descriptions were often of

inconsistent quality and accuracy, accompanying sometimes brief and insufficiently clear texts. Furthermore, the available literature is scattered and the classification confused. The conditions for investigating the biological aspects of Antarctic octocorals are far worse. The adversities and difficult access of the study site is the main limitation, adding to the lack of continuous sampling.

1.1 - GENERAL CHARACTERISTICS OF ANTARCTIC BENTHIC FAUNA

The "Challenger" expedition (1873-76) was the first to collect substantial benthic samples from Antarctic waters. The Belgian Antarctica Expedition (1897-1899) was the first Antarctic expedition to make systematic collections of benthic material (White, 1984). Since then, numerous scientific expeditions have ventured to explore Antarctica. The first studies concentrated mainly on taxonomy but more recently the focus of research shifted to studies of life history, behaviour, and physiology of the near-shore benthic fauna (Arntz, *et al.*, in press).

The initial investigations of the Antarctic benthic communities lead researchers to draw general patterns about the environment and its inhabitants. With the accumulation of additional information, some of these patterns had to be reformulated.

It had been thought that the Antarctic environment had constancy (or stability) of physical conditions (Lipps & Hickman, 1982; Dayton, 1990, amongst others). Recent research has shown that the environment has constant conditions of low, but stable temperature and low fluctuation in salinity. Furthermore, the Antarctic has less input from terrestrial sediments than the Arctic and is isolated by deep sea, circumantarctic current systems and the Antarctic Convergence, which contribute to the constancy of conditions of this environment. However, it has as sources of variability and disturbance: the highly seasonal light regime, the sea ice cover, anchor ice causing physical variation in shallow water, fresh iceberg scours, ice shelves, long-term/ large scale modification of circulation patterns, and volcanic eruptions. The Antarctic benthic environment is more exposed to physical variability and disturbance than was considered in the past (Arntz, *et al.*, in press).

Overall, this region is marked by some significant characteristics such as the stable cold temperature, strong seasonality, and short period of summer primary production. These environmental conditions suggest selected feeding, reproductive, biochemical and behavioral

patterns. A traditional view of Antarctica is that it is an ecosystem with high biomass in many areas, high endemism owing to the limited dispersal ability of many species, and within-site diversity which can be as high as anywhere in the world (Clarke & Crame, 1989).

The Antarctic environment is highly seasonal considering daylight, ice and temperature which results in a seasonality of primary production which then affects most of the rest of the system (Clarke, 1988). This seasonality of food supply may control physiological processes such as reproduction and growth. Such seasonality conditions particular adaptive responses (Pearson & Rosenberg, 1987) and this factor might have selected for organisms that sustain long starvation periods or live on food resources other than primary production (Arntz *et al.*, in press).

The intense austral summer productivity of shallow Antarctic seas is responsible for a misconception concerning the productivity of these waters, considering the short duration of the bloom (8-10 weeks). The claim that Antarctic waters are "highly productive" may pertain more to coastal waters than to the whole Southern Ocean (Tanner, 1985). The overall primary production is much lower than previously supposed (Hempel, 1985; Tanner, 1985; Le Jean, 1985; Tiler, 1985; Order, 1985).

Much of the phytoplankton goes to the benthos which acts largely as a sink (Hempel, 1985). Ice algae have also been shown to contribute substantially (30%) to the total biomass production of the Southern Ocean (Spindler & Dieckmann, 1991) and the detrital fallout from the sea ice may play an important part as an energetic source for benthic communities (Dayton, 1990; Arntz *et al.*,in press). The food input from the water column might be insufficient to cover the requirements of epibenthic suspension feeders for most of the year. Lateral advection of suspended matter might be an additional food source for some of these organisms (Berkman *et al.*, 1986; Dayton, 1990). As stated by Arntz *et al.* (in press), a large proportion of Antarctic benthic organisms have developed a high degree of independence from fluctuating food conditions. Necrophagy is widespread amongst these organisms. Scavengers, predators and omnivorous can probably feed all year round.

The species that base their energy economy upon the food supply will have this energy shared between a number of processes, such as somatic growth, gonadal growth, activity and heat loss. If the energy input is low and the activities have to be maintained, one of these processes will be restricted, depending on the demand of the processes concerned (Dunbar, 1968). Thus, it is thought that in Antarctica the low food supply results in slow growth. Slow growth is, therefore, a result of food availability rather than temperature (Clarke, 1988). As

a consequence, for many species, growth occurs mostly in the eutrophic summer months (Clarke, 1990). Despite slow growth being a common tendency amongst Antarctic invertebrates, a large variability in growth performance of benthic populations has been documented (Arntz *et al.*, in press).

Three major modes of reproduction have been observed in antarctic invertebrates: pelagic planktotrophy; pelagic lecithotrophy and non-pelagic lecithotrophy, which involves the production of demersal or crawling planulae. Species which develop pelagic planktotrophic larvae, in general, produce many small eggs, assuring that at least a few will survive the hazards of the pelagic life. Although these feeding larvae appear to have an advantage if released during the eutrophic months when particulate food is abundant, some species release their larvae before the phytoplankton bloom, avoiding predatory zooplankton. Others release their larvae at any time of the year since these larvae feed upon bacterioplankton and uptake of dissolved organic material and do not depend on the summer production (Pearse *et al.,* 1991; Clarke, 1992). The species which produce pelagic planktotrophic larvae have high dispersal capability and are widespread in the Antarctic Ocean.

Species which produce pelagic lecithotrophic larvae will produce fewer but larger eggs. These larvae have been considered as advantageous in environments of limited food resources (Thorson, 1950; Jablonski & Lutz, 1983; McClintock & Pearse, 1986). It is more costly to produce many small eggs than a few large and more nourishable ones. To produce such large and nutritious eggs whilst depending on the available resources may make development slow and may take more than a year. Vitellogenesis takes place in one summer season and full development and spawning the subsequent summer (Clarke, 1992). The larvae are non-feeding and therefore do not depend on summer production, and can hence be released at any time of the year. The survival of the juveniles might, however, depend on the available food (Pearse *et al.*, 1991). Species which produce pelagic lecithotrophic larvae can have reasonable dispersal capacity but are believed to have shorter pelagic life than planktotrophic larvae (Emlet *et al.*, 1987).

Species which produce non-feeding, non-pelagic, lecithotrophic larvae will produce a few large eggs that will be brooded until the larva is mature. The larvae will have demersal habits or will settle soon after being released. This will ensure early survival compensating for the low fecundity but will decrease the dispersal capability which in many cases will suggest patchy distribution and endemism.

1.2 - DEVELOPMENT OF THE RESEARCH OUTLINES AND OBJECTIVES

The present study is part of a Brazilian project PROANTAR, No. 9616, "Bionomy of the Antarctic Benthic Fauna" that was established in 1982. Brazil was formally recognized as an adhering member of the Antarctic Treaty in 1975. It became an effective member in 1984, at the beginning of its participation in the "Scientific Committee on Antarctic Research" (SCAR).

Eleven scientific expeditions have already been undertaken. From those expeditions a significant amount of benthic material has been accumulated. For the past few years, the program "Antarctic Near-shore Benthos" has been carried out and marine *in situ* studies have been included in the program.

The first aim of this study was to prepare a report on the Octocorallia collected by the Brazilian Antarctic Expeditions. Amongst the specimens collected, those that could be referred to the family Primnoidae showed a remarkable dominance. This family is abundant in deep water fauna, and it is the most important gorgonian family of the Antarctic and Subantarctic waters. Despite its importance, it has been poorly investigated and has become the main area of interest for this thesis.

The predominant genus within the family Primnoidae was *Thouarella*. 34 nominal species and 3 varieties have been assigned to this genus. The characters that have been applied to distinguish these species are not fully reliable because of substantial variation. An investigation of all the aspects which could cause these variations was carried out so that the validity of these characters could be evaluated. In order to accomplish this objective, a detailed study of the species *Thouarella variabilis*, that makes up the largest part of the collection, was conducted. An investigation of colony morphology; structure, formation and function of the sclerites; growth and age determination; internal morphology; and reproductive biology of *Th. variabilis* was undertaken.

The subsequent sections of this chapter briefly describe the study area and the materials used in this study. The remainder of this thesis is in six chapters:

Chapter 2 - gives a general description of the external features and colony morphology of *Thouarella variabilis* with special reference to the architectural design of sclerites, describing details of their morphological features; arrangement and number of operculum, marginal, adaxial and body scales; development of sclerites throughout growth, and their function as

anti-predator defence; and aspects of branching patterns, arrangement and distribution of polyps. It was also considered to be of interest to give a brief account on age determination through the examination of growth layers present in the axes of the studied specimens.

Chapter 3 - gives a brief description of the internal morphology of the polyps of *Th. variabilis*. This study was based on sections examined under light and electron microscopes. Unfortunately, the material was badly preserved and only limited information could be obtained. The available information is, however, sufficient to meet the objectives of this study.

Chapter 4 - describes the main features of the reproductive biology of *Th. variabilis*, concerning sexuality, reproductive mode, fecundity, gametogenesis, gametogenic cycles, distribution of the reproductive stages throughout the colony, ultrastructure of eggs, sperm sacs and larvae and the influence of the reproductive state on the external morphological features of the polyps.

Chapter 5 - evaluates the taxonomic status of *Thouarella variabilis* and of the varieties attributed to this species. A description based on the examined specimens is presented together with the diagnosis for the species. The synonymy, habitat, geographic and bathymetric distribution of the species are also included.

Chapter 6 - presents a taxonomic review of the genera *Thouarella* and *Amphilaphis* (Family Primnoidae), with discussions on the taxonomic position of these genera, on the incorporation of the genus *Amphilaphis* into *Thouarella* and on the grouping of the species of the genus *Thouarella* into subgenera. A description of all the nominal species and varieties attributed to this genus is included.

Chapter 7 - summarizes the conclusions of this study and discusses possible avenues of future work.

1.3 - DESCRIPTION OF THE AREA

During the first Brazilian Expeditions to Antarctica, specimens of octocorals were collected in the Bransfield Strait, Antarctic Peninsula (Fig. 1). During the last Expedition the work was concentrated in a more specific area, Admiralty Bay (Fig. 2), King George Island, South Shetland Archipelago.

The Scotia Arc comprises the South Shetland Archipelago, South Orkney, South Sandwich and South Georgia, and occupies the Western Antarctic Subregion. It displays distinctive features from the other Antarctic areas, such as more complex water circulation, higher temperature, closer position to the American continent and, for this reason, shows peculiarities in zoogeography.

Bransfield Strait (Fig. 1) is located between the Antarctic Peninsula and the South Shetland Islands, with the Bellinghausen Sea to the west and the Weddell Sea to the east and to the north of the South Shetland Islands the western entrance to Drake Passage. The Bransfield Strait has three basins and an interconnecting series of deep water passages. Of the three basins the easternmost is over 1500 m deep, the central is 1900 m deep and the westernmost is 1100 m deep (Huntley *et al.*, 1991).

The region of the South Shetland Archipelago is characterised by complex meteorological conditions and intricate oceanic current patterns (Pruszak, 1980). There are eight distinct water masses in this region, and the Bransfield Strait lies at the western end of the confluence of two principal water masses, the Weddell Sea and the Scotia Sea. Under the influence of local winds, each island of the South Shetlands has complicated circulation patterns (Huntley *et al.*, 1991).

King George Island is the largest island (length of 130 km) of the South Shetland Archipelago (Presler, 1980). Admiralty Bay, its largest embayment, covers an area of 120 km^2 and a volume of 18 km^3 (Jazdzewski *et al*, 1986; Arnaud *et al*, 1986) distributed over its three inlets, Eszcurra, Martel and Mackellar Inlets and the main part of the Bay (Pruszak, 1980).

The coast line of Admiralty Bay is varied with sandy, gravelly, stony and rocky beaches (Presler, 1980; Arnaud *et al*, 1986), and most of its length is protected from wave action by the presence of glaciers or pack ice in winter (Bullivant & Dearborn, 1967). A part of the eastern and northern shores of the Bay is formed by glaciers (Arnaud *et al*, 1986).

The configuration of the bottom relief shows steep falls of the bottom slope and great topographic diversity (Arnaud *et al*, 1986). Coarse sediments, prevailing at the shore, can occur down to a depth of about 50m; as depth increases the sediments become increasingly mixed with muddy sand and the deeper parts of the bottom are covered by a fine mud and only single drop-stones can be found (Jazdzewski *et al*, 1986; Wägele & Brito, 1990; Nonato *et al.*, 1992a,b). Macro-algal beds fringe Admiralty Bay down to a depth of 90m.

The annual water temperature ranges from +2.5°C to -2.0°C (Jazdzewski *et al*, 1986). The water temperature observed by Pruszak (1980), in the austral summer, ranged from + 1.76°C (surface) to -0.24°C (at the sea bed ≈ 500m) in the entrance to the Bay and from +1.76°C (surface) to 0.2°C (at the sea bed ≈300m) inside the Bay. In winter, under the ice, the temperature varies from -1.6°C to -2.0°C (Jazdzewski *et al*, 1986).

The freezing process of water in the Bay takes place from May to August with freezing of the entire surface occurring in July. Freezing is associated with frosty and windless weather and no waves. Prior to November the ice is disrupted and disappears as a result of intensive waves from the sea and strong north winds (Jazdzewski *et al*, 1986). The weather is calm again by late December (Bodungen, 1986).

The most frequent winds blow from the SW, W and N sectors (Pruszak, 1980). The mean value of the wind speed in the Bay in summer is 7m s^{-1}. Winds generate an extensive outflow of the surface water from Admiralty Bay into the Bransfield Strait, with the occurrence of katabatic winds increasing directly beyond the exit of the Bay (Pruszak, 1980).

The surface flows along the inlets show an average flow velocity of 0.2-0.3m s^{-1} and a maximum of 0.5m s^{-1} whilst in the main part of the Bay the average velocities range from 0.3 to 0.5 or 0.6m s^{-1} with a maximum of 1m s^{-1} or even more, increasing directly beyond the exit of the Bay. Along the shores of the Bay the current speeds are slightly lower than the mean values because of the wind field, which is influenced by fairly steep shores and a series of other boundary conditions produced by the coastline profile. As a result, the currents generated have a velocity of only about 0.3m s^{-1} to 0.15m s^{-1} or even less (Pruszak, 1980). Admiralty Bay has a semi-diurnal tidal pattern with a maximum tidal range of 2.5m (Pruszak, 1980).

Salinity, like temperature, is relatively stable through the whole Bay and has no effect upon general current circulation in the area. In summer the salinity decreases at the surface (32.9 - 34.0 ppt) increasing with depth (34.5 ppt) and varying from 16.4 to 34.2 ppt in glaciers (Pruszak, 1980; Szfranski & Lipski, 1982; Jazdzewski *et al*, 1986; Arnaud *et al*, 1986).

The waters of the Bay are rich in oxygen, phosphorous and nitrogen compounds. Concentrations are typical for the Antarctic water southwards of the convergence zone. This is a consequence of the good circulation of the waters between the Bay and the open sea (Samp, 1980; Pecherzewski, 1980).

The waters of Admiralty Bay are connected with those of the Bransfield Strait by a 500m deep trench (that forms a branch of the deep waters of the Bransfield Strait) (Szafranski & Lipski, 1982; Rakuza-Suszczewski, 1980; White, 1984). In general, one third of the bottom of the Bay is deeper than 200m, with the deepest area being over 600m deep (Nonato *et al.*, 1992b).

The quantity of suspended matter drifting from the land into the Bay during the austral summer is estimated as averaging about 2,000 tons per day, ie. about 200-240 thousand tons per year. 20-30% of the suspended matter is brought into the Bay from the Bransfield Strait each year by winds and feeding penguins. According to the turbidity the visibility recorded by Pecherzewski (1980) ranged from 0.2 to 7.6m.

The four main sources of dissolved and particulate detritus are; the run-off of nitrogen waste products, from penguins and seals; freshwater contributions from glacier melts (Dawson *et al*, 1985); primary production of phytoplankton and associated modification by secondary consumers; and contributions from substantial areas of macro-algal beds.

The guano produced by penguins, forms ornithogenic soils and is extensively and thoroughly washed out to sea. In some areas these run-off solutions can enrich levels of nutrients in coastal waters, leading to significant increases in primary and secondary production (Myrcha *et al*, 1985).

The shelf waters of the Bransfield Strait are among the most productive in the Antarctic with regards to phytoplankton and krill. The phytoplankton in this region exhibits a distinct seasonal cycle, with high standing stocks in spring, late November, declining to low levels in summer, late December (Hart 1942; Horne 1969; Sakshang & Holm-Hans, 1984; Bodungen, 1986). Sea-ice micro-algae contribute a significant fraction of primary production in some Antarctic pack-ice regions (Palmisano & Sullivan, 1985).

The bacteria, which play a crucial role in the cycling of nutrients, are freely suspended in the water, attached to other organisms, to particles of organic debris and in the superficial layer above the sea-floor where they are abundant (Tanner, 1985).

The occurrence of a rich and diverse benthic faunal community in association with algae in Admiralty Bay (Rakusa, 1980) points to the existence of a detritus-based food chain.

The benthic communities of the Bay have been surveyed in the deeper parts by means of dredge and trawls. In shallow places the survey has been based on SCUBA observations. Six localities were investigated around the Bay.

In front of the Brazilian station, Martel Inlet, a more protected area, the strong summer sedimentation seems to be prejudicial to the formation of a high biomass of suspension feeding epifauna such as sponge mats or coelenterate mats that are characteristic of other areas in Antarctica. The benthic community is represented, in those areas, mainly by grazers and predators with exceptions of some abundant filter-feeders.

Hard-bottom communities are a minor element of the benthos as most of the sea-floor in the Bay is formed by soft sediments. Owing to the frequent occurrence of drop-stones, larvae of sessile species have a chance to find a suitable place for colonization virtually everywhere.

The few areas of hard bottom are the most species rich sites of the Bay. The benthic communities in these places are well represented by most of the groups that occur in Antarctica. Octocorals were present in only three of the sites surveyed, the southeastern coast of Martel Fjord, near Wanda Glacier, Napier Rock and Punta Ullman.

On the southeastern coast of Martel Fjord, near Wanda Glacier, the upper sublittoral is formed by a slope with an inclination of about 25-35°. Stones, bare rocks and (in gaps and flat depressions) mud form the bottom. On sheltered rocks sponges and bryozoans were found. The number of sessile animals increases with increasing depth, with an important population of large ascidians. Below 20m many Anthozoa, especially Actiniaria, grow on rocks or burrow in mud. On the rocks, the epiphytic fauna is rich, with several species of sponges, Hydrozoa and Bryozoa, accompanied by Nudibranchia, Crinoidea, Asteroidea and Polychaeta.

Napier Rock, located in the entrance to the Bay, is a rock that arises from close to the 160m line steeply to the water surface. The nearly vertical slopes are densely covered with algae, down to 30m deep with *Desmarestia* sp. dominating the vegetation. On these plants live some gastropods, amphipods and isopods. Between the rhizoids and on the bare rock a rich epiphytic community grows, which, in contrast to the foregoing places, is free of sediments. In shallow water archaeogastropods are very frequent, grazing on the encrusting algae. Below 6 to 8m depth the first Bryozoa, Hydrozoa and Porifera appear. At 10m depth a large number of animals are competing for space: many Porifera, solitary and colonial Ascidiacea, accompanied by climbing Holothurioidea, Nudibranchia, Isopoda, other gastropods, crustaceans and Polychaeta. Here, the Cnidaria, mainly the Octocorallia are found. Several

species of the orders Alcyonacea and Gorgonacea are found at this site and some colonies were collected for taxonomic and biological studies. The most abundant family is Primnoidae (Gorgonacea) and the genus *Ascolepis* can often be seen covering the most exposed side of the rock.

Punta Ullman, on the northeastern side of the Martel Inlet, is a more sheltered site. Specimens of *Thouarella variabilis* were only found in this site at a depth of 53 m. The environmental conditions at this sampling site were not recorded because the sampling was conducted from on board a ship. Most of the samples were obtained from the Bransfield Strait. The distribution of the specimens of *Th. variabilis* studied is shown in Figure 1.

1.4 - MATERIALS

Most material for this study was collected by recent Brazilian expeditions to the South Shetland Archipelago and Bransfield Strait, Antarctica. Deep water sampling was conducted aboard a ship by means of dredge and nets, beam-trawl and otter-trawl. Additional sampling was also conducted by SCUBA diving in the shallow waters of Admiralty Bay, King George Island. This material is part of the collection held by the Instituto Oceanográfico - Universidade de São Paulo, Brazil. Material used for comparison was collected by German colleagues at Maxwell Bay, King George Island, by SCUBA diving. Further comparison was based on specimens collected during former expeditions to the Antarctic and which are held in the collections of some museums (Natural History Museum, London; Museum für Naturkunde, Berlin; Senckenberg Museum, Frankfurt; NMNH, Smithsonian Institution, Washington D.C.; and Zoologisk Museum, Oslo). The location of the investigated specimens is given in Chapter 5 together with the description of the species *Thouarella variabilis*.

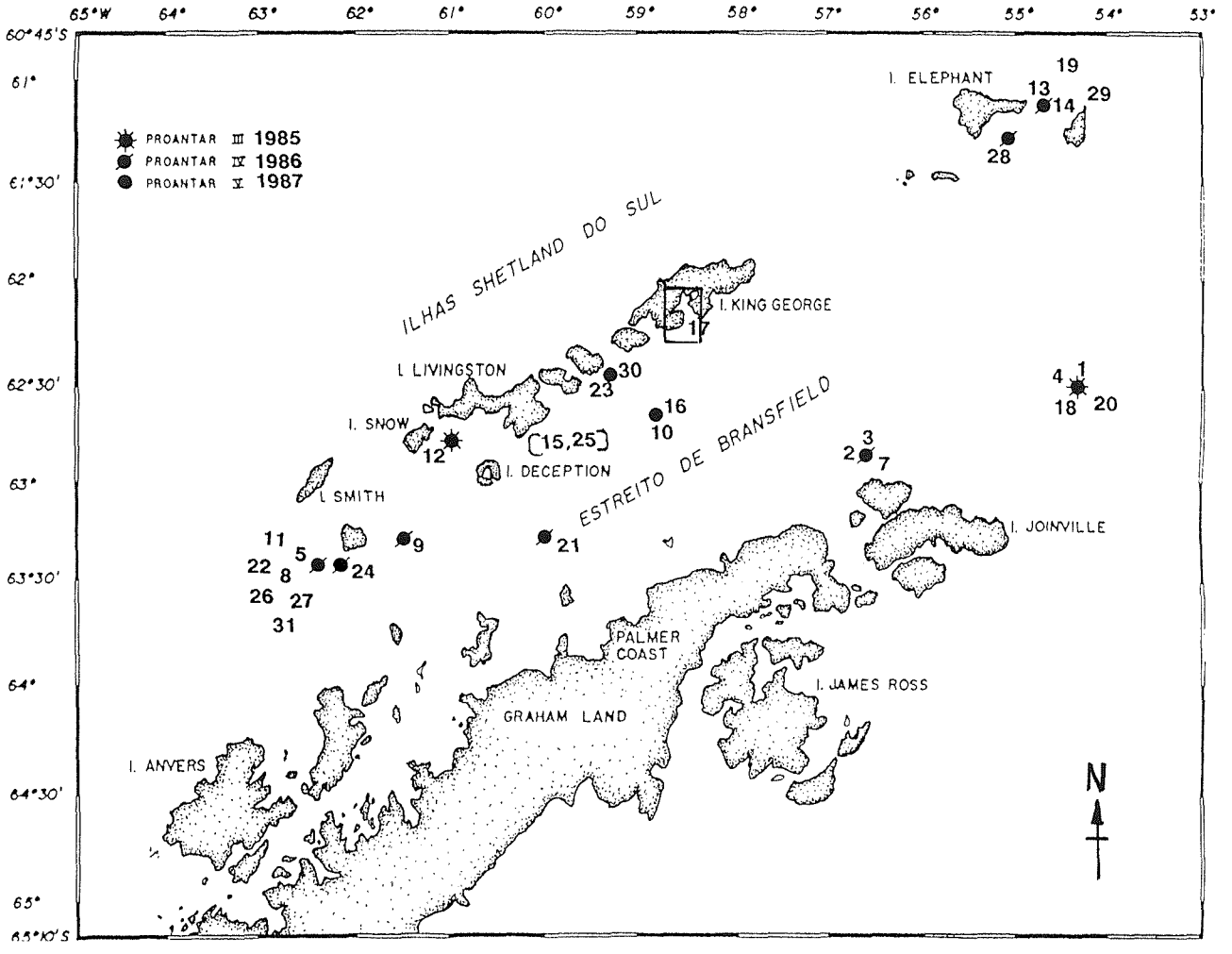


Fig. 1 - Bransfield Strait, Antarctic Peninsula and South Shetland Archipelago, with the location of the sampling sites. Marked area - Admiralty Bay, King George Island (see Fig. 2).

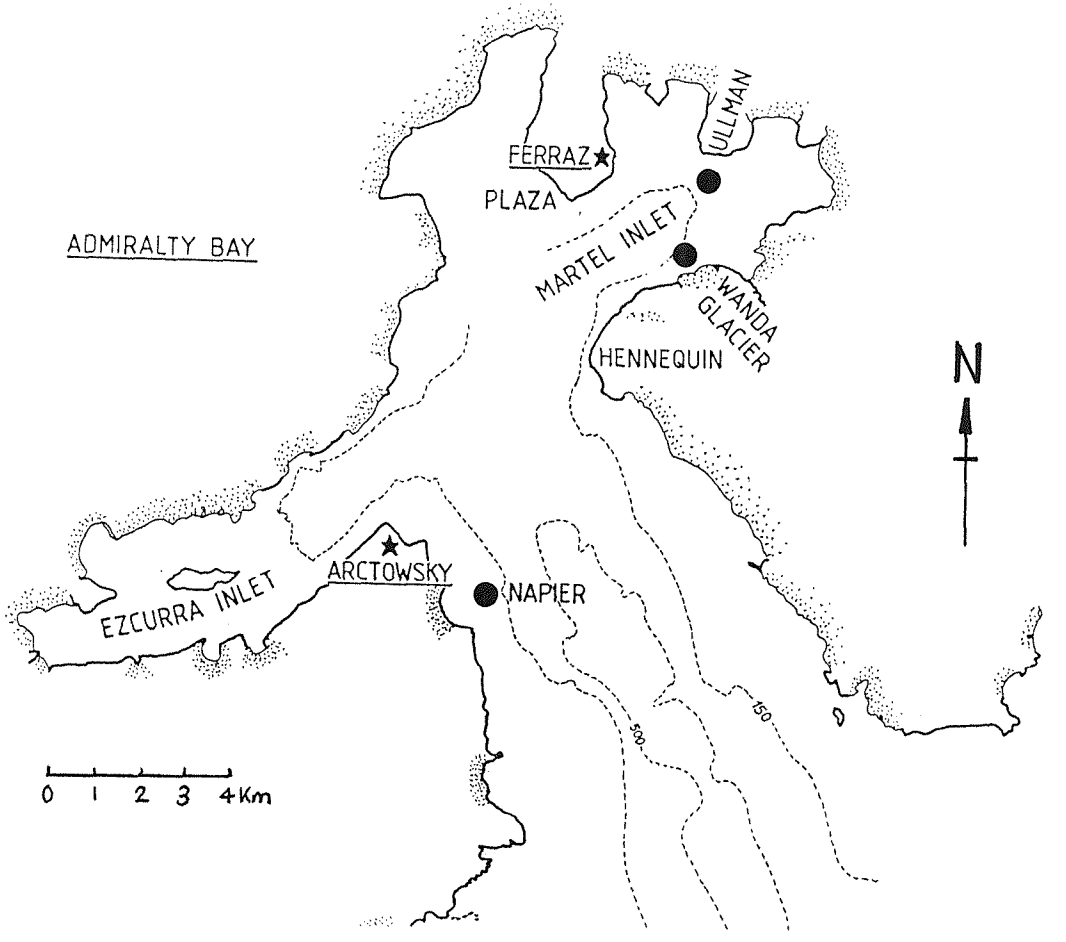


Fig. 2 - Admiralty Bay, King George Island, showing the sampling sites with the positions of the Brazilian Station "Comandante Ferraz" and the Polish Station "Arctowsky".

CHAPTER 2

MORPHOLOGY OF COLONY; STRUCTURE, FORMATION AND FUNCTION OF SCLERITES; AND ESTIMATES OF AGE IN *Thouarella variabilis*

2.1 - INTRODUCTION

2.1.1 - FORMATION AND CHEMICAL COMPOSITION OF SCLERITES AND AXIAL SKELETON

Unlike the majority of scleractinians, octocorals are mostly ahermatypic. The exceptions are the reef-building helioporacean *Heliopora coerulea* and, to a lesser extent, the stoloniferan *Tubipora musica*. The skeletal structure of octocorals is composed essentially of calcareous sclerites whereas some species have in addition an axial skeleton, which consists of horny proteinaceous material called gorgonin. Gorgonin is occasionally permeated with calcareous structures as seen in the species of the order Gorgonacea, suborder Holaxonia. Some other species have compact calcareous axes which may be continuous (as in *Tubipora* and *Corallium*) or interrupted by horny nodes as seen in the species of the family Isididae. Species which have horny axes are largely composed of organic matter and species which possess compacted calcareous axes have little organic matter (Clarke & Wheeler, 1922).

The major inorganic constituents of octocoral skeleton and tissues are Ca, Na, Mg and Si, together with minor concentrations of Ti, Al, Fe, K, P, Mn, Sr, Ba, Cr, Co, V and Zn (Clarke & Wheeler, 1922; Buddemeier *et al.*, 1981; Ohde & Kitano, 1981; Velimirov, 1980). Most of the mineral content of octocorals is made up of $CaCO_3$ and $MgCO_3$ (up to 83.5 % in *Eunicella* spp, Velimirov *et al.*, 1976; and almost 100% in *Plexaura grandiflora*, Clarke & Wheeler, 1922). Calcium carbonate can be deposited in octocorals as aragonite, Mg-poor calcite, or Mg-rich calcite. When the $MgCO_3$ content exceeds 4% in the calcite lattice, the crystal form is referred to as high magnesian calcite (Chaves, 1954, in: Velimirov & Böhm, 1976). The crystalline character of the skeleton depends on its chemical composition. Aragonitic organisms are nonmagnesian whereas organisms rich in magnesium are calcitic. Magnesium carbonate (magnesite) and calcite are isomorphous, therefore it is more probable that Mg associates with calcite rather than with aragonite which has a different crystalline form. *Heliopora coerulea* shares more similarities in chemical composition with scleractinians by

exhibiting less magnesium carbonate (nearly nonmagnesian) and more calcium carbonate than any other octocoral (Clarke & Wheeler, 1922). The studies of Clarke & Wheeler (1922) and Chaves (1954) showed that $MgCO_3$ concentrations increased linearly with water temperature. Since warm water favours the assimilation of magnesium, cold water organisms from high latitudes or from deep waters are low in magnesium when compared to warm water organisms (Clarke & Wheeler, 1922). Velimirov (1980) demonstrated, however, that although a minimum temperature is required, the major factor controlling the incorporation of Mg into the carbonate skeletons was the variability of temperature rather than the temperature itself.

The skeleton of a colony is not necessarily formed of only one calcium carbonate polymorph. Rather, the colony is a mosaic of varied proportions of the different polymorphs and recrystallization might eventually occur. The proportions vary also among the different taxonomic groups. In the families Primnoidae and Chrysogorgiidae, aragonite is said to be the main crystal deposited whereas the Helioporacea (= Coenothecalia) *Heliopora coerulea* and *Lithotelesto micropora* are purely aragonitic (Meigen, 1903, in: Bayer, 1981c) as are most of the recent reef-building scleractinian corals. Apart from these examples of aragonitic octocorals, octocorals are known to deposit calcium carbonate mostly as calcite (Velimirov, 1980; Bayer, 1981c; Druffel *et al.*, 1990).

Calcite is a low pressure $CaCO_3$ whose solubility in water increases with increasing pressure and decreasing temperature (Deer *et al.*, 1966). Calcite has a rhombohedral structure, projecting at the surface of the sclerites in a "hound's tooth pattern" (Bayer, 1981c) or as a "dog-tooth spar" (Read, 1970). Aragonite is distinguished from calcite by the orthorhombic structure of the crystals, by the different cleavage, by being harder, and by having higher specific gravity (Deer *et al.*, 1966; Read, 1970). Magnesite ($MgCO_3$) with its trigonal structure, is similar to calcite but with a smaller cell because of the small size of the magnesium ion (Deer *et al.*, 1966). Aragonite is less stable than calcite and at normal temperatures and pressures, is metastable and frequently inverts to calcite. In fossil shells, for instance, the aragonite is gradually converted into calcite and fossilised corals often have their aragonite completely transformed into calcite whilst retaining the same original appearance (Ohde & Kitano, 1981). In some reef-building corals, the upper parts are composed of aragonite and the lower parts which are under higher hydrostatic pressure are composed of calcite (Deer *et al.*, 1966; Read, 1970; Velimirov, 1979). Magnesium and strontium ions might hinder the transformation of aragonite into calcite (Deer *et al.*, 1966; Ohde & Kitano, 1981). The content of Sr, Mn and Na is higher in aragonite than in calcite (Kitano *et al.*, 1971a,b, 1975; in: Ohde & Kitano, 1981). The concentration of Sr and Na might decrease as aragonite is converted into calcite (Ohde & Kitano, 1981).

Calcium is first absorbed by the tissue and posteriorly transported to the spicule-forming vesicles or to the site of axis formation (Allemand & Grillo, 1992). The ions enter the cells passively and are transported to the site of mineral deposition through a transcellular route via an energetic process (Velimirov and King, 1979). The uptake of Ca is initially rapid but once an equilibrium is achieved, the influx becomes slow (Velimirov & King, 1979; Allemand & Grillo, 1992). Velimirov and King (1979), working on *Eunicella papillosa*, noted that the rate at which Ca is incorporated depends on the activity of the octocorals, with the lowest rates occurring when the polyps are expanded for short periods or when only a few polyps are expanded. The uptake and deposition of calcium increases with the increase of temperature and Alemmand & Grillo, (1992) pointed out that this effect is saturable for temperatures higher than 20°C. A back-flux of Ca into the seawater was also observed (Velimirov & King, 1979) and was interpreted by Allemand & Grillo, (1992) as a passive efflux by leakage or an active efflux from cells by calcium pumps. The composition and physiology of the living tissue may affect the relationship between the surrounding water and the deposition of calcium carbonate (Buddemeier *et al.*, 1981). The living tissue can act as a barrier and regulator for Ca uptake (Velimirov & King, 1979).

The soluble bicarbonates from seawater and the carbon dioxide from the metabolism of the tissues are thought to be the sources of carbonate for the production of calcium carbonate (Wilbur, 1976, in: Allemand & Grillo, 1992). Metabolic CO_2 appears to be the main source of carbon for skeletogenesis in *Corallium rubrum*. Allemand & Grillo, (1992) demonstrated that CO_2 production by respiration in this gorgonian is more than enough to support all calcification. The limiting step for the biocalcification rate appears to be the hydration of metabolic CO_2 by carbonic anhydrase to HCO_3. For *Corallium niobe,* a deep-sea species, Druffel *et al.* (1990) observed that dissolved inorganic carbon rather than respiration was the primary source of carbon for the skeleton calcite.

Calcification processes are essentially the same in hermatypic and ahermatypic corals (Buddemeier *et al.*, 1981). For both, the calcification rate is known to vary in the different growth regions of the colony, being higher in branch tips than in lower branch regions (Velimirov & King, 1979). Goldberg (1991) noted that the organic skeleton is a direct product of the skeletogenic epithelia. The organic content is higher in the branches than in the lower portions of the stem, suggesting that mineral matter and organic content vary with age, resulting in tissue regression and increased mineralization, supporting the findings of Velimirov & Böhm (1976) that mineralization is higher at the base than at the branch tips. The same authors suggested that the number and/or size of spicules increase towards the base, although in sites of strong water movement the spicules tend to be larger in the branch

regions than at the base. Calcification rates tend to increase with the increasing speed of water movement (Velimirov & King, 1979). The rate of calcium deposition is higher in the spicules rather than in the axial skeleton (Allemand & Grillo, 1992)

The presence of a basal stalk or holdfast, with which the colony is attached to the substratum, is a character common to many species of octocorals. The stalk becomes consolidated and fixed to the substrate through cementation of the coenenchymal spicules (Konishi, 1981). In some gorgonaceans, whereas the colony might be composed of calcitic spicules, the stalk consists of different proportions of aragonite and calcite. Lowestam (1964) suggested this variation to be a function of water temperature in the habitat. Konishi (1981) observed, in *Sinularia,* that the proportion of aragonite decreases from the lower portion of the stalk to the upper portion where it contacts the coenenchyme.

2.1.2 - NATURE OF THE AXIAL SKELETON AND VARIATION IN THE COLONY MORPHOLOGY RELATED TO WATER MOVEMENT

The compacted axial skeleton of species of the suborder Scleraxonia had been thought to be formed by spicules fused by a calcareous cement (Bayer, 1955, 1956, 1981d; Muzik and Wainwright, 1977; Wainwright and Koehl, 1976). Allemand & Grillo (1992), however, demonstrated that active calcium secretion rather than migration of spicules is responsible for the growth of the axial skeleton, although some spicules might be incorporated into the skeleton.

The axes of holaxonian gorgonaceans are composed of gorgonin, which consists of a proteinaceous matrix in which flexible collagen fibres are embedded (Lewis *et al.*, 1992). Gorgonin is deposited extracellularly in concentric layers and collagen, the major component, originates from the ectoderm (Jeyasuria & Lewis, 1987; Esford & Lewis, 1990; Lewis *et al.*, 1992). The organization of the collagen, together with the chemical and macromolecular composition of the axis, will influence the mechanical properties of the axis (Goldberg, 1976; Lewis *et al.*, 1992). The gorgonin is stiffened by sclerotization and by mineralization (Jeyasuria & Lewis, 1987; Esford & Lewis, 1990). Minerals in a variety of crystal forms and aggregate shapes will be deposited extracellularly within the collagen interstitial spaces (Lowestam, 1964; Kingsley & Watabe, 1984; Jeyasuria & Lewis, 1987; Lewis *et al.*, 1992). The sclerotization of the collagen controls the flexibility of the axis and mineralization its stiffness (Lewis *et al.*, 1992).

Some authors (Jeyasuria & Lewis, 1987; Lewis *et al.*,1992) have stressed the importance of the calcareous material in modulating the mechanical properties of the axial skeleton. Calcium carbonate, deposited as calcite, has been mostly recorded for gorgonaceans (Lewis *et al.*, 1992). The hardness of the carbonate is increased by incorporating small Mg ions into the calcium carbonate, increasing the density of ionic packing (Jeyasuria & Lewis, 1987). The nature of the axes of gorgonaceans studied by Lewis *et al.* (1992) varied from unmineralized gorgonin to heavily mineralized axes. In the unmineralized and moderately mineralized axes, the mineral bodies occurred in the matrix between and around collagen fibres and in heavily mineralized axes the collagen fibres perforate the crystalline components. In the latter case, the crystalline components provide high resistance to compressional forces whereas the flexible uncalcified collagen fibres accommodate the tensional forces; furthermore, the incorporation of collagen into the mineral component increases stiffness (Lewis *et al.* 1992).

Gorgonians are adapted to specific depths and hydrodynamic regimes (Esford & Lewis, 1990). The axial stiffness might define the water movement related zonation: stiffest axes appear to be confined to deeper water and no wave surge; most flexible axes to shallower water and moderate surge; and intermediate stiffness with shallow water and high energy habitats (Jeyasuria & Lewis, 1987). Among the species studied by Jeyasuria & Lewis (1987); Esford & Lewis (1990); and Lewis *et al.* (1992), the species which had heavily mineralized axes were the stiffest and inhabited areas of current-generated water movement, below wave zones. These currents can be unidirectional or multidirectional. Species with moderately mineralized axes had moderately high stiffness and torsion resistance and inhabited the surge zone subjected to wave and bidirectional current-generated water movements. Unmineralized axes or low mineralized axes had low stiffness and low torsion resistance and inhabited areas below the wave affected region.

Not only the nature of axis, its stiffness and torsion resistance are related to hydrodynamic forces, but also the shape of the colony. The orientation of gorgonians related to water movement has been examined by many authors (Hickson, 1940; Laborel, 1960; Carlisle *et al.*, 1964; Theodor & Denizot, 1965; Wainwright & Dillon, 1969; Svoboda, 1970, 1973, 1976; Riedl, 1966, 1971; Grigg, 1970, 1972; Rees, 1972; Kinzie, 1973; Velimirov, 1973; 1976; Leversee, 1976; Wainwright & Koehl, 1976; Muzik & Wainwright, 1977; Koehl, 1984; Jeyasuria & Lewis, 1987; Esford & Lewis, 1990). Theodor (1963) noted that in turbulent water flow the loosely branched and whip-like colonies were commonly found whilst in unidirectional and constant currents the concave form, facing the current was the usual pattern. Grigg (1972) observed that when there is no dominant current direction bushy forms predominate and in the presence of bidirectional currents, fan-shaped forms are more typical.

The juveniles and small colonies develop in the sheltered boundary layer, which has a slower current than the main stream (Wainwright & Kohel, 1976). Once the colony has reached a certain size, it will be subject to the drag forces of the main stream. The smallest fans are, therefore, randomly oriented whilst the older higher ones, have favourable orientation (Wainwright & Dillon, 1969). If the base of the stem is subjected to a different flow than that at the distal portion of the colony, the colony grows helically (Svoboda, 1976). Grigg (1972) suggested that torsional forces produced by water movement increase growth in the direction of stress. Progressive changes in orientation occur during growth with the colonies gradually adjusting their growth pattern to the direction of the water movement (Grigg, 1972; Velimirov, 1973). If the colony is passively twisted and held by the current during periods of subsequent addition of skeletal material, the colony will be kept in its new orientation (Wainwright & Dillon, 1969). Fan-shaped colonies attain mechanical stability when perpendicular to the current flow (Theodor and Denizot, 1965; Wainwright & Koehl, 1976) since the drag forces are minimized (Grigg, 1972). Several advantages have been pointed out for the nature and orientation of the axis. As the main supportive structure of the colony, the axis maintains the elevation of the branches (Jeyasuria & Lewis, 1987), and keeps the polyps off the substratum (Lewis *et al.*, 1992). By separating the branches of the colony (Jeyasuria & Lewis, 1987) and being perpendicular to the current, the interbranch abrasion is reduced (Grigg, 1972). The nature of the axis and shape of the colony enables the organism to withstand (Music & Wainwright, 1977) and to modify the water velocities (Wainwright & Koehl, 1976). By facing the current flow, the colony exposes its maximum area to the current, increasing in this way its feeding efficiency (Wainwright & Dillon, 1969) by maximizing the contact between feeding surfaces and suspended food (Grigg, 1972).

Velimirov (1976) evaluated the differences in growth form of the gorgonian *Eunicella cavolini* according to the peculiarity of the area the colonies inhabited. Species from exposed areas developed longer and thicker end branches, a greater number of polyps and narrower fans with few ramifications. Species from sheltered areas had thinner branches with sparsely distributed polyps and large ramified fans. Velimirov suggested that availability of food is higher in exposed areas favouring colonies with long branches, few ramifications and numerous polyps. In sheltered areas the feeding efficiency would be improved by presenting large fans with highly ramified narrow branches bearing fewer polyps.

Wainwright & Koehl (1976) suggested that the arrangement of polyps could be correlated to the ways in which the morphological features of colonies modify the current flows. Leversee (1976) proposed that the patterns of polyp distribution may affect feeding success. Following the same idea, Jeyasuria & Lewis (1987) hypothesized that the arrangement of polyps on the

branches was influenced by the torsion resistance of the axis. They observed that in axes with low resistance to twisting forces the polyps were distributed around the whole circumference of the branches. Since the axes twist easily, the polyps were always exposed to the food bearing current. Species with axes highly resistant to twisting forces showed, conversely, polyps in a single row on one or both sides of a branch, which might be the most favourable feeding position. The feeding response of colonial suspension feeders appears to be stimulated by changes in current velocity and in prey density (Leversee, 1976).

Water movement through the colony will also depend on the colony porosity, where porosity is the ratio of the average distance between nearest neighbour branches to average branch diameter (Chamberlain & Graus, 1975a). Water flow is also affected by the relative branch size and by the arrangement of branches or branching pattern, since in closely spaced branches some parts will be adequately nourished while other parts may struggle for survival (Chamberlain & Graus, 1975a; Brazeau & Lasker, 1988). These hydromechanical properties of the colony form will alter the velocity of the flow, reducing drag forces and generating eddy currents. Leversee (1976) suggested that eddy currents may contribute to the feeding success of colonies which are multiplanar.

The axial skeleton of species of the order Antipatharia (subclass - Ceriantipatharia, Anthozoa), have a similar structure to the axial skeleton of octocorals. Antipatharians, also called black corals, are known for their lustrous, semi-precious skeleton. Unlike octocorals, the non-calcareous skeletal axis of antipatharians are not composed of gorgonin but of antipathin, which is a composite of chitin and scleroprotein (Goldberg, 1978, 1991; Schwartzman & Opresko, 1992). Another peculiarity of the anthipatharians is the presence of spines on the surface of their skeletons, considered an important taxonomic character (Schwartzman & Opresko, 1992; Kim *et al.*, 1992). In his work on *Anthipathes fiordensis,* Goldberg (1991) found that the overall protein content of the skeleton was about 60% of the dry organic weight and that the proportion of protein was higher in the terminal portions of the branchlets than in the basal parts. The three main amino acids detected in his study were, in order of reducing concentration, glycine, alanine and histidine. Goldberg (loc. cit.) suggested that the amino acid composition of the rings has an electrostatic binding function acting as an interlaminar glue and that lipid, found in low concentrations, is also associated with cement. The overall chitin content found by Goldberg was about 15%, with lower values for the tip than for the base. The spines present on the surface of the axis are considered by Kim *et al.* (1992) to have a significant reinforcing effect as they apparently increase the surface area for cementing the skeletal layers, preventing delamination by shear forces, thus adding to the strength of the skeleton.

The chitin-protein architecture of black corals is composed of chitin fibrils embedded in an amorphous protein matrix, where the chitin is expected to stiffen the structure (Kim *et al.*, 1992). Kim *et al.* (1992) found relatively low proportions of chitin in black coral skeletons compared to other more stiff biological materials (shell, bone), and deduced that greater flexibility should be more important than stiffness for antipatharians.

2.1.3 - GROWTH LAYERS AND ESTIMATES OF AGE

Growth in gorgonians accounts for increases in the axial skeleton and in spicule size or number (Velimirov & King, 1979). The axial skeleton grows in length and density or thickness by adding growth layers or lamellae (Kim *et al.*, 1992). Even if extension growth ceases or reduces, the calcification remains the same, increasing the skeletal density (Dustan, 1975; Velimirov, 1980). The lower parts of the colony have low extension rates, but calcification still occurs increasing the thickness (Velimirov, 1980). Growth rates in ahermatypic corals are low (Table 2.1) and calcification appears to be slow (Velimirov, 1975; Velimirov & King, 1979). Reef-building corals are, conversely, known to be fast growing (Velimirov, 1980). Grigg (1974) observed, in *Muricea californica*, that growth rate decreased as a function of height. This author also indicated that individual colonies differed in annual growth rate as a consequence of extrinsic (abrasion by attached algae, predation and food supply related to orientation) and intrinsic factors. Growth rates increased with higher water velocities (Sebens, 1984; Esford & Lewis, 1990).

The increase in density of the axial skeleton is apparently periodic (Goldberg, 1991) because if it was continuous and regular, no lines, bands or layers would be detected. A variation or discontinuity in deposition defines the lamellae or layers, also called growth rings (Clark, 1974; Grigg, 1970), and the depositional sequences may provide an index to growth rate (Goldberg, 1991). In antipatharians each lamella is composed of several sublayers of fibrils, separated from each other by a cement layer (Goldberg, 1991; Kim *et al.*, 1992). Kim *et al.* (1992) observed that the fibrils intersect and overlap one another gradually changing in orientation and that the spines are the major cause of this reorientation. This author also suggested that the layers correspond to a particular arrangement of fibrils, the darkest regions being composed of densely packed fibrils arranged in parallel and the lightest regions consisting of fibrils arranged obliquely (helicoidal arrangement). The intermediate bands contain fibrils in different planes, including parallel. In species which deposit calcified material, the growth layers might be formed by an alternation of dark-coloured organic layer and of lighter carbonate-rich increment (Goldberg, 1991).

Table 2.1 - Growth rates and maximum age of some octocorals and antipatharians

	GROWTH RATE (mm/yr)	AGE (YR)	AUTHOR
Black coral (unidentified)	29 NZ 64 Hawaii 45-93 Palau	15	Grange, 1985 Grigg, 1976 Grigg, 1975
Heliopora coerulea	42 mm/6 months		Velimirov, 1980
Eunicella singularis	33	25-30	Weinberg & Weinberg, 1979
Lophogorgia ceratophyta	28.5	35	Weinberg & Weinberg, 1979
Eunicella cavolini	21.5		Velimirov, 1975
Paramuricea clavata	18.3	50	Weinberg & Weinberg, 1979
Muricea californica	16.9	50; 30cm high = 20yr old	Grigg, 1974
Corallium japonicum	3		Grigg, 1974
Corallium niobe	0.11 ± 0.02	180 +- 40	Druffel et al., 1990

Goldberg (1991) defined two areas: the growth rings and the between-ring areas. He found it difficult to interpret the numerous sub-layers which exist between the main layers. Grigg (1974) had already pointed out the existence of poorly differentiated layers between well-marked layers. In older colonies it is difficult to distinguish the peripheral layers because they are thin and close together (Grigg, 1974). The thickness of growth layers was varied and, based on this information, Grigg (loc. cit.) deduced that the growth rate was irregular.

The causes for seasonal differences or periodicity in deposition in invertebrates have been suggested by many authors. In some insect groups, a daily periodicity in deposition of the cuticle was observed, where the parallel chitin fibrils (dark bands) are formed during the day and the helicoidal layers (light bands), which rotate gradually from parallel, are deposited during the night (Neville and Luke, 1969; Kim *et al.*, 1992). The growth rate of *Muricea californica* and *M. fruticosa* was shown to be faster in the first half of the year than in the second, perhaps related to seasonal differences in temperature, food supply or annual breeding cycle (Grigg, 1970; 1974). Lewis *et al.* (1992) considered the different tidal regimes as the cause for the daily accretion rate. They hypothesised that if currents from both directions contributed to the food supply, there would be four accretions per day (assuming a semi-diurnal tide). If currents in only one direction carried enough food, whether nocturnal or not, then only two accretions in growth would occur per day. Periodicity in growth influenced by daily tidal currents had already been recognized by Wainwright & Dillon (1969) who also considered the influence of different currents in summer and winter months. Thus, growth rings may represent daily, seasonal and annual periods (Wells, 1963; Wainwright & Dillon, 1969). This may suggest the possible presence of intermediate rings between major rings.

Druffel *et al.* (1990) considered the absence of known-aged skeletal bands in deep water octocorals, since seasonality in water temperature and other trigger factors do not exist in the deep-sea. However, banding had already been observed in solitary corals from the deep sea (Grigg, 1974).

2.1.4 - ARCHITECTURAL PROPERTIES AND FUNCTION OF SURFACE SCLERITES

Sclerites are present in varied sizes and forms amongst the different species of Octocorallia or even within the same species. Club, capstan, rod, spindles, double-disc, double-heads, radiate, needle, plate, scaphoid, spicules and scales are just a sample of the numerous forms of sclerites described in the glossary of terminology in octocorals by Bayer, Verseveldt and Grasshoff (1983). The surface of sclerites is ornamented with granules, tubercles, ridges, wrinkles or other more complex features (Bayer *et al*, 1983). The function of these ornaments and of the sclerite form have been examined recently by Lewis and Wallis, 1991; van Alstyne and Paul, 1992; van Alstyne *et al.*, 1992.

Sclerites have an important function in the structural support of colonies (Lewis & Wallis, 1991; van Alstyne & Paul, 1992). Lewis & Wallis (1991) suggested that the sclerite-containing coenenchyme modifies the mechanical properties of the axes, since the architecture and orientation of these sclerites will limit movement, increasing stiffness. Under tension, the mesoglea that surrounds the sclerites will stretch and the sclerites come in contact with each other and their tubercles will catch, keeping them engaged. When the colony is under compression the tubercles will disengage and the sclerites move close together, rotating according to their disposition and shape. Some forms will limit compression, some will limit extension and others will limit both. In a single colony many forms can be present, disposed in different layers. In this way, the sclerites can act as passive devices for limiting the freedom of movement of the entire structure contributing to the increase of colony stiffness (Lewis & Wallis, 1991). Besides their function as structural support, the sclerites also have a defensive function.

The soft body of octocorals is susceptible to predation. Besides being sessile the colony is potentially rich in nutritional source of protein, fat, and carbohydrate (Sammarco & Coll, 1988). Despite this, the incidence of predation by generalist predators such as fish, molluscs, echinoderms and crustaceans is negligible (1-2%) (Sammarco and Coll, 1988). Octocorals must, therefore, have evolved highly efficient anti-predator defences. Chemical and morphological defences have been observed in this group. The existence of toxic compounds

in octocorals has been well recognized (Tursch, *et al.*, 1978; Bakus & Thun, 1979; Bakus, 1981; Coll *et al.*, 1982a, 1982b; Sammarco *et al.*, 1983; Sammarco & Coll, 1988; van Alstyne & Paul, 1992; van Alstyne *et al.*, 1992). The secondary metabolites, usually terpenes, produced by octocorals will be feeding deterrents towards generalist predators, being distasteful or toxic, with effects which range from narcotization to death (Coll *et al.*, 1982a). Toxicity has been known not only for its role as anti-predatory, but also in competition for space, causing mortality upon contact with competitors (scleractinians or other octocorals) or even causing tissue necrosis under conditions of non-contact, where toxic compounds are released to the surrounding water (Sammarco *et al*, 1983). Structural modification, such as sweeper tentacles with an agonistic function was observed in the octocoral *Erythropodium caribaeorum* (Sebens and Miles, 1988). The sweeper tentacles are very elongated, lack pinnules and are densely packed with nematocysts, which are used to cause tissue necrosis in neighbouring corals (Sebens & Miles, 1988). Octocorals which lack stinging nematocysts might produce chemical compounds as a defence instead (Vermeij, 1978 in: Bakus, 1981). In nature there seems to be a balance in the presence of different defensive mechanisms. In holothurians, for instance, the exposed species are toxic and the cryptic species are nontoxic (Bakus, 1981). Sammarco and Coll (1988) proposed for octocorals an inverse relationship between toxicological defence characteristics and structural defence characteristics. The more efficient the morphological defence the fewer toxic compounds are produced. There are several morphological properties which contribute to colony defence: gross colony form, consistency or texture of the colony, presence of mucus, colour, ability to withdraw polyps, and the nature and distribution of sclerites throughout the colony (Sammarco & Coll, 1988).

The function of sclerites as anti-predator defence appears to be very effective as they can reduce feeding by fish by 95 % (van Alstyne & Paul, 1992). Studying the genus *Sinularia*, van Alstyne *et al.* (1992) observed that the performance of sclerites as feeding deterrents depends on their size, shape and concentration, although concentration has shown to be the most effective deterrent. Sclerites increase in both size and concentration from the tips to the bases of the colonies (van Alstyne & Paul, 1992; van Alstyne *et al.*, 1992). Therefore, the sclerites at the basal portion of the colony are more effective feeding deterrents when compared to the sclerites at the tips.

Some species will have polyps which retract into the coenenchyme and are protected against predation by the surface sclerites of the coenenchyme and calyx. In other species the polyps are non-retractile and permanently exposed. This latter case occurs in primnoids, where the polyps are non-retractile but well protected by a calcareous armature. The species of the

genus *Thourella* have scale-like sclerites, which can be ornamented by long and sharp thorns surrounding an operculum formed by small triangular scales. The operculum closes the well protected soft parts of the polyp. The coenenchyme is also covered by scales which overlap each other. Knowledge of sclerite function and mechanism has been limited to sclerites present in tropical gorgonians and alcyonaceans which show varied forms such as inter alia spindles, clubs, rods, scaphoids, radiates and double-heads. The scale-like sclerites, characteristic of primnoids, have been investigated only as a taxonomic character. Details of the surface structures of these scales have been well documented since they are important for distinguishing species. However, as most of the studies on this family were done at the end of last century and beginning of this century, the investigation was limited compared with present day standards. With the advances in technology now available (for example SEM), it has been possible to identify more structures.

2.1.5 - OBJECTIVES OF THE CURRENT STUDY

The objectives of the research presented in this chapter are to give a general description of the colony morphology of *Thouarella variabilis* with special reference to the architectural design of sclerites, describing details of their morphological features; arrangement and number of operculum, marginal, adaxial and body scales; formation of sclerites throughout the polyp development; and their function as anti-predator defence; and aspects of branching patterns, arrangement and distribution of polyps. It was also considered to be of interest to give a brief account on age determination through the examination of growth layers present in the axes of the studied specimens.

2.2 - METHODOLOGY

The methodology that will be described in this section was applied for both taxonomic studies and studies on the skeletal parts of the colony.

2.2.1 - FIXATION AND PRESERVATION

Some of the specimens, mainly the ones collected during the first Brazilian expeditions, were fixed in unbuffered formalin in which they were left for several years. The consequence of the use of this fixative was damage to the calcareous elements of the skeletal structure of the

colonies. Most of the specimens were, however, appropriately fixed in buffered formalin (10%) and then transferred to alcohol (70°GL) in which they remained stored for many years prior to further processing. No specimen was preserved dry because of the fragility of the calcareous sclerites presented by this species, which can be easily damaged.

2.2.2 - ORGANISATION OF STUDY

Surveying taxonomic literature, it was possible to identify the major characters used to describe the morphological features of the studied species. Based on this, a programme of details to be observed was established. It included shape, size and thickness of the colonies; branching pattern of colony and branches; distribution of polyps in the different parts of the colony and branches; inclination of polyp in relation to the stem; size of polyps; anthocodial armature, set of sclerites on the polyp; number, arrangement, size and details of the different types of sclerites; colour; structural components, diameter and appearance of axis; growth rings; holdfast; and presence of commensals or any other peculiarity which could be observed. From these observations, a better understanding of the general morphology of *Thouarella variabilis* could be attained.

2.2.3 - MACROSCOPIC OBSERVATIONS

Measurement of the length and width of the colonies were performed by means of a flexible ruler and a calliper. The larger branches were measured with a calliper and the smaller branches, polyps and sclerites were measured with an eyepiece micrometer. The position, orientations and distance between branches were also observed, initially unaided and then were subsequently confirmed and measured under a stereomicroscope.

The presence, aspect and diameter of the holdfast and the colour and shape of the colony were also registered. Colour is a dubious character since the colony had been preserved in alcohol for many years, though some seem to have the original colour well preserved. The colonies were also examined for the presence of skeletal modification caused by commensals. Polychaete annelids have commonly been observed to induce the branches of octocorals to bend forming a tube which the worms inhabit. The presence of epibionts such as bryozoans, hydrozoans and sponges was also noted.

2.2.4 - MICROSCOPIC OBSERVATIONS

The polyps of *Thouarella variabilis* are small (the average length being around one millimetre) and extremely fragile, being easily damaged when handled. It was advisable, therefore, to extract a fragment of the colony for examination, preserving the rest of the specimen. In order to undertake the measurements of the branches and polyps in a satisfactory way and to observed properly the distribution of polyps and the arrangement of sclerites on the polyps, the fragments were attached to the bottom of a petri dish, which had been covered with stained wax. To fix the fragment in a desirable position, the most practical method was to use needles employed for auricular acupuncture, which are made of slender and flexible material, but strong enough to allow fixation of the animals.

To determine the branching pattern, the branches were classified according to a branch ordering system proposed by Brazeau & Lasker (1988). The most distal branches were defined as first order branches and a second order branch arises when two first order branches join. Following the same logic, a fourth order branch appears only when a third order branch is joined by another third order. A branch which joins another branch of equal order is called a source branch and a branch which joins a branch of higher order is called a tributary (Brazeau & Lasker, 1988). The ordering system of Brazeau and Lasker (loc. cit.) was posteriorly abandoned and the more traditional ordering system, where sensibly the first order branchlet is the most proximal branchlet, was adopted instead. Branches of all orders were measured with an eyepiece micrometer.

To count the sclerites from different parts of the specimen, the polyp had to be turned slowly under a stereomicroscope. To manipulate it efficiently, the polyp was embedded in a dense gelatinous substance. Although glycerin is advisable because it guarantees the preservation of the specimen, "hair styling gel" has a denser consistency and was a better solution if polyps could be spared.

For the investigation of the fine structures of sclerites and to obtain their measurements, the sclerites had to be freed from the tissues. For that, the specimens were treated with concentrated sodium hypochlorite bleach for the dissolution of tissues. This treatment is rapid with the sclerites being exposed almost instantly. When the objective was to examine the arrangement of opercular elements whilst dissolution was in process, the specimen was placed in distilled water and small quantities of sodium hypochlorite solution were added gradually. In this way the speed of dissolution could be controlled as desired. This procedure also avoided the presence of bubbles that could disturb the observation. If they persisted they

could be removed with filter paper. Once free from tissue, the sclerites were washed in distilled water and observed in a light microscope. Sclerites were measured by using an eyepiece micrometer. The sclerites could, therefore, have their position, number, shape and size recorded.

2.2.5 - ULTRAMICROSCOPY

Polyps, branches and sclerites were prepared for examination by scanning electron microscopy (SEM). Undamaged polyps and branches were carefully chosen and were meticulously cleaned of surrounding tissue. The specimens were dipped in sodium hypochlorite solution, transferred to hydrogen peroxide for a few seconds and then placed in distilled water for thorough washing. They were eventually rinsed in 70 % ethanol and dried. Caution was taken to avoid excess exposure of the specimen to the aforementioned solutions as they might destroy the polyp. When dried properly the specimens were mounted on aluminium stubs to which they were adhered by a carbon impregnated film. The polyps were arranged in a desired position and sometimes they were attached with araldite to the tips of needles for auricular acupuncture which allowed better manipulation.

For the preparation of sclerites the whole tissue was removed in sodium hypochlorite bleach following the procedure mentioned in Section 2.2.4. After complete dissolution, the spicules were brought to the centre of the petri dish by careful spinning movements and the solution was removed with a pasteur pipette. The sclerites were then washed four times in distilled water following the same method. They were left to dry afterwards until further processing. Once clean and dry, the sclerites were mounted on aluminium stubs. They were transferred to the carbon impregnated film covering the stub with an eyelash attached to the tip of a wooden stick.

Some preparations were coated with gold/palladium but most were coated with 20nm of gold in a Hummer VI sputter coater. The preparations were examined with two scanning electron microscopes (SEM): ISI 60A and JSM 6400 (JEOL) at an acceleration voltage of 20 kv.

2.2.6 - STAINING AND ILLUSTRATION

The colour of the studied colonies varied from pinkish cream to milky white after preservation; consequently, individual sclerites are not delineated and the structures and arrangement of

the anthocodial armature cannot be visualised. By staining the polyps with methylene blue, it was possible to emphasize the spicular architecture. This is a non-destructive method because the staining is easily washed away in alcohol.

Drawings were used together with photography to illustrate the polyps and sclerites. They were drawn by means of a camera lucida. This being a limited and time-consuming technique, macro-, micro- and ultramicrographs were used instead to illustrate the morphological features.

2.2.7 - GROWTH STUDY

To evaluate the morphological development of scales through the different phases of growth, five classes of polyp size were defined: ≤ 0.30mm; 0.31-0.50mm; 0.51-0.80mm; 0.81 - 1.20mm; ≥ 1.21mm. Sclerites of polyps from these different classes were examined by SEM. Sets of scales for each class were prepared and compared.

To study the development of polyps along the colony, branches were removed from three regions of the colony: distal, middle and proximal. All the polyps on branches from each region had their position located and their length measured. They were separated according to the classes mentioned above.

In an attempt to determine the age of the colonies, several methods were employed for counting the growth rings. Most of these were unsuccessful. The major difficulty was to get satisfactory transverse sections for examination. The most successful method consisted in embedding portion of the axes in a mixture of acetone (5 - 10 %), araldite AY105 and hardener HZ18. The preparations were kept in a refrigerator, to prevent the acetone evaporating, for at least three days. When solidified, the blocks were sectioned in a fine low-speed saw, ISOMET. Some samples were polished with a diamante plate but as the result was the same, and being an expensive technique, this step was abandoned. The samples were adhered to an aluminium stub by a carbon impregnated film, coated with carbon using an Edwards 306 auto unit and eventually observed in a scanning electron microscope (SEM) JSM 6400 (JEOL) at an acceleration voltage of 20 kv.

2.3 - RESULTS

2.3.1 - COLONY SHAPE AND BRANCHING PATTERN

The colony shape is mostly superficially similar to a bottlebrush with short ramified branches rising all around the main stem (Figures 1 to 6). The branches rise in opposing pairs alternating in four directions. Sometimes the pairs arise from the same point at the main stem, being perfectly opposite, but at other times they are offset. In some parts of the colony the branches arise in three directions and the fourth one arises in the same direction as the first, not in pairs. The distance between two branches of the same direction is on average 6mm, decreasing towards the distal region of the colony. The branches are perpendicular or directed slightly upward.

The branching pattern described above was observed in colonies which were apparently unaffected by any disturbing factor, such as the presence of a commensal or a change in the direction of water movement. In some specimens the common pattern presented above could be misinterpreted with the stem being twisted, giving the impression that the branches arise in all directions. If the stem is untwisted, the colony shows a biseriate arrangement with the branches bent laterally forming a frontal and versal side. In other specimens both patterns can be seen in the same colony, where the upper portion is bottlebrush and the lower portion biseriate.

The presence of annelid commensals has been seen to induce the formation of tubes (Figures 11 and 12) which evidently affects the ramification of the colony. The branches are bent to form the tube and, therefore, the ramification is altered into a biseriate arrangement. Evidence of this can be seen in colonies which have a bottlebrush pattern up to where the tube starts and from there become biseriate.

Branch length varies along the colony, the longest branches are positioned in the middle of the colony decreasing in length toward the distal and proximal regions of the colony. The shortest branches are found at the terminal portion of the colony. This difference in branch length gives the colony a spindly shape (Figure 5).

Only a few colonies were intact, without any part lost, which made assumptions about size, thickness or correlated measurements difficult. The largest size observed amongst the studied specimens was 130 mm (260 mm in a specimen held in the collection in Brazil) and

the thickest colony was 37.5 mm in diameter. There is no apparent correlation between length and thickness. Some high colonies have a slim appearance whereas some short colonies are relatively thick.

The branches are divided into branchlets, which are dichotomously branched in one plane early in development. The poorly ramified branches are perfectly dichotomous, where the branching pattern is a repeated bifurcation and only source branchlets of up to four or even five orders are found, no tributary branchlets are observed. The more ramified branches are complex and show tributary branchlets. When the colony grows to a certain extent, side stems arise irregularly along the colony (Figures 4 and 6).

2.3.2 - DISTRIBUTION AND NUMBER OF POLYPS

The polyps tend to be biserially distributed on the branchlets, although polyps rising irregularly from all directions are also common. They are isolated, alternating with each other, occasionally in pairs, never in whorls (Figures 19 and 68). The distance between them varies along the same colony and between colonies. The average distance being 1.8 mm, diminishing toward the tips of the branches (Figure 13), where they might be considerably aggregated making the branchlet heavy and with a loose appearance. The length of the branchlets might also influence the distance between the polyps. On the less ramified branches, the polyps are more separated and on the more ramified branches, which present shorter branchlets, they are closer together. The main stem also bears polyps but they are absent at the base of the stem and along the polychaete tube where a free path is left for the worm (Figure 11).

The typical number of polyps per 10 mm of branch is 10. In the middle region of the colony where the polyps are more scattered, the common number is 8 polyps per 10 mm, sometimes reduced to 5 or 6. At the proximal region, the number varies between 8 and 10 and at the distal region, where the polyps might be found closely crowded, 20 to 30 polyps/10 mm have been recorded.

2.3.3 - SHAPE AND SIZE OF POLYPS

The shape of the polyps does not differ much, unless the age or reproductive phase are considered. They are pear-,cup- or club-shaped, with a narrow neck and a broadened calyx

(Figures 17, 21 and 67). Because of the inward position of the polyp on the stem, the side of the body turned toward the axis is called adaxial and the side facing outward is called abaxial (Bayer, 1982). Some polyps are so narrow in the lower part that only one longitudinal row of large scales can be observed on the abaxial side (Figure 21) whereas in polyps bearing eggs several longitudinal rows are apparent as a consequence of the enlargement of the base and neck (Figure 20).

In relation to the axis, the polyps are directed outward or slightly upward (Figures 13, 17 and 19). They are often seen bent towards the stem (Figures 19 and 67 to 69), especially the polyps at the bases of the branches, which are bearing eggs. At the terminal portions of the branchlets, the polyps are mostly directed outwards (Figure 13).

The mean polyp length was 1.08 ± 0.02 mm and the longest polyp observed was 2.00mm. The mean diameter of the calyx is 0.60 mm, although the variance of this estimate is high owing to the difficulties in measuring the calyx precisely.

The colonies are mostly mature and the sturdy appearance of the polyps bearing eggs makes the branches look crowded and the branchlets thick, since the distance between the polyps diminishes as they expand.

2.3.4 - SPICULAR ARCHITECTURE

The results of the XRD analysis showed that the sclerites of the examined specimen of *Thouarella variabilis* are composed of calcite; there is no trace of aragonite as was expected in the species of the Family Primnoidae. Sclerites in *Thouarella variabilis* are present exclusively in the form of scales which can vary considerably in shape and size. These scales are thin, fragile and can be complexly ornamented. Longitudinal rows of body scales, as registered in the literature, were not definable, unless the polyp was expanded when bearing eggs. The rows of body scales are, rather, diagonally arranged. Five to six diagonal rows were observed on the abaxial and lateral sides (Figure 66) and two longitudinal rows of small scales are present on the adaxial side.

An operculum, formed by eight triangular scales, is found at the distalmost portion of the polyp (Figure 58). The tip of these eight opercular scales can vary from being blunt to sharply pointed. They are widely separated from each other when the operculum is open and the soft

parts of the polyp are exposed. When the operculum is closed, the scales are close-fitting, overlaying each other, sealing the soft parts of the polyp. The opercular scales are surrounded by eight conspicuously thorny scales on the margin of the calyx. These marginal scales are disposed in two rings of four scales, an internal ring and an external ring, which alternate each other. The abaxial and lateral marginal scales are larger and more thorny than the marginal scales on the adaxial side of the polyp. In some specimens, the most lateral scales on the abaxial side have markedly long thorns.

The body scales on the abaxial side of the polyp can also occur in transverse rows. The first transverse row of body scales (excluding marginal scales), the submarginal scales, is located immediately below the marginal scales. These submarginal scales are slightly thorny and are disposed alternating with the marginal scales of the outer ring and coinciding with the scales of the inner ring (Figures 65 and 66). Below this first row there are two rows of medial scales which are larger than the submarginal scales and only one or two scales are present on each row (Figures 21 and 67). The basal row at the most proximal portion of the polyp, where it merges with the coenenchyme, is composed of a pair of large body scales, which seem to be curved to accommodate the polyp on the axis. These basal scales are the largest scales on the polyp and the marginal scales the longest. The scales on the lateral sides are slightly smaller. The body scales overlap each other on their proximal portion and have the distal margin free (Figures 59, 60, 71 and 72). The adaxial side of the polyp is covered by two longitudinal rows of small and plate-like scales. Unlike the other scales of the polyp, these adaxial scales are not overlapping but parallel. In some specimens the large lateral scales appear to cover the small adaxial scales (Figure 57). In expanded polyps, however, their presence can be confirmed. The scales on the coenenchyme covering the axis are similar to the body scales from the lower portion of the polyps but are smaller and less curved. They overlap each other along the whole axial skeleton (Figures 68 to 71).

2.3.5 - MORPHOLOGICAL FEATURES AND ORNAMENTATION OF SCLERITES

The inner or outer surfaces of the scales have distinct ornamentation. In most instances, the inner side of the scales present on a polyp are ornamented by tubercles and compound warts and on the outer side by spines and crests, ridges or wrinkles.

The opercular scales are triangular with the proximal portion being larger and tapering towards the distal portion (Figures 33 and 79a-b). The proximal portion or base is indented and sculptured by irregularly arranged tubercles whereas the margins of the tapered distal

end are serrated and spiky. The outer surface of the scale is relatively smooth and homogeneously covered by short spines, which are projected crystalline fibres erupting on the surface of the scale (Figure 39). The distal end of the scale is slightly concave with ridged edges (Figure 79b). The inner side of the opercular scales are covered by compound warts mostly aggregated at the convex centre of the scale (Figure 79a). The distal end of the inner side is more spiky than the outer and can present a short and stout keel (Figure 33).

The marginal scales are ornamented with more complicated processes when compared with the other scales (Figures 79c to f). The base is heavily indented and the tubercles are more numerous and pronounced than in the opercular scales (Figure 79d). The distal portion is serrated and displays a conspicuous thorn, whose length varies considerably between specimens. In some marginal scales, the region where the thorn is linked to the main body of the scale is winglike with a smooth free margin (Figures 49 and 79e) which is almost translucent in some cases. The outer side of the scale is covered by short spines homogeneously distributed throughout the surface of the scale (Figures 54, 56 and 79c, d and f). The base on the outer side is more crowded with tubercles than on the inner side. Ridged spines or crests radiate toward the serrated margins of the scales (Figure 79c). The aspect of the thorn appears to be unique to every single scale. The thorns might be similar in structure but the mode of calcification or stratification differs from scale to scale. In general, the thorn is lance-shaped, with winglike lateral expansions and an inner keel and an outer ridge. The margins are deeply serrated along the thorn. On the inner side, the thorn has a markedly strong keel (Figure 79e), slightly grooved with a relatively smooth surface, although they can occasionally possess series of aligned spines. On the outer side, the central crest is thinner and covered by a series of spines or protuberances (Figures 62 and 63). Close to the base of the keel, where it enlarges to form the main body of the scale, more series of these protuberances are formed on the laterals. The margins on the outer side of some scales, appear stratified on certain parts where parallel small winglike expansions are found (Figure 63). On the inner side of the scale numerous compound warts are found, particularly in the convex portion of the scale, where the warts are closely spaced (Figures 49, 51 and 53). The borders of the distal portion of the scale are smooth and free from warts. At the limit of the distribution of the warts, at the base of the thorn, mostly damaged warts are observed (Figures 50 and 52).

The submarginal scales are slightly rounded with a pointed distal portion (Figure 41 and 79h). They have numerous tubercles on their bases, mainly on the outer side, which is covered throughout by spines arranged, in some scales, in radial rows. The small thorn is armoured with numerous spines. The surface of the inner side is smoother, with warts equally

distributed over the whole surface apart from the margins (Figures 78i and 79g). The other body scales are proportionately larger, wider and more concave and curved to fit the body of the polyp (Figures 79i, j and l). The outer side is packed with tubercles at the base and ornamented with spines along the whole surface (Figures 79j and l). The distal portion is smoother and serrated towards the point where the radial ridges converge (Figure 79j). Some scales are so ornamented that only pointed spines can be seen on their outer surface (Figure 79l). The inner surface is packed with warts which are most developed on the outer portion of the scale (Figure 79i). Only a few damaged warts were observed on the margin of the scale.

The adaxial scales are considerably smaller, usually slightly rounded, and occasionally amorphous (Figures 30, 78k and 79k). They can also have their outer surface ornamented with well distributed spines (Figure 79k). On the inner side a few big warts are found. In some scales most are damaged (Figure 78k). The borders of the scales are not serrated as seen on the other scales.

2.3.6 - FORMATION OF SCLERITES THROUGHOUT THE POLYP DEVELOPMENT

Young polyps arise in between adult polyps. They probably appear whenever there is an extension of the axis and enough space between two polyps. They are also seen rising close to bifurcations where the extension of the axis is more likely to occur. The young polyps have been observed to grow in the same direction as the adult polyps, close to or opposite to them (Figures 18 and 19). The polyps were classified in five size classes so the development of the calcareous sclerites through growth could be evaluated. The description of the sclerites within each class is presented here. The classes were defined according to the phases where more variation was found and are therefore unproportional.

FIRST SIZE CLASS ($\leq$ 0.3 mm) - the youngest polyps observed were composed of an aggregate of scales without any proper form. When the polyp has reached around 0.3 mm (Figure 73), it has a sturdy body surrounded by scales of irregular shape. It is difficult to determine the type and position of these scales because the polyps are too small and the tissues are quickly dissolved. However, some resemblance to the adult form might still be traced. The scales positioned at the distalmost portion of the polyp have the first evidence of thorns, showing a low blunt spine on their distal region. The other scales have the features

of the body scales from adult polyps. They present a few short spines randomly distributed on their surface. The margins are smooth, not serrated and the crystalline fibres are easily distinguished (Figures 22 to 24).

SECOND SIZE CLASS (0.31 - 0.50 mm) - the polyps start displaying a more complex calcareous armature, although the scales are still poorly ornamented (Figure 74). The opercular scales are already similar to the adult opercular scales (Figures 74a and b). They are triangular with the extremity of the tapered end pointed. No tubercles are present at the base of the scales but there are already several well distributed short spines on the surface of the scale (Figures 37 to 40). The short spines where the crystalline fibres start to project are more like nodules. An opening at a central point of the scale is noticeable at this stage (Figures 37 and 74a). The marginal scales are also triangular in shape but more pointed than the operculars (Figures 74c to e). The thorn is recognisable and the body of the scale is proportionally smaller when compared to the size of the developing thorn. There are no tubercles at the base, the margins are smooth, not serrated and the short spines are at the same stage as those from the opercular scales. The body scales are wide and resemble the shape of the body scales of the adult polyps but they lack ornamentation, apart from some short spines and a few ridges (Figures 74f and g). They have smooth surfaces and margins.

THIRD SIZE CLASS (0.51 - 0.80 mm) - at this size, the polyps resemble the adult polyps but some scales are still underdeveloped (Figures 25, 75, 76 and 77). The opercular scales have the same aspect as the anterior class but have developed tubercles at the base, which now appears indented and the margins of the distal portion are serrated (Figures 75a, b and c, 76a and b and 77a). The inner side has many warts and a spiky keel is shown on its pointed end (Figures 33, 75b, 76a and 77a). On the margins of the inner side, it is still possible to distinguish the crystalline fibres (Figure 36) but on the central portion the surface appears more compacted and the calcification is more complex around the warts (Figures 34 and 35). The marginal scales are now markedly different (Figures 75d and e and 76c and f). Their bodies are larger and wider when compared to the scales of the anterior stage. Their bases are indented and crowded with tubercles and the margins of the distal portion are serrated and form ridges. The spines on the outer surface are homogeneously distributed (Figure 75e) as are the warts on the inner side (Figure 75d). The warts, at the limit of their distribution, at the base of the thorn are damaged. The thorn is reasonably developed but is not very pronounced. The submarginal scales are wide, show tubercles on their base and have serrated free margins (Figures 75g and h). On their outer side, they show radial rows of spines, which are longer than the short spines present on the scales of the anterior stages

(Figure 75g). These scales show a short thorn at the distalmost portion. The other body scales have lost their smooth margins which are now serrated and their surfaces are more ornamented with spines and warts (Figures 26, 27, 75i to l, 76d and 77d and e). The adaxial scales are rounded, with smooth edges and their inner side has no damaged warts (Figures 28, 30 and 31).

FOURTH SIZE CLASS (0.80 - 1.20 mm) - at this size, the polyp has attained the average adult size and scales are well developed (Figure 57). The opercular scales do not differentiate significantly from the anterior stage, apart from being more sharply pointed. The marginal scales are better developed (Figures 78a to d). They display long, lancet-shaped, thorns and numerous spines on their outer surface (Figure 78b) and their inner side is packed with compound warts (Figure 78b). The body scales are also well ornamented by spine and warts and the thorn of the submarginal scales are more conspicuous (Figure 78j).

FIFTH SIZE CLASS (≥ 1.21 mm) - the largest and oldest polyps are not different in general features from the previous stage but display a much heavier ornamentation and calcification (Figure 79). The thorns of the marginal scales are markedly strong, spiky and stratified. The outer side of the scale is noticeably armoured with spines and the inner side packed with compound warts. They have the same appearance as the scales of the adult polyp described in Section 2.3.5.

2.3.7 - ARCHITECTURAL DESIGN AND FUNCTION OF SCLERITES

The soft parts of the polyps of *Thouarella variabilis* do not retract into the coenenchyme, which is notably thin in this species. If these polyps did not have a protection of any sort, they would be potential prey for a variety of predators. The calcareous armature surrounding the soft parts of the polyps provides not only skeletal support but also functions as anti-predator defence.

The sclerites are arranged in such a way that the polyp is supported on the surface of the axis. Despite their rigid appearance, the sclerites provide a certain flexibility to the polyp, which can bend towards the stem and expand its volume when bearing eggs. The overlapping body scales can, to a degree, be pulled apart when the polyp is under tension. When overlapping each other, the outer tubercles of the base and the spines on the surface of the underlying scale engage the tubercles and warts of the inner surface of the overlying scale. They, therefore, interlock with each other, avoiding excessive extensions of the scales.

The tissues of the polyp have a high capacity for extension, which can be observed on fully extended reproductive polyps. To accompany this expansion of the polyp, the sclerites have to be pulled apart and, in doing so, the tubercles, spines and warts probably disengage and allow the overlapping scales to slide to a certain degree. Evidence of such movement, is the damaged warts present on the distal portion of the inner surface of the scales. These worn warts delineate the extent of the movement. On opercular scales, worn warts are rarely observed, this can be explained by the nature of their movement. They are not being pulled apart when overlapped and when the operculum is open these scales are disposed parallel to the marginal scales. The adaxial scales, which are arranged in parallel, do not present basal tubercles, and the inner warts are used to anchor them to the mesoglea. The tissue on the adaxial side is probably capable of great extension since the small adaxial scales are not limited in freedom of movement. When the polyp is fully extended, the lateral body scales are positioned on the abaxial side and, therefore, it is possible to define longitudinal rows of abaxial scales.

The scales covering the axes of the branches and main stem might contribute to the stiffness of the axial skeleton since, under tension and compression, the interlocked scales will limit extension, although allowing some flexibility of movement. Under tension, the scales are able to slide apart but remain interlocked and under compression they can move closer together.

The architectural design of the calcareous armature of the polyps appears to be an efficient anti-predator defence. The close fitting operculum enclosing the soft parts of the polyp surrounded by a circle of sharply pointed thorns is a potential deterrent against predators. The scales are strongly calcified and have their outer surface sculptured with sharp spines and serrated free margins. Since the polyps are able to lean towards the stem, their adaxial sides are properly protected and, therefore, the scales covering this side are not disadvantaged by being smaller and weaker. The lack of long thorns on the marginal scales on the adaxial side facilitates their leaning. The polyps on the periphery of the branchlets have more thorny scales than the polyps from other regions. These peripheral polyps have their adaxial side exposed as they stand erect. In non-reproductive polyps expansion to accommodate eggs is not necessary and, as already mentioned, in normal conditions the lateral scales will cover the adaxial side. Polyps on the periphery are rarely reproducing and polyps in the middle and base of branches, which are reproducing, are usually leaning towards the main stem. Disturbance, such as the presence of commensals, might induce differential calcification and improvement of the efficiency of the protective calcareous armature.

2.3.8 - PRESENCE OF COMMENSALS

Annelid commensals are commonly observed inhabiting the colonies of *Thouarella variabilis*. The polychaete often found on the colonies studied was *Hemilepidia* sp., family Polynoidae (Figure 12). These commensals induce conspicuous external modifications upon their hosts. The branches, for some unknown reason, bend to form a tube along the colony, parallel with the main stem (Figure 11). The tube usually starts at the proximal part of the colony and goes up to the middle or even to the most distal portion of the colony. Where the tube finishes, there is always an open enlarged area without branches. In some colonies two tubes are formed opposite to each other. The sides of the branches which are in contact with the tube are deprived of polyps. The polyps appear aggregated on the opposite side of the tube. These polyps which are close to the commensal tube have more thorny marginal scales and the body scales are more calcified than on the other polyps on the colony. The submarginal scales are also more thorny and so are the opercular scales. The axes of the branches and main stem forming the tube have heavily calcified and compacted scales paving the path of the worm.

2.3.9 - FEATURES OF THE AXIAL SKELETON AND GROWTH LAYERS

The axial skeleton is horny with a smooth surface in some parts and slightly grooved longitudinally in others (Figures 7 to 10 and 96 to 98). The colour varies from dark brown to yellow. The diameter or density varies according to the age of the colony. In cross section, the axis shows alternate layers, often not easily distinguished (Figures 80 to 95 and 99 to 103). The limits of the layers are poorly defined. Thick layers alternate with thin lines (Figure 102). The width of the layers is irregular and decreases towards the border with the innermost layers being the thickest. The outer layers seem to be darker than the inner layers (Figure 83 and 102). Between two thin compact light layers, there is a perpendicular deposition of skeletal material which is permeated with gaps and cavities (Figures 99 and 100). When a cross section is observed as a whole in a large scale, it is possible to define the main growth layers but when each portion of the axis is examined in detail many sublayers are detected between the main growth layers. These sublayers differ in density and can be compacted or porous.

The central core of the axis is amorphous and has a seemingly soft consistency (Figures 95, 102 and 103). The limit between the central core and the first growth layer is hard to detect. In young colonies the first growth layer can be relatively large when compared to the

subsequent layers (Figures 84 and 86). In some axes there are undulating layers formed around the central core. This is observed mainly in young colonies whose few growth layers are not compressed by many subsequent ones.

If it is assumed that the wider layers are a result of the higher availability of food in the eutrophic summer months and the thinner layers represent the low growth during the energy-poor winter months, the count of a wide and a thin layer would indicate a complete year. In young colonies, up to 5 or 6 years old, the wide growth layers are easily detectable since, by being uncompressed, they leave gaps between them. In old colonies, the outer growth layers are thin and apparently compressed against each other (Figures 94 and 95). They are not easily distinguishable, which makes the determination of age in old colonies unreliable. The outermost layer of the axis is usually dark and crusty (Figure 83). In *Ascolepis* sp., a species used for comparison, a relatively thick outer dark cortex was seen permeated with calcareous material (Figure 89).

The axes of the branches are slender and fragile usually with a pale yellow colour. The growth layers observed in such axes are smoother and better delineated when compared to the growth layers of the main stem (Figure 88).

2.3.10 - HOLDFAST

The holdfast is a cone-shaped enlargement of the base of the main stem (Figures 7 to 10). This base is composed of aggregated calcareous material, probably sclerites. In young colonies, the horny base of the main stem is reinforced with some calcareous components through which it is cemented to the substratum. The older the colony, the heavier the calcification of the holdfast. In old colonies, the holdfast appears compacted (Figure 8) whereas in younger colonies the crystalline components are less packed and irregularly distributed around the bases of the main stem (Figure 10). The lower portion of the stem lacks coenenchyme, probably because of heavier abrasion by suspended material on this portion. The colonies can adhere to virtually any solid substratum, but they have been mainly observed on stones and shells.

2.4 - DISCUSSION

Thouarella variabilis has not yet been recorded in areas of shallow water in Antarctica. The shallowest record in the existing literature was from a depth of 50 m (Port-Lockroy, Chenal de Roosen, Gravier, 1914) and amongst the specimens examined for this study, 53 m (King George Island, Admiralty Bay). The investigated colonies were, therefore, not exposed to wave surge but instead inhabited areas of current-generated water movement. The nature of the currents is unknown but could be unidirectional or multidirectional. *Th. variabilis* is predominately bottlebrush, with branches distributed in all direction. According to Grigg (1979) these bushy forms predominate when there is no dominant current direction. Some colonies have, however, been found to have a biseriate branching pattern where the branches are distributed on two sides of the colony only. The reason for this variation in orientation might be the change in water direction. In unidirectional or bidirectional current-generated water movements, the colonies gradually change their orientation facing the current. A bottlebrush colony would be at a disadvantage in a bidirectional current since their fragile branches could be damaged by interbranch abrasion and some polyps could be in unfavourable positions for feeding. Furthermore, the axial skeleton of *Th. variabilis* is reasonably flexible and has a low torsional resistance, allowing passive twisting. Some biseriate colonies are continuously twisted assuming a bottlebrush appearance. This could indicate inconstancy in the direction of the predominant water movement or these colonies might have been subject to different water flows during their development and grown helically. The colonies which have the upper portion bottlebrush and the lower portion biseriate, or the other way around, might have had different flows acting at the basis of the stem and at the top of the colony. This could be a result of the difference in water flows between the boundary layer and the larger drag forces of the main stream.

The bottlebrush form is of low porosity, having the branches closely spaced. Under the effect of strong unidirectional currents, the water passage through the colony could be reduced by its low porosity and, as a consequence, some parts of the colony could be poorly nourished whilst others are damaged by abrasion. In moderate multidirectional water movement, conversely, the velocity of the flow could be modified by the shape of the colony, reducing the drag forces and generating eddy currents, which facilitate the uniform nourishment of the polyps of the colony.

The colonies of *Th. variabilis* are highly ramified and the polyps are homogeneously distributed on the branches, apart from the periphery of the branches where they are closely spaced. These are the requirements for improved feeding efficiency in sheltered areas, which

have low availability of food (Velimirov, 1976). The aggregation of polyps at the periphery of the branches might be explained by this being the most favourable feeding position. At the middle and bases of the branches the polyps are more sparsely distributed and they will probably depend on the occurrence of eddy currents to grab food. Whether colonies with higher porosity or with greater availability of food will present a higher number of polyps, which can be aggregated, is unknown. Other species of the genus *Thouarella,* which are less ramified and more porous than *Thouarella variabilis*, have strongly aggregated polyps. Within *Th. variabilis,* some colonies have more aggregated polyps than others, which can be explained by the peculiarities of the environment which they inhabit. The packed appearance of the branches might as well be explained by the reproductive state of the polyps which change their shape, expanding to accommodate their eggs or sperm sacs. As the polyps expand considerably, the distance between the polyps diminishes making them more aggregated.

The appearance of the sclerites during development varies considerably. After reaching a size of 0.8 mm, the opercular and ventral scales do not vary a great deal, but the body scales, especially the marginal scales, vary substantially. In the first stages of development, when the body of the polyp is still not well defined, being more an aggregate of scales, warts and spines are limited and tubercles do not exist at the base of the scales. Once the polyp has a defined shape and the scales are well placed on the body, tubercles can be observed at the bases of the scales as they, together with the inner warts and outer spines, play the important role of anchoring the scales to the mesoglea and in holding the scales together. Above a certain size (1 mm), the polyp has an adult form and the main variation in scale form will consist in reinforcing the calcareous structure, on heavier calcification and fortification of spines and thorns. The older the polyp, the more robust its appearance.

Factors such as the environmental stress, the availability of certain chemical elements, water temperature and pressure and the presence of commensals might influence the nature of calcification and consequent shape of sclerites. The increase of speed of water movement increases the calcification rates (Velimirov *et al.*, 1979). The calcium, magnesium and soluble bicarbonate uptake will depend on the availability, temperature and activity of the polyp. The water temperature and pressure will have an influence on the nature of the crystal components present on the octocoral. The presence of annelid commensals will induce modification in the spiculation of the octocorals. The polyps located close to the commensal tube have long thorns and spines and are heavier calcified. Another factor which should be considered is the availability of food for the colony because the calcification process is a costly, demanding energy expenditure. In view of all these sources of variation on the

spiculation of octocorals, caution should be taken when using sclerites as taxonomic characters. Within the same colony, polyps with long and short thorns were observed opposite to each other; between colonies the variation on the length and strength of the thorns and spines was remarkable. A colony with short-thorned polyps looks more fragile than a colony whose polyps are bearing long and strong thorns and heavily calcified and spiky body scales. At a first glance, these differences in appearance could lead to the separation of the two colonies into different groups.

The presence of toxic compounds in the studied species has not been investigated. The tentacles of the polyps were shown to have a large amount of nematocysts. Whether the nematocysts are of offensive or defensive nature is not known. According to Sammarco and Coll (1988), there is an inverse relationship between toxicological defense characteristics and structural defence characteristics. If so, the production of toxic compounds in *Th. variabilis* might be unnecessary since the morphological defenses presented by this species seem to be an efficient deterrent. In none of the specimens examined was there evidence of polyps being damaged by predators. The architecture design of the calcareous armature enveloping the polyp, where an operculum enclosing the soft body of the polyp is surrounded by long thorns and spines, appears to be an efficient defence mechanism. The large calcareous scale-like sclerites covering the polyps and coenenchyme might also be a potential antipredator defence. Van Alstyne and Paul (1992) demonstrated that sclerites are feeding deterrents, depending on their concentration, shape and size. The role of the scales on the structural support of the colony by increasing the stiffness of the axis was not properly investigated but the mechanism of interlocking and sliding shown by the scales adding to their high concentration covering the stem, suggests some influence of the scales on the mechanical properties of the axes.

The growth in *Th. variabilis* accounts for the increase in density and in extension of the axial skeleton and for the extension and higher ramification of branches, with the continuous bifurcation and consequent formation of new branchlets. In young colonies the branches are poorly ramified and the branching pattern is perfectly dichotomous where only source branchlets are formed. An uniform length of the branch is always kept and the last order branchlets seem to grow simultaneously and at the same rate as the adjacent branchlet, independent of its order. The first order of the branch is usually a single stalk which links the branch to the main stem. In some parts of the colony this stalk seems doubled indicating that two branches could rise from the same point on the main stem becoming separated with successive growth. In older branches the ramification is more complex and some tributary branchlets might be observed, where the branches are not equal, being pseudodichotomous.

In some old colonies, it was also observed that one side of the branch is more ramified than the other side. This could be a result of differential feeding positions on the two sides. The side which will be supplied with least food will be induced to increase the surface area for feeding by being more ramified or, conversely, the side which is more exposed to food has more favourable conditions for axial growth and polyp development and therefore increment in its branch pattern. Another hypothesis is the simple availability of interbranch space allowing higher ramification. The aggregation of polyps at the last order branchlets could be minimized by continuous growth of the axial skeleton between the polyps and consequent increase in distance between them. The highest growth rate in extension of branchlets seems to be, however, close to the bifurcations where areas devoid of polyps are observed and young polyps are preferentially arising. The higher ramified branches are distributed at the middle portion of the colony and the poorly ramified or simple branches are located at the proximal and the distalmost portions of the colonies. The branches at the distalmost portion of the colony are the youngest, consequently they show poor branching pattern and high aggregation of polyps.

The growth of the main axial skeleton occurs by the addition of layers of skeletal material. A thin layer of compacted material overlies a thicker layer of unconsolidated material deposited perpendicularly. The thickness of these layers is varied suggesting that the growth rate is irregular. The causes for the discontinuity in deposition of skeletal material are unknown. In Antarctic waters the major trigger for growth is known to be the availability of food in the eutrophic summer months. This could be a potential explanation for the periodicity in deposition of skeletal material, where the larger layers could be formed in the months of high energy and the thin compact layer in the energy-poor winter months. The existing subrings between the main rings are not as easily explained but could be associated with daily tidal currents or even activity of the colony. Based on the alternation of growth layers, it was possible to estimate the age of young colonies, which were up to 10 years old. It was, however, difficult to interpret the peripheral rings which were very closely disposed. It was also not possible to associate the estimated age of the colony to its height or thickness because many of the examined colonies lacked their bases and their exact high could not be estimated.

The examination and evaluation of all the main morphological aspects of the colonies of *Thouarella variabilis* enables some understanding of the environment where these colonies inhabit. Their shape, branching pattern, orientation, distribution and number of polyps, spicular architecture of polyps, and seasonal deposition of skeletal material or even the nature of the substratum where the colony is attached via a holdfast have been proved to be affected

to a certain degree by the physical, chemical and biological conditions of the surrounding environment. In view of this, it is difficult to deny the influence of the environment in the direction of the selected characters, available within a range of tolerance of the organism. Therefore, the different environmental conditions cause a spectrum of variations. This environmental influence was not greatly considered in the past because of the lack of ecological surveys at that time. It is, thus, not hard to believe that many nominal species were based on these variations and that a careful review should be undertaken.

LIST OF FIGURES FOR CHAPTER 2

Fig. 18 - Polyps from the middle region of the branch, with different size and shape. Larger and more sturdy polyps bearing eggs. They show less pronounced marginal scales than the terminal polyps shown in Figs 15 and 17. Ad, adaxial side of the polyp (towards the axis); Ab, abaxial side of the polyp (outwards from the axis). 18 X.

Fig. 19 - Sequence of different sizes of polyps, from very young polyps to mature adult polyps. 8 X.

Fig. 20 - Polyp expanded by bearing reproductive products. Polyp scales placed in longitudinal adaxial rows (LR). 21 X.

Fig. 21 - Non-reproducing polyp, with polyp scales placed in diagonal abaxial rows (DR). Polyp from Figures 20 and 21 taken from the same colony. 21 X.

Fig. 22 - Body scale from a polyp of the first class size. Outer side, showing developing short spines; proximal portion smooth and distal portion showing the loose crystalline fibres (calcite) (see Figs 23 and 24). 700 X. Scale bar: 10 μm. (SP1SC1).

Fig. 23 - Detail from the scale shown in Fig. 22, showing the crystalline fibres (calcite). 3,500 X. Scale bar: 1 μm. (SP1SC1).

Fig. 24 - Detail from the scale shown in Fig. 22, showing the crystalline fibres (calcite) on a damaged part. 5,000 X. Scale bar: 10 μm. (SP1SC1).

Fig. 25 - Third size-class polyp, showing underdeveloped scales. 84 X. Scale bar: 100μm. (SP1SC1).

Fig. 26 - Body scale from a second size-class polyp. Outer side, showing short spines, slightly indented proximal portion and slightly serrated free margin. 280 X. Scale bar: 10μm. (SP1SC2).

Fig. 27 - Detail from the scale shown in Fig. 26, showing the short spines on the outer side of the scale. 1,900 X. Scale bar: 10 μm. (SP1SC2).

Fig. 28 - Detail from the scale shown in Fig. 30, showing the compound warts on the inner side of the scale. 3,200 X. Scale bar: 1 μm. (SP18SC2V).

Fig. 29 - Marginal scale from a third size-class polyp. Outer surface, showing short spines, indented proximal portion, slightly serrated free margin, and underdeveloped median thorn. 180 X. Scale bar: 100 μm. (SP1SC3).

Fig. 30 - Adaxial scale from a second size-class polyp. Inner surface, showing compound warts and smooth outline. 280 X. Scale bar: 10 μm. (SP18C2V).

Fig. 31 - Detail from the scale shown in Fig. 30, showing a compound wart on the inner side of the scale. 2,300 X. Scale bar: 10 μm. (SP18SC2V).

Fig. 32 - Detail of the calcification of the margin of the scale shown in Fig. 30. 14,000 X. Scale bar:1 μm. (SP18SC2V).

Fig. 33 - Opercular scale from a third size-class polyp. Inner side showing warts, an indented proximal portion and a median keel. 400 X. Scale bar: 10 μm. (S1C3O).

Fig. 34 - Detail from the scale shown on Fig. 33, showing the formation of warts and the slightly compacted appearance of the surface. 2,500 X. Scale bar: 10 μm. (S1C3O).

Fig. 35 - Inner side of a marginal scale showing the compacted surface and a damaged wart. 2,600 X. Scale bar: 10 μm. (S1C2M).

Fig. 36 - Detail of the margin of the scale shown in Fig. 33, showing the crystalline fibres (calcite). 4,600 X. Scale bar:1 μm. (S1C3O).

Fig. 37 - Opercular scale from a second size class polyp. Outer surface, showing a few short spines distributed around a central opening; with smooth proximal portion and free margin. 500 X. Scale bar: 10 μm. (S18C2O).

Fig. 38 - Detail of the margin of the scale shown in Fig. 37, showing the crystalline fibres. 2,800 X. Scale bar: 1 μm. (S18C2O).

Fig. 39 - Detail from the scale shown in Fig. 37, showing the crystalline fibres and the erupting short spines. 2,800 X. Scale bar: 1 μm. (S18C2O).

Fig. 40 - Detail from the scale shown in Fig. 37, showing the crystalline fibres and the erupting short spines. 3,500 X. Scale bar: 1 μm. (S18C2O).

Fig. 41 - Submarginal scale from a third size class polyp. Outer surface showing short spines, a serrated free margin, an indented base with tubercles, and a short thorn. 230X. Scale bar: 100 μm. (S1C3P).

Fig. 42 - Detail from the scale shown in Fig. 41, showing a developed short spine. 4,200X. Scale bar: 1 μm. (S1C3P).

Fig. 43 - Detail from the scale shown in Fig. 41, showing the crystalline fibres (calcite). 9,000 X. Scale bar: 1 μm. (S1C3P).

Fig. 44 - Detail of the serrated margin of the scale shown in Fig. 41, showing the crystalline fibres (calcite). 4,900 X. Scale bar: 1 μm. (S1C3P).

Fig. 45 - Crystalline fibres from a submarginal scale of a second size class polyp. 7,000X. Scale bar: 1 μm. (S18C2P).

Fig. 46 - Short spine erupting on the outer surface of a submarginal scale from a second size class polyp. 5,600 X. Scale bar: 1 μm. (S18C2P).

Fig. 47 - Wart starting to form on the inner surface of a body scale from a first size class polyp. 4,900 X. Scale bar: 1 μm. (S18C1B).

Fig. 48 - Detail of the margin of the scale shown in Fig. 41, showing the crystalline fibres (calcite). 4,900 X. Scale bar: 1 μm. (S1C3P).

Fig. 49 - Marginal scale from a third size class polyp. Inner surface, with numerous warts, a serrated free margin and a median keel. 130 X. Scale bar: 100 μm. (S18C3M).

Fig. 50 - Detail of the marginal scale shown in Fig. 49, showing the undamaged warts in the convex portion of the scale and the damaged warts more distally distributed. 420 X. Scale bar: 10 μm. (S18C3M).

Fig. 51 - Detail of the marginal scale shown in Fig. 49, showing the compound warts in the inner surface of the scale. 770 X. Scale bar: 10 μm. (S18C3M).

Fig. 52 - Detail of the marginal scale shown in Fig. 49, showing the damaged warts distally distributed on the inner surface of the scale. 1,300 X. Scale bar: 10 μm. (S18C3M).

Fig. 53 - Detail of the marginal scale shown in Fig. 49, showing the warts on the inner surface of the scale. 2,100 X. Scale bar: 10 μm. (S18C3M).

Fig. 54 - Detail of the short spines and calcification of the outer surface of a submarginal scale from a third size class polyp. 1,300 X. Scale bar: 10 μm. (S18C3P).

Fig. 55 - Crystalline fibres from a marginal scale of a second size class polyp. 2,800 X. Scale bar: 1 μm. (S18C2M).

Fig. 56 - Detail of the short spines and calcification of the outer surface of a body scale from a third size class polyp. 2,600 X. Scale bar: 10 μm. (S1C2B).

Fig. 57 - Adaxial side of a fourth size class polyp, showing the arrangement of opercular (Op), marginal (Ms) and body scales (Bs). Adaxial scales (Ad) are covered by the lateral body scales. 35 X. Scale bar: 100 μm. (S1C4I).

Fig. 58 - Detail of the arrangement of the opercular (Os) and marginal scales. External ring of marginal scales, Me; internal ring of marginal scales, Mi. 70 X. Scale bar: 100 μm. (S1C4I).

Fig. 59 - Side view of the polyp shown in Fig. 57, showing the abaxial body scales (arrows) and lateral body scales. 100 X. Scale bar: 100 μm. (S1C4I).

Fig. 60 - Inner side of two body scales, showing the warts and the indented margins. 130 X. Scale bar: 100 μm. (S18C3B).

Fig. 61 - Terminal portion of a marginal thorn from a fourth size class polyp. 630 X. Scale bar: 10 μm. (S18C4MV).

Fig. 62 - Proximal portion of the thorn shown in Fig. 61, with serrated margins and a series of short spines in the middle portion. 420 X. Scale bar: 10 μm. (S18C4M).

Fig. 63 - Proximal portion of the thorn shown in Fig. 61, with serrated margins and two series of short spines in the middle portion. 1,100 X. Scale bar: 10 μm. (S18C4M).

Fig. 64 - Serrated margin of a marginal scale from a fourth size class polyp. 1,200 X. Scale bar: 10 μm. (S18C4M).

Fig. 65 - Abaxial side of an adaxially incurved young polyp, showing the underdeveloped polyp scales. 120 X.

Fig. 66 - Abaxial side of a mature polyp, showing the arrangement of body scales and well developed marginal scales. 100 X.

Fig. 67 - Side view of a mature polyp adaxially incurved towards the axis, showing the arrangement of basal and lateral body scales. 100 X.

Fig. 68 - Arrangement of polyps on the branch, coincidently in pairs. Detail of their inclination towards the axis. 75 X.

Fig. 69 - Arrangement of the body scales, showing the way they fit in with the coenenchyme scales. 75 X.

Fig. 70 - Arrangement of coenenchyme scales on the branch. 100 X.

Fig. 71 - Coenenchyme scales overlapping one another, with serrated free margins and radially arranged ridges. 320 X.

Fig. 72 - Arrangement of body scales showing the way they overlap one another. 430X.

Fig. 73 - Inset - first size class polyp, showing the arrangement of underdeveloped scales. a - g, polyp scales with positions not defined; free margins slightly serrated with crystalline fibres easily recognized and smooth proximal portion. Scale bar: 20 µm. (SP1SC1).

Fig. 74 - Set of scales from a second size class polyp. a and b, opercular scales; c-e, marginal scales; f and g, body scales. Scale bar: 50 µm. (S18C1O).

Fig. 75 - Set of scales from a third size class polyp. a and c, opercular scales, outer side; b, opercular scale, inner side; d, marginal scale, inner side; e, marginal scale, outer side; f-h, submarginal scales, outer side; i and k, body scales, outer side; j and l, body scales, inner side. Scale bar: 50 µm. (S1C3).

Fig. 76 - Set of scales from a third size class polyp. a, opercular scales, inner side; b, opercular scale, outer side; c, marginal scale, inner side; f, marginal scale, outer side; d, body scales, inner side; e, adaxial scale outer side. Scale bar: 50 µm. (S18C3).

Fig. 77 - Set of scales from a third size class polyp. a, opercular scales, inner side; b, marginal scale, outer side; c, marginal scale, inner side; d, body scale, inner side; e, body scale, outer side; f, adaxial scale inner side. Scale bar: 50 µm. (S1C3).

Fig. 78 - Set of scales from a fourth size class polyp. a (missing letter) and d, marginal scales, inner side; b and c, marginal scale, outer side; e, f, h and j, submarginal scales, outer side; g and i, submarginal scales, inner side; k, adaxial scale, inner side. Scale bar: 100 µm. (S18C4).

Fig. 79 - Set of scales from a fifth size class polyp. a, opercular scale, inner side; b, opercular scale, outer side; c, d, and f, marginal scales, outer side; e, marginal scale, inner side; g, submarginal scales, inner side; h, submarginal scale, outer side; i, body scale, inner side; j and l, body scales, outer side; k, adaxial scale, outer side. Scale bar: 100 µm.

Figs 80-89 are cross sections from young colonies.

Fig. 80 - Cross section of main stem showing the growth layers. 60 X. Scale bar: 100µm. (Th7)

Fig. 81 - Detail of the cross section shown in Figure 80 showing the growth layers (arrows) and the central core (c). 120 X. (Th7)

Fig. 82 - Cross section of main stem showing the growth layers. 80 X. Scale bar: 100µm. (Th12)

Fig. 83 - Detail of the cross section shown in Figure 82 showing the dark outer layer. 180 X. (Th12)

Fig. 84 - Cross section of main stem showing a large first growth layer. 30 X. Scale bar: 100µm. (Asc88s)

Fig. 85 - Cross section of main stem showing the growth layers. 35 X. Scale bar: 100µm. (Th2)

Fig. 86 - Cross section of main stem showing a large first growth layer. 50 X. Scale bar: 100µm. (Th3)

Fig. 87 - Detail of the cross section shown in Figure 86 showing the large first growth layer. 180 X. Scale bar: 100µm. (Th3)

Fig. 88 - Cross section of main stem (s) and branch (b) showing the growth layers. 50 X. Scale bar: 100µm. (Th18)

Fig. 89 - Detail of the cross section of the main stem shown in Figure 88 showing a considerably large outer layer impregnated with calcareous elements. 16 X. Scale bar: 1mm. (Asc90s)

Figures 90-95 are cross sections from older colonies.

Fig. 90 - 40 X. Scale bar: 100µm. (Thx)

Fig. 91 - 140 X. (Thx)

Fig. 92 - 40 X. Scale bar: 100µm. (Th6)

Fig. 93 - 80 X. Scale bar: 100µm. (Th6)

Fig. 94 - 70 X. Scale bar: 100µm. (Th9)

Fig. 95 - 130 X. Scale bar: 100µm. (Th9)

Fig. 96 - Growth layers (arrow) and surface of the main stem. 110 X.

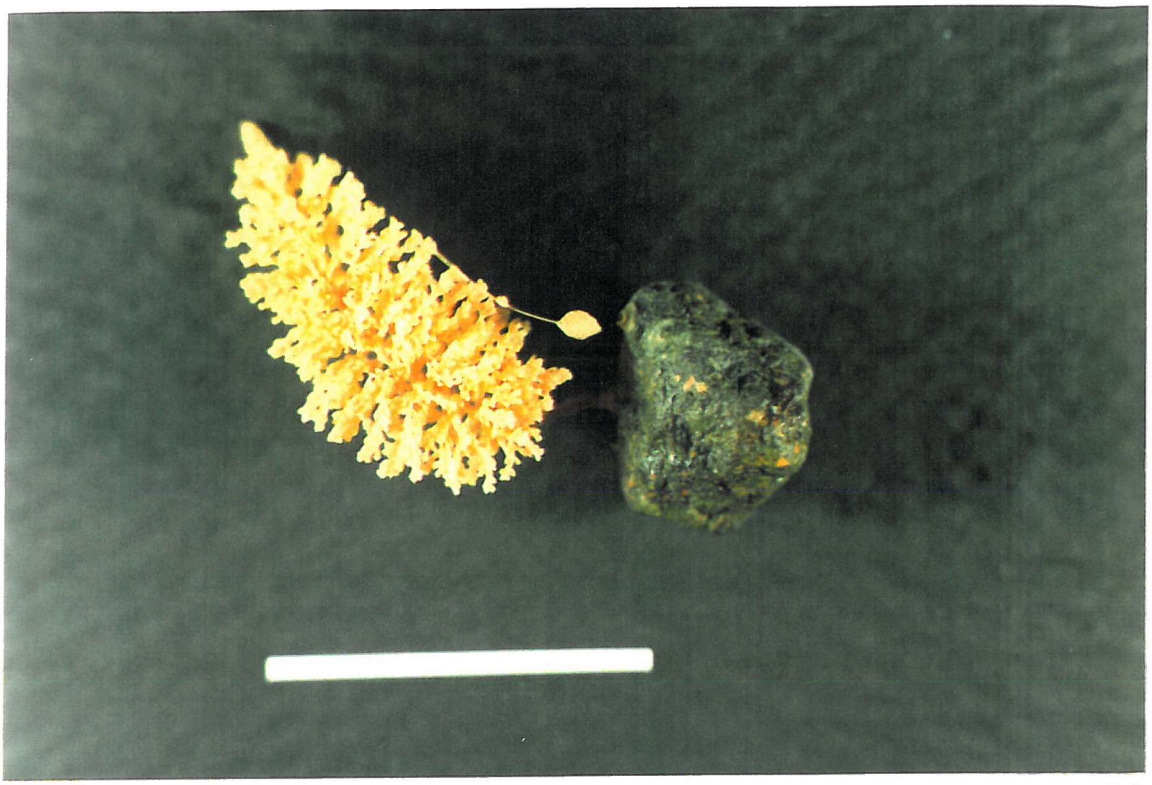
Fig. 97 - Surface of the main stem. 750 X.

Fig. 98 - Detail of the grooved surface of the main stem. 2,100 X.

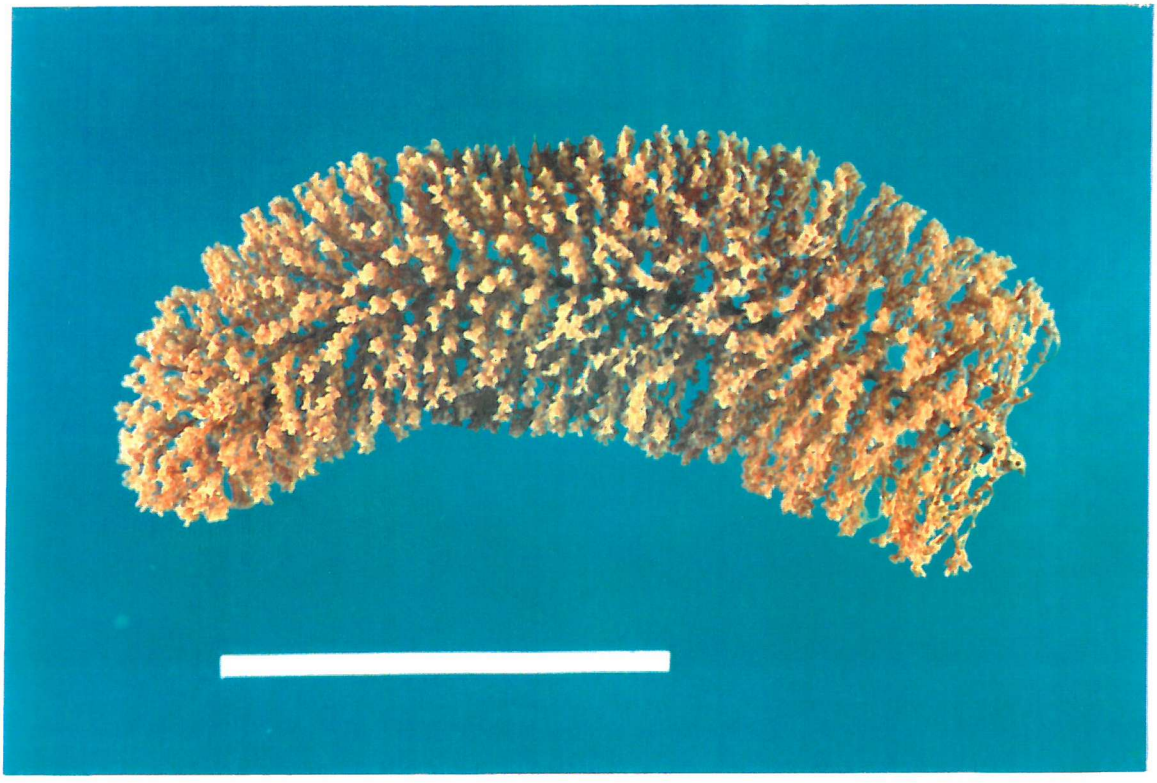
Fig. 99 - Cross section of the main stem showing thin compacted and larger non-compacted growth layers. 200 X.

Fig. 100 - Cross section of the main stem showing thin compacted and larger non-compacted (arrow) growth layers.

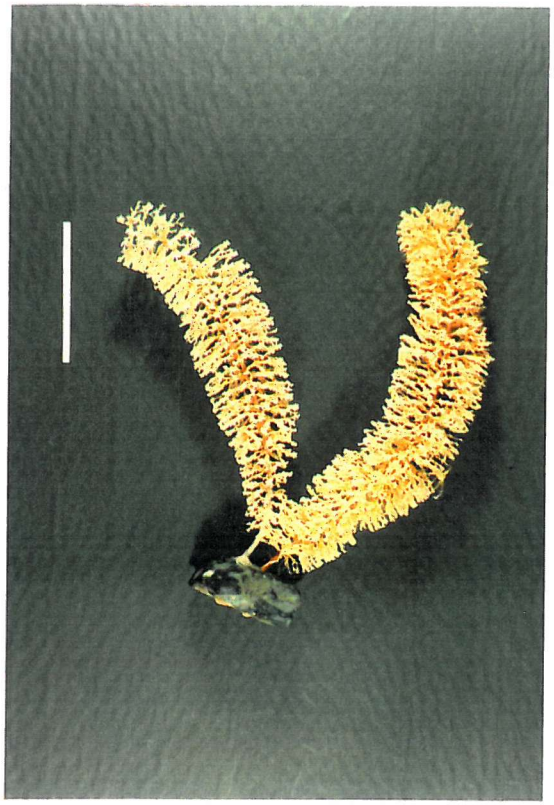
Figures 101-103 - Cross sections of the main stem showing the growth layers. 101: 100 X, (Th32), 102: 130 X, (Th18) and 103: 90 X, (Th8).



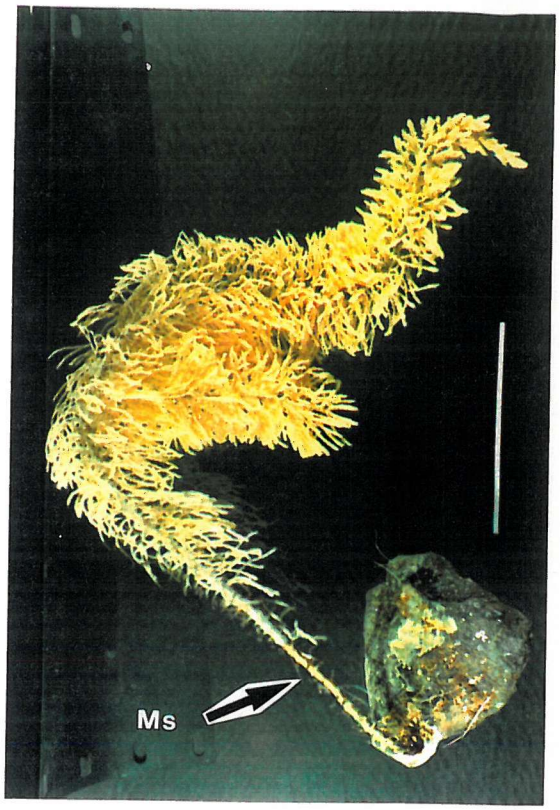
2



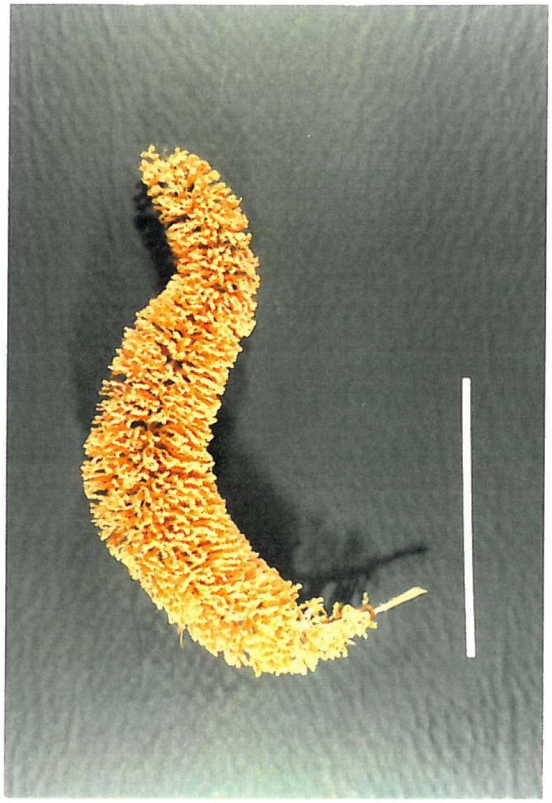
1



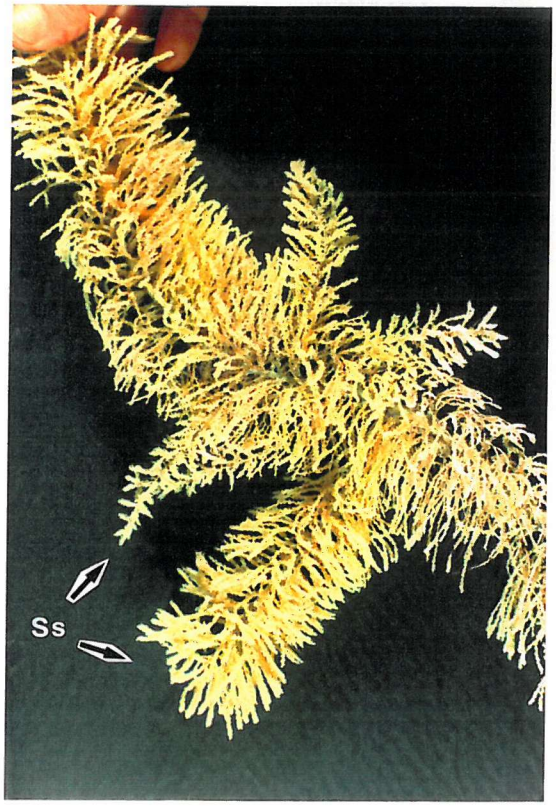
3



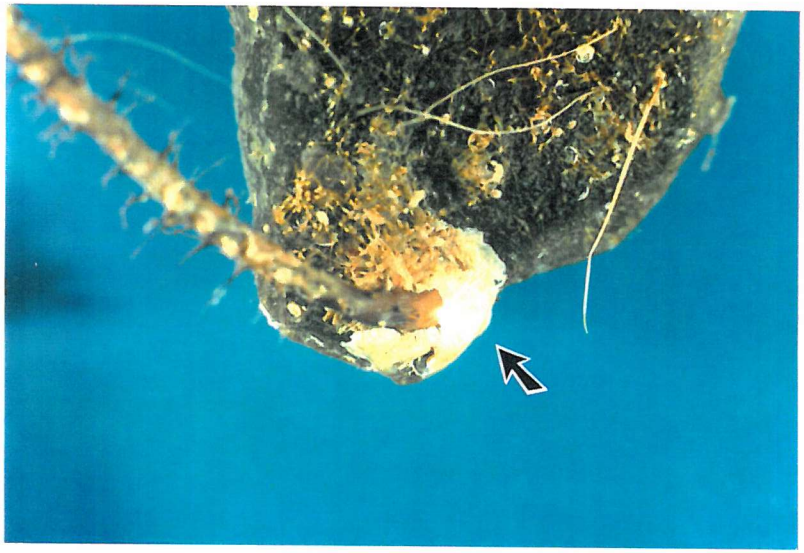
4



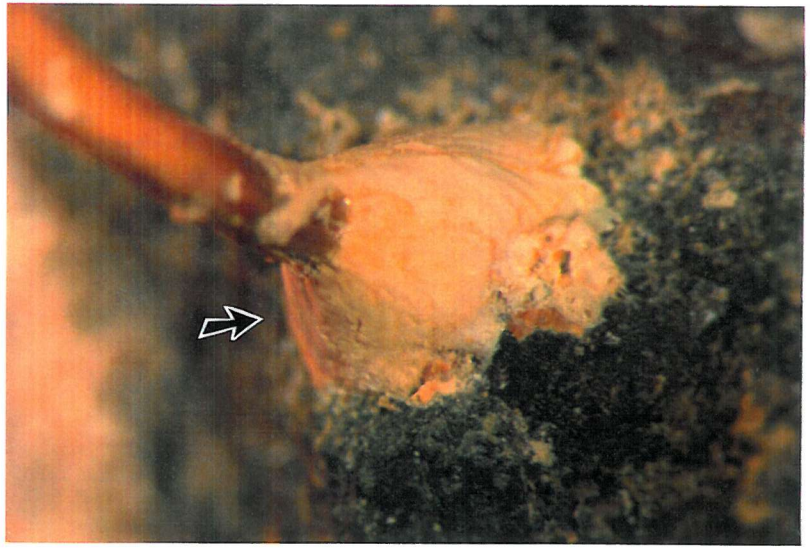
5



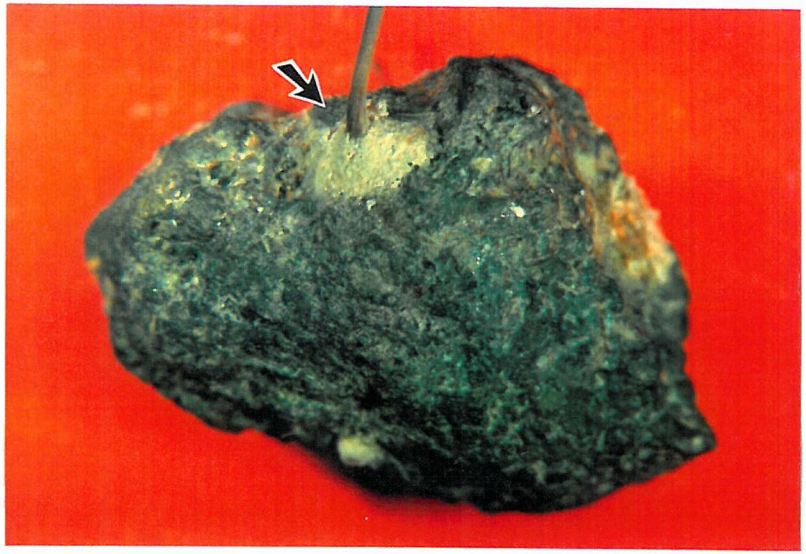
6



7

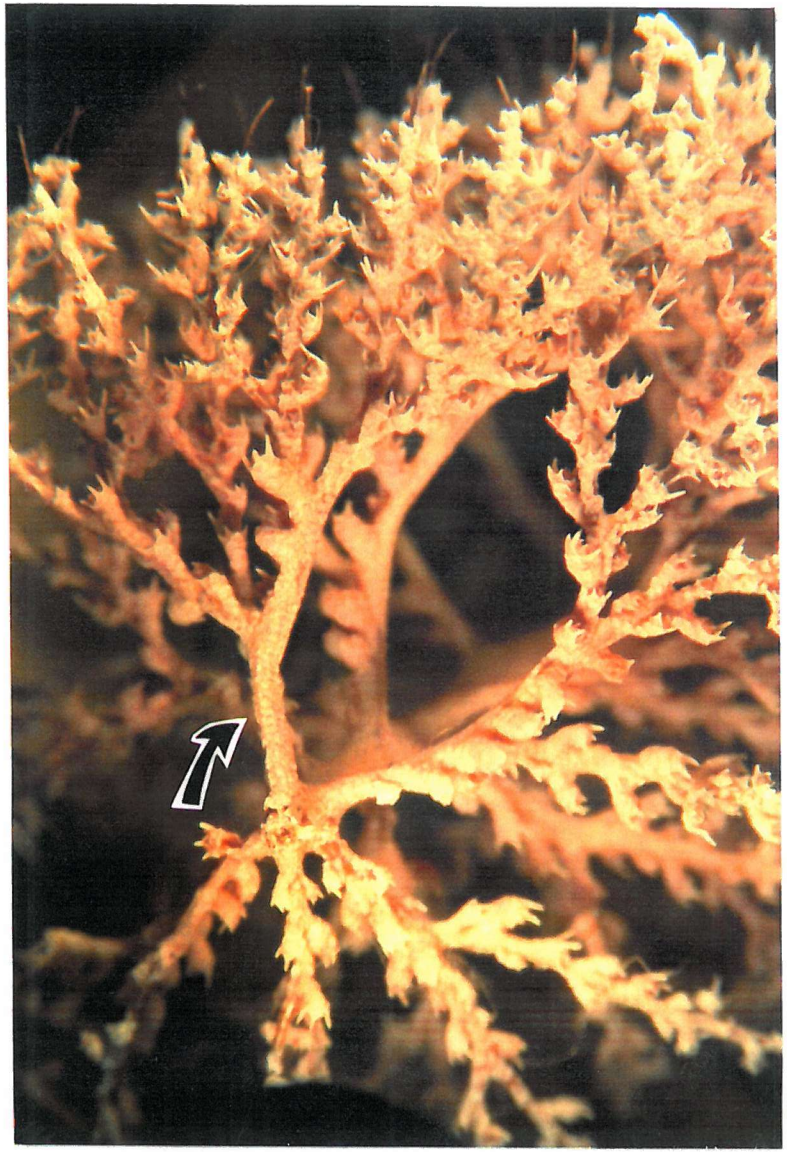


8



9

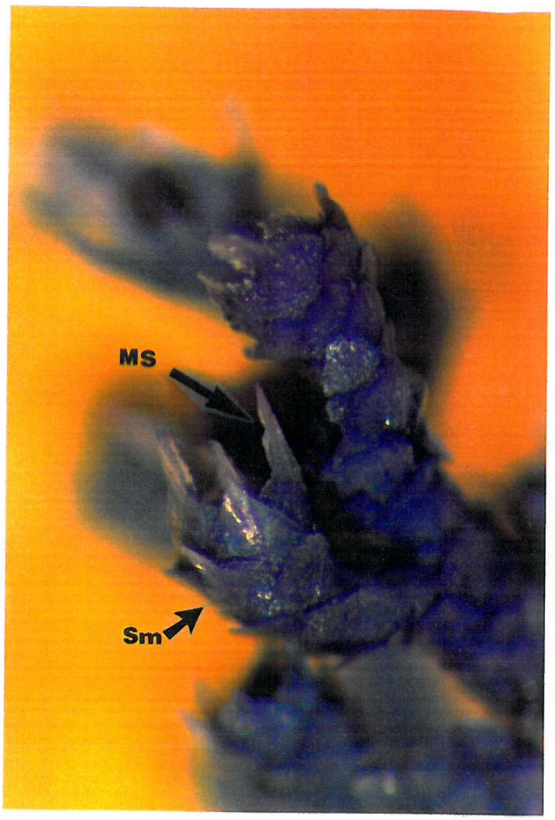
10



11

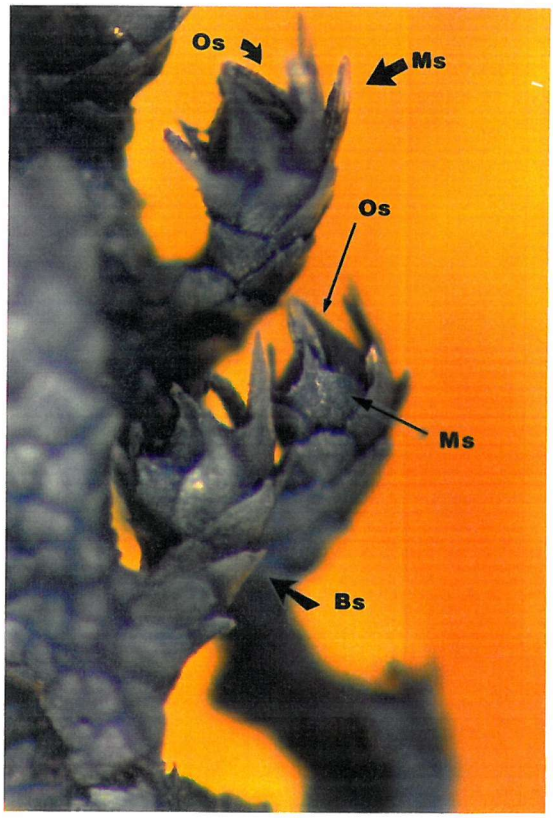
12

14

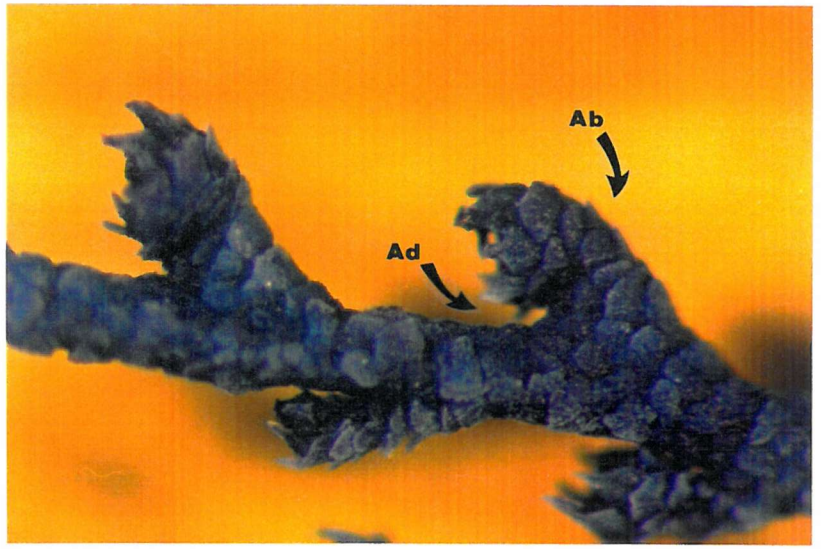


15

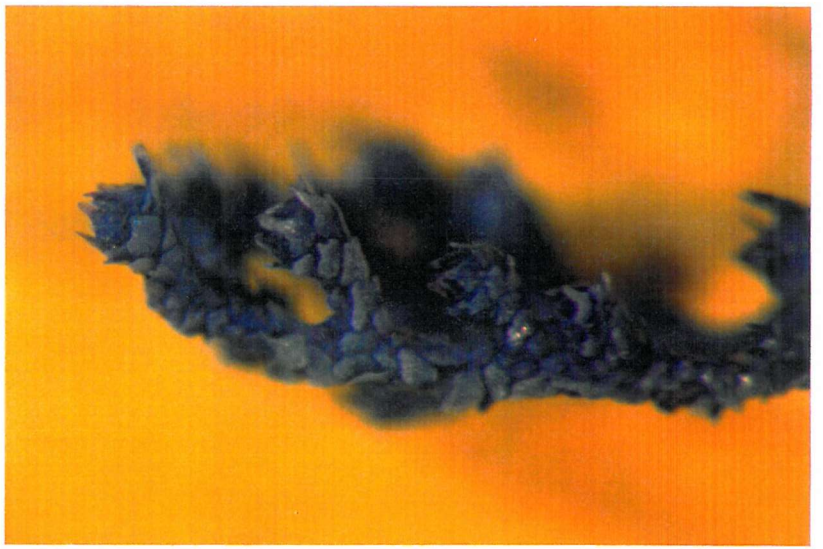
16



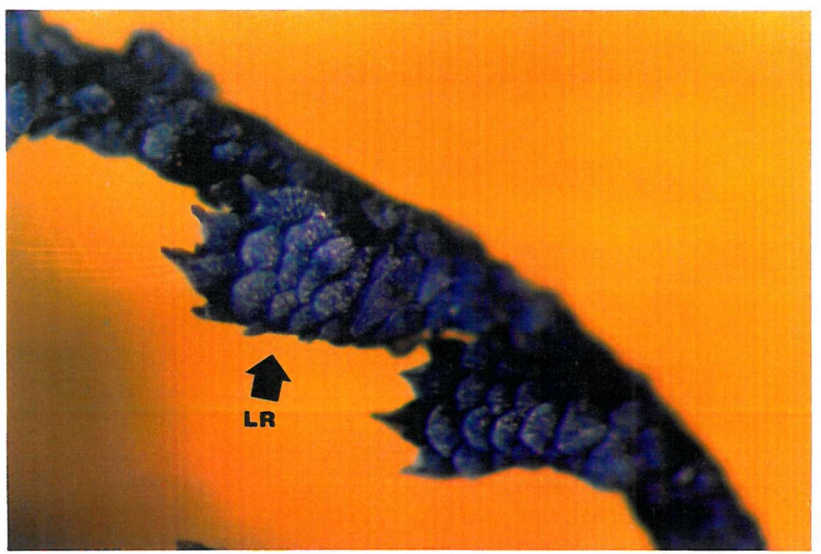
17



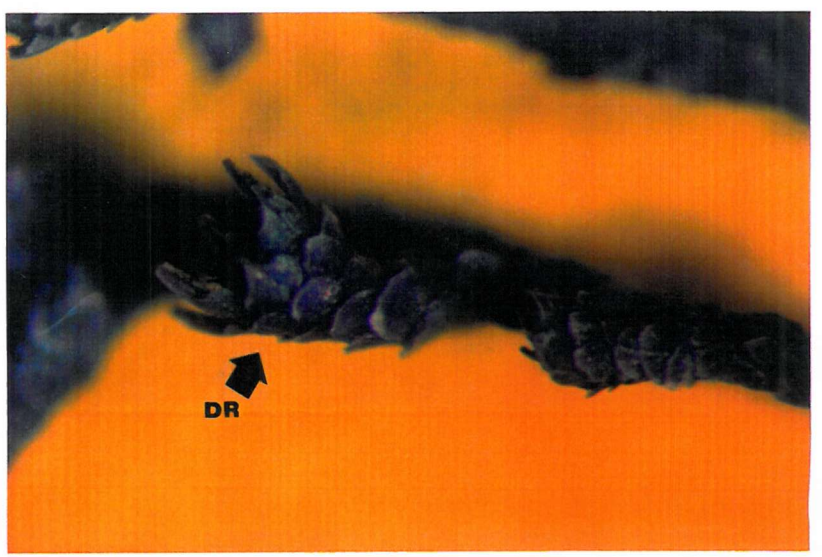
18



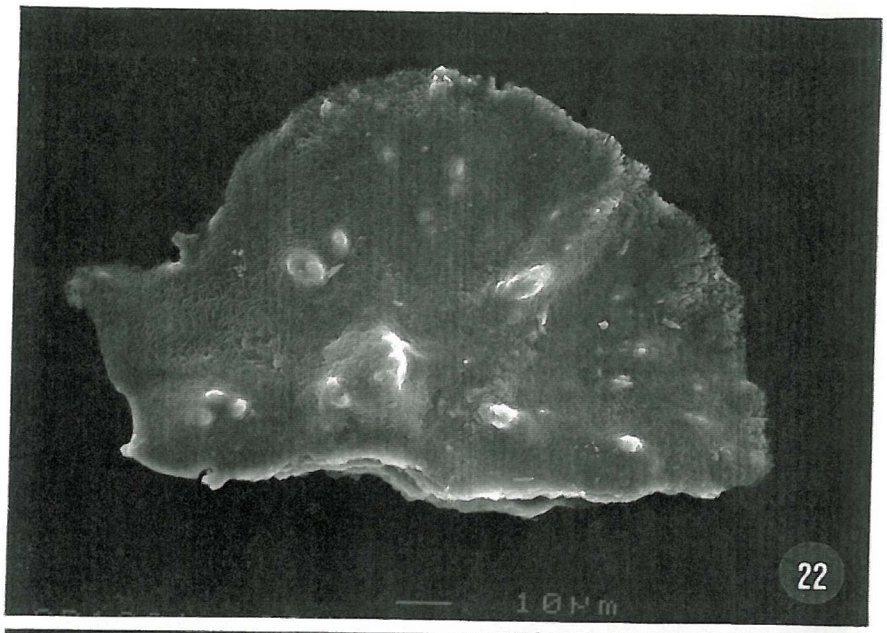
19

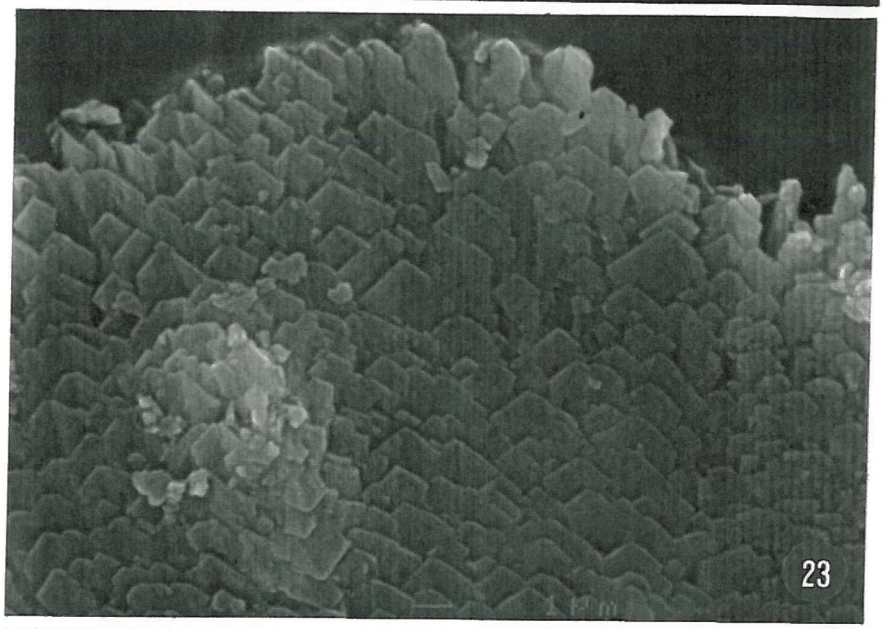


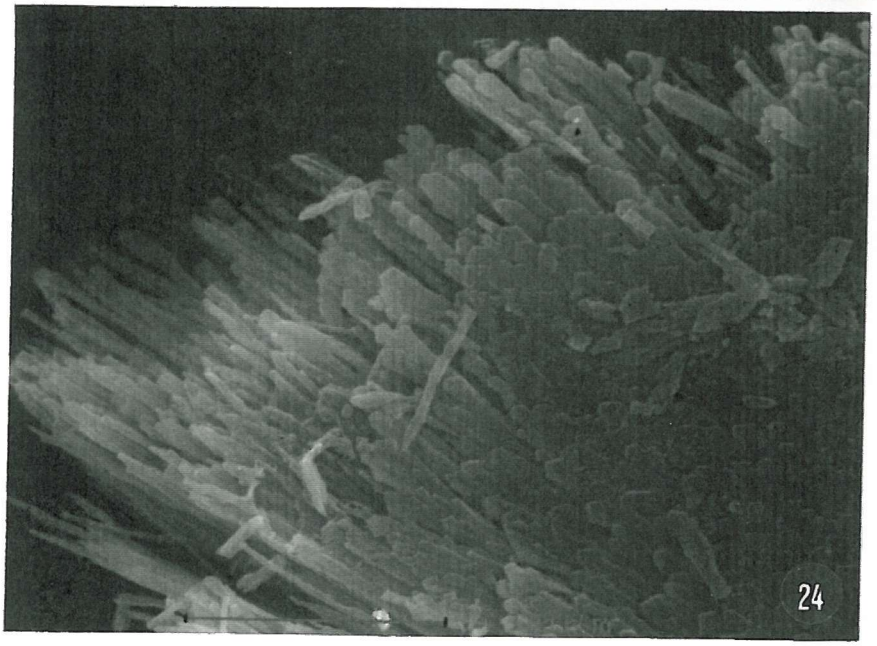
20

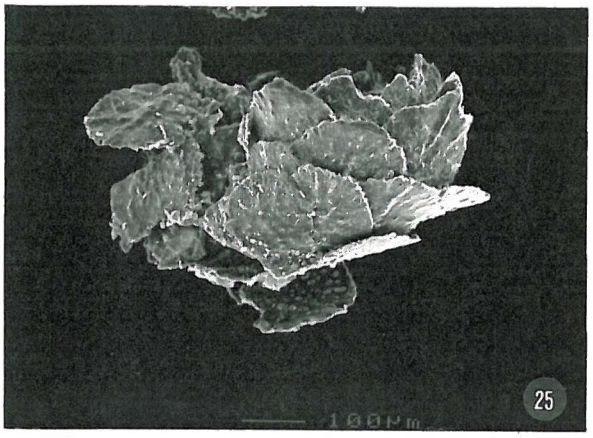


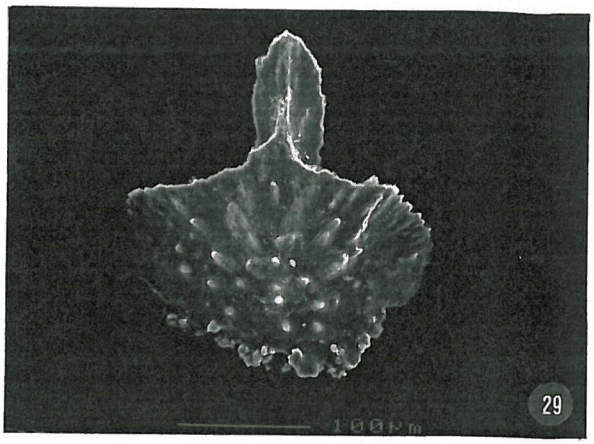
21

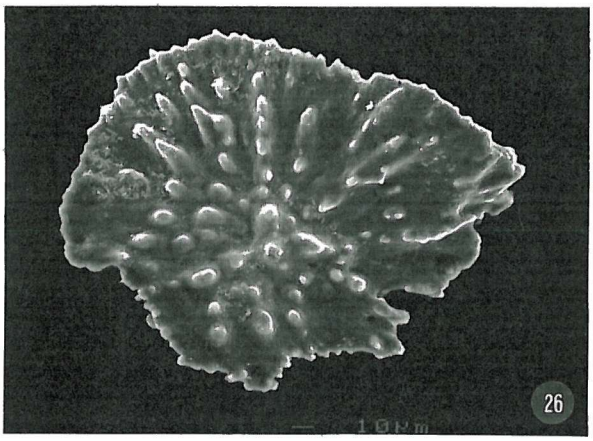

10μm
22

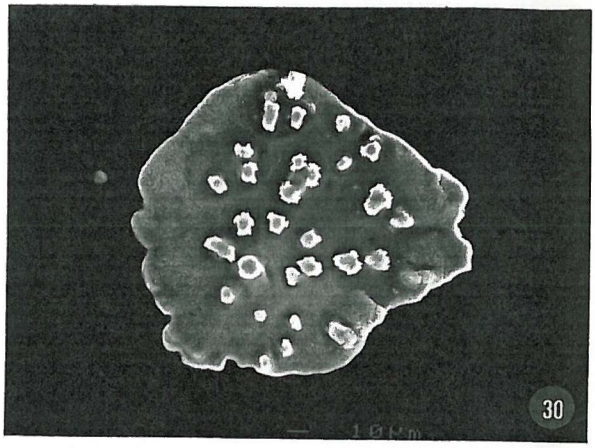

23

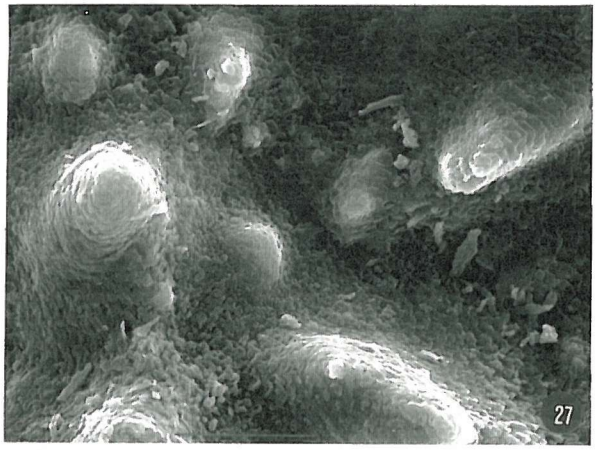

24

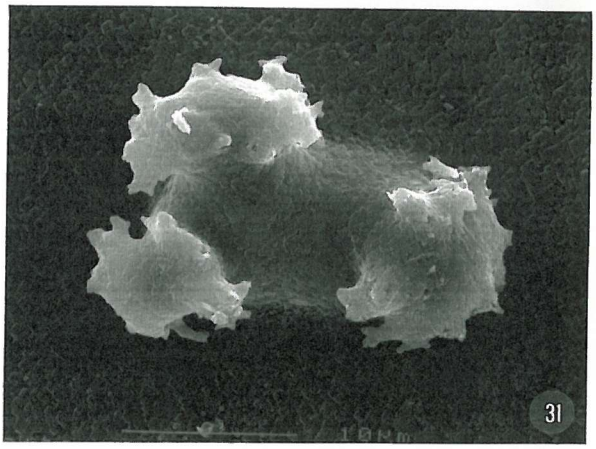

100µm
25

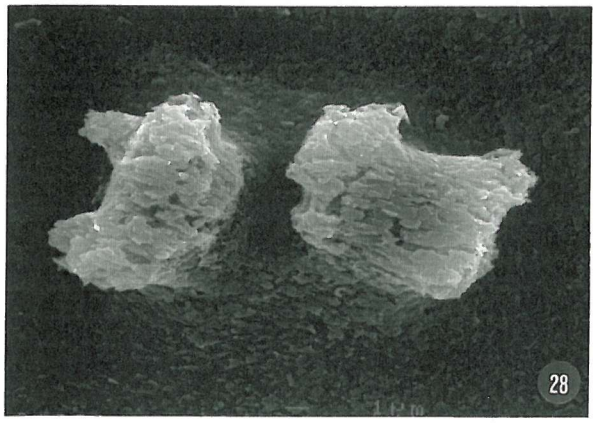

100µm
29

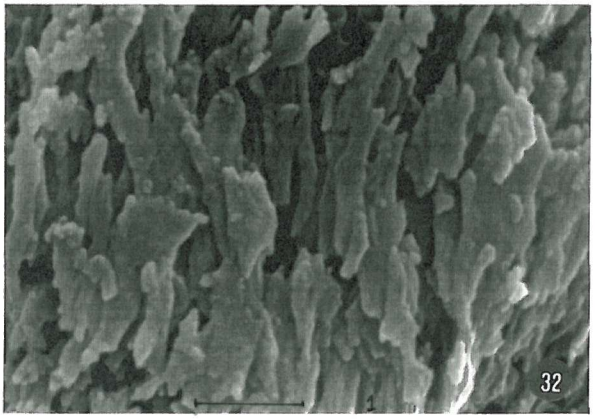

10µm
26

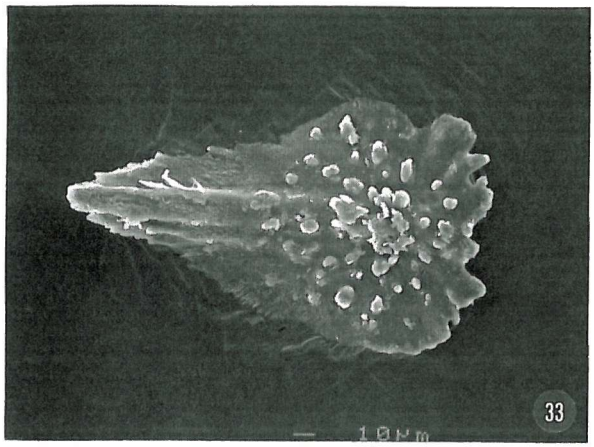

10µm
30

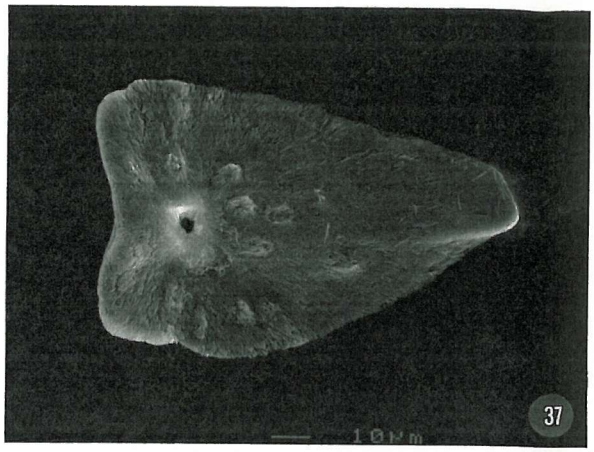

27

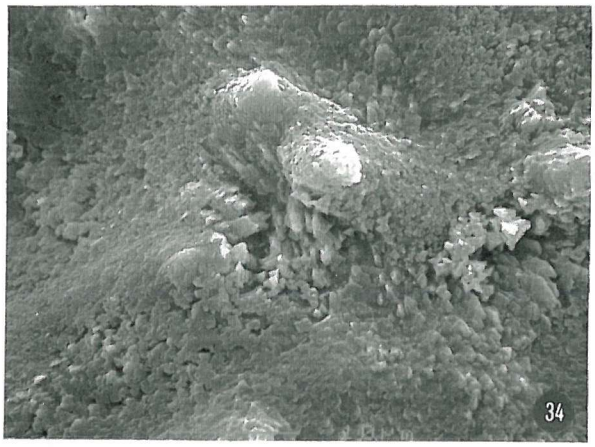

10µm
31

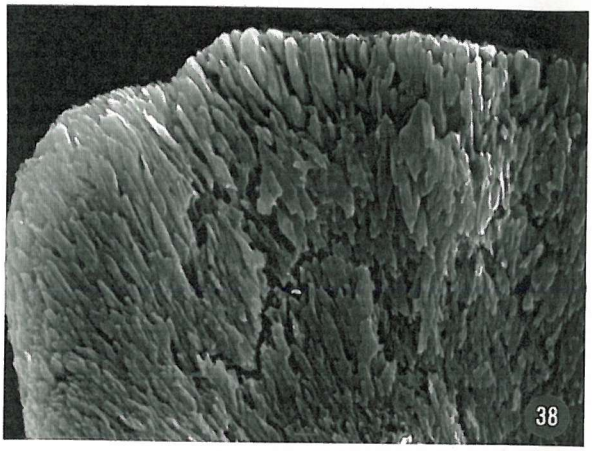

10µm
28

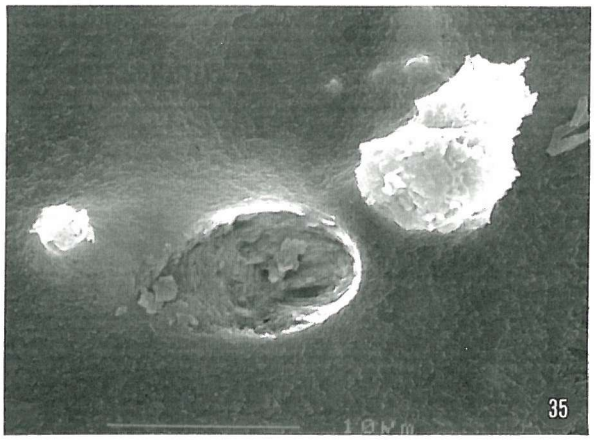

1µm
32

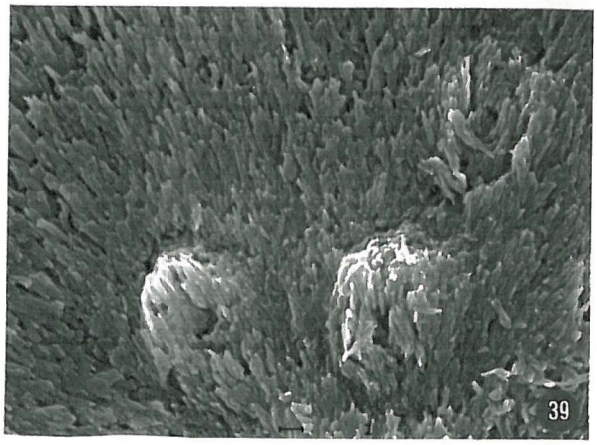

10μm
33

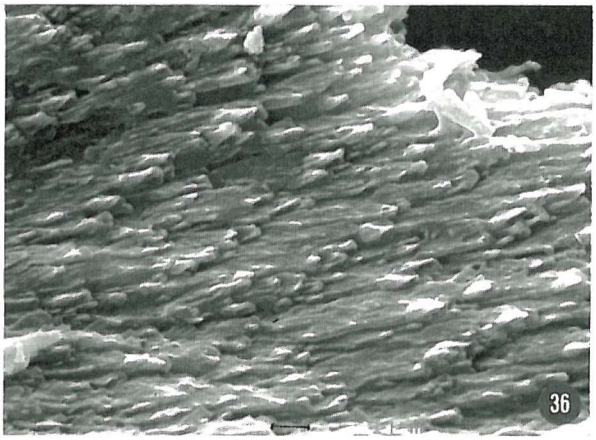

10μm
37

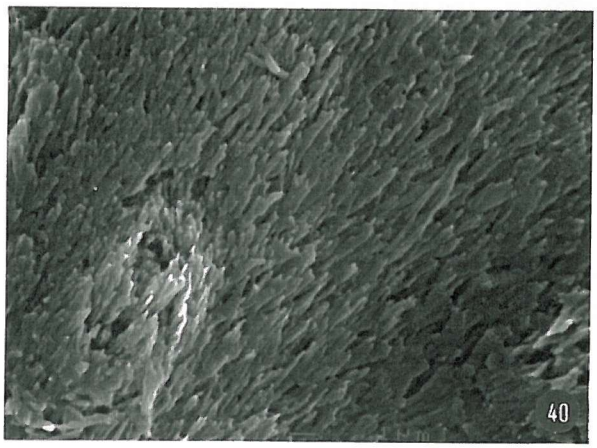

34

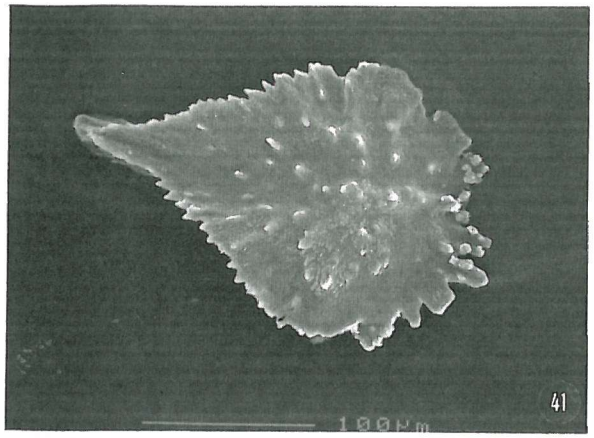

38

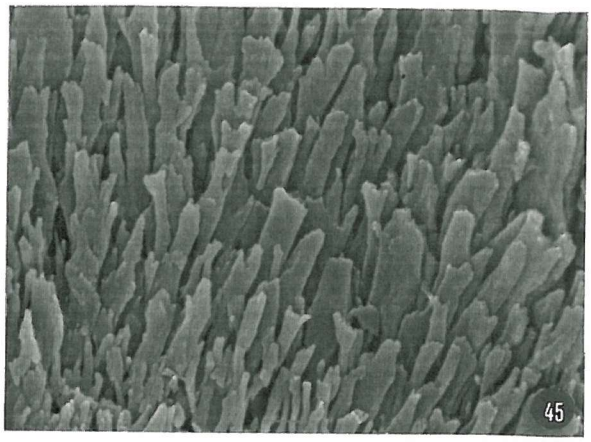

10μm
35

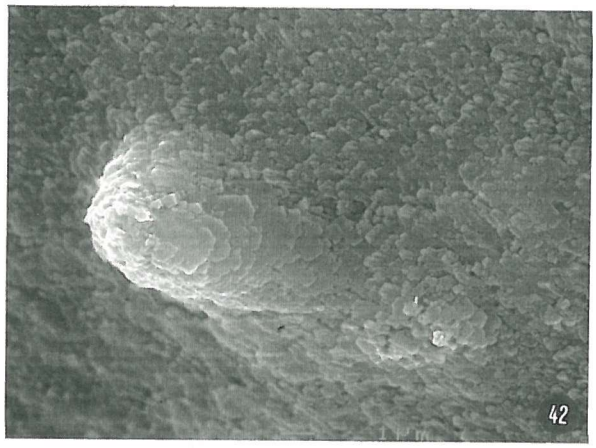

39

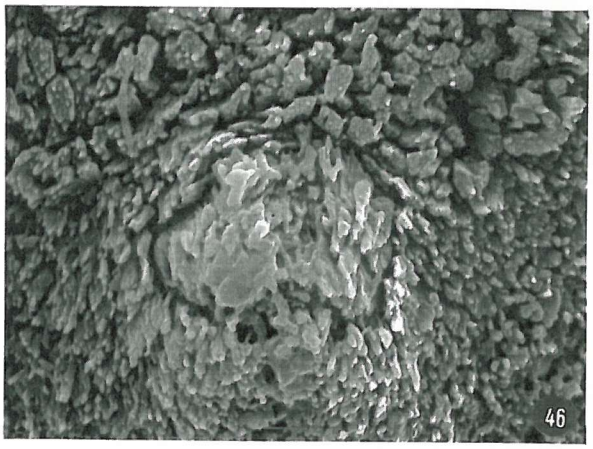

36

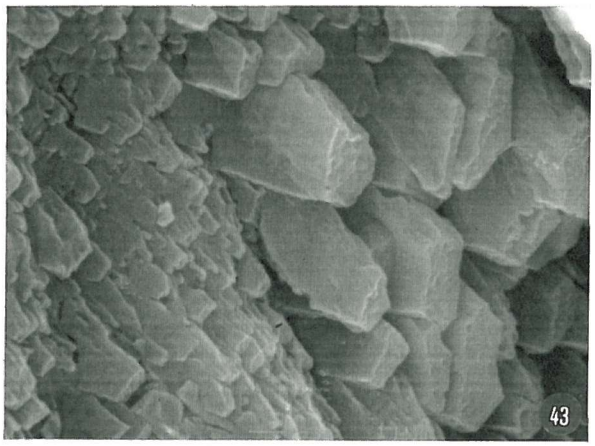

40

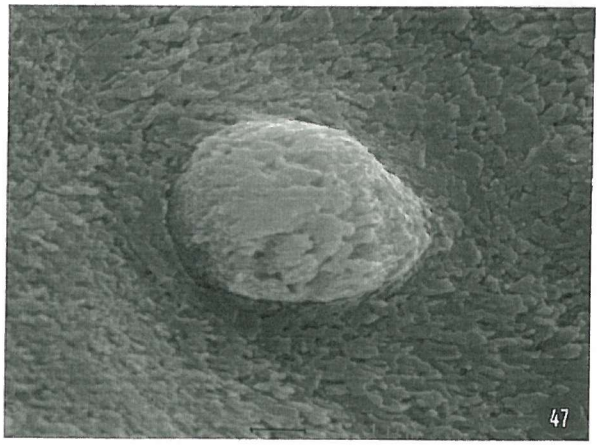

100µm
41

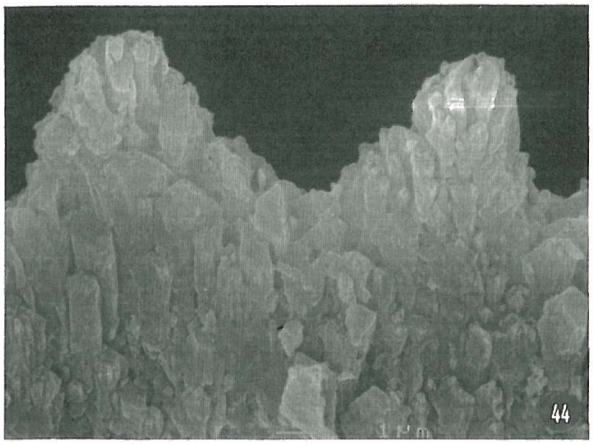

45

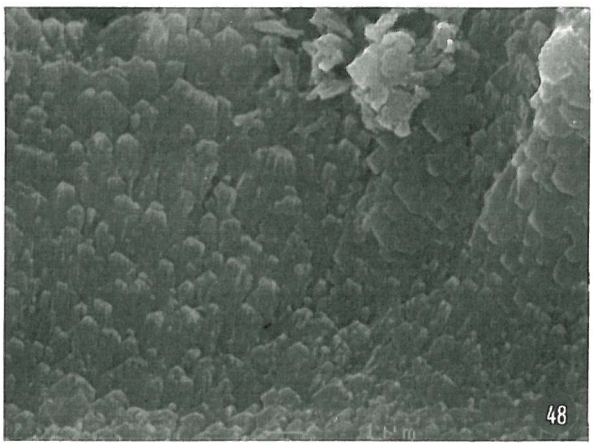

42

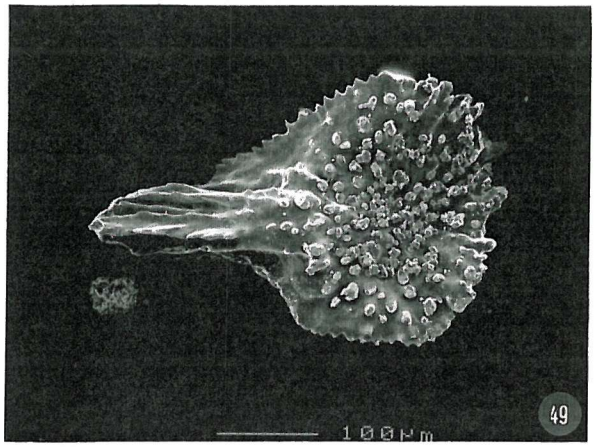

46

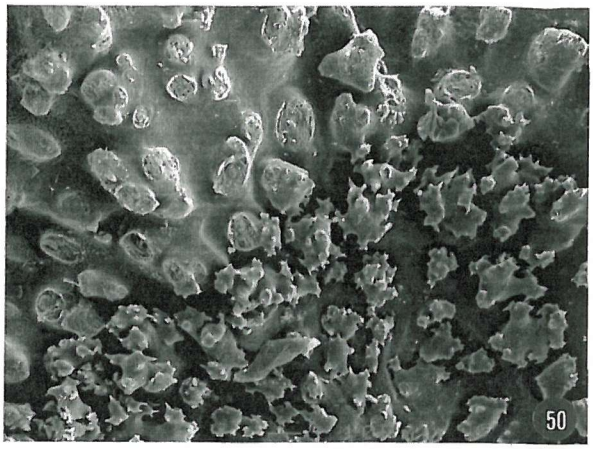

43

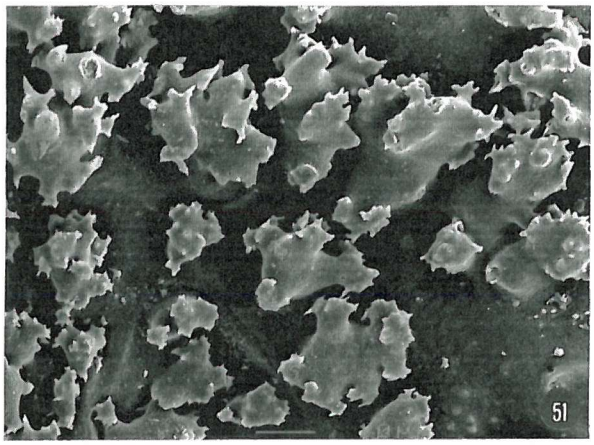

47

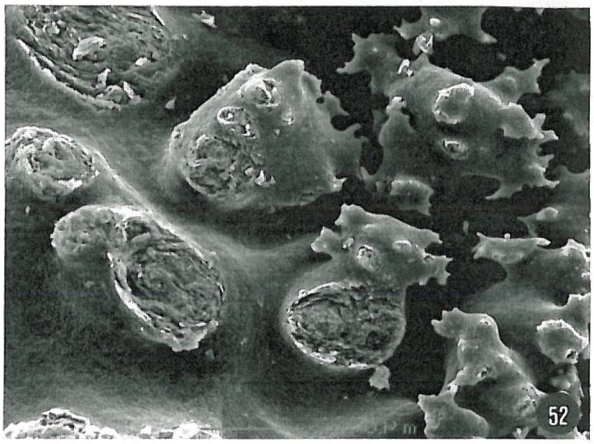

44

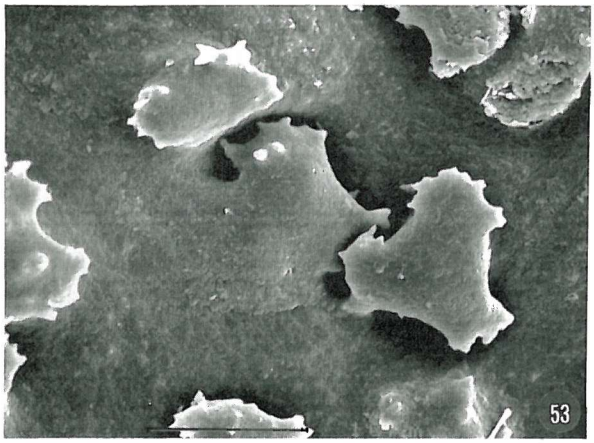

48

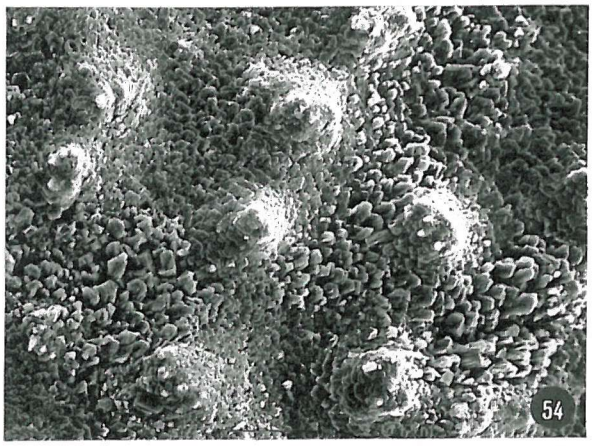

100µm
49

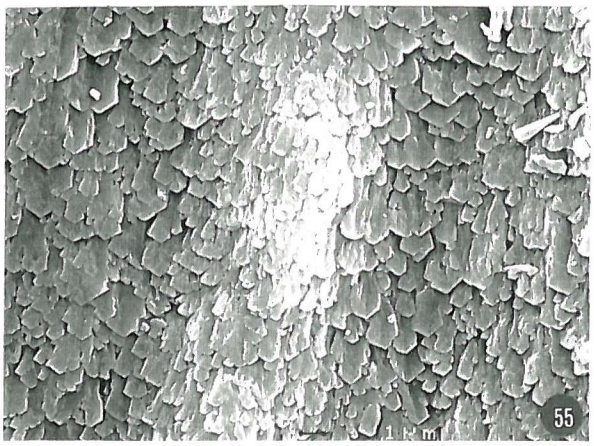

50

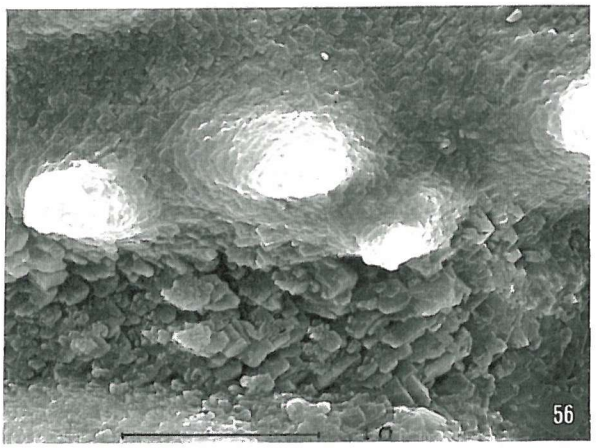

51

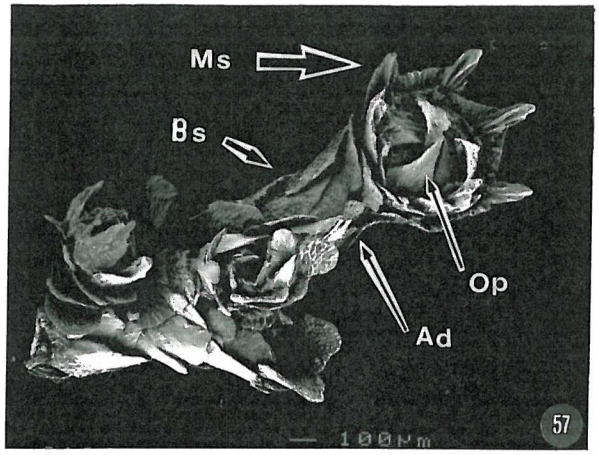

52

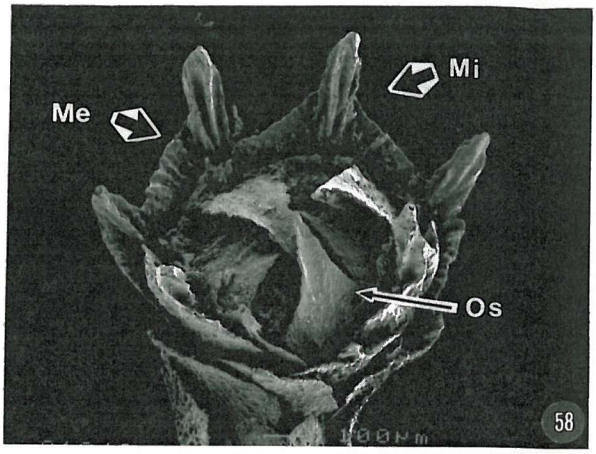

53

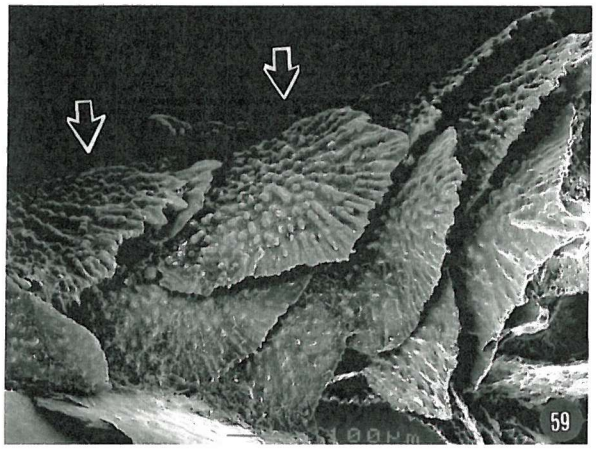

54

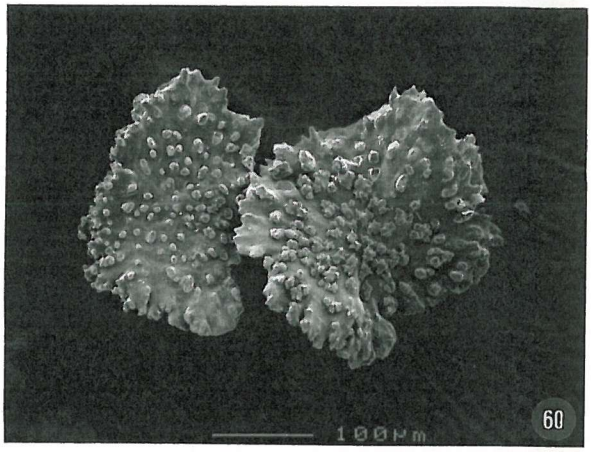

55

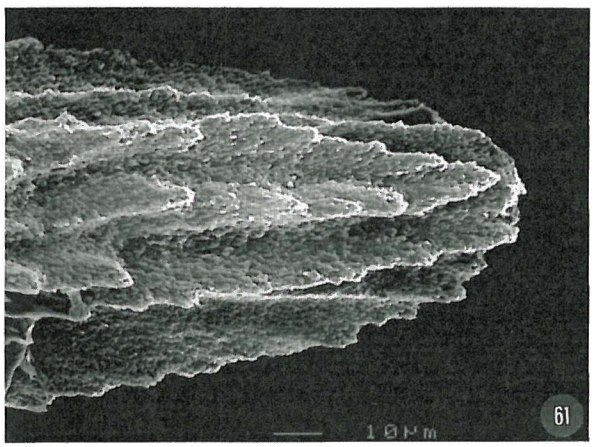

56

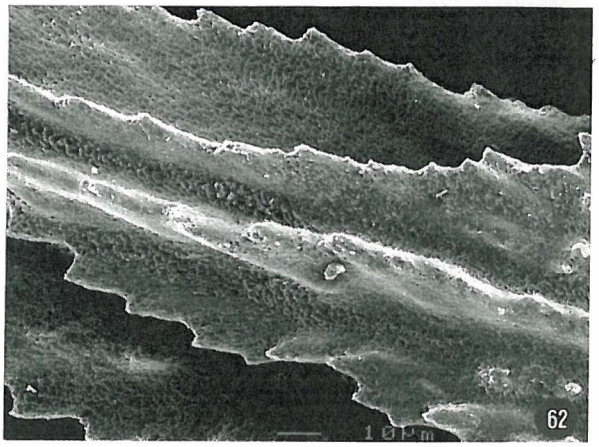

Ms
Bs
Op
Ad
100μm
57

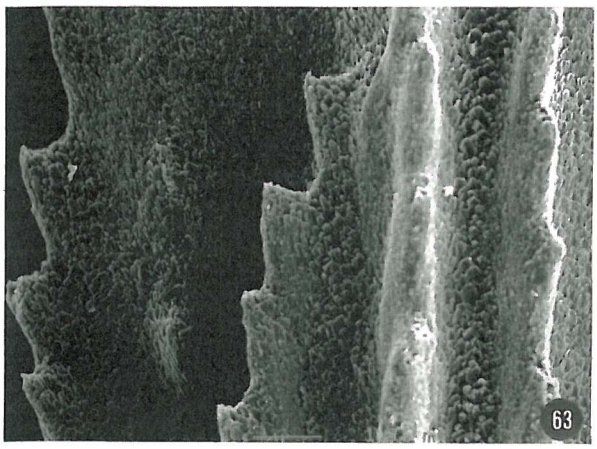

Me
Mi
Os
100μm
58

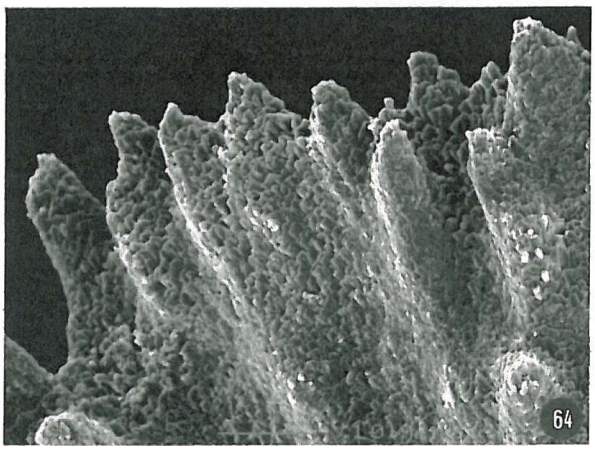

100μm
59

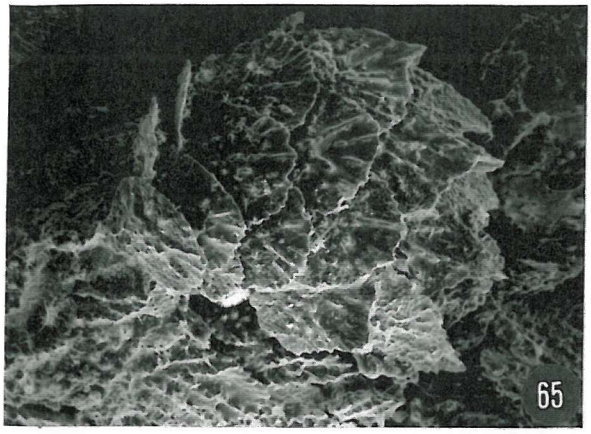

100μm
60

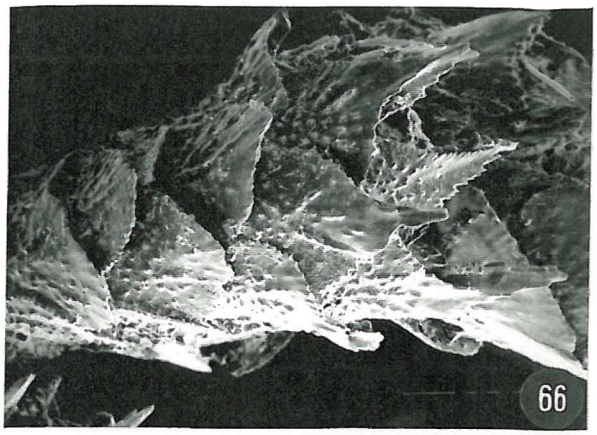

10μm
61

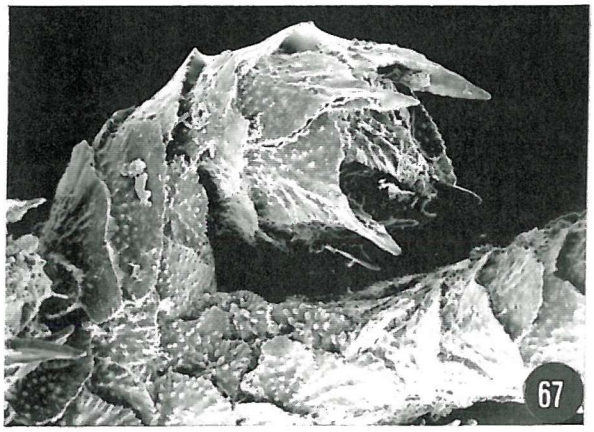

10μm
62

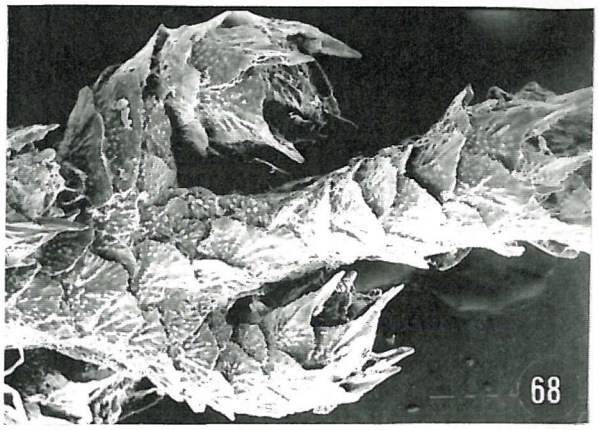

63

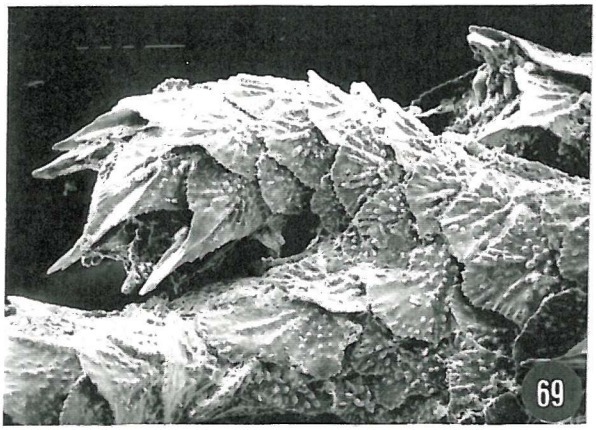

64

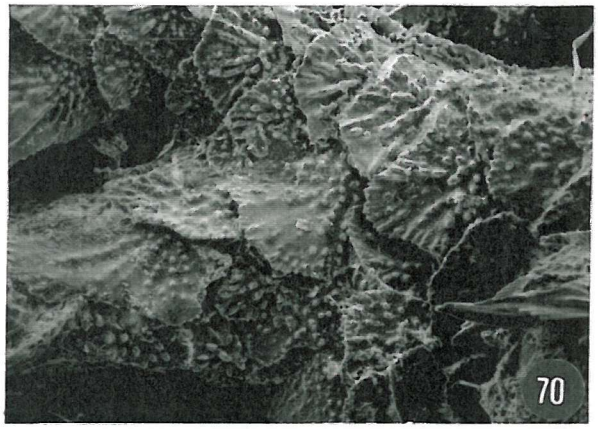

65

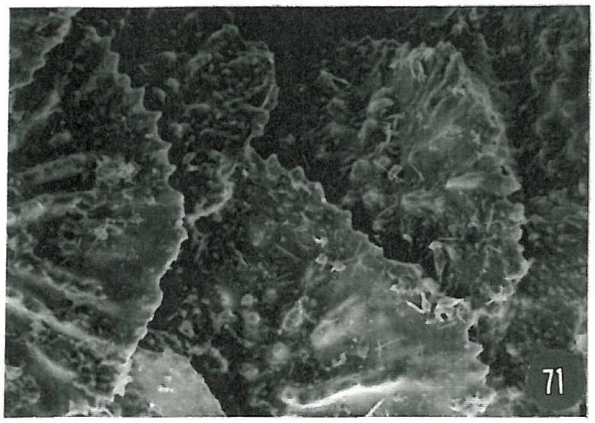

66

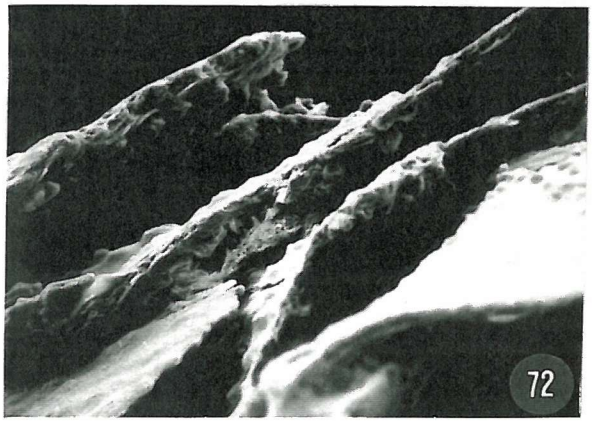

67

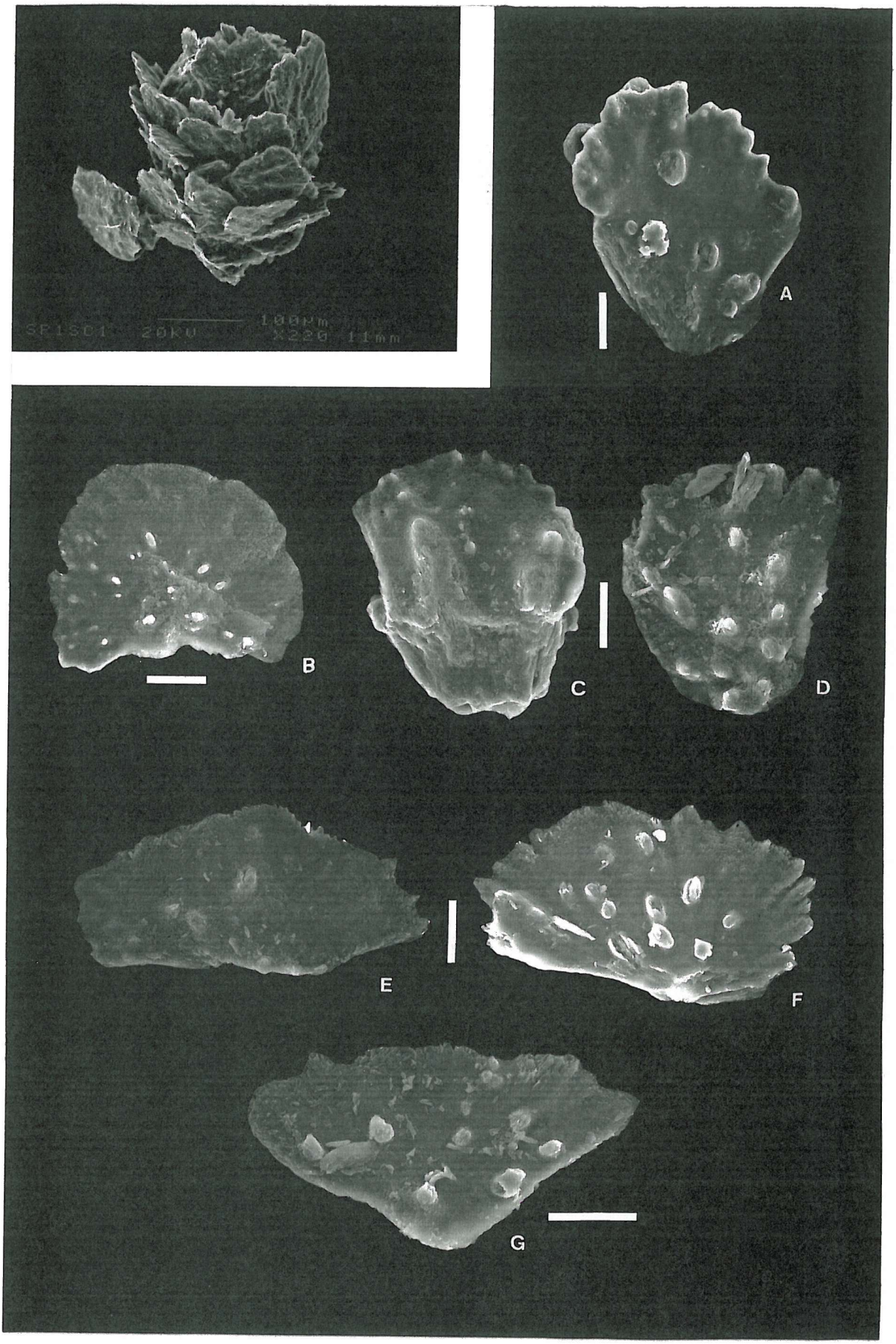

68

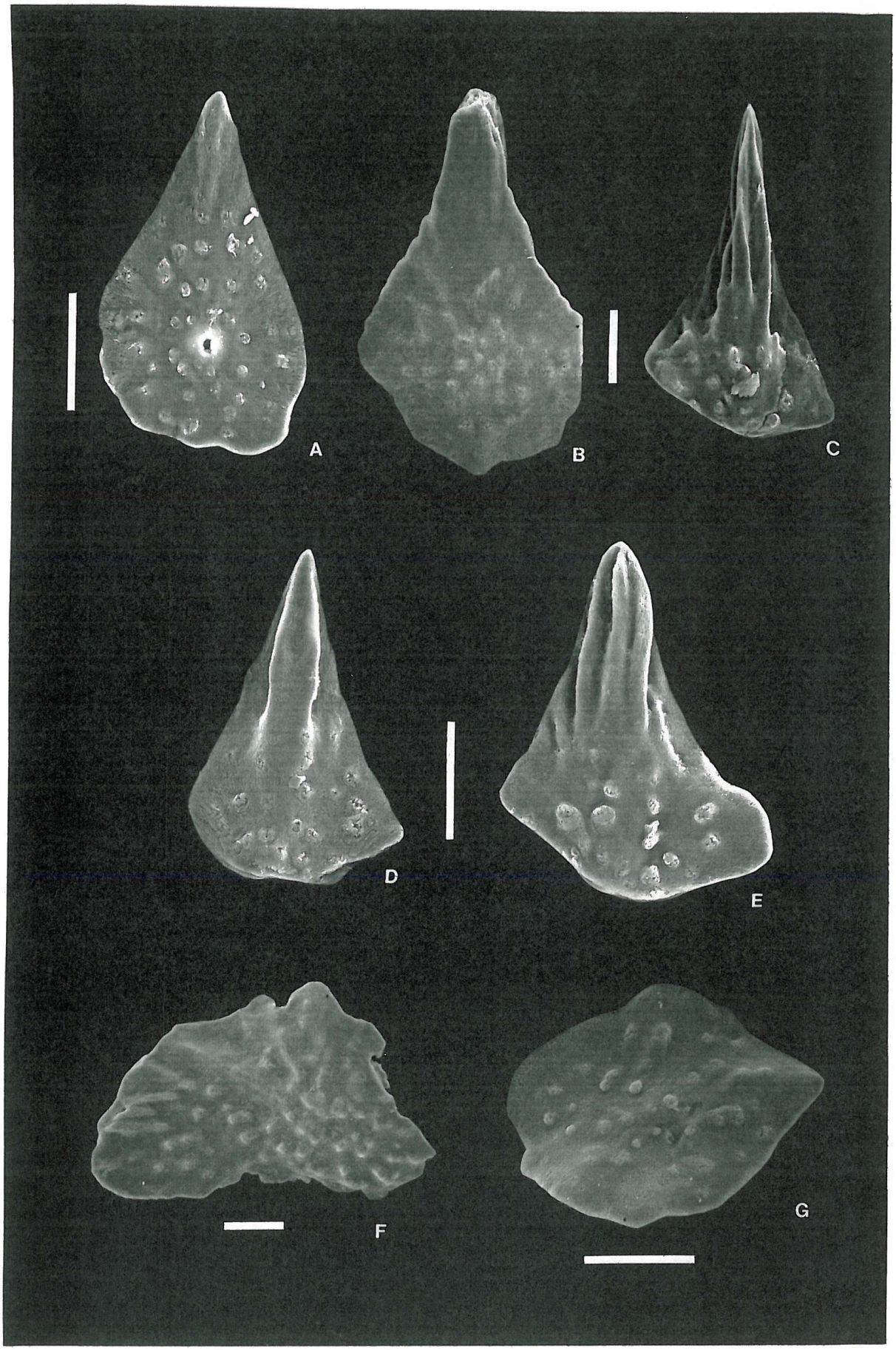

69

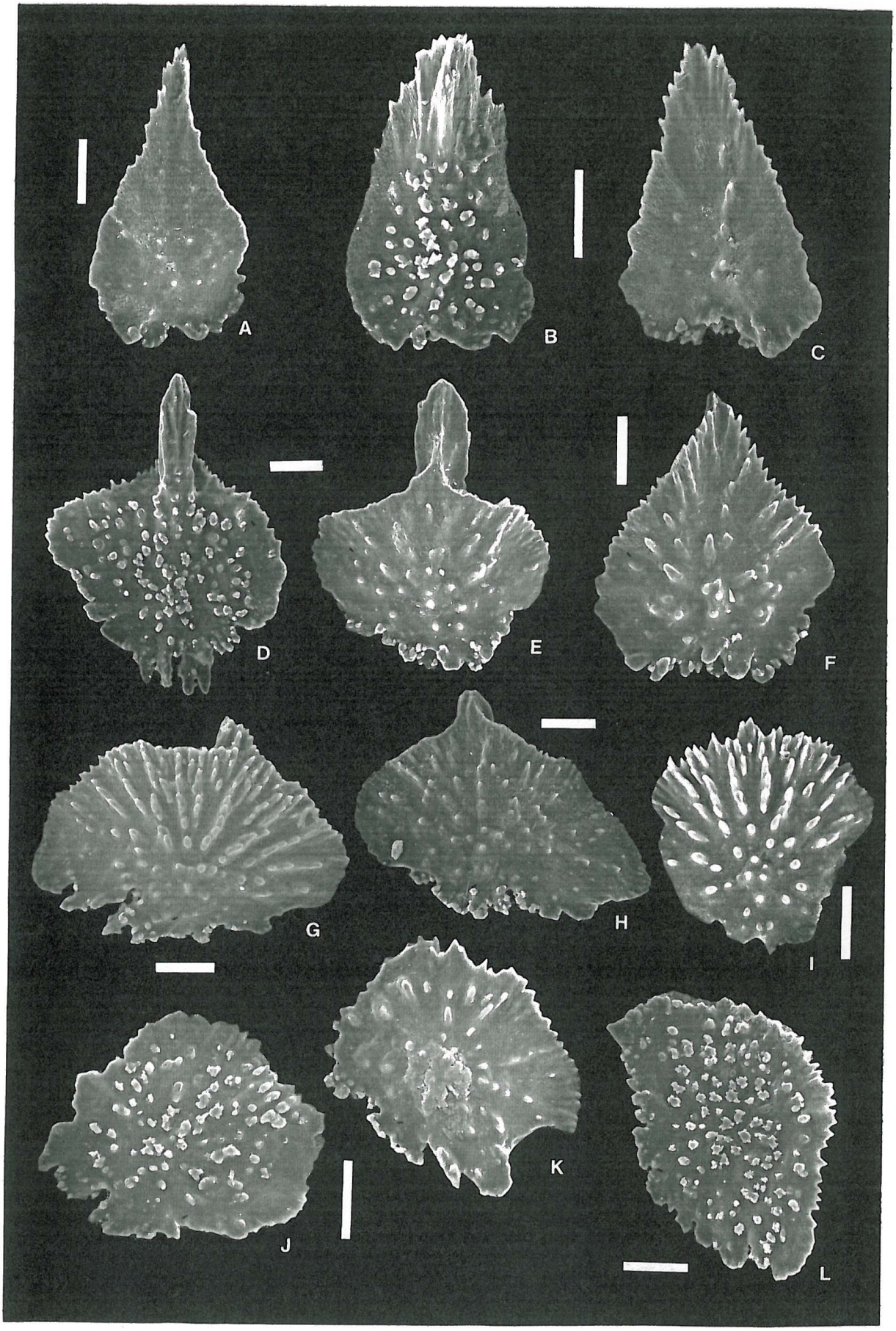

70

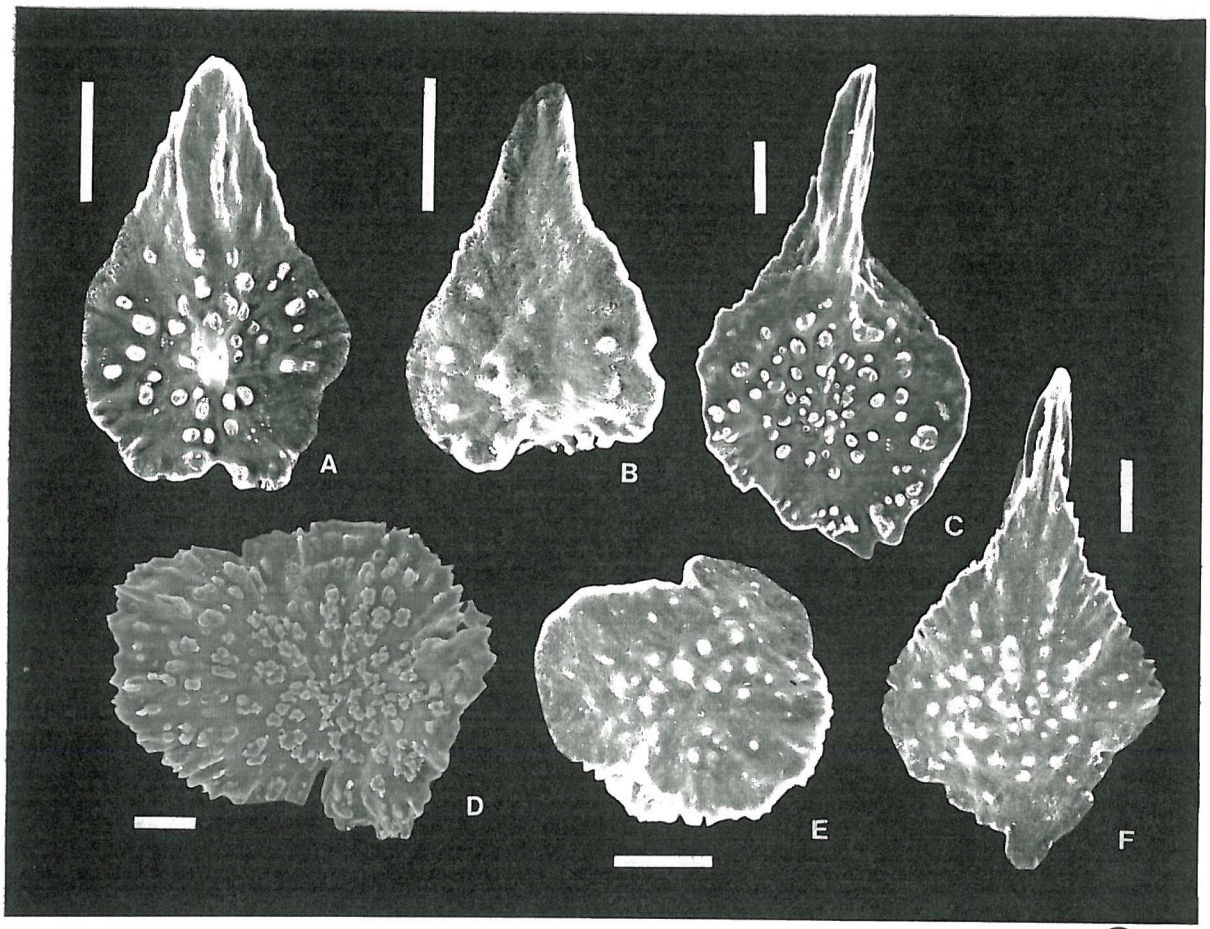

71


72

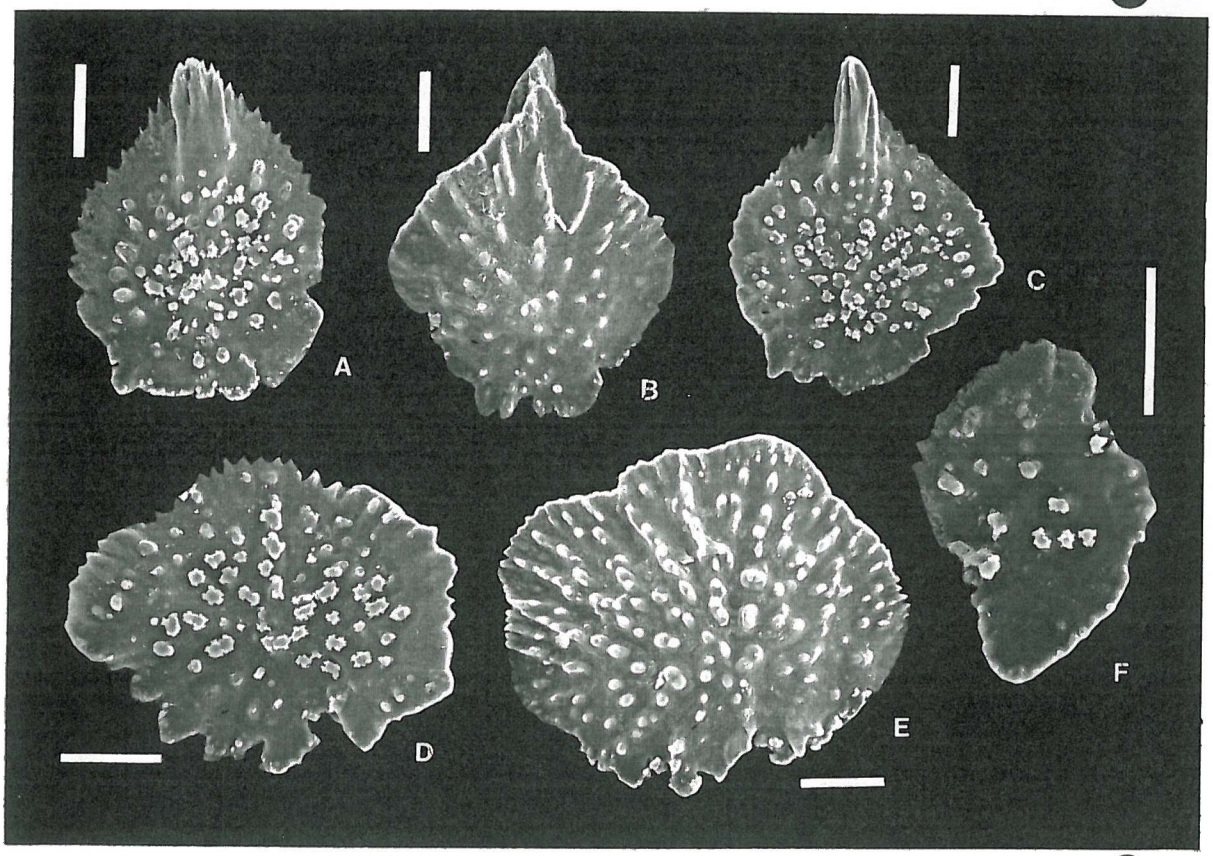

SP1SC1 20kV 100µm X220 11mm
A
B
C
D
E
F
G


A
B
C
D
E
F
G

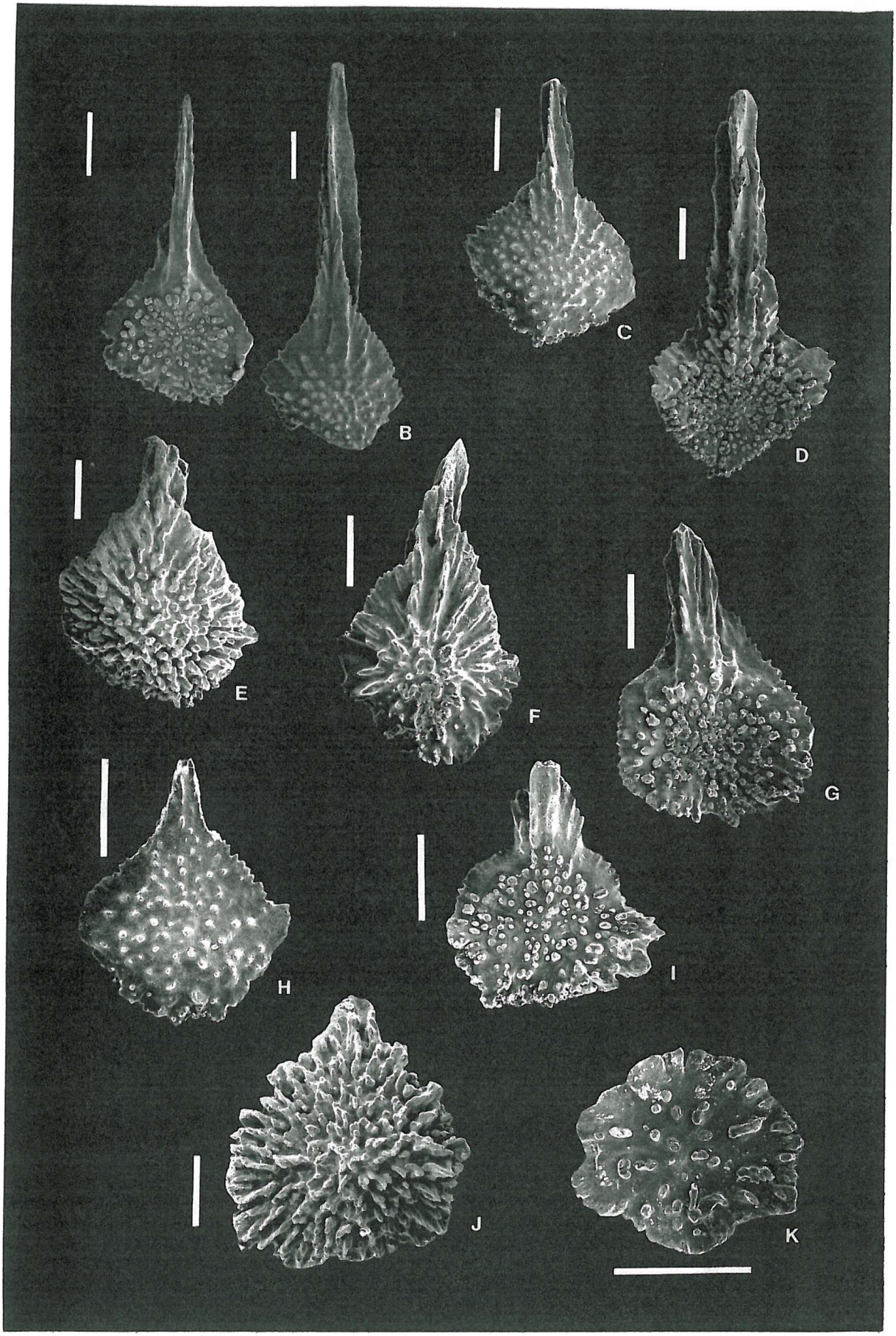

A
B
C
D
E
F
G
H
I
J
K
L

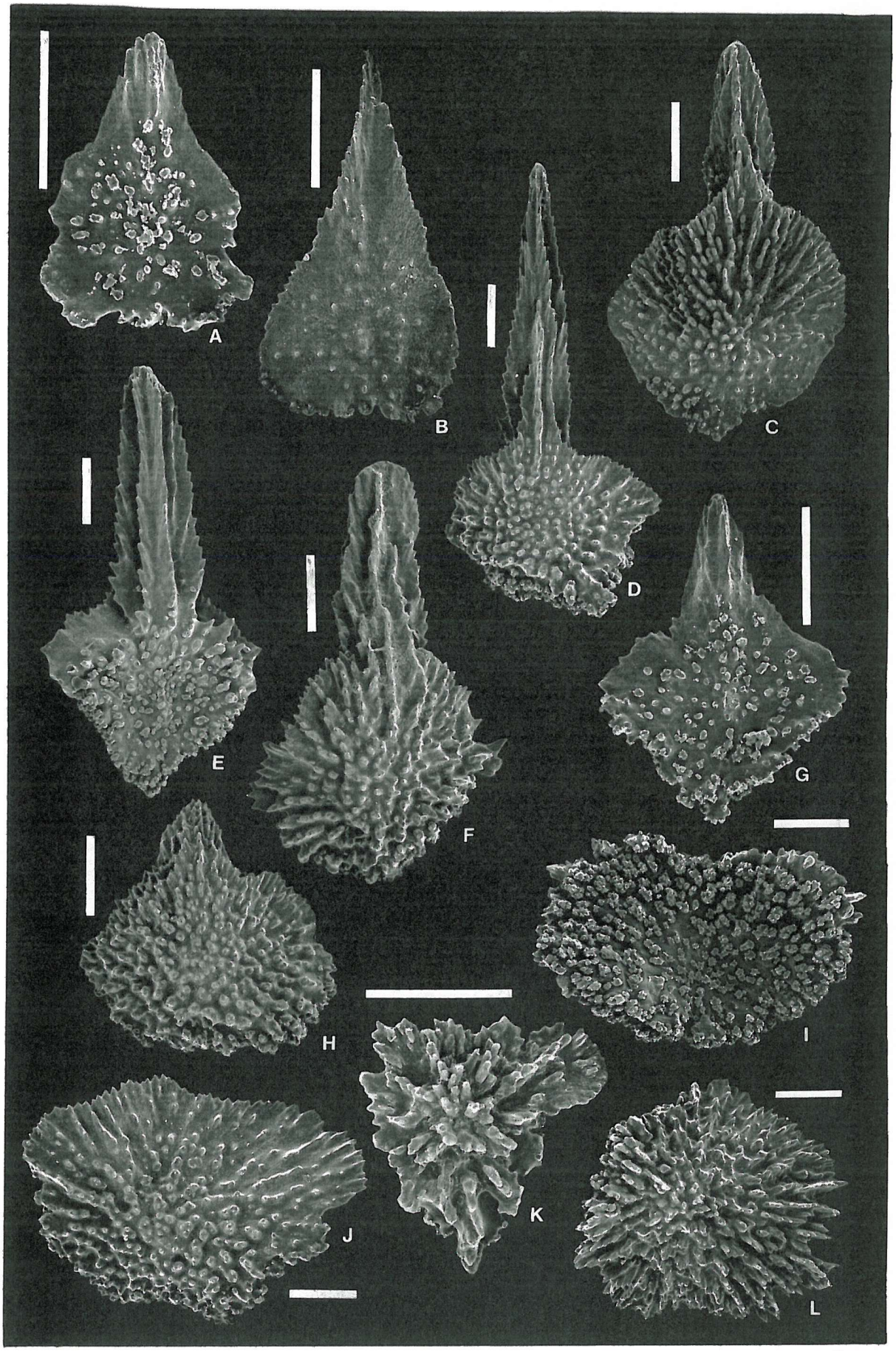


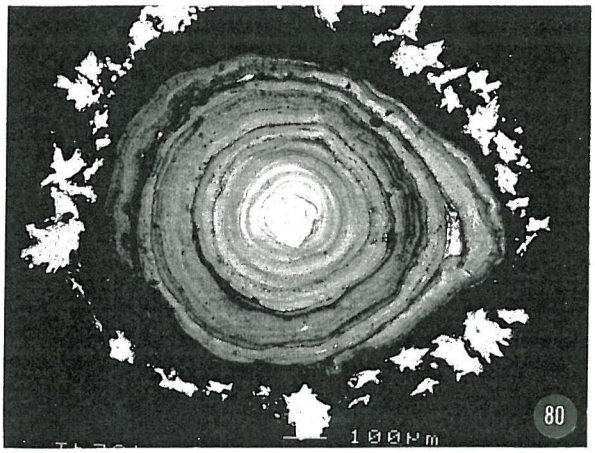
76

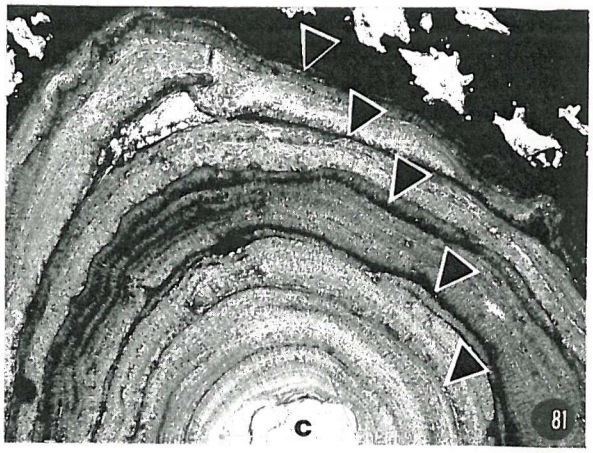


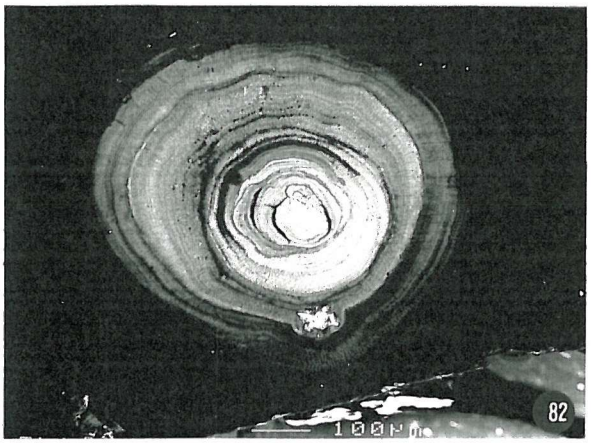
77


B
C
D
E
F
G
H
I
J
K

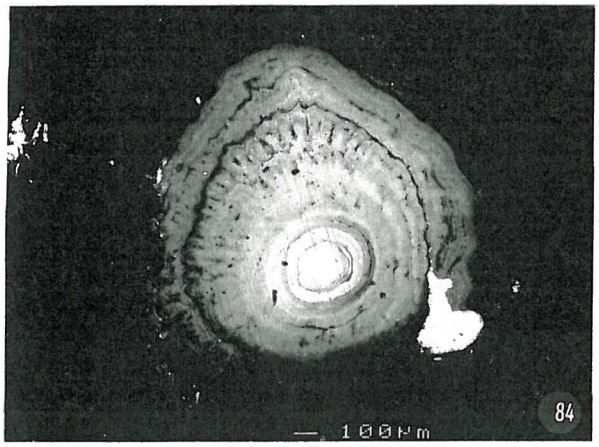

A
B
C
D
E
F
G
H
I
J
K
L

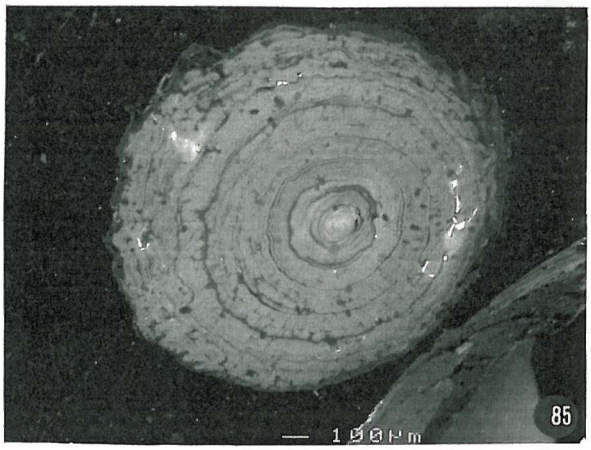

100μm
80

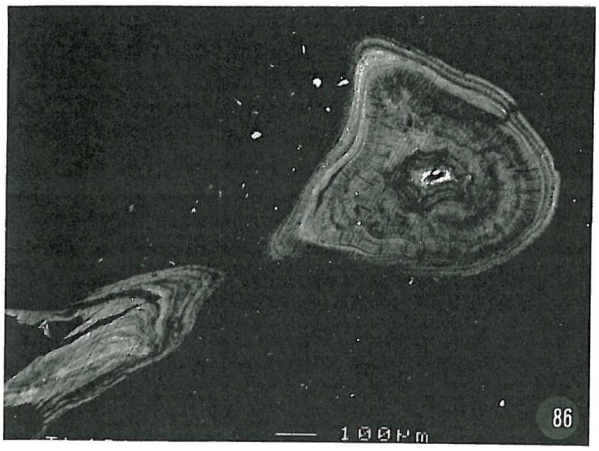

c
81

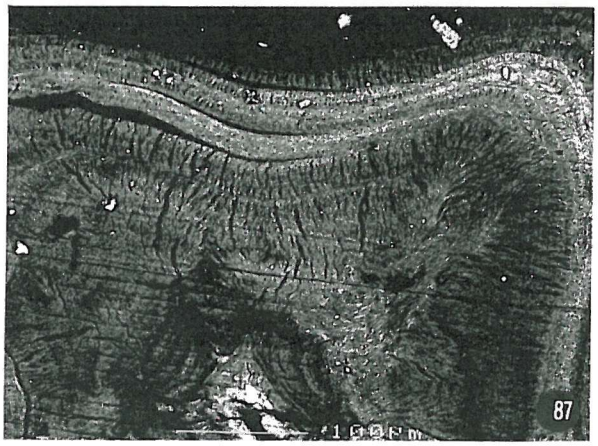

100μm
82

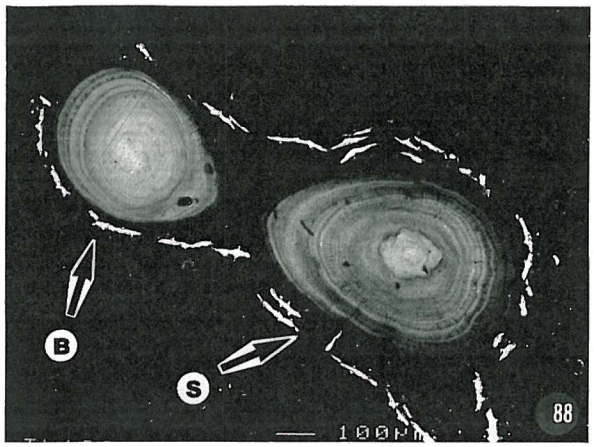

83

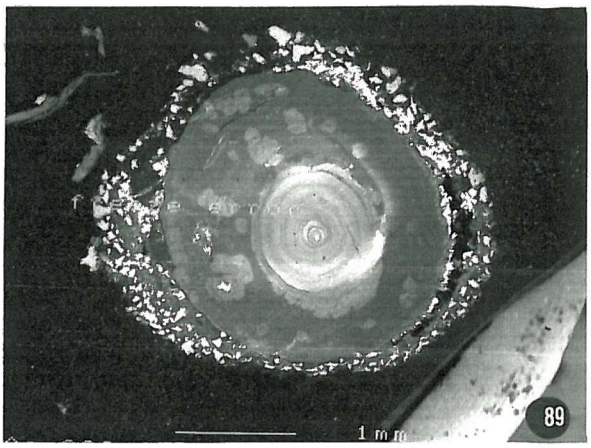

100μm
84

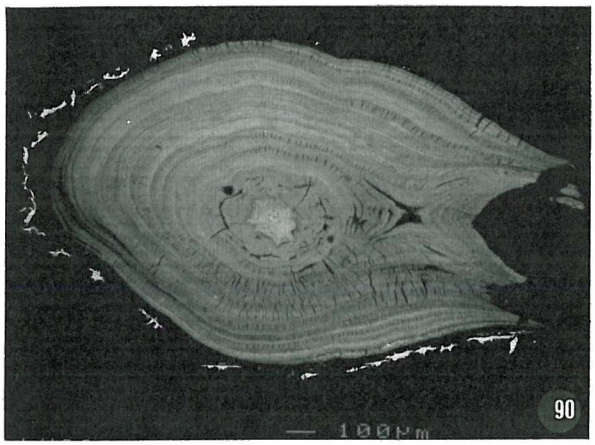

100μm
85

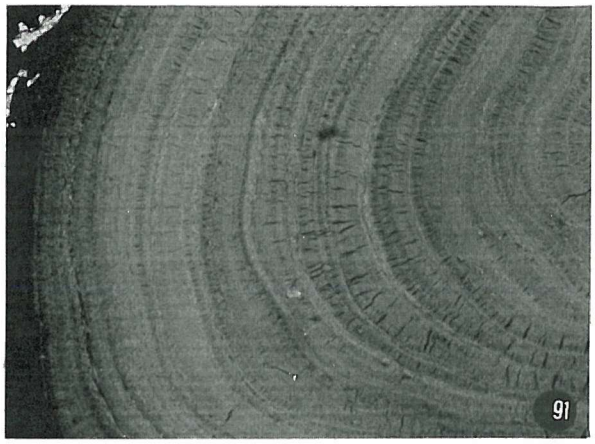

100μm
86

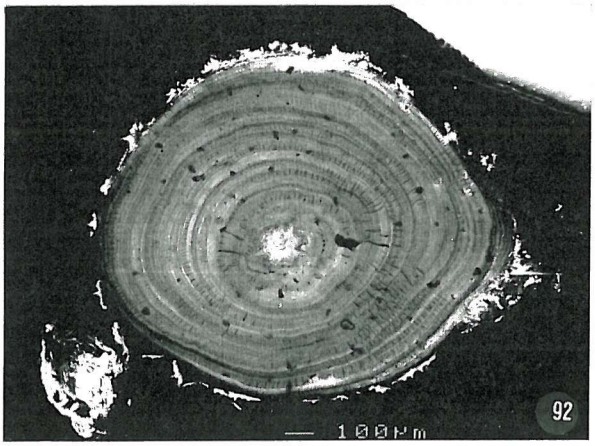

100μm
87

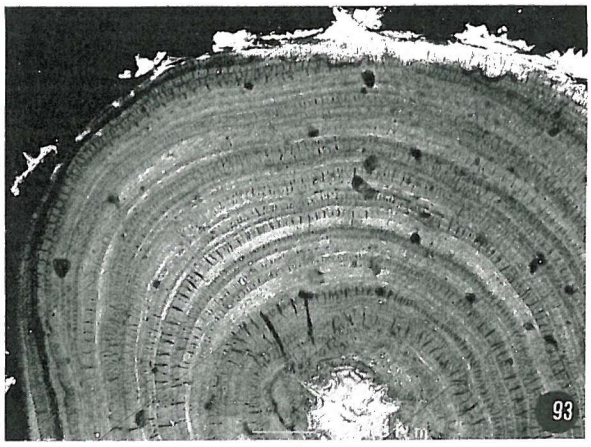

B
S
100µm
88

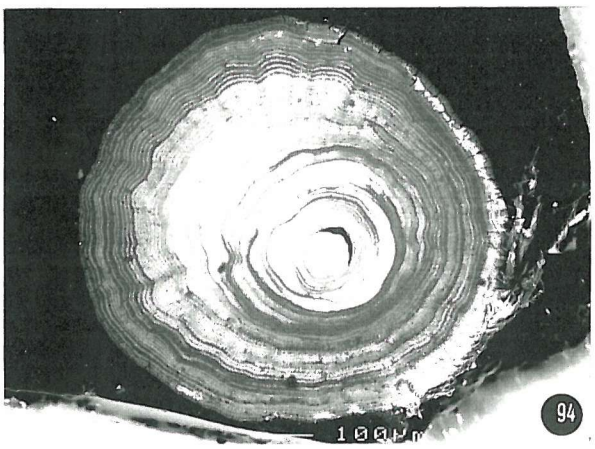

1mm
89

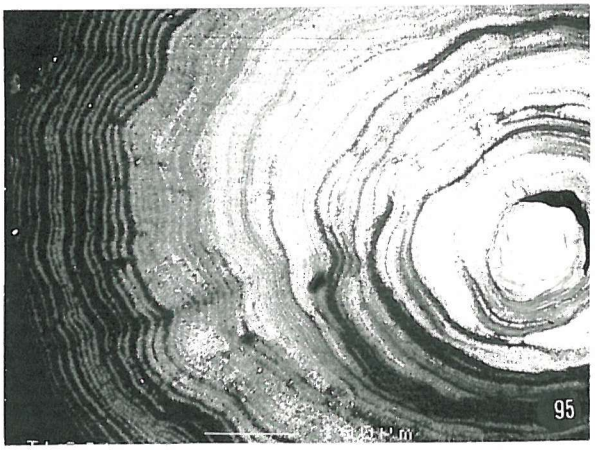

100µm
90

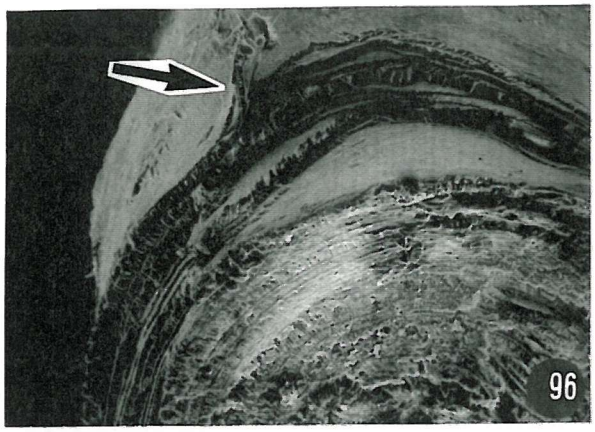

91

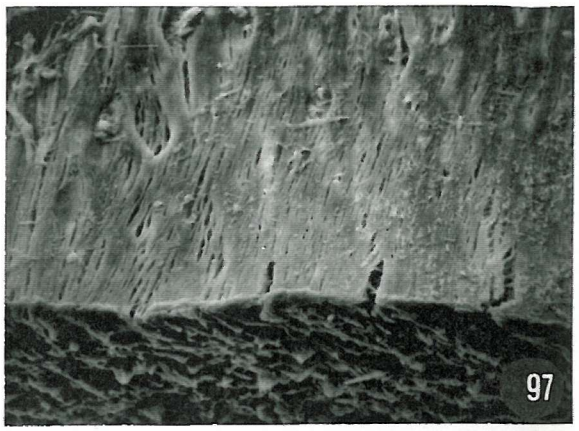

100µm
92

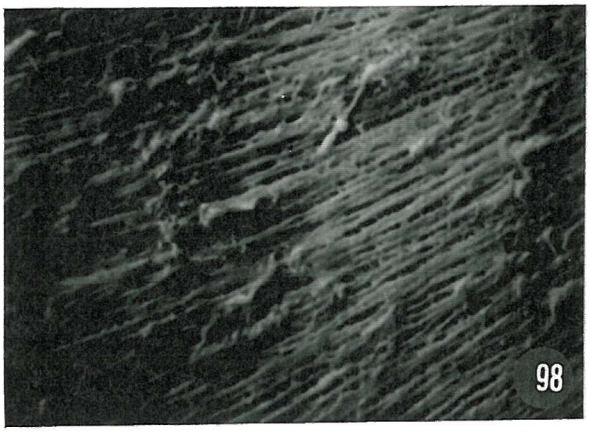

93

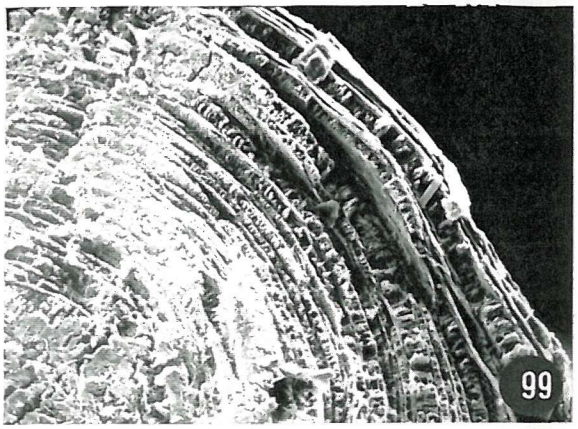

100µm
94

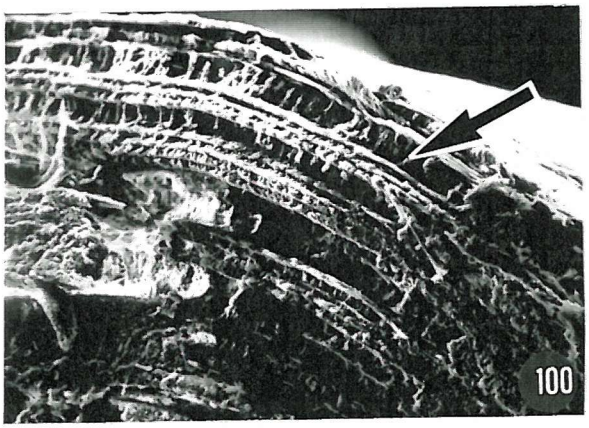

95


96

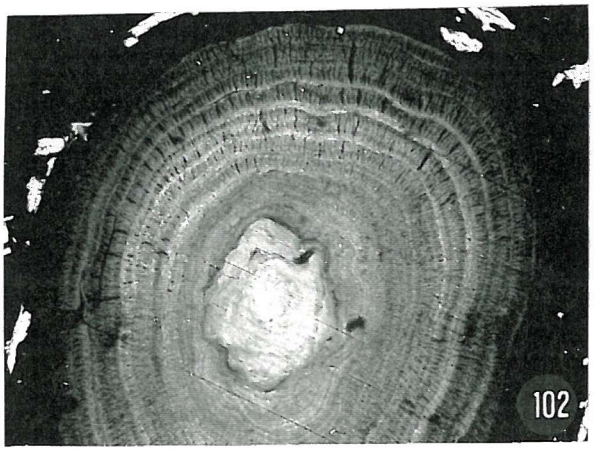

97

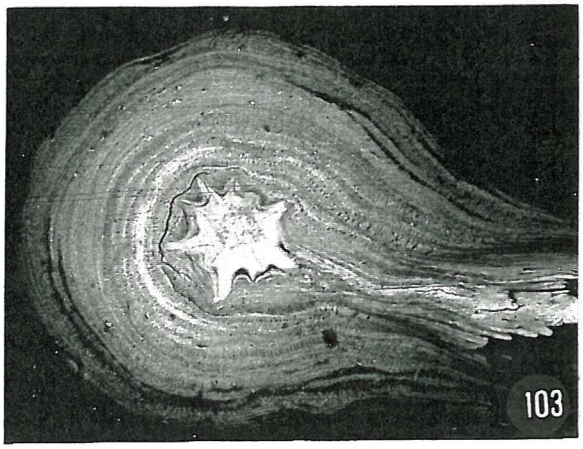

98


99


100


101


102


103

CHAPTER 3
INTERNAL MORPHOLOGY OF *Thouarella variabilis*

3.1 - INTRODUCTION

The fundamental structure of octocorals comprises a polypoid body with eight pinnate tentacles distributed around the oral disc where the mouth invaginates to form a pharynx (stomodeum) leading to a gastric cavity.

The polyp is formed by two epithelial layers, an ectoderm and an endoderm separated by a connective tissue layer, the mesoglea. Eight thin vertical sheets, the mesenteries, stretch radially into the body cavity from the body wall, dividing the coelenteron into eight incomplete intermesenteric compartments. Each mesentery extends as a free septal edge into the gastric cavity (Figs 13, 47, 56, 64 and 65 in Chapter 4). On one side of the pharynx, there is a strongly ciliated longitudinal tract, the siphonoglyph. The two mesenteries opposite the siphonoglyph are heavily ciliated and long compared to the other six. A narrow neck, or sphincter, located in the middle portion of the polyp, separates the pharynx from the gastric cavity.

The side where the siphonoglyph is located is widely referred to in the literature as the ventral side of the polyp and the dorsal side of the polyp is the opposite side of the siphonoglyph. *Thouarella variabilis* has its polyps inclined towards the axis creating, as a consequence, an adaxial side (towards the axis) and an abaxial side (outwards from the axis) (Fig. 18 in Chapter 2). The siphonoglyph is located on the abaxial side and the gonads on the adaxial side. If the adopted terminology is used, in this case, it could be said that the ventral side of the animal is on its back. To avoid such confusion, the terms ventral and dorsal as indicative of the side of the polyp, have been avoided in this study and asulcal, sulcal, adaxial and abaxial will be used instead.

The different parts of the polyp are named: sulcal or abaxial side - the side of the polyp nearest the siphonoglyph and away from the axis; asulcal or adaxial side - the side opposite the siphonoglyph and towards the axis; anthocodia -the distal part of the polyp, bearing the mouth and the tentacles; coenenchyme -the colonial tissue between the polyps, covering the axis and stem; gastric cavity, gastrovascular cavity, coelenteron -the interior space of a polyp; and the body-wall is the wall of polyp body enclosing the gastric cavity (Bayer *et al.*, 1983).

This chapter will provide a detailed description of each of the features mentioned above.

3.2 - ECTODERM

The ectoderm consists of an outer epithelial and an inner subepithelial layer which covers the polyp and coenenchyme, although the two layers are not easily distinguished. The ectodermal cells vary in shape according to their position on the polyp or to the tension or compression to which they are subjected. The cells can have a stretched and squamous resemblance in portions of the polyp which are under tension and are thick, folded and seemingly stratified, in compressed portions. The ectoderm is thicker in areas between scales (Figs 5, 6), on the asulcal side towards which the polyp is bent, on the calyx (Figs 1, 2), on the tentacles (Figs 25, 26, 33, 37, 38) and specially on the mouth and pharynx (Figs 9-15). The ectoderm is thinner when covering portions of the scales (Figs 3, 4).

Between the scales the supporting ectodermal cells are cuboidal or low-columnar in shape (Figs 5, 6), have a large nucleus, granular cytoplasm, several small vacuoles and contractile fibres. Among the supporting cells are granular cells, sometimes packed with large granules. Cells bearing filamentous cytoplasmic processes are also commonly observed in the ectoderm (Fig. 2). Large intercellular spaces are observed separating the cells. Cnidoblasts are abundant in the ectoderm surrounding the calyx, on the most distal portion of the calyx (Figs 1, 2) and on the ectoderm of the tentacles, especially at the base of the tentacles (Figs 25-27). The thin ectoderm layer covering the scales is composed of considerably stretched cells with large nuclei and granular cytoplasm (see Figs 3, 4).

The ectoderm is known to bear supporting, glandular and sensory elements (Bayer, 1956; 1974; Lang da Silveira, 1977). The ectoderm also contributes to the nutrition of the octocoral through direct uptake of dissolved organic compounds (Schlichter, 1982; Fautin & Mariscal, 1991).

3.3 - ENDODERM

The endoderm is the inner epithelial layer. It covers the body wall and the mesenteries on both sides and it is generally formed by low-cuboidal cells, which have a large nucleus, granulous cytoplasm and are ciliated (Figs 7, 8, 13, 15,16, 35). When the polyp is in the first reproductive stages, the endodermal cells on the free edge of the mesenteries in the gastric

cavity are tall and expanded since they are packed with fat granules and germ cells (Figs 19-24). Close to the bases of the mesenteries large cells bearing long cilia are found (Figs 21, 22; Chapter 4: Figs 55-56). The endoderm in the gastric cavity is known to be glandular, absorptive and responsible for ingesting food (Matthews, 1916). The endoderm which extends from the gastric cavity into the coenenchyme maintains the link between the polyps along the colony by lining the gastrodermal tubes, or solenia.

3.4 - MESOGLEA

The mesoglea is a structureless, jelly-like substance which separates the ectoderm and the endoderm. It contains amoeboid cells, precursors of the cnidoblasts (which form nematocysts) and scleroblasts (responsible for the production of calcareous sclerites). Long cells containing granules are also observed in the mesoglea (Fig. 7). The mesoglea is thin in the mesenteries (Figs 19, 20), tentacles (Figs 25, 37), pinnules (Figs 39, 40) and pharynx (Figs 9, 13, 14) and thick in the base of the tentacles (Fig. 38) and in the body wall (Figs 7, 8, 34), especially between scales (Fig. 5)

3.5 - TENTACLES, MOUTH, PHARYNX AND SIPHONOGLYPH

The eight pinnate tentacles, which surround the oral disc, grow as an extension of the intermesenteric cavities (Figs 25, 33). The same endoderm layer lining the mesenteries covers the inner portion of the tentacle wall. However, the flat cuboidal endodermal cells present on the mesenteries are replaced by tall columnar, sometimes very elongate, cells in the endoderm of the tentacles. In some portions of the endoderm of the tentacles more than one layer of elongated cells are observed (Figs 25, 33, 35). The mesoglea is thinner around the pinnules (Figs 39, 40) and thicker at the tentacles, especially at the bases (Fig. 38). The ectoderm is formed by tall-columnar cells, which are sometimes fairly stratified (Figs 25-27). The pinnules are surrounded by a thick ectodermic layer (Fig. 40). Some considerably large globular cells are observed amongst the ectodermic cells (Fig. 25). In the lower portion of the tentacles and between the folded tentacles, stratified layers of cuboidal or ovoid cells are found and the majority of these cells present intracellular corpuscules which resemble developing nematocysts (Figs 25-27, 29-32). The nematocysts are probably used to capture prey, however, no particulate food was observed on the tentacles or in the body cavities of the examined specimens.

The mouth and pharynx are formed by an invagination of the ectoderm towards the coelenteron (Fig. 47 in Chapter 4). They are, therefore, lined by the ectoderm and not by the endoderm like most of the body cavity. The pharynx is formed by several ectodermic layers, a thin mesoglea and a flat endoderm composed of cuboidal or elongated cells (Figs 9-15). The pharynx is cone-shaped, being wider in its outer portion and becoming narrower towards the middle portion of the polyp, where it gives way to the gastric cavity. On one side of the pharynx a longitudinal groove, the siphonoglyph, is present. In its upper region, the siphonoglyph is composed of several layers of cells, the deeper layers are formed of cuboidal or ovoid cells, and the outer portion of columnar, heavily ciliated cells (Fig. 11). Along the pharynx, the siphonoglyph is thinner, being formed by one or two layers of long columnar and heavily ciliated cells (Figs 12-14). The nuclei of these cells are positioned at their lower portion close to the mesoglea and the free edge of the cells are clear. On the other side of the siphonoglyph, the pharynx is thicker and formed of longer cells (Figs 9, 10). This is a very granular epithelium and many clusters of large fat granules are observed in the cells. Some cells are markedly stretched from being packed with granules and their nuclei are restricted to the deeper portion of the cells. Large intercellular spaces are observed between these cells, which might be an artefact of the methodology. The outer portion of these cells appear to be ciliated. Cnidoblasts are seldom observed in the pharynx (Fig. 28).

The opening of the pharynx into the body cavity acts as a sphincter and it is more stratified than any other part of the polyp (Fig. 15). It has many layers of cuboidal or small ovoid cells, with large nuclei and little cytoplasm. The endoderm in this region is formed on one side by large and vacuolated cells, and on the other by low cuboidal cells.

3.6 - MESENTERIES

The mesenteries consist of thin sheets of mesogleal substance arising from the supporting lamella of the body wall, and covered on both sides by endodermic cells. The mesenteries are attached to the body wall up to the base of the polyp (Figs 13 and 65 in Chapter 4). During the breeding season, the lower portion of the mesentery might be free since they expand when bearing clusters of germ cells (Fig. 66 in Chapter 4). In the gastric cavity each mesentery is free along its inner edge.

The two mesenteries opposite the siphonoglyph, named asulcal mesenteries, have their free edge slightly concave and heavily ciliated (Figs 17, 18, 36). These mesenteries, like the mouth and pharynx, are lined by ectoderm whilst the other six mesenteries are covered by

the endoderm. The asulcal mesenteries are composed of tall-columnar cells, each one bearing several cilia (Fig. 18). Their nuclei are located at their lower portion and the outer portion of the cells contain various granules. The cells at the centre of the mesentery are the longest and narrowest and do not bear cilia (Figs 17, 18). Towards the sides the cells are shorter, wider, less ciliated and not as compressed against each other as the cells in the central portion. Together with the siphonoglyph, the asulcal mesenteries are responsible for maintaining the water circulation inside the colony. They create an upward current of water in the coelenteron by the active lashing of the cilia borne on their cells.

The other six mesenteries are known to be secretory and absorptive, completing the digestion begun in the pharynx. However, Schlichter (1982) observed that in *Heteroxenia fuscescens*, the degradation of particulate food takes place in the asulcal mesenteries rather than in the other six mesenteries. The two asulcal mesenteries do not bear germ cells or fat granules, only the six other mesenteries form gonads. The endodermic cells covering the other six mesenteries are flat cuboidal or elongated (Fig. 20) but when they are bearing germ cells and are packed with lipid droplets, they can be long-columnar or considerably expanded (Figs 16, 21, 23). The endodermic cells are in general ciliated (Fig. 22). They can be extensively vacuolated and have large intercellular spaces (Figs 16, 23). Sometimes only one side of the mesentery is shown expanded whereas the other is low cuboidal (Fig. 20). At the base of the mesenteries there are some large and globular cells with large ovoid nuclei (Figs 21, 22). Amongst these cells, other elongated or spindle-like cells bearing considerably long cilia are found. Once the endodermic cells loose their lipid and germ cell content, they resemble empty flaccid bags (Fig. 24).

3.7 - MUSCLES

Longitudinal muscle fibres are observed all along the tentacle between the mesoglea and the ectoderm (Figs 25, 33, 38). On the pinnules the muscle fibres are disposed on their inner side (Fig. 40). Tentacles and pinnules are highly contractile (Fig. 72 in Chapter 4). Circular muscle fibres are found in the pharynx where the epithelium connects the mesoglea and in the base of the endoderm in the gastric cavity. The mesenteries bear longitudinal retractor muscle which will be used to retract the polyp inside its calcareous armature. These fibres are present in only one face of the mesenteries (Figs 33-35).

LIST OF FIGURES FOR CHAPTER 3

Fig. 20 - Mesentery showing endodermal cells (Ed) on both sides. Cells on one side packed with granules (G). m, mesoglea. 4,200 X. (32679)

Fig. 21 - Lower portion of the mesentery (BM) showing endodermal cells packed with granules (G). 1,800 X. (32669)

Fig. 22 - Endodermal cells at the lower portion of the mesentery showing a large globular cell and cells with long cilia. 5,600 X. (32670)

Fig. 23 - Mesentery showing expanded granular endodermal cells (Ed). m, mesoglea. 3,500 X. (32665)

Fig. 24 - Flaccid endodermal cells (Ed) which have lost part of their granular content. m, mesoglea. 2,100 X (32723)

Fig. 25 - Tentacle showing endodermal cells (Ed), muscle fibres (ms) globular ectodermal cells (Ec) and numerous cnidoblasts. 1,400 X. (32713)

Fig. 26 - Region at the base of the contracted tentacles showing cnidoblasts. 1,100 X (32712)

Fig. 27 - Detail of the cnidoblasts shown in Figure 26. 2,100 X. (32711).

Fig. 28 - Cells in the pharynx. G, granules; N, cnidoblasts. 4,900 X. (32676)

Fig. 29 - Cnidoblasts (N) in the ectoderm of the calyx region. 3,500 X. (32684)

Fig. 30 - Cnidoblasts (N) in the ectoderm of the calyx region. n, nucleus. 12,000 X. (32671)

Fig. 31 - Cnidoblasts (N) in the ectoderm of the tentacles. n, nucleii. 14,000 X. (32605)

Fig. 32 - Cnidoblasts (N) in the ectoderm of the tentacles. n, nucleii. 11,000 X. (32604)

Fig. 33 - Tentacles and intermesenteric cavities (mc) showing the ectoderm (Ec), endoderm (Ed), muscle fibres (ms) in the mesentery and mesoglea (m) in the mesentery. 140 X. (9a)

Fig. 34 - Intermesenteric cavities (mc). 140 X. (9b)

Fig. 35 - Intermesenteric cavity (mc) showing muscle fibres (ms), endoderm (Ed) and mesoglea (m). 280X. (9c)

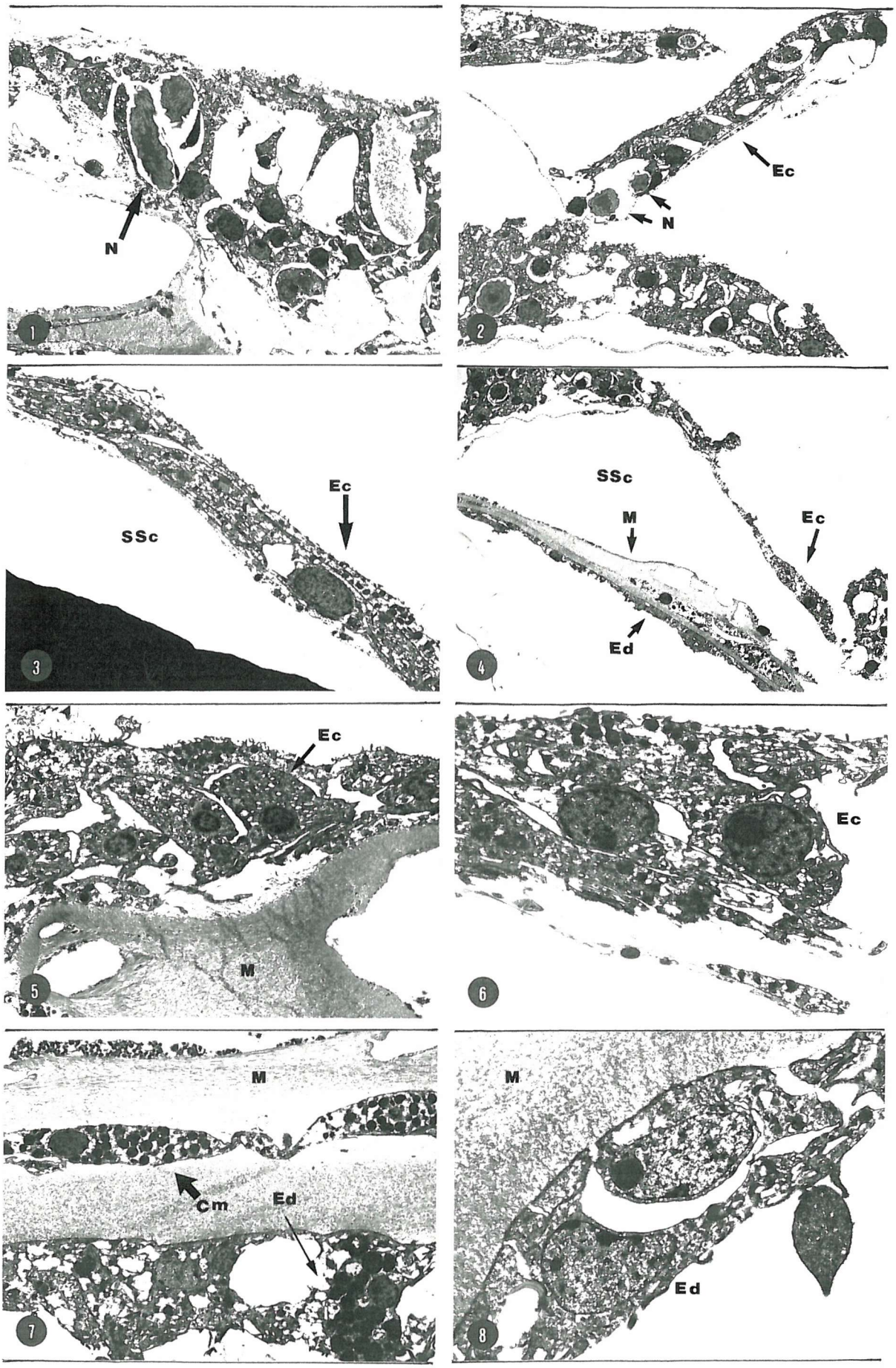
Fig. 36 - Asulcal mesentery (DM). 280 X. (9b)

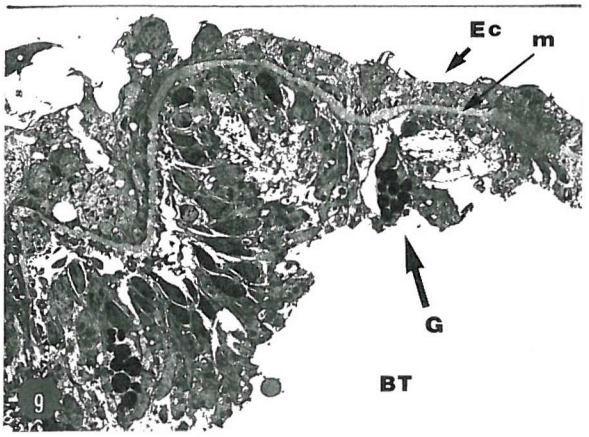
Fig. 37 - Region of the anthocodia showing the tentacles (arrow). 70 X. (9b)

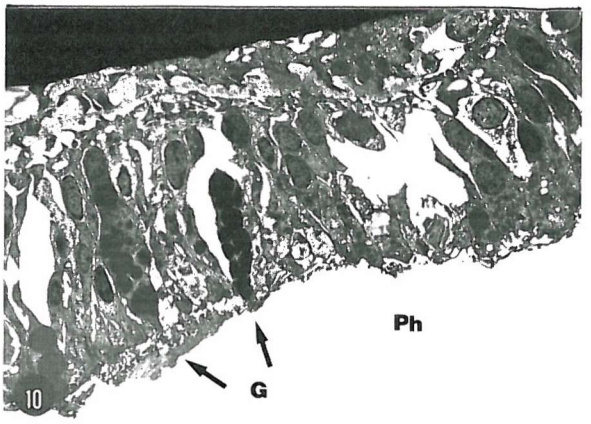
Fig. 38 - Longitudinal section of tentacle showing pinnule (Pi) and mesoglea (m) and muscle (red staining). 280 X. (9a)

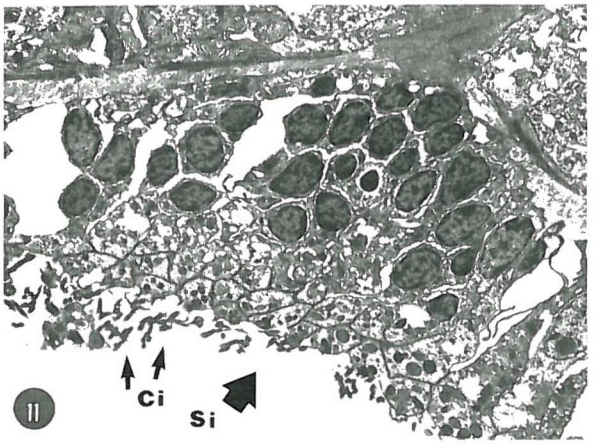
Fig. 39 - Longitudinal section of tentacle showing pinnule (Pi). 280 X. (9a)

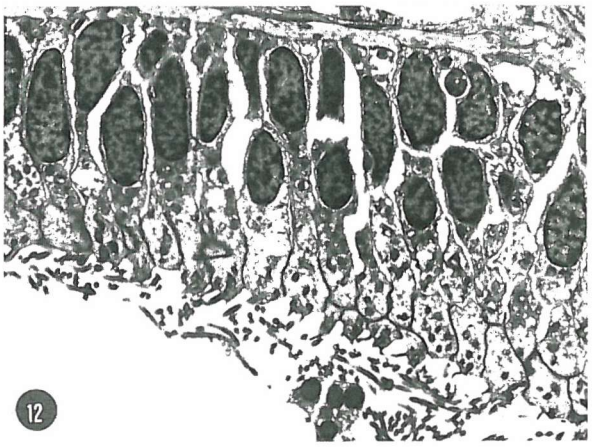
Fig. 40 - Tentacle showing pinnule (Pi) in cross section. 280 X. (9a)

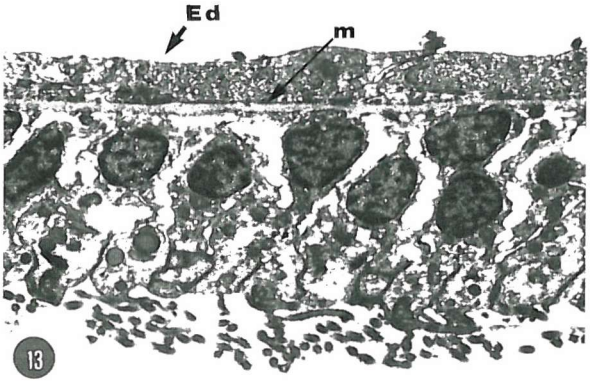

N
1
Ec
N
2
Ec
SSc
3
SSc
M
Ec
Ed
4
Ec
M
5
Ec
6
M
Cm
Ed
7
M
Ed
8


Ec
m
G
BT
9

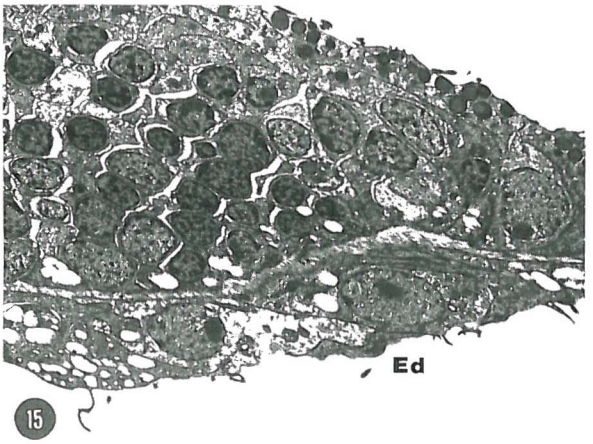

Ph
G
10

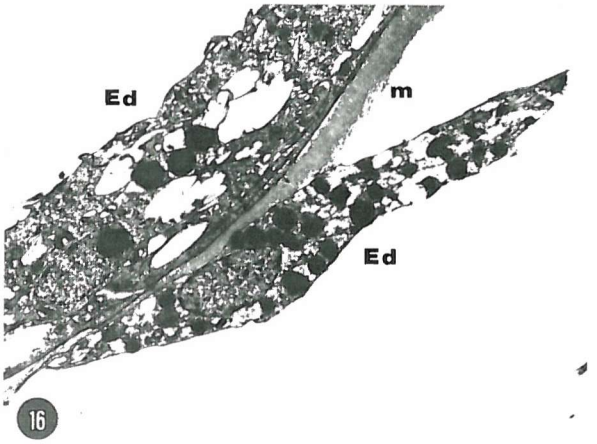

Ci
Si
11

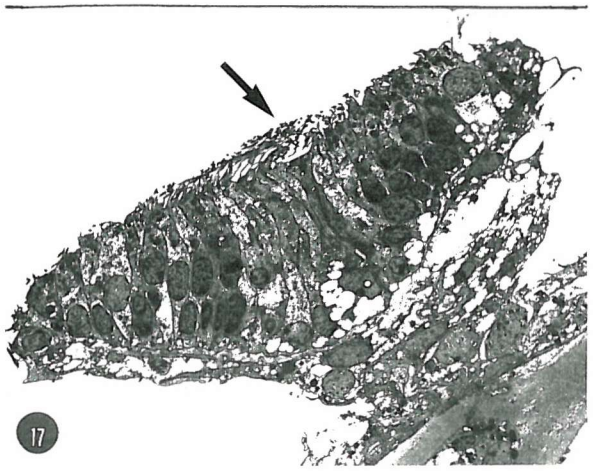

12


Ed
m
13

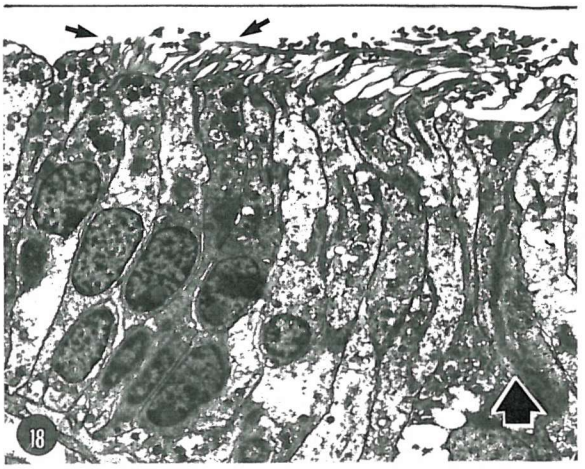

14


Ed
15

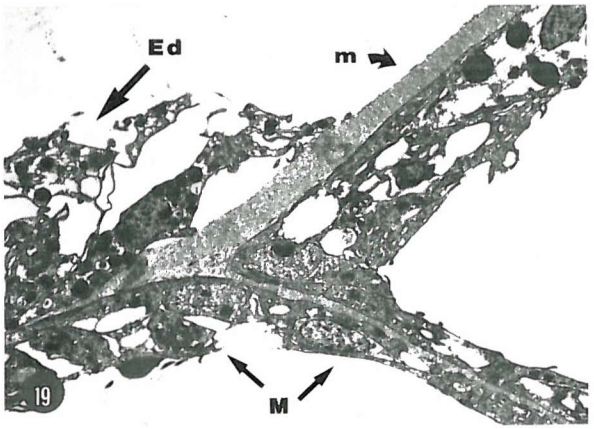

Ed
m
Ed
16



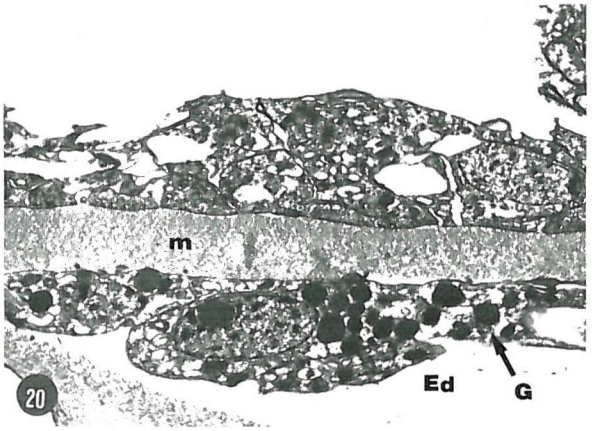
17



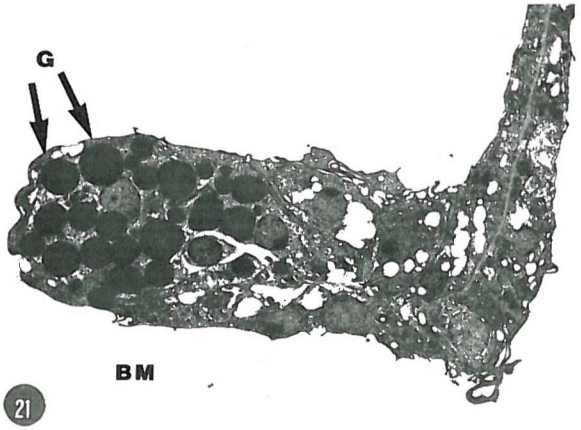
18



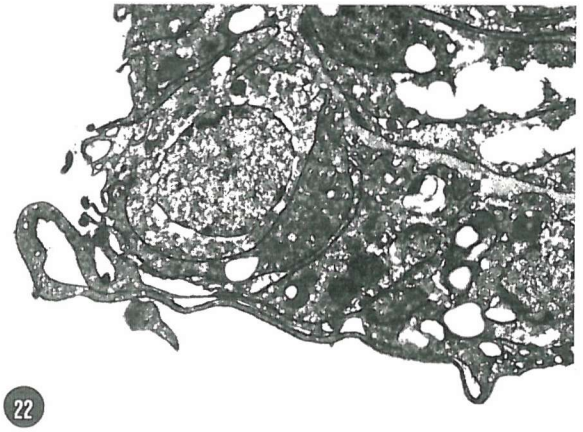
19



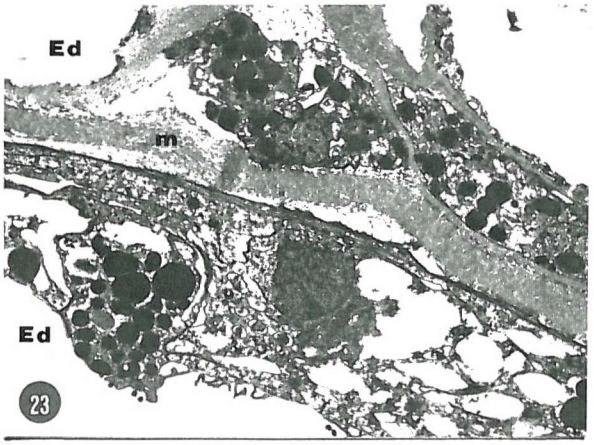
20



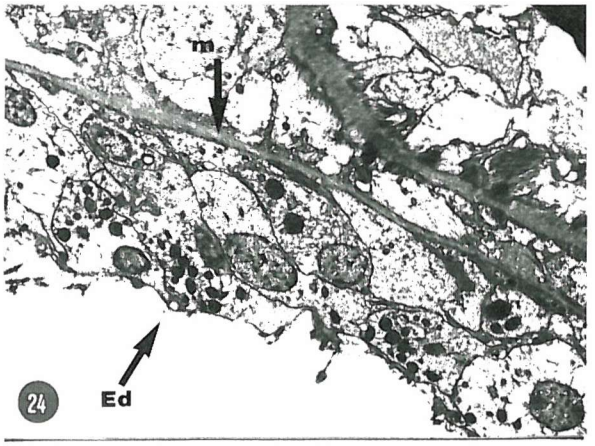
21



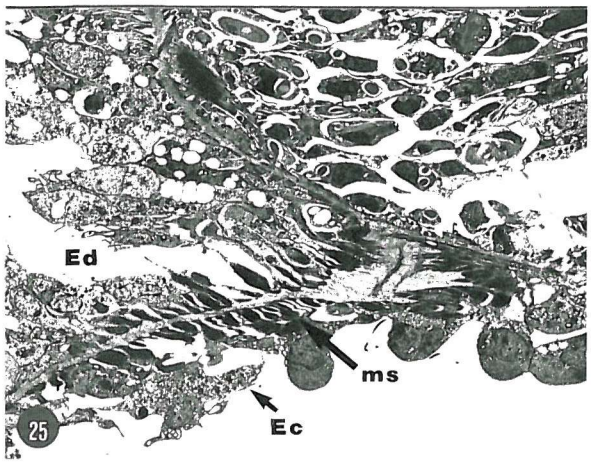
22

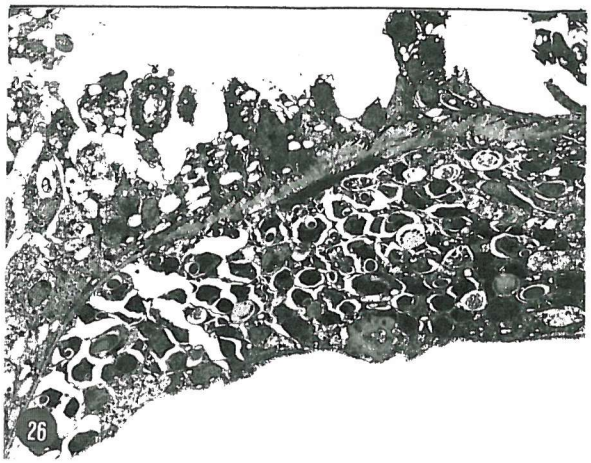


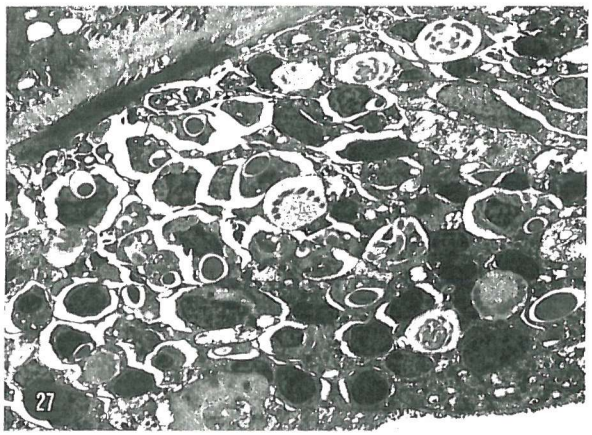
23

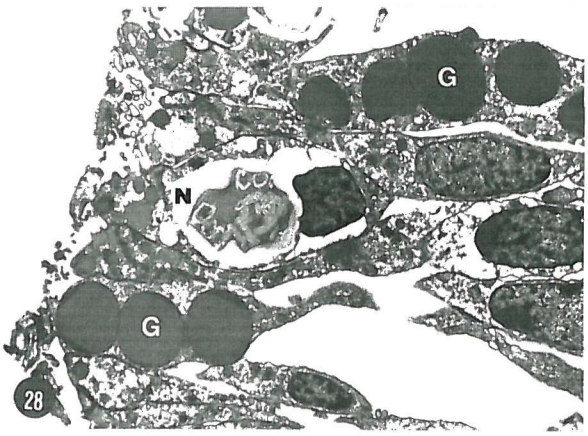


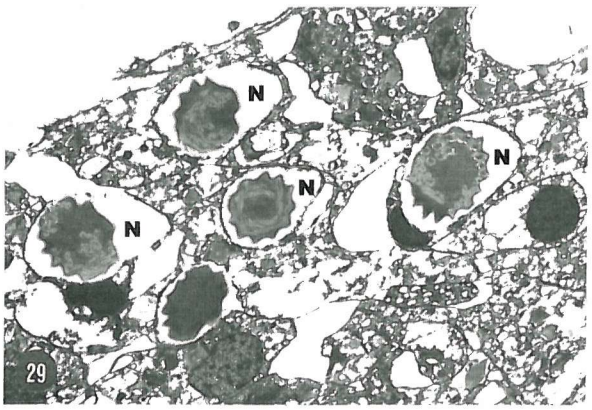
24

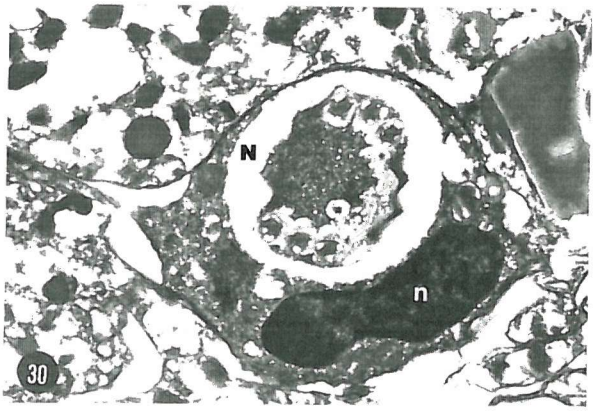

Ed
ms
Ec
25

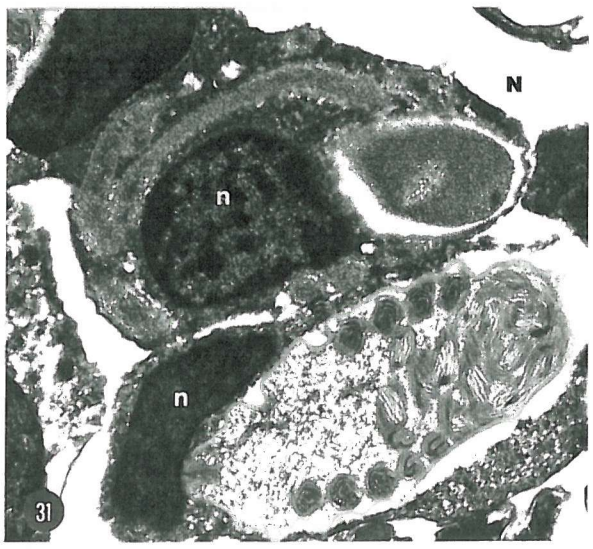

26

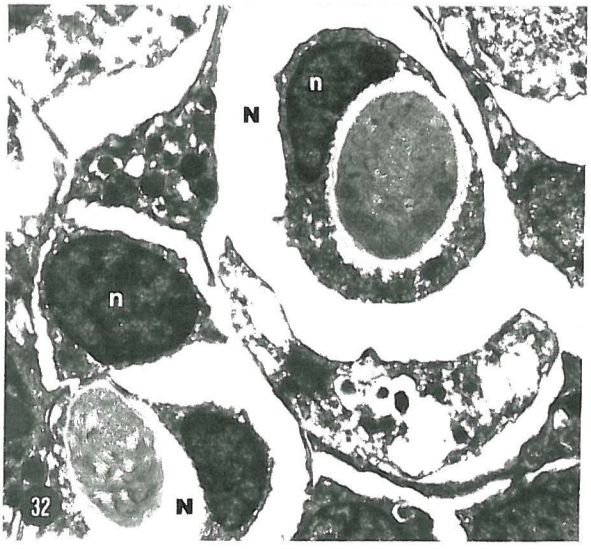

27

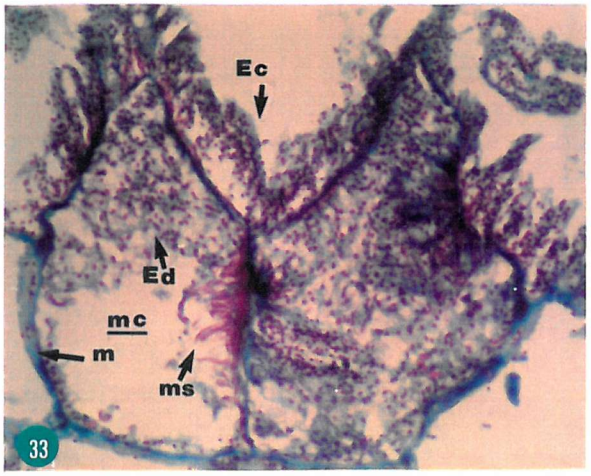

G
N
G
28

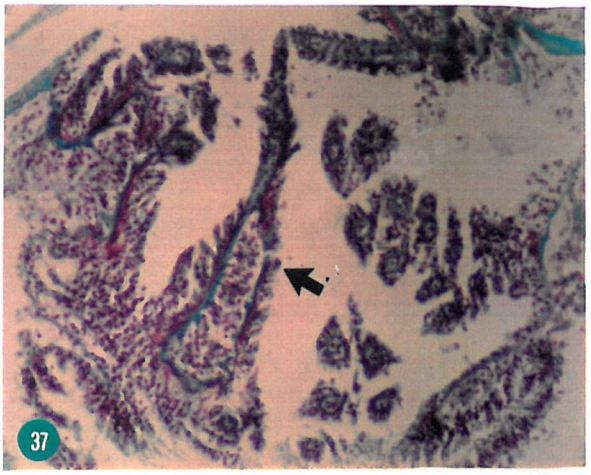

N
N
N
N
29

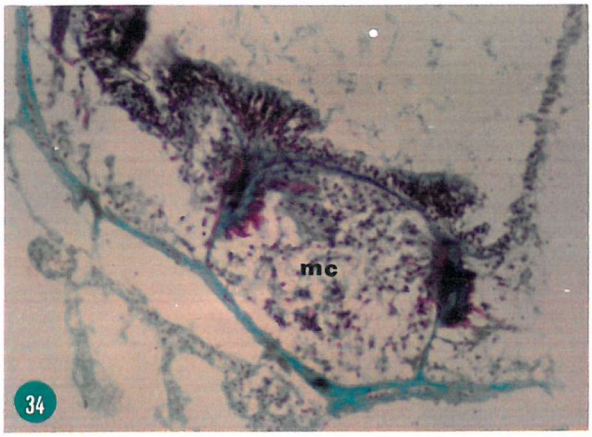

N
n
30

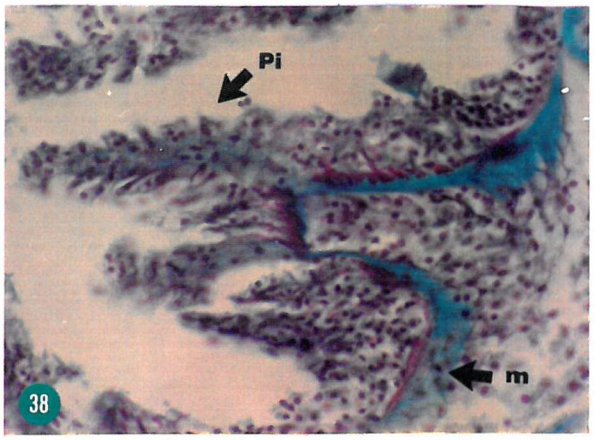

N
n
n
31

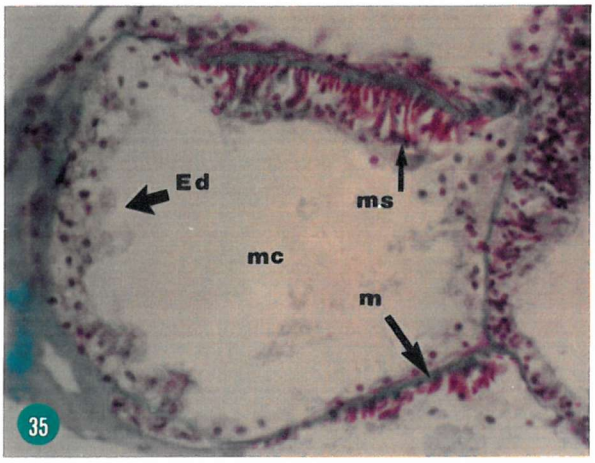

n
N
n
N
32

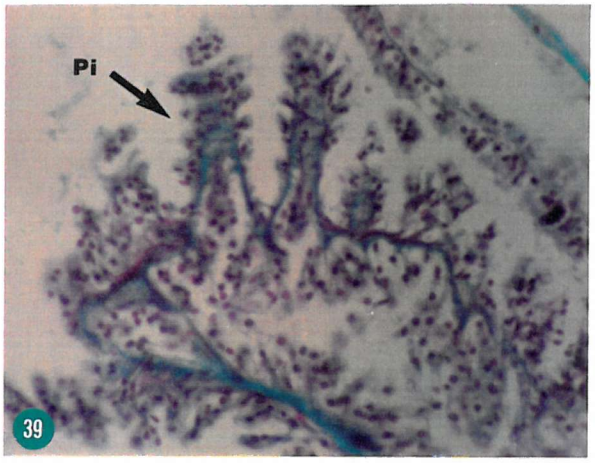

Ec
Ed
mc
m
ms
33

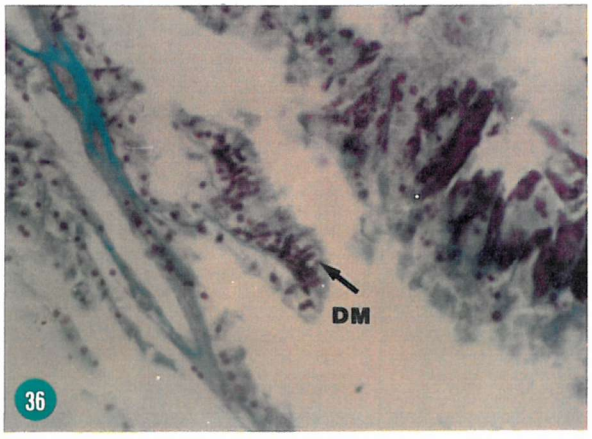

37

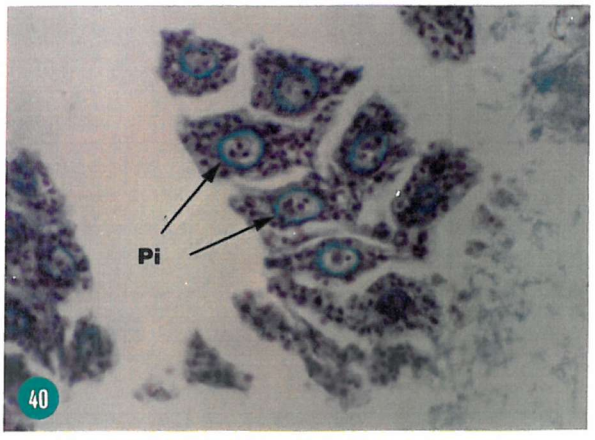

mc
34


Pi
m
38


Ed
ms
mc
m
35


Pi
39


DM
36


Pi
40

CHAPTER 4
REPRODUCTIVE BIOLOGY OF *Thouarella variabilis*

4.1 - INTRODUCTION

Our knowledge of the reproductive biology of octocorals is still very limited, although their significance, accounting for much of the biomass and species diversity on coral reefs is well recognized. Until recently, the available information on reproduction of octocorals was based on a few bulky reports from the last century and the first half of this century, adding to the cursory observations existing in taxonomic descriptions. For the past few years the investigation of the reproductive biology of octocorals has increased.

Among the six orders of Octocorallia, the Telestacea has been the least investigated on this aspect and remains poorly known. For Stolonifera, the detailed reports of Kowalevsky & Marion (1882, 1883) and Gohar (1948) on *Clavularia* were followed by more recent work (Suzuki, 1971: working on Cornulariidae; Weinberg, 1986; Benayahu, 1989). The Alcyonacea has been better investigated, particularly the families Alcyoniidae and Xeniidae, as a result of work on *Alcyonium* (Hickson, 1895; Hill, 1906; Muller, 1910; Matthews, 1916; Hartnoll, 1975, 1977; and Sebens, 1983), *Lobophytum* (Yamazato *et al.*, 1981; Uehara *et al.*, 1987), *Sarcophytum*, *Parerythropodium* (Benayahu & Loya, 1983, 1984, 1986), *Xenia, Heteroxenia* and *Efflatounaria* (Ashworth, 1899, 1900; Gohar, 1940; Gohar & Roushdy, 1961; Dinesen, 1985; Benayahu & Loya, 1984, 1985; Benayahu *et al.*, 1988; 1989; Benayahu, 1990). Apart from the early work of Kinoshita (1910) on *Anthoplexaura*, the order Gorgonacea has mostly been investigated in recent years (Theodor, 1967; Grigg, 1970, 1977; Vighi, 1970; Goldberg, 1974; Weinberg, 1979; Beheti-Gonzalez & Guardiola, 1979; Walker & Bull, 1983; Gili & Garcia, 1985; Brazeau & Lasker, 1989, 1990). Were it not for a recent paper by Babcock (1990), the reproductive biology of the Helioporacea (= Coenothecalia) would be virtually unknown. The Pennatulacea was of interest to some early workers (Grant, 1826, 1827, 1829; Dalyell, 1838; Wilson, 1880; 1883; Lacaze-Duthiers, 1887; Jungersen, 1888; Roule, 1932) but there is little recent contribution to the knowledge of their reproduction (Birkeland, 1969; Chia & Crawford, 1973; Rice *et al.*, 1992).

The early works dealt mainly with broad outlines of the group, natural history, development and colony formation. The more recent lines of research have led to more detailed studies on development, life cycle, reproductive patterns and larval ecology.

The colony morphology, polyp size, patterns of gamete development, habitat and levels of disturbance are thought to determine patterns of reproduction in reef anthozoans (Stimson, 1978; Rinkevich & Loya, 1979; Fadlallah, 1983; Fadlallah & Pearse, 1982a, b,; Szmant, 1986; Brazeau & Lasker, 1990). This assumption, however, was based on a few data and requires further study. In order to facilitate the understanding of the influence of colony morphology on the variety of reproductive patterns existing among the different orders of octocorals, a brief description of the main features of each order, based on Bayer's review (1973) of the colonial organization in octocorals, is presented here.

The colony morphology of octocorals will vary according to the level of organization and integration between the individual polyps of the colony. The degree of physiological interconnection in the colony, via the solenia and gastrodermal canal, differs in the six orders of the subclass Octocorallia. The more the interdependence of individual polyps, the more complex is the organisation of the group. This colonial integration ranges from simple forms, with poorly interconnected polyps, to highly organized colonies with polyps specialized for respiratory, circulatory, digestive, sexual and locomotory functions.

The orders Stolonifera and Telestacea have the most simple forms. In the Stolonifera the individuals develop vegetatively from basal or lateral stolonal outgrowths. They have the endodermal canals of the stolons linked but not the gastric cavities. In some species of the order Telestacea the gastric cavity is partially penetrated by the solenia and in more complex species the polyps arise from the network of solenia of the body wall of the zooids showing two patterns of ramification, monopodial and sympodial.

Species of the order Alcyonacea have their polyps embedded in a common coenenchyme with the gastrovascular cavities close together and the distal portions independent. Dimorphism is an important feature of this order. Besides the autozooids, which exist also in the other orders, they have siphonozooids that have the siphonoglyph as the main morphologic feature. The function of its siphonozooids is to improve the efficiency of the transport of water through the colony, making possible the development of massive forms, common in the Alcyonacea. The monomorphic forms existing in this order are usually branched, lobed or arborescent, but the polyps are always involved in a common coenenchyme. In some dimorphic alcyonaceans, gonads develop in both autozooids and siphonozooids but in others they are confined to the siphonozooids as seen in the rare deep-sea alcyonacean

Bathyalcyon robustum whose colony consists of a single giant autozooid with numerous siphonozooids embedded in its body wall. The autozooid is sterile and the siphonozooids bear the gonads (Bayer, 1956, 1973).

The order Gorgonacea is characterized by mainly arborescent forms with regular branching patterns. In the species of this order the polyps are united by coenenchyme only in the bases and the solenia do not penetrate deeply into the gastric cavity. The main feature of this order is the support of the colonial coenenchyme by a horny or calcareous axial structure. The nature of the axis is one of the important characters for the taxonomic classification of the families of this order. Based on this character the gorgonaceans are divided into two suborders, the Scleraxonia and the Holaxonia. Gorgonians are usually erect and the colonial architecture is designed in such a way that a large surface area for feeding is provided. The colonial shape varies considerably from unbranched to regular branching pattern, such as one plane pinnate; bushy dichotomous or pinnate; or spirally arranged. The polyps can be retractile into the coenenchyme, or non-retractile, being in the latter case supported and protected by an anthocodial armature of calcareous sclerites that can even develop into an operculum closing the soft parts of the zooid. The individuals are arranged on the stem and on the branches disposed in some cases on the sides, alternating, in pairs, or forming regular whorls.

The Pennatulacea is the most complex order of Octocorallia, with a high level of colonial integration, polymorphism and extensive functional specialization of zooids. The primary individual (oozooid) is generally or bilaterally surrounded by daughter polyps (autozooids and siphonozooids). The founding polyp only functions as support to the daughter polyps and does not reproduce sexually, nor feed or take water. These functions are fulfilled by the specialized vegetative polyps.

The last order, the Helioporacea (= Coenothecalia), is a special case among the octocorals. It can be easily mistaken as a scleractinian because, unlike the other species of the subclass Octocorallia, the species of Helioporacea build up a massive skeleton and are able to form reefs. Despite its hermatypic characteristic, the Helioporacea is referred to the subclass Octocorallia as it shares the same anatomical characters of the other orders of Octocorallia. This order is known to be represented by only two Recent species, *Heliopora coerulea* (Bayer, 1973; Babcock, 1990) and *Lithotelesto micropora* (Bayer, 1981c).

4.1.1. - ASEXUAL REPRODUCTION

Several modes of asexual reproduction have been observed in octocorals and include budding, pinnitomy, transverse fission and autotomy. The most widely known is budding, in which the new individual derives from outgrowth of the solenia of the parent colony. In this case no new colonies are formed, and only the colony size is increased.

Asexual reproduction not only contributes to the enlargement of a colony by increasing the number of zooids but can also have part of the same function as sexual reproduction in the formation of new colonies. Pinnitomy has only been observed in *Anthelia* and involves the production of new colonies from detached pinnules (Gohar, 1940). Transverse fission was observed by the same author in *Heteroxenia fuscescens*. In this mode of reproduction a polyp becomes constricted and the lower half develops a new set of tentacles and the upper portion will settle down and form a new colony. Autotomy differs from other forms of asexual reproduction because it involves constriction or fragmentation of colonies rather than individual polyps.

Autotomy or colony fragmentation have been registered for many octocorals, as well as antipatharians, by natural fragmentation of branches (*Antipathes dichotoma*), fragmentation by tidal exposure or storms (*Plexaura* spp) or by longitudinal fission (*Lobophytum confertum, L. rigidum, Sinularia and Sarcophyton*) (Cary, 1919; Fishelson, 1973; Grigg, 1976; in Walker & Bull, 1983). Autotomy was also observed by Walker & Bull (1983) in the unbranched gorgonian *Juncella fragilis* which constricts a subterminal point of its axis releasing a daughter colony and avoiding excessive growth. Another peculiar mode of reproduction is the "migratory behaviour" of *Xenia macrospiculata* which multiplies asexually by colony fission and opportunistically captures space, by detaching from the original position and adhering to adjacent substrata (Benayahu & Loya, 1985).

4.1.2. - SEXUALITY

Unlike scleractinians, which have been known to present a large variety of reproductive patterns, octocorals had, until recent years, been thought to be exclusively gonochoric. With the growing interest in the reproductive biology of octocorals, knowledge of the variety of reproductive patterns in this group has increased. Hermaphoditism has been reported for several species or within species from the same area (previously found only to be gonochoric), and this mixed breeding was also observed for the same species inhabiting

different areas. The occurrence of parthenogenesis was also registered for some members of this subclass. Gonochorism, however, has been shown to be the dominant pattern among all the orders of Octocorallia.

Our knowledge of sexual reproduction among the order Stolonifera is limited to the genus *Clavularia*, which has been recorded as gonochoric (Gohar, 1948; Benayahu, 1989). The order Telestacea remains poorly known and no record is available. For the order Alcyonacea, the families Alcyoniidae, Xeniidae and Nephtheiidae have received better attention and gonochorism, hermaphroditism and even parthenogenesis have been observed for these families.

For the family Alcyoniidae, gonochorism has been the most commonly observed sexual feature as demonstrated in *Alcyonium digitatum, Lobophytum crassum, Sarcophyton glaucum, and Parerythropodium fulvum fulvum* (Hickson, 1895; Matthews, 1916; Hartnoll, 1975; Yamazato *et al.,* 1981; Benayahu & Loya, 1983; 1986; and Uehara *et al.,* 1987) although hermaphrodite specimens of *Alcyonium digitatum* have occasionally been found (Matthews, 1916; Hartnoll, 1975). In *Alcyonium hibernicum* only female colonies were found and in this case the ova developed parthenogenically (Hartnoll, 1977).

The family Xeniidae is mostly represented by gonochoric species but hermaphoditism is not uncommon and to date has been recorded in six species: *Xenia viridis* (Gohar, 1940), *Xenia novaebritanniae, Xenia grasshoffi, Heteroxenia fuscescens, H. elizabethae,* and *H. coheni* (Benayahu, 1991). In *H. fuscescens* hermaphroditism is related to dimorphism and siphonozooids develop in connection to female gonads (Gohar & Roushdy, 1961; Benayahu *et al.,* 1989). The development of siphonozooids in the colony is size-dependent owing to the allocation of energetic reserves initially to growth and subsequently to reproduction (Achituv & Benayahu, 1990). Small colonies are monomorphic and male, and when the colony has attained a certain size, siphonozooids and female gonads develop. Achituv & Benayahu (1990) suggested that dimorphism in this species is essential for brood care as the larvae will be brooded externally in the intersiphonozooid spaces. Siphonozooids are also believed to increase the colony surface-area and consequently the energy assimilation, which can then be allocated to planula development. In dimorphic colonies female gonads are predominant and male gonads are relatively sparse (Achituv & Benayahu, 1990).

In the genera *Anthelia, Sympodium, Efflatounaria* and *Xenia,* gonochorism associated with monomorphism is the common pattern with the exception of the *Xenia* spp mentioned above,

which are hermaphrodite (Gohar, 1940; Dinesen, 1985; Benayahu & Loya, 1989; Benayahu, 1990; Benayahu, 1991).

The occurrence of a mixed breeding system has been reported for the species *Heteroxenia elizabethae* which was found to be hermaphrodite in the Red Sea and gonochoric at the Great Barrier Reef, Australia (Benayahu *et al.*, 1989). The same mixed breeding was suggested for *Heteroxenia ghardaqensis* (Benayahu, 1991).

In the species of the order Gorgonacea studied to date, gonochorism has been the dominant sexual status (Theodor, 1967; Grigg, 1970; 1977; Goldberg & Hamilton, 1974; Beheti Gonzalez & Guardiola, 1979; Weinberg & Weinberg, 1979; Brazeau & Lasker, 1990), although parthenogenesis and hermaphroditism were not ruled out. Brazeau & Lasker (1989) studying an undetermined species of *Plexaura (Plexaura* A) noticed a complete absence of males among the examined colonies and assumed that female colonies probably reproduced parthenogenically. A similar species, *Plexaura homomalla*, has only been recorded as gonochoric (Goldberg *et al.*, 1974; Beheti-Gonzalez *et al.*, 1979). Goldberg & Hamilton (1974) found little evidence for "transsexuality" in *P. homomalla* and preferred to refer it to collecting error. Reproduction of primnoids will be discussed later in this chapter (see Section 4.1.6).

For the order Pennatulacea, only gonochoric species have been reported (Wilson, 1883; Hyman, 1940; Chia & Crawford, 1973; Achituv & Benayahu, 1990; Rice *et al.*, 1992) and the helioporacean *Heliopora coerulea,* was also found to be gonochoric (Babcock, 1990).

The sex-ratio in alcyonaceans varies between species, between different geographic area within the same species and even with different bathymetric distributions. For sessile brooding species it seems to be advantageous if the male colonies outnumber the female colonies increasing the chance of success of internal fertilization, which is thought to be the case for *Xenia macrospiculata* and the same is shown for the Nephtheidae *Capnella gaboensis* where the sex-ratio favours male colonies (Farrant, 1985). In *Parerythropodium fulvum fulvum* the sex-ratio in deeper waters is skewed towards male colonies and in shallow waters favours female colonies. Benayahu & Loya (1983) believe the latter case to be caused by the formation of local colony aggregation via asexual reproduction.

4.1.3. - REPRODUCTIVE MODE

Based on the available data, it is hard to define the dominant reproductive mode among octocorals. Brooding, internal fertilization with surface brooding, broadcasting and parthenogenesis with broadcasting have been reported for the different orders of Octocorallia. The stoloniferan *Clavularia crassa* studied by Kowalevsky and Marion (1883) was found to be a brooder and another species of the same genus, *Clavularia hamra* was reported by Gohar (1948) as broadcaster and by Benayahu (1989) as surface brooder. Still within the same order, the Cornulariidae *Cornularia sagamiensis and C. komaii* also showed surface brooding as their reproductive mode (Benayahu, 1989).

In alcyonaceans, the reproductive mode can be broadcasting, internal brooding or surface brooding. *Alcyonium digitatum* spawns, releasing the eggs to the surrounding water where fertilization takes place and a pelagic planula develops. The fleshy and massive species *Sarcophyton glaucum and Lobophytum crassum* are also broadcasters and produce pelagic planulae (Yamazato *et al.,* 1981; Benayahu & Loya, 1983). *Alcyonium hibernicum and A. siderium,* conversely, brood their embryos, releasing an already mature crawling demersal planula. *Parerythropodium f. fulvum,* an encrusting subspecies, is a surface brooder which keeps its embryos entrapped in mucus on the surface of the colony (Benayahu & Loya, 1983).

In Xeniidae, internal brooding and surface brooding are found. *Xenia umbellata* develops its embryos in endodermal brooding chambers located at the distal part of the polyp cavity (Benayahu *et al.*, 1988). The embryos of *Heteroxenia fuscescens* mature externally between the siphonozooids (Achituv & Benayahu, 1990) and in the monomorphic genus *Efflatounaria,* the eggs remain attached to the surface of the colony between the swollen polyps (Dinesen, 1985). Surface brooding was also recorded for the nephtheiid *Capnella gaboensis* (Farrant, 1985). Benayahu (1990) stated that no hermaphrodite spawners have been found among alcyonaceans.

Internal fertilization with surface brooding has been the common reproductive mode found in the order Gorgonacea as demonstrated for *Briarium asbestinum* (Brazeau & Lasker, 1990); *Eunicella singulares* (Weinberg & Weinberg, 1979); *Muricea californica, M. fruticosa* (Grigg, 1970; 1977). In these species, the embryo is released from the polyp early in development, shortly after fertilization, and it remains on the surface of the colony until development is accomplished. The embryos of the parthenogenic species *Plexaura* A initiate their larval development while still inside the polyp and are then released from the polyp to the surrounding water without being brooded, being considered by Brazeau & Lasker (1989) as

broadcasters. An unidentified species of the genus *Leptogorgia* studied by Wilson (1883) was also found to be a broadcaster. The same was said for the genus *Gorgonia* (Grant, 1826).

From the few, but noteworthy, reports on reproduction of pennatulaceans, one might gather that the common mode of reproduction is broadcasting. The studies of Grant[1] (1829) on *Virgularia miriabilis,* despite not being very clear, provide evidence of this species being a broadcaster. The same was reported for *Renilla reniformis* (Wilson, 1883) and for *Ptilosarcus guerneyi* (Chia & Crawford, 1973)

The embryos of the helioporacean *Heliopora coerulea* start development internally and are posteriorly brooded on the surface of the polyp within a temporary brood-space created by highly distended polyps (Babcock, 1990).

Recent studies on secondary metabolites in octocorals have suggested that the presence of secondary compounds, primarily terpenes, contribute to the reproductive success of these animals and might be important in the evolution of the different reproductive patterns presented in this group. In asexual reproduction, toxic secondary metabolites, which function as anti-predator and anti-competitor adaptations and in reproduction, are transferred from the parent directly to the new individuals through fragmentation, production of stolons, or simple division and colony growth. In sexual reproduction little is known, but there is some evidence that some compounds (e.g. diterpene thunbergol, 3,4-epoxynephthenol) are associated specifically with the process of reproduction and are just found in the eggs and not in the tissue of the parent colony. These compounds, absent in the coral tissue of some species, increase concentration during the period immediately preceding ovulation and disappear after egg release. They also disappear from the larva within a week. Despite the limited information available, it is suggested that these compounds might act as chemical trigger for release of the eggs or as sperm attractant (Sammarco & Coll, 1988).

4.1.4. - ONSET OF MATURITY AND FECUNDITY

The onset of maturity is believed to vary according to the division of the limited energetic resources between metabolic activities, somatic growth and reproduction. Hartnoll (1975) has shown in his studies of *Alcyonium digitatum* that specimens which inhabited exposed areas

---

[1] Grant (1827) stated: "These ova...are certainly not generated by the polypi themselves, as we might be led to believe by some authors, as Pallas (*El. Zooph. 362*) who state as a character of these animals that their polypi are oviparous. The ova in almost every known zoophyte are formed by the common connecting substance of the animal, and not by the polypi, which appear to be only the mouths or organs of digestion" - which he refers to later as "faecal orifices". Grant goes further in this reasoning in his next paper (1829) where he suggests that these "faecal orifices" or "vesicles" were developed to optimize fecundity.

mature later (on the third year or later) but attain larger size because the available energy was preferentially allocated to somatic growth. Conversely, specimens in more sheltered areas reach maturity earlier (in their second year) in a smaller size. Male colonies might also reach maturity at a smaller size than female colonies as seen in the gorgonian *Briarium asbestinum*. Based on this fact, Brazeau & Lasker (1990) assumed that it is less costly to produce a functional spermary than to produce a single large egg. The dimorphic Xeniidae *Heteroxenia fuscescens* develops male colonies in less than one year but they are only dimorphic every four years when female gonads are developed (Achituv & Benayahu, 1990). The gorgonians *Plexaura* A and *Muricea californica*, and *M. fruticosa* attain maturity at a later age than the species mentioned above, varying from 5 to 10 years as seen in Table 4.1.

Table 4.1 - Onset of maturity and fecundity of some octocorals.

SP	ONSET OF MATURITY (yr)	FECUNDITY	AUTHOR
Clavularia hamra	_	14-26 eggs/pol/yr	Benayahu, 1989
Alcyonium digitatum	2 - 3	_	Hartnoll, 1975
Alcyonium hibernicum	1	_	Hartnoll, 1977
Heteroxenia fuscescens	- less than a year; - 4 years	_	Achituv & Benayahu, 1990
Xenia macrospiculata	_	15-30 embryos/pol/yr	Achituv & Benayahu, 1990
Capnella gaboensis	2 - 3	5-10 eggs/pol/yr	Farrant, 1985, 1986
Briarium asbestinum	2 - 3	2.73 eggs/pol/yr	Brazeau *et al.*, 1990
Plexaura A	5	1.9± 0.99 eggs/pol/yr	Brazeau & Lasker, 1989
Muricea californica	5 - 10	1.6 eggs/pol/yr	Grigg, 1977
Muricea fruticosa	5 - 10	3.8 eggs/pol/yr	Grigg, 1977
Pseudopterogorgia	_	6-10 embryos/pol/yr	Brazeau & Lasker, 1989
Eunicella singulares	_	6000 eggs/colony/yr	Weinberg & Weinberg, 1979
Ptilosarcus guerneyi	_	200.000 eggs/col/yr	Chia & Crawford, 1973
Heliopora coerulea	_	1.34 ± 0.8 embryos/pol/yr	Babcock, 1990

Colony fecundity will depend on the size of the colony and the number of eggs per polyp. The number of eggs per polyp will be determined by the length of the polyp cavity (Benayahu & Loya, 1984). For convenience, fecundity is usually based on number of eggs per polyp, rather than the overall number of eggs per colony. Fecundity estimates would be more reliable if

based upon the number of larvae produced rather than egg number (as pointed out by Brazeau & Lasker, 1990), since the number of eggs produced initially (potential fecundity) is not necessarily the same number produced eventually (actual fecundity). The stoloniferan *Clavularia hamra*, produces several eggs but only a certain number will reach maturation while some others will be absorbed before maturation (Benayahu, 1989). The number of oocytes per polyp in *Briarium asbestinum* also decreases prior to spawning which indicates that females increase reproductive effort and fitness by increasing egg size rather than the number of eggs per polyp (Brazeau & Lasker, 1990).

Low reproductive output in some species is offset by improved reproductive efficiency that will ensure higher survival rates. In *Alcyonium hibernicum*, early maturity, brood care, suppression of male colonies as it develops its ova parthenogenically, and benthic habit of the planulae compensate its low fecundity optimizing its survival capacity (Hartnoll, 1977). The same was shown for the gorgonians *Plexaura* A, *Muricea californica* and *M. fruticosa* which are known to have low fecundity (Grigg, 1977) (Table 4.1). However, low fecundity in *Plexaura* A might be offset by developing ova parthenogenically and in *Muricea* spp by surface brooding their embryos. Babcock (1990) suggests that brooding is an alternative to enhance early survival avoiding larval predation and compensating for low fecundity.

Some gorgonians such as the winter breeding *Pseudopterogorgia*, *Paramuricea clavata* (Gili & Garcia, 1985) and *Eunicella singulares* (Weinberg & Weinberg, 1979) have higher fecundity (Table 4.1). Some pennatulaceans are believed to be also considerably fecund (*Ptilosarcus guerneyi:* Chia & Crawford, 1973; *Kophobelemnon stelliferum:* Rice *et al.*, 1992). Fecundity in this case is more sensibly measured per colony rather than per polyp, as the integration of the colony is more complex and the individual polyps are less independent than in the other orders of this subclass.

4.1.5. - GAMETOGENIC CYCLES

Annual cycles of development are known to be dominant among octocorals (Table 4.2), although in the orders Alcyonacea and Gorgonacea there are some cases of prolonged two-year period of oogenesis with overlapping generations, as seen in *Lobophytum crassum* (Yamazato *et al.*, 1981; Benayahu *et al.*, 1990); *Corallium rubrum* (Vighi, 1972) and in the undetermined parthenogenic species *Plexaura* A (Brazeau & Lasker, 1990). *Sinularia humesi, S. leptoclados, S. mayi* and *Litophyton arboreum* have oogenic cycles of 18-24 months

(Benayahu *et al.*, 1990). Prolonged oogenesis as suggested by Benayahu *et al.* (1990), leads to high parental energy expenditure in sexual offspring, increasing the yolky content of the eggs and probably contributing to planula dispersal.

The spermatogenic cycle is usually shorter than the oogenic cycle as shown for the summer breeding species *Plexaura homomalla* (Goldberg & Hamilton, 1974; Beheti-Gonzalez & Guardiola, 1979) and *Briarium asbestinum* (Brazeau & Lasker, 1990). Summer breeding has been more commonly observed, although some species breed in winter eg *Alcyonium digitatum* and *Pseudopterogorgia* sp. For *Alcyonium digitatum,* Hartnoll (1975) suggests that since the planulae do not feed they do not suffer by the scarcity of plankton in winter and benefit by the decreased number of predatory zooplankton. They have a long pelagic life and high dispersal capability, and when they settle and metamorphose they will take the advantage of the abundant food brought with spring. The summer breeding species, conversely, do not usually have pelagic larvae, being mostly brooders or surface brooders, releasing demersal planulae and avoiding planktonic predators.

Differences in breeding periods have also been recorded, as seen in the two forms of *P. homomalla, P. h. typica* (Florida: Goldberg & Hamilton, 1974) and *P. h. kuekenthali* (Cuba: Beheti-Gonzalez & Guardiola, 1979). The Caribbean species has a longer reproductive period, probably influenced by a difference in temperature in the two areas as suggested by Beheti-Gonzalez *& Guardiola* (1979). The breeding season might vary as well with depth but still related to temperature, with surface colonies spawning earlier than deeper ones as seen in the summer breeding species *Eunicella singulares* (Weinberg & Weinberg, 1979) and in *Muricea californica* and *M. fruticosa* (Grigg, 1970; 1977). Grigg (1977) considered temperature as the major factor controlling spawning and affecting maturation.

The pennatulacean *Ptilosarcus guerneyi* is reported to spawn in spring or summer (Chia & Crawford, 1973). For *Renilla reniformis*, Wilson (1883) observed successive broods of eggs in a single year.

The development of oocytes and spermaries varies throughout the year between different species. There are periods of slow growth and periods of rapid development. In *A. digitatum* little happens during the first months (winter) of development of sperm sacs, and in spring and summer they develop rapidly in size followed by a period of slow growth (4-5 months; autumn-winter). The oocyte reaches maximum size and is stored for four months before being released. During this period the colonies enter a quiescent phase, remaining contracted

and do not feed (Hartnoll, 1975). The ova of *A. hibernicum* has a development period of 12 months, spawning in late summer. It develops rapidly in autumn and has a slow growth in winter and spring, attaining its maximum size in early summer. The larva, which develops parthogenetically, is stored for a month until late summer when it is then released (Hartnoll, 1977).

In xeniids the life cycle is varied. *Xenia umbellata* follows an annual cycle of development with seasonal spermatogenesis, continuous oogenesis and a seven month period of planulation per year. *Xenia macrospiculata* also reproduces annually, the spermatozoan are released in May and the planulation period lasts for 4 to 5 months (May-September) (Benayahu, 1991). *Heteroxenia fuscescens* does not follow any annual pattern having continuous gametogenesis and planulae are released all year round (Benayahu *et al.,* 1989; Achituv & Benayahu, 1990; Benayahu, 1991).

Octocorals are known to produce large eggs, compared to the size of the polyps. Some oocyte diameters are listed in Table 4.2. The eggs of the broadcaster *Renilla reniformis* seem to have one of the smallest diameters (350 µm, Wilson, 1883). The average oocyte diameter seems to range from 500 to 600 µm and is mainly reported for internal brooders. Most of the largest oocytes mentioned in Table 4.2. are reported for the surface brooders *Heteroxenia fuscescens* (900 µm - Benayahu *et al.*, 1989); *Briarium asbestinum* (up to 900 µm - Brazeau & Lasker, 1990); *Heliopora coerulea* (840 to 920 µm - Babcock, 1990) and in the genus *Efflatounaria* the oocytes can reach a diameter of 1 mm (Dinesen, 1985).

Surface brooding is commonly seen in whip-like or encrusting colonies that have a characteristic thin coenenchyme, e.g. *Pararerythropodium fulvum fulvum, Clavularia crassa, Cornularia komaii, and C. sagamiensis* (Kowalewsky *et al.*, 1883; Suzuki, 1971; Benayahu & Loya, 1985). In these encrusting species the polyp cavities are short and, since their eggs are large, if embryogenesis were internal the fecundity of the animal would be even lower (Benayahu & Loya, 1985). These authors deduced that space limitation and consequent low fecundity would be compensated by the protection provided by this reproductive strategy, maximizing the survival of the few larvae produced.

Sperm sacs can be considerably large as well seen in *A. hibernicum* where the maximum size registered for sperm sac was 960 µm (Hartnoll, 1975).

Table 4.2 - Development cycle of some octocorals. Data extracted from existing literature.

SP	DEVELOP. CYCLE	OOGENESIS	SPERMATOGENESIS	SPAWNING	EGG SIZE (μm)	AUTHOR
Clavularia hamra	ANNUAL	11 months (SEP-AUG)	8 - 10 months (OCT-DEC)	SUMMER (AUG)	600-750 (: 250-550)	1, 2
Alcyonium digitatum	ANNUAL	9(3) - 12(4) months	-	MID-WINTER DEC-JAN (4,5) DEC-FEB (6)	500(3,6) 600 (-960)(4)	3, 4, 5, 6
Alcyonium hibernicum	ANNUAL	12 months	-	LATE SUMMER (AUG-SEPT)	530	7
Heteroxenia fuscescens	continuous gametogenesis; planulae released all year round				900	8, 9
Xenia macrospiculata	ANNUAL	spermatozoon released in May; planulation period lasts for 4-5 months (May-Sept)				9
Efflatounaria	-	-	-	-	1 mm	10
Capnella gaboensis	ANNUAL	6 - 7 months	-	EARLY WINTER (MAY-JUNE)	492±69	20, 21
Briarium asbestinum	ANNUAL	12 months	3-5 months	-	600-900	11, 12
Plexaura homomalla	ANNUAL	11 - 12 months	3 months			13, 14
Plexaura A	2 YR CYCLE	-	-	-	500-600	11
Corallium rubrum	2 YR CYCLE	-	-	-	-	15
Renilla reniformis	successive broods of eggs were observed in a single year				350	16
Ptilosarcus guerneyi	-	-	-	SPRING OR SUMMER	500-600	17
Kophobelemnon stelliferum	-	-	-	-	800	18
Heliopora coerulea	ANNUAL	-	-	SUMMER	840-920	19

1 - Gohar, 1948
2 - Benayahu, 1989
3 - Hickson, 1895
4 - Hartnoll, 1975
5 - Hill, 1906
6 - Matthew, 1916
7 - Hartnoll, 1977
8 - Benayahu *et al.*, 1989
9 - Benayahu, 1991
10 - Dinesen, 1985
11 - Brazeau & Lasker, 1989
12 - Brazeau & Lasker, 1990
13 - Goldberg & Hamilton, 1974
14 - Beheti-Gonzalez & Guardiola, 1979
15 - Vighi, 1972
16 - Wilson, 1883
17 - Chia *et al.*, 1973
18 - Rice *et al.*, 1992
19 - Babcock, 1990
20 - Farrant, 1985
21 - Farrant, 1986

4.1.6 - NOTES ON REPRODUCTIVE ASPECTS OF ANTARCTIC AND DEEP-SEA OCTOCORALS

The reproductive biology of the deep-water and Antarctic octocorals is virtually unknown apart from a few notes. The species of *Dasydogorgia,* abundant in deep waters, have developing sexual cells in the basal portion of the polyp, which are apparently distended (Wright & Studer, 1889). These authors stated that a dioecious condition appeared to be predominant and in some colonies all the polyps contained eggs. In *Dasydogorgia flexilis*, Wright & Studer noticed that the shape of the polyp was affected by its reproductive state and pointed out that "this form...cannot therefore be regarded as specifically characteristic, since it is dependent

upon the maturity of the sexual organs at the time". In the Primnoidae *Primnoella* sp and *Primnoa flagellum,* also studied by Wright & Studer (1889), the colonies were shown to be dioecious.

Thomson (1907) described the primnoid *Primnoa reseda* as a brooder. He observed clusters of ova at different stages of development with a very definite envelope attached to the mesentery via a stalk. The larva attained a length of 800 µm and width of 400 µm. Kukenthal (1924 in: Goldberg & Hamilton, 1974) found the same species, then named *Primnoa reseadaeformis,* to be hermaphrodite. Gravier (1913) gave a brief account of the reproductive biology of Antarctic octocorals based on existing data, showing that brooding is the common reproductive mode recorded for Antarctic octocorals. *Mopsea gracilis* was observed to have large eggs occupying almost the whole polyp cavity and *Mopsea elongata* produced eggs which were 650 µm in diameter (Roule, 1908). Other brooding species recorded were *Plumarella delicatissima* (Verluys, 1906), *Rhapalonella pendulina* and *Pellastisis* (Nutting, 1910). Notes by some other authors only suggested the presence of ova or the swollen state of the polyp but this sort of information does not contribute at all to the knowledge of reproductive biology of these animals. From the limited data presented above, one might consider that gonochorism associated to brooding is the trend in Antarctic and deep-sea octocorals.

4.1.7 - OBJECTIVES OF THE CURRENT STUDY

The primary objectives of this study were to define the main features of the reproductive biology of the primnoid *Thouarella variabilis*, concerning sexuality, reproductive mode, onset of maturity, fecundity, gametogenesis, gametogenic cycles, distribution of reproductive stages throughout the colony, morphology of eggs, sperm sacs and larvae and the influence of the reproductive state on the external morphological features of the polyps. Lack of sufficient material hindered extensive studies of some of these aspects. The research was concentrated mainly on major areas where sufficient data were available such as gametogenesis and morphology of the reproductive products.

4.2 - METHODOLOGY

4.2.1 - PRESERVATION

As mentioned in Chapter 2, most of the specimens had been fixed in formalin (10%) and then transferred to alcohol (70°GL) in which they remained stored for many years prior to further processing. For histological studies the damage of the sclerites caused by formalin is not an obstacle for the investigation since the calcareous structure will have to be removed anyway. For ultrastructural studies, however, the ideal procedure would be to have the samples fixed in gluteraldehyde and postfixed in osmium tetroxide; unfortunately, there was no such special interest at the time the samples were collected. Formalin-fixed specimens were the only available material for this study.

4.2.2 - DISSECTION

The polyps were dissected and examined under a binocular stereomicroscope for the presence of oocytes, sperm sacs or embryos. Based on this investigation it was possible to obtain, the following information: colonial and polyp sex; reproductive mode; stage of reproduction; distribution of the different reproductive stages along the colony; onset of sexual maturity and fecundity.

The colonies were systematically divided into three regions: distal (top of the colony); middle; and proximal (base). The branches from each region were divided into two portions: peripheral (tip) and internal (middle and base of the branches). Five polyps were removed from each portion, therefore, ten polyps were taken from each region and thirty from each colony. If the information acquired was ambiguous and depending on the state of preservation of the colonies, more polyps were examined under the dissecting microscope or confirmed histologically.

The polyps were decalcified in Bouin's solution for at least 12 hours and then transferred to alcohol. Some polyps were so transparent when decalcified that all the internal structures were perfectly visible and dissection was unnecessary. These intact polyps were saved for further microscopical study.

Under a stereomicroscope the small and fragile decalcified polyps were longitudinally torn apart by means of very flexible forceps, microscissors and microscalpels. The presence, number and position of the oocytes, sperm sacs or embryos were recorded. The male and female reproductive development were classified into five and seven stages respectively, according to their size and external appearance. The diameter of oocytes, sperm sacs and embryos were measured with an eyepiece micrometer.

To determine the relationship between the external appearance of the polyp and its reproductive stage, measurements of all the external and internal features of the polyp were recorded with an eyepiece micrometer. Measurements of the length of the polyp, diameter of the calyx, width of the neck, or median region and width of the base were used in an attempt to build a pattern of external proportions of the polyp. Internal measurements such as the diameter of the pharynx, median region and gastric cavity can demonstrate any the correlation between the consequent expansion of the internal structures and the number and size of eggs or sperm sacs. The study of the expansion and consequent external appearance of the polyp could be used to infer, only through external observation, whether the polyp bears reproductive products or not; although it does not make possible a reliable definition of the exact reproductive stage. Further microscopical investigation is necessary to ensure the definition of the reproductive stage. The data collected in this part of the study could provide additional information about the capability of expansion of the soft parts of the polyp and of its calcareous armature.

4.2.3 - LIGHT MICROSCOPE HISTOCHEMISTRY

Histological sections were used for the determination or confirmation of sex, development state and investigation of the structure of the gonad. The system of study was the same as that used for the dissection. Polyps of different regions of the colony, portions of the branches and of different sexes and reproductive stages were examined.

Considering the small size (around 1 mm long) and fragility of the polyps, special histological treatment and processing techniques were adapted. The samples were kept in the same container during the whole treatment to facilitate the handling. Small jars (1cm x 4 cm) with plan base were chosen as containers and not more than three polyps were placed in the same jar.

EMBEDDING - After being decalcified in Bouin's solution, the tissue was dehydrated through ascending concentrations of ethanol: 30% (30 min.); 50% (30 min.); 70% (30 min.); 90% (30 min.); twice in 100% (30 min.); and twice in histoclear (30 and 45 min. respectively). The samples were embedded in paraffin wax, which was replaced four times at intervals of 1 hour, whilst kept in an oven at 65°C. The samples were then removed from the oven to a hot plate where the polyps were positioned at the desired angle by using a hot pin. When the wax was solidified, the jars were carefully broken with a nut-cracker so the block of paraffin could be released. The blocks of paraffin were then lapidated to a suitable size and attached to wooden blocks by melting their bases. The preparations were sectioned transversely or longitudinally at 5 μm (occasionally at 10 μm) and mounted on glass slides.

STAINING - As a first attempt, some sections were stained with haematoxylin and eosin, but better results were attained with Masson's trichrome. The staining procedure was as follows: the sections were cleared in histoclear for 5 min.; gradually hydrated from 100% to 90% alcohol; and transferred to: haematoxylin (15 min.); running water (15 min.); ponceau and acetic acid (5 min.); rinsed in distilled water; phosphomolybdic acid and phosphotungstic acid (1 min.); rinsed in distilled water; fast green (F.C.F.) and acetic acid (2 min.); distilled water; gradually dehydrated to 100% alcohol; cleared in histoclear and mounted as permanent slides with DPX.

4.2.4 - SCANNING ELECTRON MICROSCOPY (S.E.M.)

Electron microscopy was used to improve the understanding of the ultrastructure of oocytes, sperm sacs, embryos, and interior features of the polyps. The observations made under an electron microscope clarify and complete the information obtained in dissections and light microscopy. Polyps of different sexes and reproductive stages were examined.

After being decalcified, the specimens were dissected longitudinally under a stereomicroscope to expose the gastric cavity, mesenteries and the reproductive products present. Some eggs and embryos were left intact and some were cut transversely or just had their membrane torn apart so the internal structures could be observed and sperm sacs were opened to expose the spermatozoa. Fine microscalpels and a pin made of optical fibre were used to handle such minute structures. Eye-lashes were used to clean the surface of the eggs and sperm-sacs which were then prepared for further processing.

The specimens preserved in 70% alcohol were dehydrated gradually to 100% alcohol and critically point dried with CO_2. Once dry, the polyps, eggs, embryos and sperm sacs were mounted and positioned on aluminium stubs and were adhered to it by a carbon impregnated film. Some preparations were coated with gold/palladium but most were coated with 20 nm of gold in a Hummer VI sputter coater. The preparations were examined with three different scanning electron microscopes (S.E.M.): JSM - P15 (JEOL); ISI 60A and JSM 6400 (JEOL) at an acceleration voltage of 20 kv.

4.2.5 - TRANSMISSION ELECTRON MICROSCOPY (T.E.M.)

Transmission electron microscopy was used for obtaining supplementary information about the ultrastructure of germ cells, nurse cells and the first stages of oocytes. It was important as well to clarify the peculiarities of the different membranes. It was not possible to obtain such information using light microscopy. The material was, however, badly preserved and only limited information could be obtained.

The TEM processing involves the embedding of the tissues in epoxy resin, which is hard enough to enable thin sections to be cut, and the staining of the tissues with heavy metal salts to improve contrast.

Embedding procedure: The tissue was placed in 2% osmium tetroxide solution (2 hours); rinsed in distilled water; transferred to 2% uranyl acetate (30 min.); rinsed in distilled water; dehydrated with ethanol - 70% (10 min.), 90% (10 min.), absolute ethanol (3 X 10 min.); placed in histosol (30 min.); transferred to 50% histosol: 50% resin (1 hour); and soaked in spurr's resin (24 hours). The resin was polymerised by being heated in an oven at 60°C for 16 hours.

The solid block of resin was posteriorly sectioned (around 70 nm) and the sections examined with a transmission electron microscope, Hitachi H - 7000.

The data recorded from the dissection studies added to the information obtained from the microscopical studies were analysed statistically and are presented in the following section.

4.3 - RESULTS

4.3.1. - GENERAL FEATURES OF THE REPRODUCTIVE BIOLOGY

Those specimens of *Thouarella variabilis* examined showed negligible evidence of hermaphroditism, both colony and polyps being prevalently gonochoric. Only one colony had both male and female polyps. The colonies were, with a few exceptions, in reproductive phase. Not all the polyps in a ripe colony were breeding, but the polyps which were bearing gonads presented similar reproductive stages all along the colony.

The presence of larvae inside the polyp cavities of many examined colonies indicates that brooding is the reproductive mode. The fecundity of this species seems to be very low, with only one larva being released per female polyp at a time. However, brood care provides protection to the embryo optimizing its survival, which might compensate for the low fecundity, ensuring successful colonization. The presence of oocytes in different stages of development within the same polyp suggests a two year cycle of oogenesis or continuous gametogenesis with release of larvae occurring throughout the year.

The first reproductive stage of either male or female polyps could be identified by the notably swollen appearance of the mesenteries in the gastric cavities, where the gonads will be developed (Figs. 1-3; 65-69; 98-101; 106). The germ cells, present in the mesenterial endoderm, migrate into the mesenterial mesoglea. Gonads can be defined for this species as thickenings of the mesentery which will eventually give rise to the sperm sacs in the male polyps and will support the development of the oocyte in female polyps. Only the four lateral mesenteries were observed to bear developing genital cells. The sperm sacs and oocytes seem to be developed preferentially on the two lateral mesenteries which are close to the asulcal mesenteries on the adaxial side of the polyp (Figs. 13-15; 56, 57; 72-74) although sperm sacs have also been observed in other lateral mesenteries (Fig. 16).

4.3.2 - DEVELOPMENT OF MALE GONADS AND SPERMATOGENESIS

Gametogenesis was classified in different stages, according to difference in size and external and internal structures.

FIRST REPRODUCTIVE STAGE

The small membrane-bound aggregates of spermatogonia present in the mesenterial mesoglea increase in number as the spermatogonia divide mitotically into primary spermatocytes. As a consequence, the spermatocytes, searching for space, bulge into the coelenteric cavity drawing around themselves the mesoglea in which they are embedded and the outer layer of mesenterial endoderm. This expansion of the mesentery happens at the lowest portion of the two lateral mesenteries which are next to the asulcal mesenteries (Figs. 2, 3).

From being an expansion of the mesentery, the sperm sac starts its development already with considerable size (around 100 μm). The first recognizable sperm sac was oval, up to 120 μm in length and around 80 μm in width, and surrounded by a wrinkled endoderm which is continuous with the mesenterial endoderm (Fig. 4).

The comportment of the endoderm during the process of development of the sperm sac is not clear. The endoderm of the primary sperm sac resembles the stretched layer of endodermic cells covering the dilated mesenteries. The cells cannot easily be distinguished from each other and cilia are present along the whole surface of the sperm sac.

SECOND REPRODUCTIVE STAGE

As development progresses, the sperm sac becomes annular or cylindrical, slightly flattened on the top and with a central invagination (Figs. 5, 6). It is still strongly linked to the mesentery and the sequence of development shown in Figures 4 to 9 suggests a continuous expansion of the oval or annular sperm sac into a rounded structure. A hollow central cavity is gradually developed as spermatogenesis progresses.

A thin mesoglea is clearly seen between the mass of developing gametes and the endoderm layer (Fig. 48). The endoderm looks very different when compared to the earlier stage (Figs. 10-12). The wrinkled and smooth endoderm present in the primary sperm sac gives way to a densely aggregated layer of cells. These endodermic cells, clearly distinguished from each other, are cuboidal, slightly rounded or oval, and heavily ciliated (Fig. 42b). They vary in size and are not uniform in shape. These cells are disposed very close together, which might influence their variation in shape.

Apparently, the endodermic cells of the sperm sac do not retain any similarity to the endodermic cells involving the dilated mesenteries; However, they are like the endodermic cells, covering those parts of the gastric cavity which did not suffer dilatation. Their different appearance could, therefore, be explained by the facts that these cells compose part of the endoderm which has recovered from expansion; and that the developing sperm sac is not completely filled with migrating spermatocytes.

The size of a developing sperm sac does not vary considerably while it is transformed into a spherical structure. It retains the longest dimension found in the oval structure. The size in which this transformation takes place varies, however, among individual gonads and ranges from 100 to 200 μm.

THIRD REPRODUCTIVE STAGE

By the time the sperm sac has reached a diameter of 200 μm, it is spherical in shape and has become delimited from the mesentery, although it is still continuous with it through a stalk, which is filled with spermatocytes (Figs. 20, 46). The endodermic cells are less close together than in the anterior stages but still markedly visible (Fig. 48). A hollow central cavity is clearly developed by now (Figs. 29, 46, 48), the spermatocytes have already started to divide meiotically into spermatids, and spermiogenesis is in progress. Spermatocytes show very large nuclei and sparse coarsely granular cytoplasm (Fig. 35). Spermatogonia and spermatocytes are not easily distinguished from each other and these cells described could indeed be either of them. The differentiation of spermatids into spermatozoa is shown in Figures 36 to 42. The central cavity is filled with spermatozoon tails as spermiogenesis progresses (Fig. 31).

The cytoplasmic continuity between the developing genital cells retained through the spermatogenesis might be responsible for the synchrony of development of these cells (Figs. 27, 28). Within the same sperm sac the cells share in most cases the same stage of development (Figs. 30, 49) but this synchrony is not kept amongst sperm sacs within the same polyp. Sperm sacs in different stages of development are commonly observed (Figs. 52-54). The spermatocytes present in the stalk offer a convenient base for comparison with the spermatids in the sperm sac (Fig. 51).

Initially, only two gonads are developed per polyp but when they have reached their later stages of development one or two others might start growing (Fig. 16). The first two developed gonads, preferentially risen on the lowest part of the most adaxial lateral

mesenteries, might grow simultaneously as seen in figs. 14 and 56 or might have unequal growth as seen in Figures 15 and 57 (which are taken from the same specimen). Depending on the enlargement of the gastric cavity, the gonads will be kept apart or compressed against each other as in Figures 15, 48 and 60. If space is available they will keep their spherical shape, otherwise the limited space will define their shape (Figs. 46, 48).

FOURTH REPRODUCTIVE STAGE

When the sperm sac reaches a diameter of around 300 µm, it already has a much thinner and stretched endoderm (Figs. 16, 34, 51). As size progresses the endoderm becomes more and more fragile and breakable. The central cavity, previously filled with spermatozoon tails, gradually disappears and spermatozoa are distributed all around the gonad (Figs. 31, 52-53). Despite spermiogenesis being mostly synchronized within a gonad, immature spermatozoa might be seen in parts of the periphery of the gonad while mature spermatozoa make most of the gamete mass. The mature spermatozoon has an elongated head and neck that together can reach a length of 7 µm (Figs. 32, 41 and 42).

In the later stages of gonad maturation the spermatozoa are distributed in radial rows parting from a certain point (Fig. 54). The gonadal endoderm is broken in some parts exposing the mesoglea which is thin and delicate (Figs. 33, 44). Developing spermatozoa can be seen forcing the mesoglea and exposing themselves in parts of the gonad surface (Figs. 20, 53).

FIFTH REPRODUCTIVE STAGE

The maximum size observed for a sperm sac was 740 µm, although maturation might be reached when over 500 µm, since mature spermatozoa are already developed at this size. The sperm sac is then ready to spawn. Examined under the stereo-microscope, the mature sperm sacs have a fluffy appearance and are easily damaged when handled (Figs. 18, 22, 53, 62, 63).

The aspect of the mesenteries changes considerably as the gonads grow. The dilated mesenteries (Fig. 1) are now barely noticeable (Figs. 19, 22). Whether the genital cells have all migrated to the formed gonads is not clear but the mesenteries have certainly lost most of their reproductive cell content. The pressure caused by the well developed gonads causes the mesenteries and the endoderm covering the gastric cavity to take on a compact appearance as if they were just one membrane protecting the reproductive products.

The sperm sacs are most probably expelled through the mouth owing to their large size and to the absence of gonopores or any other structure open temporarily for this function. Considering that most of the internal structures of the polyp were compressed to accommodate the reproductive products (Fig. 53), it is not difficult to understand the lack of any internal structure shown by some polyps, which are almost transparent (Fig. 55). The recovery of the polyps to a normal condition after spawning is an intriguing and unknown process. It is not rare to find sperm sacs compressed to the walls of the gastric cavity, as seen in Figures 59 and 60. This observation added to the transparency displayed by the polyp suggest that these two are not the first gonads produced by this polyp this season and that former gonads have been released. It is possible, therefore, to conclude that the polyp keeps its good condition after spawning being able to continue developing new gonads. Figures 55 to 63 show the stages of maturity (from 0 to 5) of male polyps.

SUMMARY OF THE STAGES OF DEVELOPMENT OF SPERM SAC AND SPERMATOGENESIS

I - Presence of clusters of spermatogonia in the mesenterial mesoglea; mesenteries with swollen appearance; mesenterial endoderm stretched and packed with fat globules.

II - Formation of gonad; expansion of the lowermost part of mesentery into an oval, annular and finally rounded sperm sac; endodermic cells are cuboidal or slightly rounded and heavily ciliated; sperm sac size ranges from 100 to 190 µm in diameter; a central cavity gradually develops; spermatocytes divided into spermatids.

III - Spherical sperm sac, delimited from the mesentery by a thin stalk; sperm sac size ranges from 200 to 300 µm; marked central cavity filled with spermatozoons tails; spermatozoa reaching maturation; endodermic cells cuboidal but becoming so stretched that it is difficult to tell them apart.

IV - Sperm sac size over 300 µm; usually oval-shaped depending on the space availability in the gastric cavity of the polyp; central cavity disappears and the already mature spermatozoons are disposed in radial rows; endoderm fragile and breakable.

V - Sperm sac reaches maturation when over 500 µm and is ready to spawn.

A sequence of sperm sacs in different stages of development is shown in Figure 129.

4.3.3 - DEVELOPMENT OF FEMALE GONADS AND OOGENESIS

FIRST REPRODUCTIVE STAGE

The mesenteries in the female polyps are even more markedly dilated during the first reproductive stages than the mesenteries in the male polyps. At first, the mesenteries have their bases clearly linked to the wall of the gastric cavity. As can be observed in Figure 65, the lateral mesentery, beside the asulcal mesentery, is more swollen and more packed with fat globules and germ cells than the others. On this mesentery the oocyte will be preferentially developed.

With the agglomeration of fat globules and increase in size and in number of oogonia, the six mesenteries, bearing these cells, can expand so much that their endoderm and mesoglea might tear off as seen in Figures 66 to 68. The endodermic cells are stretched and elongated making it difficult to distinguish one cell from the other. The lateral mesenteries where the oocyte will be developed are still the thickest, although they do not differ considerably from the others (Fig. 66). With the limited availability of space inside the gastric cavity, the swollen mesenteries are compressed against each other becoming flattened on the sides. Their bases expand substantially detaching from the wall of the gastric cavity. The oogonia are slightly rounded, with sparse granulous cytoplasm, a large nucleus (comprising 2/3 of the cell) and a single spherical nucleolus. The nucleus is eccentrically placed and basophilic staining (Fig. 105). The oogonia eventually migrate to the base of the swollen mesenteries where only one or few oocytes will develop and reach maturation.

SECOND REPRODUCTIVE STAGE - PREVITELLOGENIC

The oocyte at a diameter of around 20 µm is at previtellogenic stage. It is spherical in shape and has a flaccid but smooth oolemma. Cytoplasmic and nuclear growth has begun and the germinal vesicle dislocates more to the periphery of the oocyte. A short pedicel links the oocyte to the mesentery (Fig. 97d). An exposed oocyte at this stage as shown in Figure 97d was rarely seen. Oocytes in even later stages were commonly observed still surrounded by the parental endoderm and mesoglea.

THIRD REPRODUCTIVE STAGE - VITELLOGENIC

At a diameter of 70 to 100 µm, the oocyte is elongated or oval in shape and connected to the very base of the mesentery as a continuation of it (Figs. 72-75, 107, 117). The oolemma

might have already started developing into a series of microvilli which cover the surface of the oocyte. Besides the oolemma, the oocyte is also surrounded by a vitelline membrane, the mesoglea, and by a layer of endoderm, both derived from the parental mesentery. The endoderm of the oocytes observed under scanning electron microscope seems to be similar to the mesenterial endoderm observed in the very early stage of the sperm sac. The endoderm is wrinkled and the endodermic cells are stretched and ciliated (Figs. 74-76).

The germinal vesicle is located close to the attachment of the oocyte to the mesentery, where it probably contacts specialized cells which will aid the nourishing of the oocyte. Once one oocyte starts developing the other germ cells might function as "nurse cells" being absorbed by the developing oocyte. At this stage vitellogenesis has begun and yolk deposition takes place.

FOURTH REPRODUCTIVE STAGE

By the time the oocyte is over 200 µm, it is spherical in shape and is surrounded by a parental endoderm similar to the endoderm of the early stages of the sperm sac, with heavily ciliated cuboidal or slightly rounded endodermic cells (Figs. 77-79, 108). The endodermic or follicle cells in this case, contribute to the nourishment of the egg. The mesoglea or vitelline membrane situated underneath the follicle is smooth and porous, maybe to allow the microvilli, already formed, to function as well for the nourishment of the egg. The oocyte shown in Figures 138, 139 and 141 is surrounded by a porous membrane interwoven by crests. This membrane appears to be the exposed mesoglea enclosing the egg. Oocytes in this condition are uncommon.

Since only one oocyte is developed to a mature stage at once, the space limitation is defined by the size and expansion capacity of the gastric cavity of the polyp. The egg at this stage already has a solid resemblance and occupies a considerable portion of the gastric cavity (Figs. 77, 119, 127).

FIFTH REPRODUCTIVE STAGE

When the oocyte is over 300 µm the follicle cells are columnar in shape and bear cilia which intermingle and are irregularly disposed in tufts (Figs. 80-83, 102, 112, 113). If the egg is the second generation to be matured, whilst the polyp already bears a mature oocyte or larva, it will be confined to the lowermost part of the gastric cavity (Figs. 110, 111). The developing

oocyte might even share the available space with other oocytes of third generation. Such an oocyte will have an irregular shape and its follicle will also vary in aspect. The portion of the follicle which is forced against the wall of the gastric cavity will have short cuboidal cells and the vitelline membrane will be continuous and smooth as if it was compressed (Figs 100b.c,d). The free portion of the follicle will have columnar cells and a less continuous and more porous vitelline membrane (Figs 100b.a,b).

The germinal vesicle present in this stage is relatively small compared with the size of the egg (1/3 of the egg) against the 2/3 observed in the first stages. The single and spherical nucleolus is placed on the periphery of the nucleus which is close to the attachment of the egg to the mesentery (Fig. 110).

SIXTH REPRODUCTIVE STAGE

At a diameter of about 500 μm or more, the oocyte displays a more compacted follicle with marked tufts pointed in three or more directions (Figs. 84, 85). If free space is available the spherical oocyte might have the position of the follicle tufts determined by the pressure of the mesenteries as seen in Figure 112. The vitelline membrane is continuous, smooth and it is loose enveloping the microvilli layer existing on the surface of the egg, which is completely filled with yolk (Figs. 84-86). The egg is still attached to the mesentery and the germinal vesicle which is placed close to this attachment is by now much smaller (1/6 of the egg) when compared to the size of the egg (Fig. 112).

SEVENTH REPRODUCTIVE STAGE

The maximum size registered for oocytes was 660 μm in length and 500 μm in width. The mature oocyte is densely packed with yolk and surrounded by a loose and smooth mesoglea or vitelline membrane. The follicular, endodermal layer is stretched as a consequence of the expansion of the oocyte and the endodermal cells are elongated and ciliated (Figs. 87, 88, 114). The whole aspect of the endoderm is remarkably different from the follicle present in early oogenesis. The oocyte detaches from the mesentery and is loose in the gastric cavity where fertilization probably occurs. The oocyte is estimated to occupy over 60 % of the total polyp volume based on analysis of cross sections (Fig. 114).

The mesenteries as well as the other internal structure of the polyp have been compressed by the growing oocyte. Virtually all the fat granules have been consumed during the

oogenesis and what is left is concentrated at the base of the gastric cavity where further oocyte development might be in process.

SUMMARY OF THE SEVEN STAGES OF OOGENESIS

I - Mesenteries markedly dilated, mainly at their bases; endodermic cells stretched and packed with fat globules; Oogonia rounded with sparse granulous cytoplasm, large nucleus and single spherical nucleolus.

II - Previtellogenic stage - oocyte diameter 20 μm and spherical in shape; germinal vesicle at the periphery of the oocyte occupying 2/3 of the egg; smooth oolemma.

III - Vitellogenic stage - stalked oocyte: 70 to 100 μm in diameter; elongated or oval in shape; starts developing microvilli; surrounded by the parental mesoglea and endoderm; endoderm wrinkled and ciliated.

IV - Oocyte over 200 μm in diameter; shape depending on the space availability, varying from spherical to oval; follicular endoderm with heavily ciliated cuboidal or slightly rounded endodermic cells; smooth and porous vitelline membrane; microvilli layer surrounding the egg.

V - Oocyte over 300 μm in diameter; follicle cells are columnar and bear cilia disposed in tufts; second or third generation oocytes may be present; peripheral germinal vesicle 1/3 of the egg.

VI - Oocyte around or over 500 μm; more compacted follicle with marked tufts; continuous vitelline membrane enveloping the microvilli layer on the surface of the egg; egg densely packed with yolk; peripheral germinal vesicle 1/6 of the egg.

VII - Oocytes in late maturation stage; maximum length 660 μm; and around 500 μm in width; loose and smooth vitelline membrane; follicular endoderm thin and stretched with elongated and ciliated cells; oocyte detaches from the mesentery and loose in the gastric cavity; occupies 60% of the volume of the polyp and it is ready to be fertilized; mesenteries of the polyp compressed.

4.3.4 - ULTRASTRUCTURAL FEATURES OF THE PLANULA LARVA

Whether the egg is fertilized while still enclosed within the follicular epithelium and the underlying mesoglea or after these two layers are detached is not known. Some oocytes were observed enclosed within only a very wrinkled mesoglea devoid of the surrounding follicular tissue (Figs. 132-137). There has been insufficient investigation to clarify whether the embryo is enveloped by the parental mesogleal coat during cleavage and when and how it looses this mesogleal integument. The growth of surface microvilli is observed on the smooth surface of a developing planula (Figs. 142, 143) suggesting that further embryonic development occurs with the embryo free from the mesogleal integument. Further examination is necessary to confirm such a suggestion.

After fertilization the egg moves to the upper part of the polyp cavity where the larva develops (Figs. 115, 116). The larva is retained by the polyp until maturation is attained. If other young oocytes are being developed at the same time, they will be confined to the lowermost part of the polyp together with the mesenteries (Fig. 116).

No cleavage was observed in the examined specimens. All the larvae observed were already in advanced embryonic development. The youngest embryo observed already had an outer layer, the ectoderm and an inner layer, the endoderm, separated by a thin mesoglea. In the larva of Figures 115 and 116 the yolk globules in the endoderm have been partly consumed and a hollow central cavity is being formed. In Figures 89, 90 and 96 the central cavity is clearly seen and the yolk granules are disposed in vertical rows in the columnar endodermal cells (Fig. 92). The surrounding mesoglea is thin and smooth (Fig. 91). The outer ectodermal layer is formed by tall and columnar cells still containing yolk globules (Fig. 90, 91). At a later stage, the ectoderm of the mature planula develops cilia and the larva passes into a free-swimming planula stage. In other species of octocorals the larva can reach this stage within three to five days after insemination but such information is not available for this species. By analogy with other polar marine invertebrates, larval development is likely to be slow.

The embryo shown in Figures 93 and 94 is already completely covered by cilia. The surface of the embryo is shown in a more advanced stage in Figures 95 and 97. In Figure 95 the embryo appears ready to leave the polyp. The eight segments clearly seen on its surface do not indicate formation of mesenteries, since they are not seen in the histological sections. These segments probably reflect pressure from the eight mesenteries on the anthocodial part of the polyp as the larvae attempts to make its way through the mouth of the polyp.

The maximum length of larva planula observed was 860 µm. The larva was estimated to occupy 80% of the total volume of the polyp (Figs. 122, 125) and sometimes almost 100% of its cavity, as shown in Figure 118. The true shape of the larva has not been recorded since no larva was observed outside the polyp. Within the cavities of the polyp the shape of the larva will depend on its position and on space availability. In Figures 118 and 140 the embryo has a broader base as its base is located at the lower cavity where more space is available and its upper part is tapered, with the space being limited on the distal portion of the polyp. In Figures 115 and 116 the embryos show the same contour displayed by the walls of the chamber. The walls surrounding the embryos are very compacted as shown in Figures 115, 145 and 146. The recovery of the female polyp after releasing the larva is even more intriguing than the recovery of the male polyp. The membrane of the polyp might have a considerable capacity of expansion and the armature of sclerites involving it might have a complex mechanism to allow such expansion. The mesoglea from *Metridium senile* was shown to have the ability to stretch three times its original length and yet have complete elastic recovery (Gosline, 1971; Lewis & Wallis, 1991).

4.3.5 - DISTRIBUTION OF REPRODUCTIVE STAGES THROUGHOUT THE COLONY

To define the distribution of maturity the colonies were systematically divided into three regions (distal, middle and proximal) and two portions (peripheral and internal). The maturity stages of the polyps within these divisions were identified and are shown graphically in Figures A1 and A2. The titles in the legend of Figs A.1.1 and A.2.1 refer to - sex/region/portion, where m-- is: male; f--: female; -1-: distal, -2-: middle, -3-: proximal; --1: peripheral, and --2: internal.

In male colonies there are relatively more polyps in the first reproductive or immature stages in the distal region with the mature stages distributed relatively evenly between the middle and proximal regions (Fig. A.1.2). The peripheral polyps were mostly in the first reproductive stage or immature (Fig. A.1.3). The later stages of maturation are found in the internal polyps (Fig. A.1.3). The same distributions were found in female colonies (Figs A.2.1., A.2.2, A.2.3).

4.3.6 - INFLUENCE OF THE REPRODUCTIVE STATE ON THE EXTERNAL MORPHOLOGICAL FEATURES OF THE POLYPS

A comparison of the external dimensions of the polyps was carried out in an attempt to understand the impact of the presence of reproductive products. The results for all polyps (male and female) are shown in Figure A3. The results for different stages of maturity are

shown in Figures A4 to A7. The overall data (Fig. A3) indicate some trend for increasing dimensions with polyp length. The lowest correlation for both male and female is with the width of the neck (correlation co-efficients of 0.16 and 0.10 respectively for male and female). However, these data are merely showing the strong relationship apparent in early stages of maturation (Figs. A4, A6). The correlation co-efficients for the data shown in Figures A3 to A7 are given in Table 4.3.

Table 4.3 - Correlation of polyp length with calyx, neck and base widths

Reproductive Stage	Correlation of male polyp length with:			Correlation of female polyp length with:		
	calyx width	neck width	base width	calyx width	neck width	base width
All data	0.27	0.16	0.27	0.34	0.10	0.26
0 (immature)	0.32	0.19	0.31	0.31	0.31	0.24
1	0.34	0.15	0.32	0.47	0.21	0.35
2	0.22	0.11	0.14	0.54	0.14	0.01
3	0.15	0.03	0.10	0.01	0.01	0.00
4	0.07	0.13	0.05	0.28	0.26	0.33
5	0.08	0.23	0.26	0.12	0.02	0.15
6				0.21	0.17	0.14
7				0.44	0.08	0.07

In immature and stage 1 polyps the size of the reproductive products are negligible and do not have any impact on the polyp dimensions, therefore the relative proportions of the polyp are maintained throughout growth in the early stages, showing a positive correlation (Table 4.3 and Figures A4 and A6). In later stages the proportions are altered by the presence of the reproductive products and less correlation is seen (Table 4.3 and Figures A5 and A7). There is less correlation between polyp length and neck width than with either base or calyx width.

Figures A8 and A9 show the correlation between sperm sac and egg size and the external dimensions of the polyp. The correlation coefficients are given in Table 4.4. In the male polyps the greatest correlation is seen between the sperm sac size and the width of the base of the polyp. In the female polyps there is some correlation between egg size and the widths of both the neck and base.

Table 4.4 - Correlation of sperm sac and egg size with polyp external dimensions.

Correlation of sperm sac size with:			Correlation of egg size with:		
polyp length	base width	neck width	polyp length	base width	neck width
0.00	0.23	0.07	0.03	0.27	0.18

A comparison was made between the average dimensions of polyps in early reproductive stages and mature stages. It was found that the polyp length and calyx width increased only by about 10% and 6% respectively. However, the width of the neck and base showed significant increases in both sexes. In the female polyps both neck and base widths increased by about 50% and in the male polyps the base width increased by about 50% whilst the neck width increased by only 25%.

An estimate of the polyp volume was made based on the average dimensions in the early and later reproductive stages. It was assumed that the polyp was composed of two truncated cones each of half the polyp length. The appropriate radii were taken from the calyx, neck and base widths. It was estimated that the volume increased by 80% and 85% respectively for the male and female polyps during development of sperm sacs and eggs.

4.3.7 - GAMETOGENIC DEVELOPMENT IN THREE DIFFERENT PHASES OF SUMMER

Samples were obtained in three successive years and in each year in a different phase of the Antarctic summer. These data were used to assess the reproductive development throughout the summer. The results are summarised in Figure A10. During the first phase of the summer the female polyps (f85) are mostly in the immature or first reproductive stage. In the second phase (f86) oocytes in different stages of maturation are observed. In the third phase (f87) oocytes in later stages of maturation are found. For the male polyps, development appears to occur earlier in the summer. In the first phase (m85) sperm sacs in the first to third stages are observed. In the second phase (m86) all stages are observed and in the last phase (m87) apart from Stage 1, the majority are in Stage 4. This analysis has been based, by necessity, upon data from successive years which may lead to interannual variations that might explain the few exceptions to the general distribution. Ideally such analysis should be carried out with data from continuous sampling.

4.4 - DISCUSSION

The reproduction of Antarctic and deep-sea octocorals has received little attention. Prior to this study, nothing was known of the reproductive biology of *Thouarella variabilis* and the lack of comparative data makes any discussion limited. Furthermore, the difficulties in the accomplishment of the proposed research were numerous. The access to the study area was limited, as was the number of samples available for the study. In addition, working with samples from different years, depths and areas, makes any statistical analysis unreliable, although it is still possible to confirm general trends. The inability to follow the gametogenic and larval development of the octocorals, compounded by the lack of continuous sampling, made it a very complex task to define the developmental stages and gametogenic cycles. Gaps in information still exist but it has been possible to describe the general aspects. Therefore, although the present study could not be done in optimal conditions it is by all means justified since our knowledge of the reproductive aspects, or any other aspects, of *Th. variabilis* is meagre.

The bottlebrush colonies of *Th. variabilis* are composed of non-retractile polyps, which attain an average length of 1 mm, half of which is occupied by the gastric cavity. The basal part of the gastric cavity of the polyps are interconnected by the solenia covered by a relative thin coenenchyme (unlike the alcyonaceans which have a thick coenenchyme and are able to develop long gastric cavities). The short polyp has its volume even more limited by the calcareous armature which surrounds it, although this armature apparently has a high capacity for expansion. Benayahu & Loya (1984) suggested that fecundity depends on the length of the polyp cavity and *Th. variabilis*, having a short gastric cavity, shows a low reproductive output, producing only one egg to maturity at a time. Like the majority of the octocorals, *Th. variabilis* develops large eggs (600 µm) that in some specimens can occupy 60% of the polyp volume. The development of other eggs might be in progress but these are restricted to the lower portion of the gastric cavity and sometimes they are compressed to the walls of the polyp as the oocyte in maturation demands additional space.

Three stages of oocyte development have been observed in a single polyp: previtellogenic oocytes; oocyte in IV or V stage; and a mature oocyte. Whether after the release of the mature oocyte, the oocyte in stage V can reach maturation still in that season is unknown. However, it is most probable that the polyp releases just one larva per summer and matures the oocyte (V) throughout the year, to reach maturation in the following year and the youngest oocyte to reach maturation in two years time. The oocyte might have a slow period of growth

during the long Antarctic winter and a rapid growth in the eutrophic summer months. It was also shown that there is a general trend of development of oocyte and sperm sacs during summer with the late stages of development being mostly observed in late summer. The considerable decrease of energetic reserves of the polyp, observed in the later stages of oocyte development, suggests that the production of a single egg is very costly. The replenishment of energetic reserves might be a slow process making it more difficult for the polyp to accomplish the maturation of a second oocyte within the same season. Unfortunately, no winter samples were available for this study and from which other hypotheses could be raised. These octocorals could have continuous gametogenesis and release larvae all year around as shown for other octocorals (Benayahu *et al.*, 1989) and for other Antarctic invertebrates (Pearse *et al.*, 1991). Despite the possibility of this hypothesis being true, it is important to reemphasize that to produce a large yolky egg is expensive and the polyp would have to continuously build up energetic reserves.

Fat granules were observed in massive concentrations in polyps producing first stage oocytes. These secretions were most abundant in the cells from the base of the tentacles to the lower part of the pharynx and even more in the endodermic cells of the mesenteries in the gastric cavity. The polyps containing oocytes at a later stage of development show very reduced amount of granules. In his studies of nutrition of *Heteroxenia fuscescens*, Schlichter (1982) showed that dissolved organic material (DOM) derived from the sea and assimilates of zooxanthellae were the most important nutritional supplies of these soft corals and particulate food was less important. The degradation of particulate food in *H. fuscescens* takes place in the dorsal mesenteries and the dissolved compounds are liberated to the gastric cavity and utilized by the polyp or transported to the rest of the colony via the solenia. Part of the DOM might be lost to the sea but can be reassimilated by the ectoderm or by the cells in the pharynx. Schlichter (1982) concluded that the energy gained from absorption of DOM should be enough to supply the energy demand of the octocoral.

Whether *Thouarella variabilis* is a secondary consumer or lives on DOM originating from the surrounding water requires additional study. Schlichter (1982) pointed out that since *H. fuscescens* does not bear cilia on its ectoderm, which would be required for filter feeding, added to the scarcity of nematocysts necessary for preying plankton, particulate food, apart from the utilized zooxanthellae, would be of less importance for this coral. However, Lewis (1981) demonstrated that octocorals are simple raptors, capturing prey by mechanical use of tentacles and Lasker (1981) described the importance of mucus on the adhesion of captured particles and the same was observed by Lawson (1990). The tentacles of *Th. variabilis* do not bear cilia and are porous as shown for the deep sea isidid *Acanella arbuscula* by Lawson

(1990), who also found dense coatings of microvilli on the tentacles of these gorgonians, suggesting that the absorption of DOM from the external environment would be an advantage to this animal, which inhabits an energy poor environment. Although, he suggested the absorption of DOM as just a supplementary diet as these octocorals were also found to feed on particulate food, phytodetritus and forams, from the flux of surface derived organic material arriving at the seabed. This author deduced that this source of food would be the trigger for synchronized reproduction and would also influence the growth in deep sea species. Particulate food was not observed on the tentacles or in the cavities of the polyps of *Thouarella variabilis*, although nematocytes were abundant in the epidermis of the tentacles of the polyps.

Similar reproductive stages were observed along the colonies of *Thouarella variabilis* but not all the polyps in a colony were bearing gonads. In contrast to Lawson's study of *A. arbuscula*, the polyps on the periphery of the branches were mostly immature although displaying adult size. The polyps which were reproducing were mainly located on the middle and base of the branches. Hartnoll (1975) noted that in the later stages of oocyte development in *Alcyonium digitatum* the colony would enter in quiescent phase and would not feed. It is not hard to believe that if the polyp has its cavities blocked with reproductive products, it would stop capturing food. If particulate food is the main source of energy for *Th. variabilis*, the polyps which have most of their volume occupied by oocytes or sperm sacs, would be disadvantaged in capturing food. This function of acquiring food could be fulfilled by the non-reproductive peripheral polyps. These "end" polyps would be in favourable feeding position as suggested by Lawson (1990) for *A. arbuscula*, although in *A. arbuscula* the "end" polyps were the ones reproducing. In *Th. variabilis* the peripheral polyps would ideally capture the particulate food, decompose it and transfer the dissolved compounds to the rest of the colony via the solenia. To define the nutritional biology of the studied species, it would be necessary to undertake additional studies of transmission and scanning electron microscopy to detect the presence of particulate food and zooxanthellae, as well morphological adaptations of tentacles of non-reproducing peripheral polyps in comparison with reproducing internal polyps.

Th. variabilis, like most of the other octocorals, has shown colonial and polyp gonochorism. Only one colony was found to contain both male and female polyps. No hermaphrodite polyp was observed. The availability of space within the polyp would alone be sufficient restriction to inhibit the production of both oocytes and sperm sacs, not withstanding the additional energetic demand.

The first reproductive stage was similar for both sexes, with the swelling of mesenteries as a result of the aggregation of fat granules and germ cells. The nucleus of these cells is large (2/3 of the cell) and stains heavily being easily mistaken for lipid droplets. Because of the poor state of preservation of the tissues, it is difficult to discern the germ cells clearly. Once the germ cells migrate from the endoderm into the mesoglea and the energetic reserves are consumed, the endodermic cells have the appearance of an empty flaccid bag. In *Clavularia hamra* the primordial generative cells wander in the mesoglea until they settle in the mesenteries (Gohar,1948). The stage when germ cells migrate to the mesoglea was not observed in the studied specimens. The gonads develop preferentially on the lateral mesenteries which are close to the asulcal mesenteries located adaxially. In the other octocorals studied to date gonads were observed on all sulcal and lateral mesenteries (Hickson, 1895; Chia & Crawford, 1973; Hartnoll, 1977; Benayahu *et al.*, 1989; Achituv & Benayahu, 1990; Benayahu, 1991; Rice *et al.*, 1992). In *Th. variabilis* gonads were never observed on the mesenteries besides the siphonoglyph, abaxially located. In *Cespitularia* sp. and *Xenia* sp. the gonads develop on the upper two thirds of the anthocodia not in the lowermost part (Gohar, 1940) and in *Heteroxenia fuscescens* the gonads, confined to the autozooids, develop on the upper end of the coelenteron occupying 30 to 60% of the length of each mesentery (Benayahu, *et al.*, 1989). In *Anthelia* sp. and *Sympodium* sp. (Gohar, 1940) the gonads develop on the whole length of the mesenteries in the gastric cavity and in *Th. variabilis* the gonads develop mainly near the base of the mesenteries in the lowermost part of the gastric cavity.

At the first stages of oogenesis, it was not possible to observe the layer of microvilli that replaces the oolemma covering the oocyte and the trophonemata or trophocytes described by Eckelbarger & Larson (1988) for *Aurelia aurita*. Since a few oocytes are developed, it is highly probable that the remaining germ cells are utilized as "nurse" cells contributing to the nourishment of the developing oocytes during vitellogenesis. Evidence of this is the reduction of germ cells at the later development stages of the oocyte. The nurse cells are absorbed or engulfed by the developing oocyte and in some species non-germinal accessory cells are also engulfed during oogenesis (Hartnoll, 1975; Eckelbarger & Larson, 1988). Some of the oocytes in contact with the one destined to develop are engulfed by it. The studies of Beams & Kessel (1983) elucidated the relationship between the nurse cells and the developing oocytes. These authors showed that after the release of one mature egg the remaining undeveloped oocytes enlarge and migrate to the cavity formerly occupied by the released egg. Once another oocyte starts to develop, chemical substances are released from this oocyte in maturation to attract the other undeveloped or developmentally blocked oocytes, which become actively ameboid and migrate towards the developing oocyte becoming part

of its ovary. When the mature oocyte is fertilized the stimulus ceases and the migration of nurse cells immediately stops. These embryonic reserve type cells, which are undifferentiated interstitial cells, can differentiate not only into germ cells but also into somatic cells and the reverse is also true. Apart from nurse cells, the oocyte is surrounded by specialised structures, such as follicle cells of somatic origin and in some species by trophonema or trophocytes. Trophonema represents the early stages of follicle or nurse cell evolution containing glycogen and lipid droplets in the ooplasm. Trophocytes have their nuclei positioned laterally and their apical surface is covered by branching microvilli as well as endocytotic pits and vesicles. Their cytoplasm contains mitochondria, glycogen particles and membrane-bound inclusions. At the beginning of vitellogenesis these cells will cover about 15% of the oocyte surface and by the end of their differentiation they will be the only cells in the germinal epithelium in contact with the developing oocyte (Eckelbarger & Larson, 1988). Further studies would be necessary to confirm the presence of trophonema or trophocytes on *Th. variabilis* but the nucleus of the developing oocyte is certainly dislocated to the periphery of the oocyte where it keeps contact with the mesentery. The nourishment of the oocytes is known to be provided mainly by lipid and proteinous yolk platelets and the process of vitellogenesis will depend on two sources of yolk platelets: heterosynthetic incorporation of precursors and autosynthesis. The period of vitellogenesis is not known for *Th. variabilis* but Eckelbarger & Larson (1988) suggest that species having the shortest vitellogenic periods utilize heterosynthetic mechanisms and those with the longest periods utilize autosynthetic mechanisms. *Th. variabilis* might utilize heterosynthetic mechanisms in the eutrophic summer months and produce its own reserves throughout the long winter.

The size at which vitellogenesis begins varies among the different anthozoan species. The subsequent stages might vary in size in which it happens amongst different species but in general they are similar in structure. However, some small variations have been observed. When the oocyte is over 100 μm in *Th. variabilis*, it is already involved by a parental endoderm, unlike *Ptilosarcus guerneyi* studied by Chia & Crawford (1973), that at this stage still lack an extracellular coat and the egg envelope consists of microvilli that covers the whole surface, providing protection for the egg. At the later stages, in some species the oocyte is covered by a jelly coat which lies under the follicular layer (Achituv *et al.*, 1990), this was not observed in *Th. variabilis*. Chia & Rostron (1970) observed that the growth of oocytes to their maximum size is rather rapid and takes about a month.

The development of sperm sacs shown in *Th. variabilis* followed the same pattern demonstrated by many other anthozoans. As mentioned by Szmant-Froelich (1980), it is difficult to distinguish between primordial germ cells and spermatogonia, and the transition of

the latter into spermatocytes. Neither can the very young spermary be distinguished from the very young ovary. Spermatocytes develop a greater affinity for haematoxylin as they grow, indicating an increase of nucleic acid content (Szmant-Froelich, 1980). The spermatids are half the volume of the secondary spermatocytes and this difference in size can be observed when comparing the contents of the young sperm sac where spermatocytes are meiotically dividing into spermatids with the spermatocytes still present on the stalk which links the sperm sac to the parental mesentery. Fadlallah (1983) suggested that spermatogenesis normally is synchronous within a cluster of cells but may be either synchronous or asynchronous amongst clusters within the same gonad or among gonads of the same polyp. Observing the sperm sacs of the specimens of *Th. variabilis* under light microscopy, one has the impression that spermatogenesis is synchronous, with all the cells sharing the same stage of development. However, when observing the sperm sacs under a transmission electron microscope many different stages can be observed in the same sperm sac, although an overall view shows that the majority of the cells are at the same developmental stage. The same central cavity which becomes occupied by the developing spermatozoon tails in the second and third stage of development was also observed by Hickson (1895) and Gohar *et al.* (1961) and the ciliated sperm sac covering the sperm mass by Benayahu (1989). Rinkevich *et al.* (1984) noted, studying scleractinians, that when colonies are at maximum reproductive development, spermaries fill up all available spaces within the polyps. They appear at the basis of the polyp, in its gastric cavity, and penetrate up into the middle of the tentacles. He also stated that it is still unknown whether polyps which are filled up with gonads are capable of preying on and ingesting zooplankton, and the same has been mentioned above for the female polyps.

The number of spermaries produced by the *Th. variabilis* is low compared to other anthozoan species. In *Capnella gaboensis*, 55 sperm sacs develop per polyp, although this number diminishes prior to spawning, interpreted by Farrant (1985) as resorption of small gonads by larger ones. *Th. variabilis* produces in general only two sperm sacs, although some young ones can be observed when the larger sperm sac are in later stages of maturation. The works of many other authors have shown spermatogenesis to be shorter than oogenesis. In *Th. variabilis*, sperm sacs of different development stage in the same polyp are not always found and even if there is they might still develop that season or be kept for next year. In any case, sperm sacs would not take two years to develop, following therefore an annual gametogenic cycle, while the oogenic cycle might take two years. The sperm sacs are relatively large (600 μm) when compared with the size of the polyp cavity. As a consequence of the lack of space, the two sperm sacs become compressed against each other. This is possible with sperm sacs because of their fluffy consistency but would not be possible with

the solid oocyte. Thereby, the polyp can cope with two mature sperm sacs but would not necessarily cope with two mature oocytes. Furthermore, it is energetically less costly to produce a functional spermary than to produce a single large egg (Brazeau & Lasker, 1990). The male polyps were found to expand to accommodate the sperm sacs increasing their average volume by 80%, mainly in the region of the base since the sperm sacs will be confined to the lower portion of the polyp. No sperm sac was observed outside the polyp, therefore, it is not known whether they burst while still inside the polyp or are discharged intact to the exterior. In the absence of a mass of spermatozoa in the polyp cavities, or of temporary gonopores, it is most probable that the sperm sacs are extruded through the mouth. At the later developmental stage the sperm sac possess a very breakable and fragile endoderm as a consequence of its expansion. Rupture might, therefore, happen only a short time after liberation, which normally takes place after complete maturity. Gohar & Roushdy (1961) points out that immature spermaries do not rupture at all.

In *Th. variabilis* fertilization is internal. The ripe egg detaches from the mesentery and is located in the middle of the gastric cavity that will enlarge to accommodate the large egg. The female polyp was found to expand, increasing its average volume by 85%, mainly in the region of the base and the neck, since the developed oocyte and larva will occupy most of cavity space. Brooding is the reproductive mode observed for this species and the embryo remains inside the polyp cavities until a mature planula larva is developed. In some other brooding species of octocorals the fertilized eggs pass to special brooding pouches situated at the upper part of the polyp cavities, near the anthocodial bases where embryogenesis takes place and the planulae are shed via temporary openings found among the polyps. These temporary openings close immediately after planulation (Benayahu *et al.*, 1989). In *Thouarella variabilis* the fertilized egg passes to the upper part of the polyp cavities but no special brooding pouch is formed. The walls of the pharynx and mesenteries are all compressed to the sides having the resemblance of a protective membrane enveloping the embryo. The polyp has a high capacity for expansion, as noted above, and the calcareous armature which surrounds it an amazing mechanism to withstand such expansion. The sclerites which normally overlap each other slide apart being juxtaposed in polyps which are reproducing. The sclerites previously placed in diagonal rows are then clearly disposed in longitudinal rows. Considering that the arrangement of sclerites in octocorals is a taxonomic descriptor, reproductive stages should be observed when a nominal species is described. If the whole colony is reproducing (which was not observed in this species), this character could lead to taxonomic mistakes.

Despite having a low reproductive output, *Th. variabilis* has evolved a reproductive mode that improves its reproductive efficiency by protecting the embryo until the planula larva is fully developed ensuring a higher survival rate. Brooding the offspring might therefore compensate for the low fecundity. This reproductive mode is common among octocorals as is the production of large eggs. The benthic habit of the planulae might add to their survival capacity. Benayahu *et al.* (1990) suggest that prolonged oogenesis increases the yolky content of the eggs and probably contribute to larval dispersal. The largest eggs seen in octocorals are from surface brooding species which also provides protection to the embryos and avoids space limitation, perhaps increasing the fecundity for the colony. No morphological adaptation for surface brooding is found on *Th. variabilis* and mature larvae are observed inside the polyp cavity confirming they are indeed brooders.

Pearse *et al.* (1991), suggest that the organic material in large lecithotrophic eggs is used primarily to form large juveniles and not as a source of nourishment for the embryos. This is based on the observation of the low consumption of organic material of eggs during development of some brooding Antarctic species. Some larvae of *Th. variabilis* were observed having the yolk content in part consumed and having eight lines on their ectoderm resembling the eight mesenteries, which would be characteristic of a juvenile rather than a larva. Nevertheless, these lines were most probably formed by the compression of the eight mesenteric cavities of the polyp since when observed internally they do not show any clear mesentery. The heavily ciliate larva leaves the polyp through the mouth and in many specimens the larva was seen extending part of the ectoderm in direction to the mouth of the polyp in an attempt to exit.

Based on the existing literature (Table 4.5), a ciliated pre-planula can be developed in 36 to 44 hours and an actively mobile planula in 3 to 5 days. The larva settles and contracts the cilia in 3 to 8 days and the mesenteries and tentacular grooves are formed in 4 to 9 days. The tentacles grow and stomodeum opens to the exterior providing the larvae with the capacity for feeding in 8 to 11 days. The pinnules are complete in about a month, the second polyp appears in 3-4 months and a young colony with 4 polyps in 5-6 months. For *Th. variabilis* such data are lacking but the available data on other species might suggest how long the larva stays in the polyp, although in *Alcyonium hibernicum*, which develops parthenogenically, the larva is stored for a month, not being released until late summer. In some brooding species of Antarctic invertebrates the embryo might be kept for months, as in the tanaid *Nototanais dimorphus* which releases juveniles 5-6 months after starting brooding the embryos (Pearse *et al.*, 1991).

In octocorals both pelagic and demersal planula larvae have been observed. The broadcaster *Alcyonium digitatum* produces a pelagic planula larvae and *A. hibernicum* and *A. siderium*, which brood their larvae produce a benthic planula (Hartnoll, 1977; Sebens, 1983). In general, in species which present brooding or surface brooding as the reproductive mode, the larvae sinks rapidly to the bottom as in *Briarium asbestinum* (Brazeau *et al.*, 1990) or is demersal lecithotrophic as in *Eunicella singulares* (Weinberg & Weinberg, 1979). *Xenia macrospiculata* broods its embryos internally until maturation and the planulae when released have a short pelagic phase and settle soon after being released (Benayahu & Loya, 1984). *Xenia umbellata* releases its planulae infected with symbionts which might also contribute to a higher survival rate (Benayahu *et al.*, 1989). The gorgonian *Paramuricea clavata* produces a short-lived lecithotrophic larva (Gili & Garcia, 1985), conversely *Muricea californica* and *M. fruticosa* produce a pelagic planula that despite being brooded on the surface of the parent colony can have a planktonic life that lasts up to 30 days (Grigg, 1977). The pennatulacean *Ptilosarcus guerneyi* produces a lecithotrophic larva 4 days after fertilization which is ready to settle in seven days but, being nutritionally independent, could remain as planula for around 30 days (Chia & Crawford, 1973).

The non-feeding, non-pelagic, lecithotrophic larvae of *Thouarella variabilis* might, like other brooding octocorals and some Antarctic invertebrates, settle soon after release, and might as a consequence have low dispersal capacity. Evidence of this is the patchy distribution shown by these octocorals in the underwater films taken in the Weddell Sea, Antarctica (Julian Gutt, pers. comm.). In these films an undetermined bottlebrush species of *Thouarella* was found to be very abundant, forming dense aggregates. The colonies within each patch were disposed very close to each other which would easily happen if the larvae settle near to the parent colonies. The same patches were recorded for another primnoid, *Ascolepis*, in Maxwell Bay, King George Island, Antarctica (Martin Rauchert, pers. comm.). However, the reproductive mode of this genus is not known. Brooding associated with low dispersion is a common characteristic of Antarctic invertebrates.

Despite its supposedly low dispersal capacity, the genus *Thouarella* is known to be widespread in Antarctic waters as it has been recorded in many different Antarctic regions. The larva of *Eunicella singularis* has a slow speed and is hence influenced by even the weakest currents (Theodor, 1967). It is not hard to believe therefore that strong currents and storms could transport the larvae of *Thouarella* to other areas distant from the parent colony. In view of its colonization success, being abundant and widespread, it is possible to state that its low fecundity is definitely compensated by the efficiency of its brooding reproductive mode, which provides protection for the embryo enhancing early survival.

TABLE 4.5 - Stages of embryogenesis in three different species of octocorals

STAGE	*Lobophytum crassum* [1]	*Xenia macrospiculata* [2]	*Alcyonium digitatum* [3]
first signs	3 hours		
first cleavage	3-5 h.		
5 cell-stage	3.5 h.		
6, 10, 12 cell-stage	3.5-4.5 h.		
16 cell-stage	4.5 h.		
MORULA	5 h.		24 h
PRE-PLANULA, cilia smooth outline	36 h		44 h
slow moving, shorter, pear-shape	43 h	2 days	4 days
actively mobile	3 days		5 days
settles	hours after		7 days
attached planulae rounded, 1/5 of original size; cilia retracted		3 days	8 days
spicules	23 days	3 days	
mesenteries; 8-10 tentacular grooves		4 days	9 days
8 primary tentacles		5 days	
body elongates stomodeum formed		6-7 days	10 days
tentacles grow; stomodeum opens to exterior		8 days	11 days
first pair of pinnules		9 days	12 days
3 pairs of pinn.	16 days		14 days
4 pairs of pinn.		15 days	16 days
5 pairs of pinn.		17-18 days	18 days
7-8 pairs of pinn.		1 month	27 days
second polyp		3-4 months	
young colony - 4 polyps		5-6 months	

1 - Benayahu & Loya, 1984

2 - Benayahu & Loya, 1989

3 - Matthews, 1916

LIST OF FIGURES FOR CHAPTER 4

Fig. 18 - Detail of the two fifth stage sperm sacs shown in Figure 17 showing the stretched endoderm and the stalk (arrows). 110 X. Scale bar: 100μm (S4)

Fig. 19 - Detail of the reduced mesenteries (M) shown in Figure 17. 210 X. Scale bar: 100μm (S4)

Fig. 20 - Detail of the stalk of the sperm sacs shown in Figure 18. 250 X. Scale bar 10μm (S4)

Fig. 21 - Detail of the reduced and compacted mesenteries shown in Figure 19. 770 X. Scale bar: 10μm (S4)

Fig. 22 - Two fifth stage sperm sacs with damaged portion showing the mass of developing spermatozoa (*). 100 X. Scale bar: 100μm

Fig. 23 - 25 not included

Fig. 26 - Second stage sperm sac, cylindrically shaped and showing ciliated endoderm. 350 X.

Fig. 27 - Developing genital cells showing cytoplasmic continuity (arrow). 2,100 X.

Fig. 28 - Detail of the genital cells continuity of the developing spermatazoa (arrow). 3,500 X.

Fig. 29 - Exposed central cavity of second stage sperm sac showing mass of developing spermatozoa. 320 X. Scale bar: 10μm (S3ST2)

Fig. 30 - Mass of developing spermatozoa. 1,000 X. Scale bar 10μm (ST2.2)

Fig. 31 - Exposed central cavity of a third stage sperm sac starting to be filled with spermatozoa tails showing mass of developing spermatozoa (S) and stretched endoderm (E). 280 X. Scale bar: 10μm (M20ST3).

Fig. 32 - Detail of the sperm sac shown in Figure 31 showing spermatozoa (S) and endoderm (E). 1,300 X. Scale bar 10μm (M20ST3).

Fig. 33 - Fourth stage sperm sac showing fragile and breakable endoderm (E) and thin mesoglea (M) and mass of mature spermatozoa (*). 770 X. Scale bar: 10μm (S3ST4).

Fig. 34 - Stretched endoderm of fourth stage sperm sac. 1,500 X. Scale bar: 10μm (M20).

Fig. 35a,b - Spermatocytes with large nucleii and granular cytoplasm. a) 10,000X (32643), b) 15,000 (32642).

Fig. 36 - Spermatids in the first stage of spermiogenesis. 60,000 X (32628).

Fig. 37-42 - Developing spermatozoa 37) 34,000 X (32634), 38) 40,000 X (32626), 39) 56,000 X (32623), 40) 56,000 X (32624), 41) 42,000 X (32625) and 42) 28,000 X (32627).

Fig. 42b - Detail from a sperm sac; A) spermatocytes (Sp) mesoglea (m) and endoderm (Ed). 1,400 X. (32652). B) spermatocytes (Sp), mesoglea (m) and endoderm (Ed). 2,500 X. (32719). C) spermatocytes (Sp), endoderm (Ed), nucleus (N) and space left by fat granules (*). 4,200 X. (32663). D) spermatocytes (Sp), endoderm (Ed), nucleus (N) and mesoglea (m). 8,400 X. (32660). E) spermatocytes (S) cytoplasmic process (arrow) and mesoglea (m). 21,000 X (32662). F) spermatocytes and nuclei (N). 8,400 X (32720). G) cilia (C) from the

endodermal cells (Ed). 28,000 X (32656) and H) cilia (C) from the endodermal cells (Ed). 21,000 X (32657).

Fig. 43 - Detail from a sperm sac showing developing spermatozoa (S), mesoglea (m) and endoderm (E). 16,000 X (32632).

Fig. 44 - Detail from a sperm sac showing stretched endoderm (E), mesoglea (m) and developing spermatozoa (S). 7,000 X (32630).

Fig. 45 - Longitudinal section of a male polyp showing the pharynx (P), and a second stage sperm sac (Sp). 100 X (4.1.2a)

Fig. 46 - Detail from Figure 45 showing the sperm sac (Sp) and the stalk (St). 200 X (4.1.2a)

Fig. 47 - Longitudinal section of a male polyp showing the pharynx (P), mesentery (m) and two sperm sacs (Sp). 40 X (4.2c).

Fig. 48 - Detail from Figure 47 showing the gastric cavity with expanded mesentery (m), two sperm sacs (Sp) with central cavity (c) mesoglea (ms) and endoderm (e). 200 X (4.2c).

Fig. 49 - Sperm sac showing endoderm (arrow) mass of developing spermatocytes (s) and central cavity (C). 280 X (2.4.2b)

Fig. 50 - Polyp with the central cavity occupied by the sperm sac shown in Figure 49. 140 X (2.4.1b)

Fig. 51 - Detail of the sperm sac shown in Figure 49 showing the exposed mesoglea (arrow) and the stalk (St). 280 X (2.4.2b).

Fig. 52 - Gastric cavity with two sperm sacs in different stages of development. Sc, spermatocytes and S, spermatozoa. 140 X (2.4.2a).

Fig. 53 - Gastric cavity with two sperm sacs in different stages of development. S, spermatozoa. 140 X (2.4.2c)

Fig. 54 - Gastric cavity with two sperm sacs in different stages of development. S, spermatozoa, radially arranged. 140 X (2.4.3a)

Fig. 55 - Non-reproductive polyps showing no internal structures. 30 X. (9a)

Fig. 56 - Male polyp in the second reproductive stage. M, mesenteries and sperm sacs (arrows). 60 X. (4.1.2).

Fig. 57 - Male polyp in the second reproductive stage showing sperm sacs (arrow) same polyp as in Figure 15. 60 X. (20.2.1.2).

Fig. 58 - Male polyp in the second reproductive stage showing sperm sacs. 60 X. (19.M.I).

Figs 59-60 - Male polyps with sperm sacs (s) in the third stage. 60 X. (19.M.I).

Fig. 61 - Two male polyps in the fourth reproductive stage. S, sperm sacs. 30 X. (27.0.1).

Fig. 62 - Male polyp in the fifth reproductive stage with two sperm sacs (S). 60 X. (25.0.2).

Fig. 63 - Polyp in the fifth reproductive stage with two sperm sacs with fluffy consistency, one of the sperm sacs is damaged. 35 X. (25.0.2).

Fig. 64 - Female polyp in the first reproductive stage showing expanded mesenteries (arrow). 60 X. Scale bar: 100μm. (1ST1).

Fig. 65 - Detail from the polyp shown in Figure 64 showing the body wall in the intermesenteric cavity surrounding the pharynx (P), expanded mesenteries in the gastric cavity (black arrow) and asulcal mesentery (white arrow). 180 X Scale bar: 100μm. (1ST1).

Fig. 66 - Exposed gastric cavity showing the markedly expanded mesenteries (black arrows) and the asulcal mesenteries (white arrows). 120 X. Scale bar: 100μm (1ST1).

Fig. 67 - Detail of Figure 66 showing the stretched endoderm (E). 600 X. Scale bar: 10μm. (1ST1).

Figs 68-70 - Detail of the content of the expanded mesentery. 68) 1,800 X. Scale bar: 10μm (1ST1), 69) 1,400 X. Scale bar 10μm (F9ST1), 70) 6,000 X. Scale bar: 1μm (F9ST1).

Fig. 71 - Content of the mesentery endodermal cells showing fat granules and germ cells (arrow). 9,800 X. (24747).

Fig. 71a - Granules in the mesenterial endoderm (arrows). 3,000 X (32666).

Fig. 71c - Fat granules (G) in the cells of the pharynx (Ph). 2,500 X.

Fig. 71d - Exposed second stage oocyte (O) showing stalk (arrow) and mesentery (M). (1,000 X ,left, 300 X, right).

Fig. 72 - Female polyp in the third reproductive stage. Oocyte (large arrow) and tentacles (thin arrow). Scale bar: 100μm

Fig. 73 - Exposed gastric cavity showing the expanded mesenteries (M), oocyte (black arrow) and asulcal mesentery (white arrow). 130 X. Scale bar: 100μm (1ST1)

Fig. 74 - Detail of the oocyte (arrow) and expanded mesenteries shown in Figure 72. 210 X. Scale bar: 100μm (S1ST1).

Fig. 75 - Detail of the oocyte shown in Figure 73 with wrinkled endoderm. 700 X. Scale bar: 10μm (1ST1).

Fig. 76 - Detail of the endoderm of the oocyte shown in Figure 75. 2,100 X. Scale bar: 10μm (S1ST1).

Fig. 77 - Female polyp in the fourth reproductive stage showing spherical oocyte (O) with ciliated and aggregated endodermal cells. 80 X. Scale bar: 100μm (S16).

Fig. 78 - Detail of the endoderm of the oocyte shown in Figure 77 showing the heavily ciliated endodermic cells. 1,800 X. Scale bar: 10μm (S16).

Fig. 79 - Detail of the oocyte shown in Figure 77. 150 X. Scale bar: 100μm (S16).

Fig. 80 - Female polyp in the fifth reproductive stage with oocyte (S). 60 X

Fig. 81 - Detail of the oocyte (S) shown in Figure 80 surrounded by follicle cells. 140 X.

Fig. 82 - Detail of the follicle cells shown in Figure 81. 700 X.

Fig. 83 - Fifth stage oocyte showing follicle cell cover. 190 X. Scale bar: 100μm.

Fig. 84 - Oocyte with exposed structures. f, follicle cell layer; M, mesoglea and mi, microvilli. 110 X. Scale bar: 100μm (S4ST5)

Fig. 85 - Oocyte with exposed structures. f, follicle cell layer; mi, microvilli and y, yolk. 110 X. Scale bar: 100μm

Fig. 86 - Detail of the microvilli layer of the oocyte shown in Figure 84. 4,600 X. Scale bar: 1μm (S4ST5)

Fig. 87 - Seventh stage oocyte (last stage of maturation). 110 X. Scale bar: 100μm (S5ST5F)

Fig. 88 - Detail of the endoderm of the oocyte shown in Figure 87. 1,500 X. Scale bar: 10μm (S5ST5F)

Fig. 89 - Female polyp with a planula larva (L) with exposed parts showing the coelenteron (*). 90 X. Scale bar: 100μm (S25T6).

Fig. 90 - Detail of the exposed parts of the larva shown in Figure 89. Ec, ectoderm; Ed, endoderm, C, coelenteron and arrow, mesoglea. 300 X. Scale bar: 100μm (S25)

Fig. 91 - Detail of the layers shown in Figure 90. Ec, ectoderm; m, mesoglea and Ed, endoderm. 1,200 X. Scale bar: 10μm (S25)

Fig. 92 - Detail of the endodermal layer shown in Figure 91. 1,600 X. Scale bar: 10μm (S25)

Fig. 93 - Developing planula larva (L) in the polyp cavity. 220 X. Scale bar: 100μm (S18)

Fig. 94 - Detail of the ciliated ectoderm of the larva shown in Figure 93. 1,300 X. Scale bar: 10μm (S16)

Fig. 95 - Mature planula larva ready to exit the polyp. 180 X. Scale bar: 100μm (S5ST6F)

Fig. 96 - Cross section of the larva shown in Figure 95 showing the coelenteric cavity (c) and ectoderm (arrow). 180 X. Scale bar: 100μm (S5)

Fig. 97 - Detail of the ectoderm of the larva shown in Figure 95. 950 X. Scale bar: 10μm (S10)

Figs 98-100 - Cells of the mesenterial endoderm (arrows) packed with fat granules (L). N, nuclei; M, mesoglea and E, endoderm. 98) 7,000 X (32614), 99) 7,000 (32612), 100) 9,800 (32610).

Fig. 100b - Cross sections of oocytes (Oc). f, follicle cells; m, mesoglea; Fs, stretched follicle; Y, yolk; g, granules; Ci, cilia and mi, microvilli. A) Uncompressed side of the oocyte. 1,100 X (32724), B) Uncompressed side of the oocyte. 2,100 X (32727), C) Oocyte side compressed against the body wall with smooth mesoglea. 700 X (32725) D) Oocyte compressed against the body wall with smooth mesoglea. 2,800 X (32725), E) 1,800 X (32735), F) Showing a stretched follicle exposing the mesoglea. 1,800 X (32734), G) Detail of mesenterial endoderm with cells packed with granules. 7,000 X (32647) and H) cilia of the follicle cells. 10,500 X (32728).

Fig. 101 - Mesenteries compressed at the lower portion of the gastric cavity. Endodermal cells (arrow) packed with granules and germ cells. M, mesoglea. 4,000 X (24746).

Fig. 102 - Fifth stage oocyte showing the follicle cells (f), mesoglea (M) and yolk (Y). 3,000 X (24745).

Fig. 103 - Cells packed with granules (arrows) in the pharynx (Ph). 400 X. (9a)

Fig. 104 - Female polyp in the first reproductive stage showing calyx (c) and expanded mesenteries in the gastric cavity (m). 100 X (10e).

Fig. 105 - Previtellogenic oocyte (Oc) with large nucleus (n) confined to the lower portion of the gastric cavity. 200 X (30.6.2b)

Fig. 106 - Detail of the expanded mesenteries (M) shown in Figure 104. 200 X (10e)

Fig. 107 - Vitellogenic oocyte (Oc) attached to the mesentery (m). 400 X (1ah).

Fig. 108 - Fourth stage oocyte showing follicle cells (f), mesoglea (m) and yolk (y). 280 X (30.6.1a)

Fig. 109 - Third stage oocyte (Oc) confined to the lower portion of the gastric cavity. 140 X (30.6.1a)

Fig. 110 - Fifth stage oocyte compressed against the mesenteries (*) and body wall at the lower portion of the gastric cavity. m, mesoglea; n, nucleus and y, yolk. 140 X (30.6.2b)

Fig. 111 - Fifth stage oocyte (O) confined to the lower portion of the gastric cavity compressed against the body wall and mesentery (*), mesoglea (arrow). 140 X (30.6.2b)

Fig. 112 - Sixth stage oocyte (O) attached to the mesentery in the gastric cavity showing follicle with marked tufts (F). 70 X

Fig. 113 - Sixth stage oocyte showing follicle layer (F), mesoglea (M) and yolk (Y). 70 X.

Fig. 114 - Female polyp in seventh reproductive stage with the oocyte (O) occupying the whole gastric cavity showing a very stretched endoderm and a thin mesoglea (m). 70 X.

Fig. 115 - Female polyp with the body cavity occupied by a planula larva (L). 100 X (30.6.2b)

Fig. 116 - Female polyp with the upper portion occupied by a developing larva (L) and the lower portion by a fifth stage oocyte (Oc) compressed against the mesenteries (mt). Ec, ectoderm, m, mesoglea. 100 X (30.6.2b)

Fig. 117-125 - Sequence of development of oocyte (arrow and o) and larva (L). 35 X (117, 119, 121, 123) and 60 X (118, 120, 122, 124, 125).

Fig. 126 - Sequence of development of sperm sacs. 60 X.

Fig. 127 - Sequence of development of oocytes. 60 X

Fig. 128-131 - not included

Fig. 132 - Female polyp with mature oocyte (arrow). 80X

Fig. 133 - Detail of the oocyte shown in Figure 132. 200 X

Fig. 134 - Detail of the wrinkled mesoglea of the oocyte shown in Figure 133. 1,000 X

Fig. 135 - Female polyp with mature oocyte (arrow). 100 X

Fig. 136 - Detail of the wrinkled mesoglea of the oocyte shown in Figure 135. 1,000 X

Fig. 137 - Detail of the oocyte shown in Figure 135, lower portion deformed to accommodate the axis of the branch. 200 X.

Fig. 138 - Oocyte involved by wrinkled and porous mesoglea with stalk (arrow). 180 X. Scale bar: 100μm.

Fig. 139 - Detail of the porous mesoglea shown in Figure 138. 1,300 X. Scale bar: 10μm

Fig. 140 - Developing larva conforming to the shape of the parent body cavity. 160 X. Scale bar: 100μm

Fig. 141 - Detail of the porous mesoglea (arrow) 3,200 X Scale bar: 1μm. (Inset: stalk of the oocyte shown in Figure 138).

Fig. 142 - Developing planula larva. 140 X Scale bar: 100μm

Fig. 143 - Detail of the surface of the larva shown in Figure 142 showing the developing microvilli. 12,000 X Scale bar: 1μm.

Fig. 144 - Alternative view of the larva shown in Figure 142. 180 X.

Fig. 145-146 - Polyp from which the larva shown in Figure 142 was extracted showing compacted body wall (c). 60 X (145), 140 X (146).

Fig. A1 - Distribution of maturity throughout male colonies.

Fig. A2 - Distribution of maturity throughout female colonies.

Fig. A3 - Comparison of polyp external dimensions, male and female, all stages of maturity.

Fig. A4 - Comparison of polyp external dimensions, female, stages 0-3.

Fig. A5 - Comparison of polyp external dimensions, female, stages 4-7.

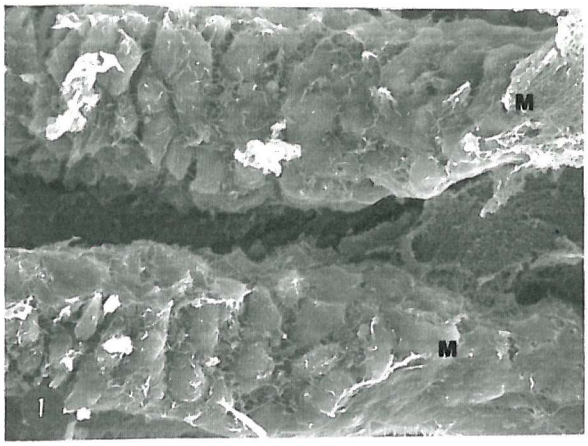
Fig. A6 - Comparison of polyp external dimensions, male, stages 0 and 1.

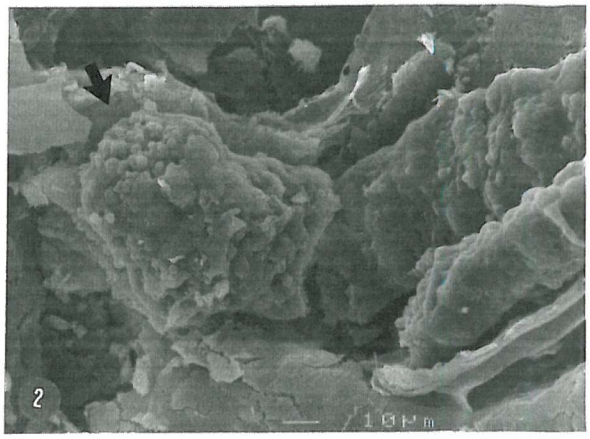
Fig. A7 - Comparison of polyp external dimensions, male, stages 2-5.

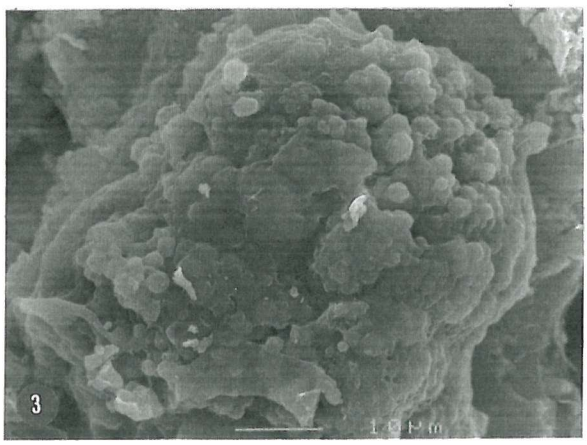
Fig. A8 - Comparison of sperm sac size and polyp dimensions.

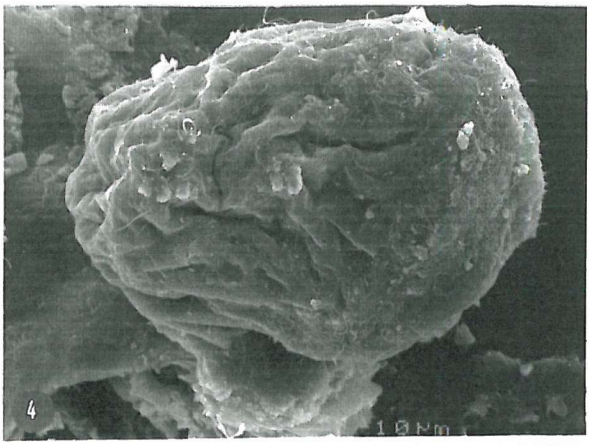
Fig. A9 - Comparison of egg size and polyp dimensions.

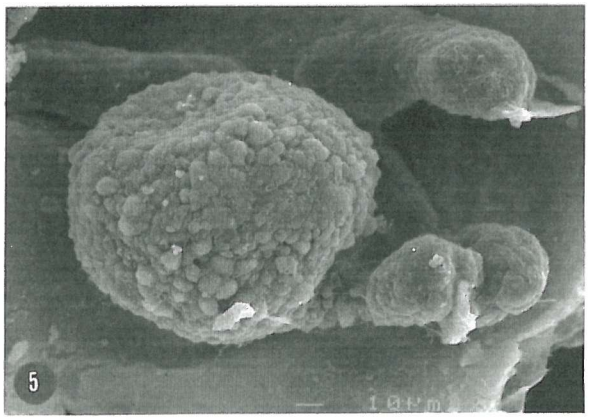
Fig. A10 - Distribution of maturity in sampling years, 1985, 1986 and 1987 (early, mid and late summers)

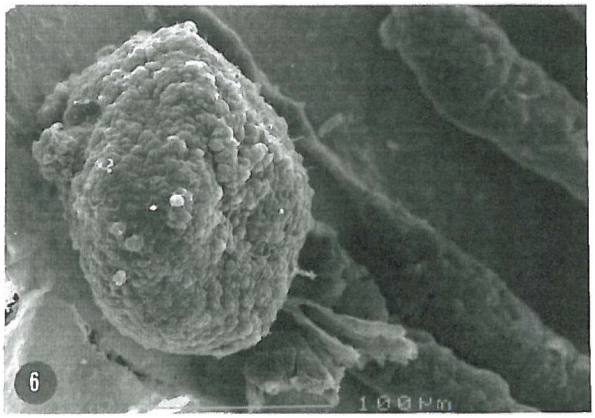

M
M
1

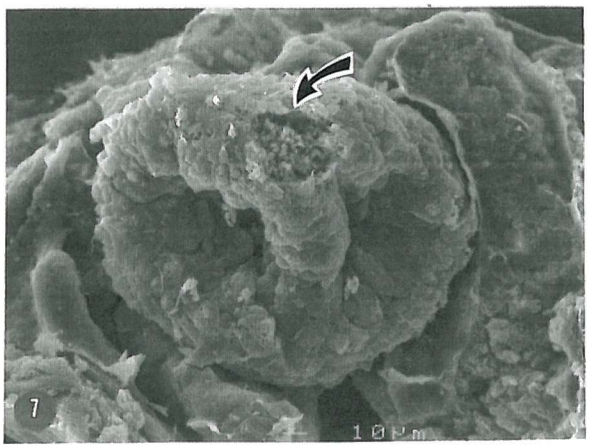

2
10μm

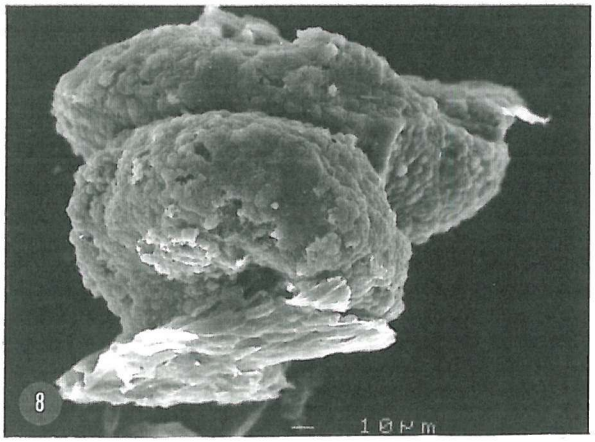

3
10μm

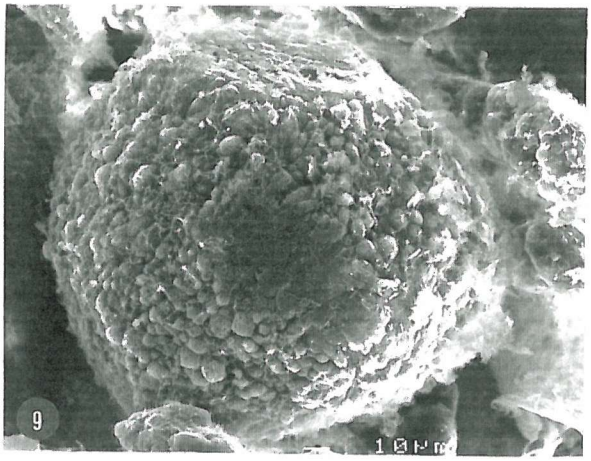

4
10μm

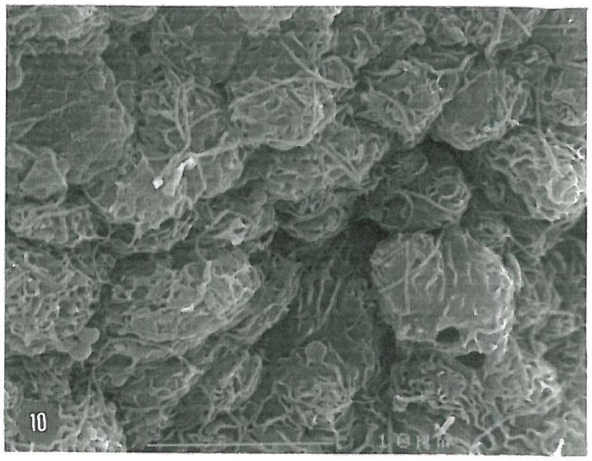

5
10μm

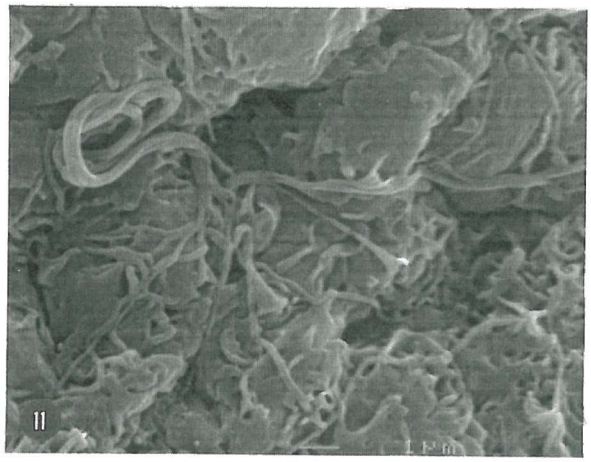

6
100μm

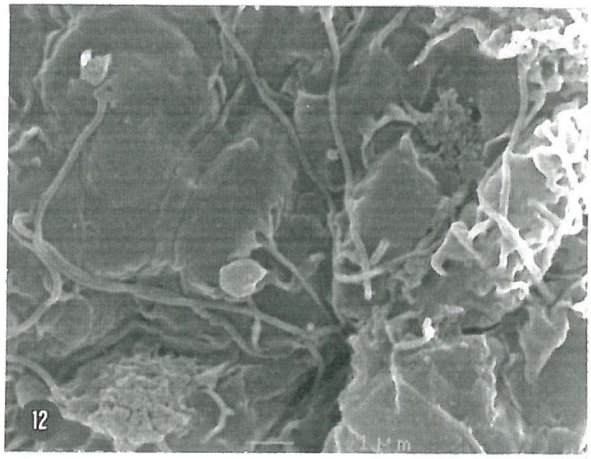

7
10μm

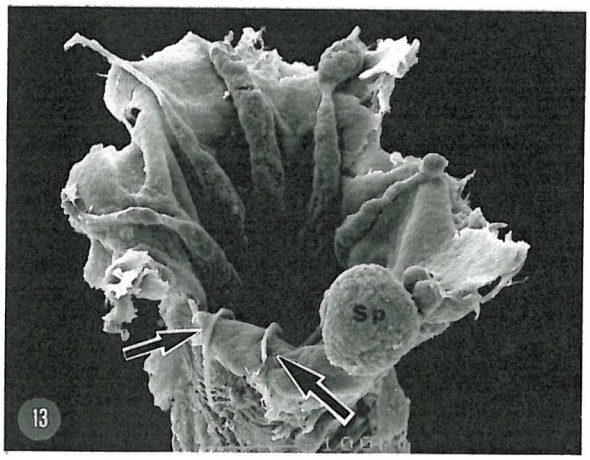

8
10μm

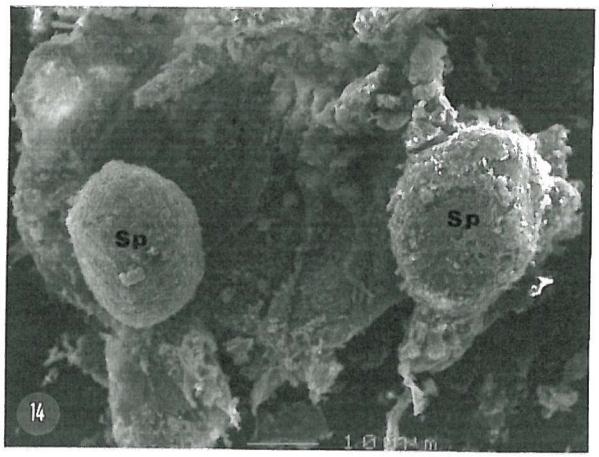

9
10μm

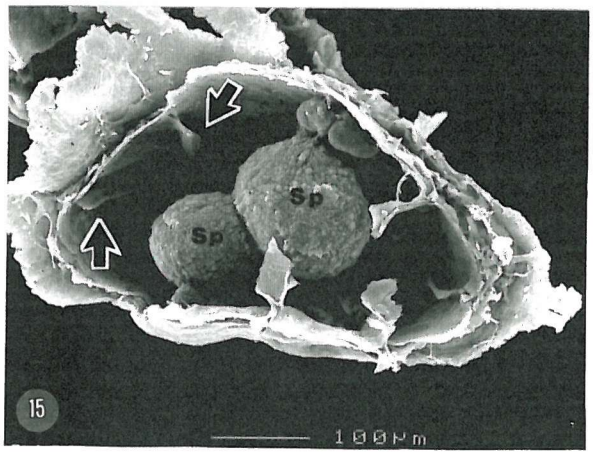

10

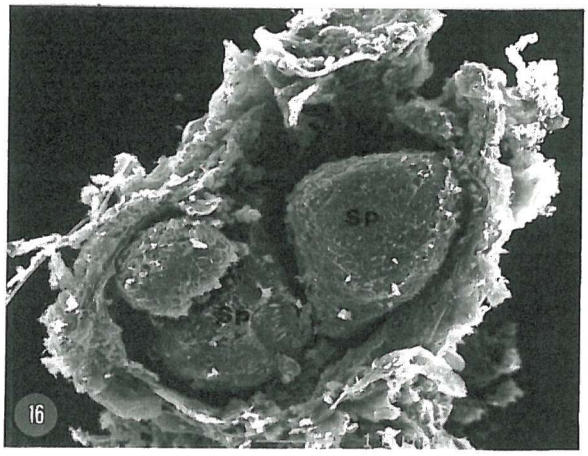

11
1μm

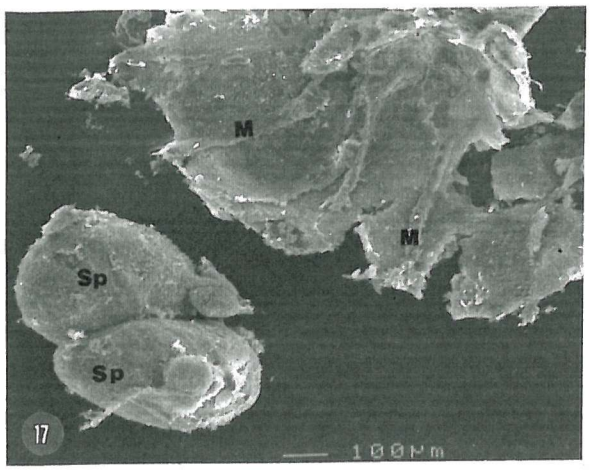

12
1μm

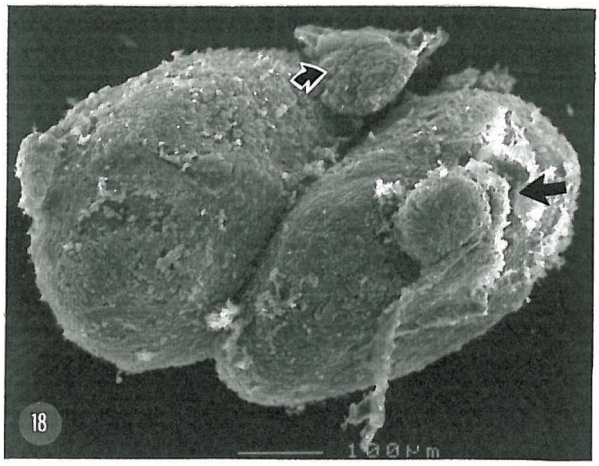

Sp
13

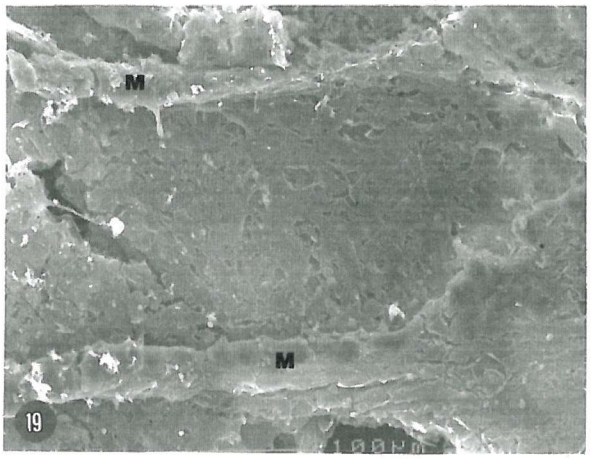

Sp
Sp
14

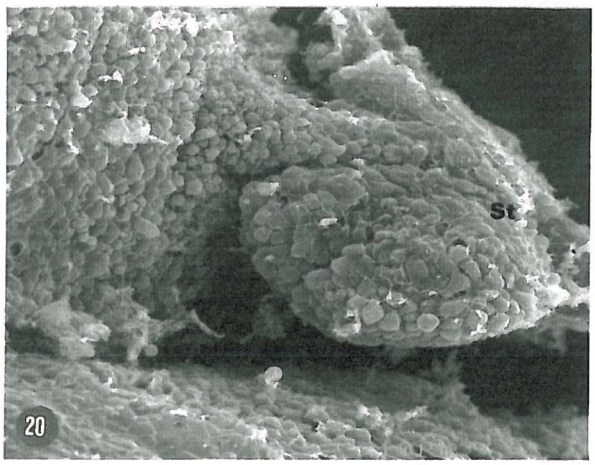

Sp
Sp
15
100μm

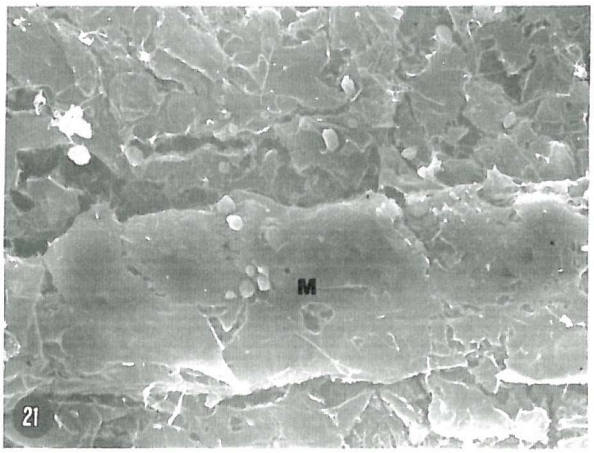

Sp
Sp
16

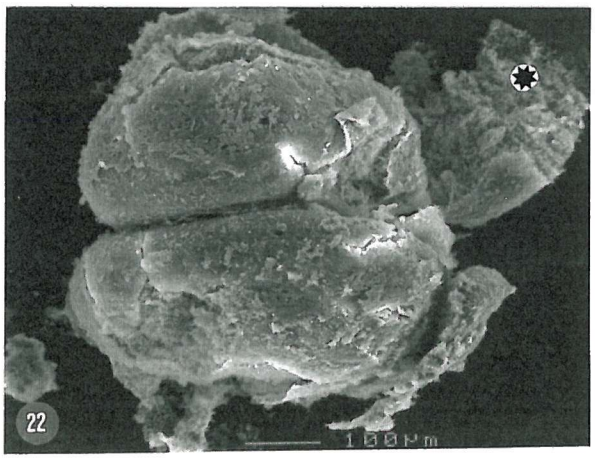

M
M
Sp
Sp
17
100µm

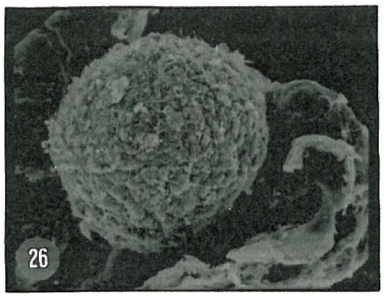

18
100µm

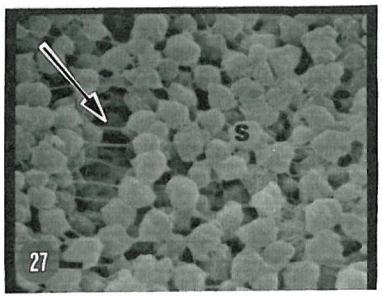

M
M
19
100µm

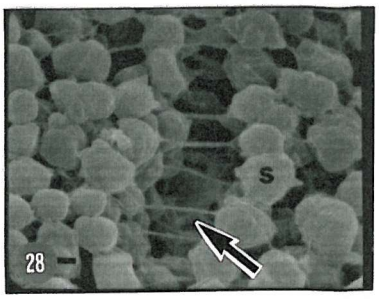

St
20

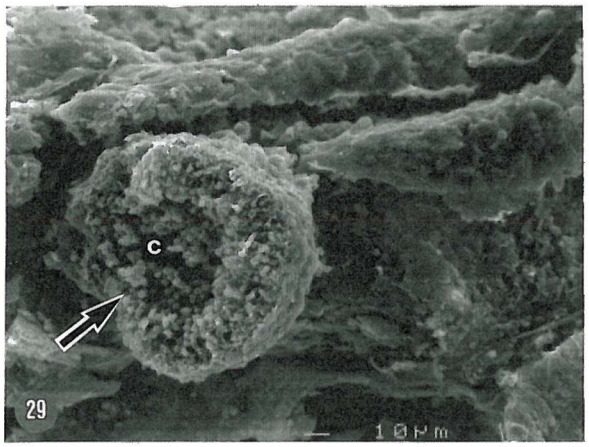

M
21

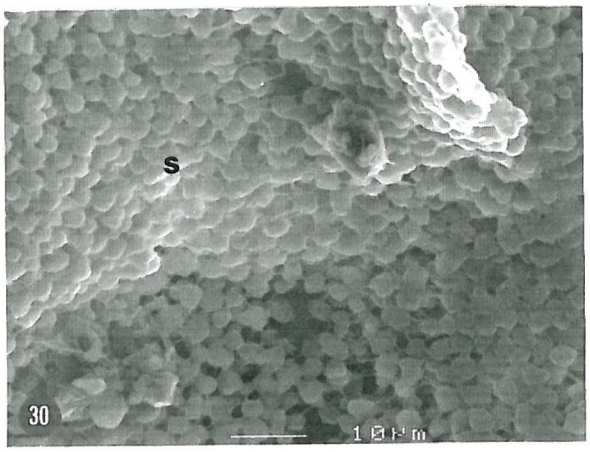

22
100µm

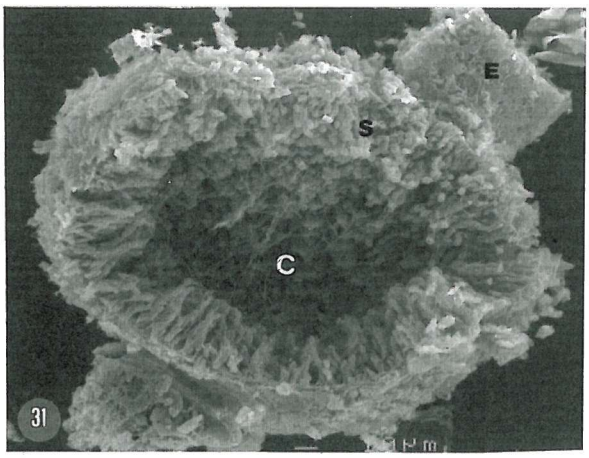

26

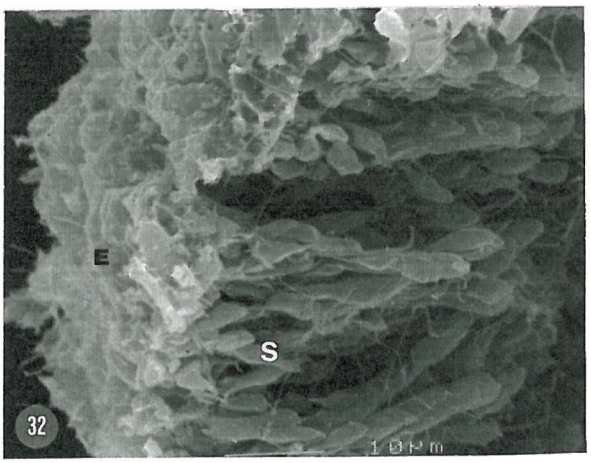

S
27

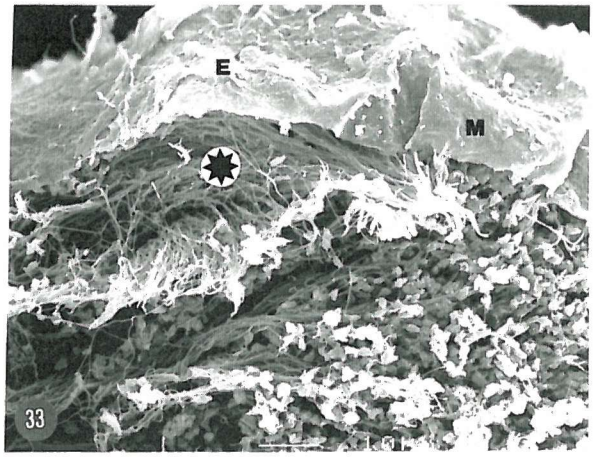

S
28

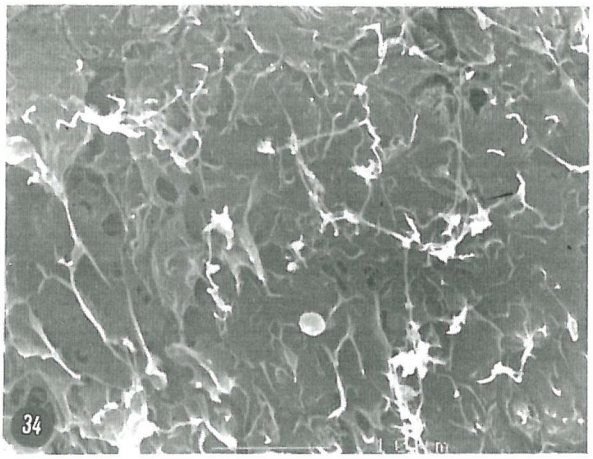

C
29
10µm

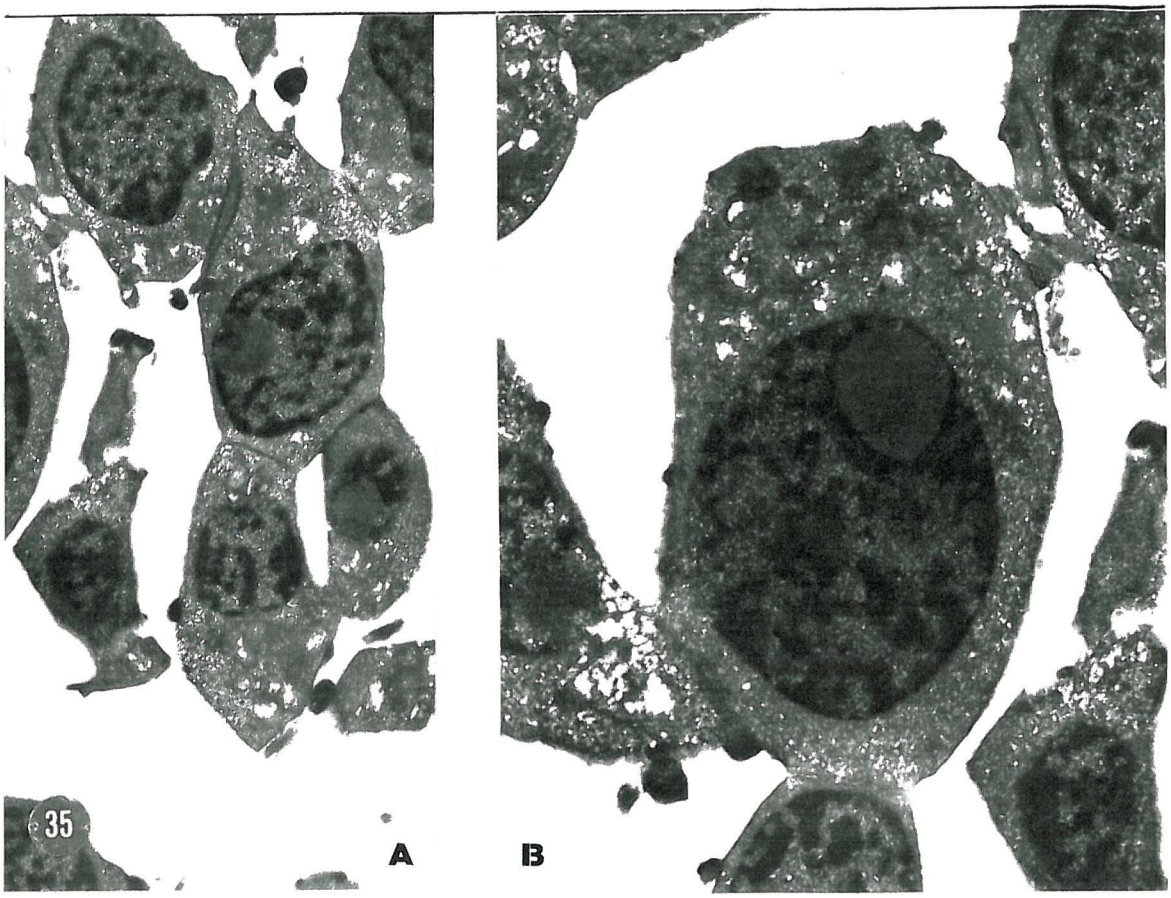

S
30
10µm

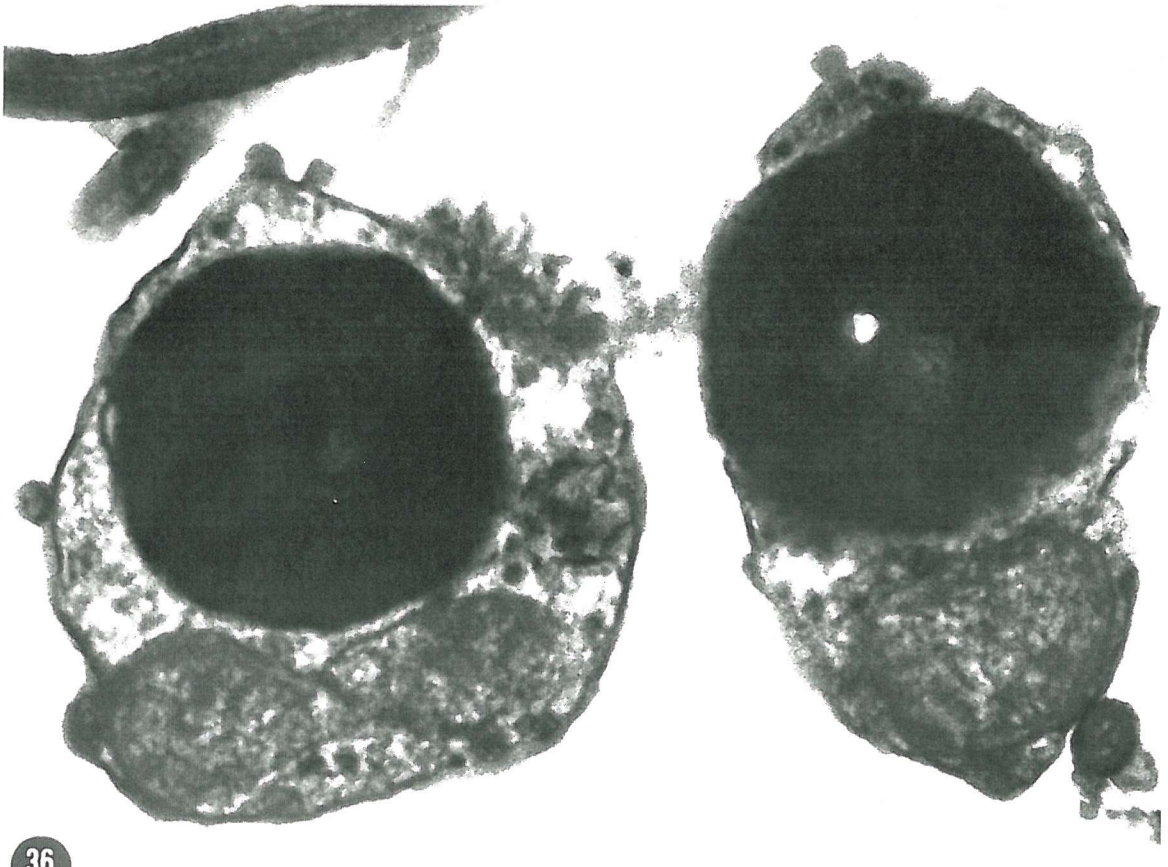

E
S
C
31

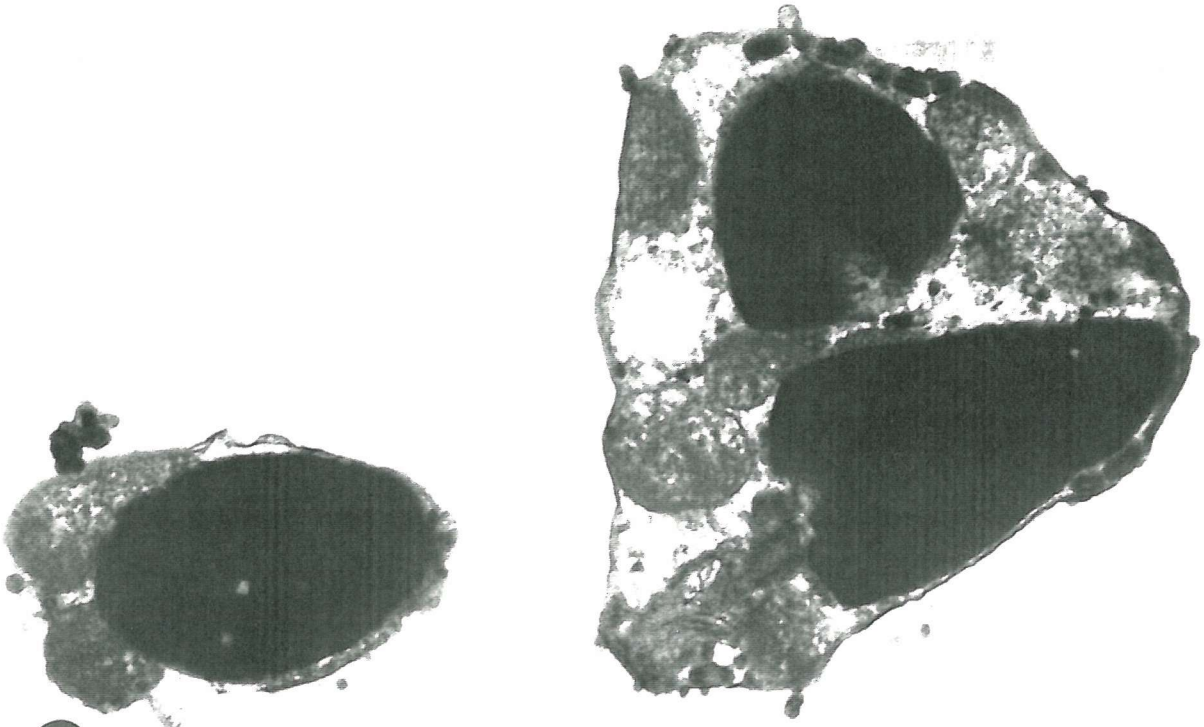

E
S
32
10µm


E
M
33

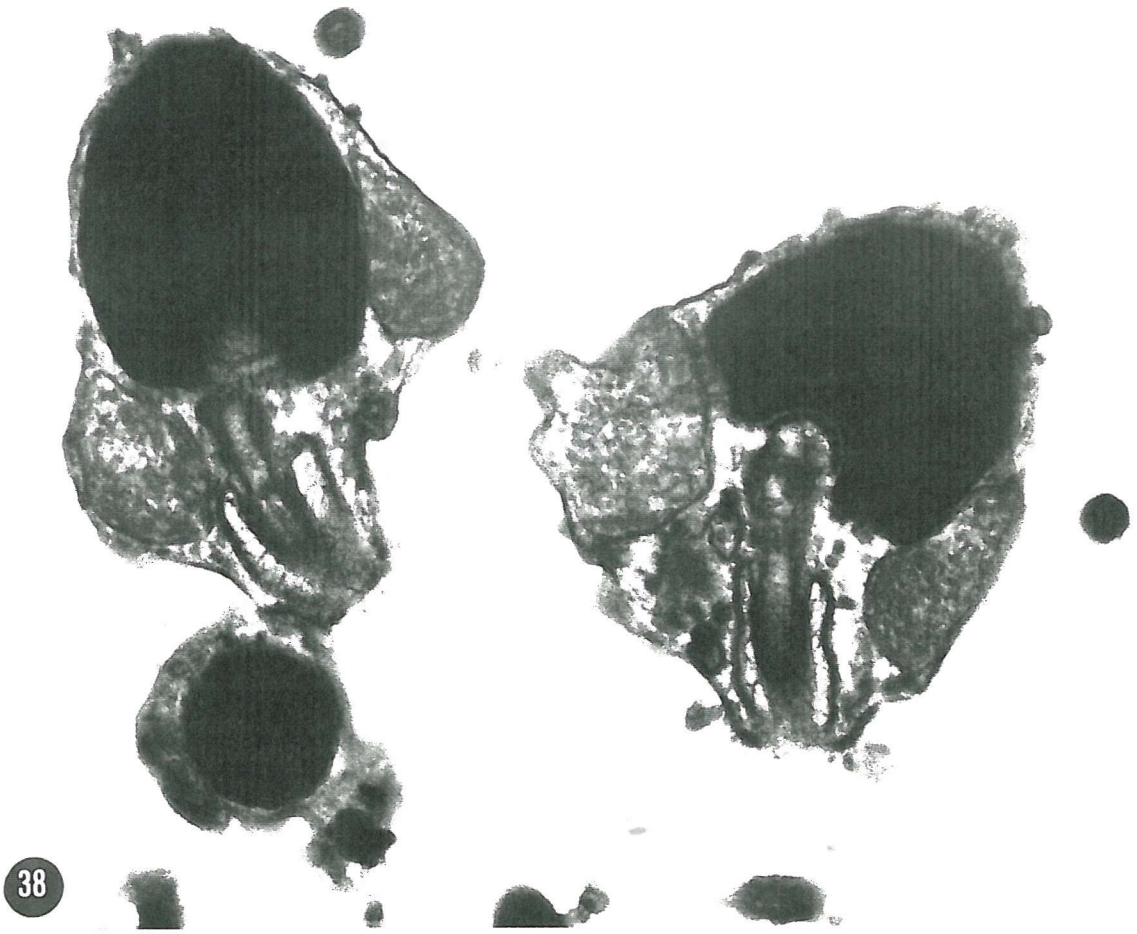

34


35
A
B


36

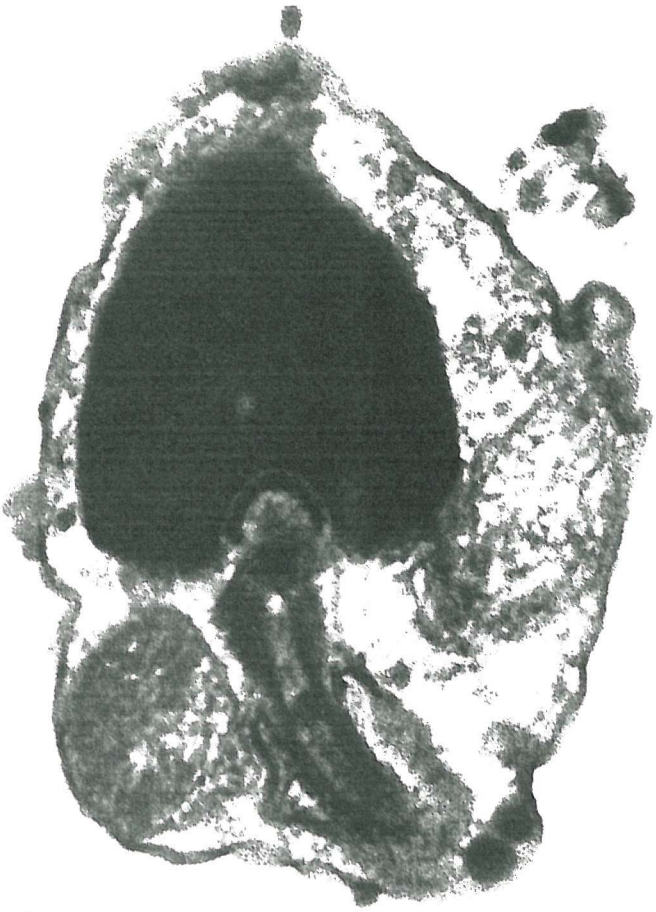


37

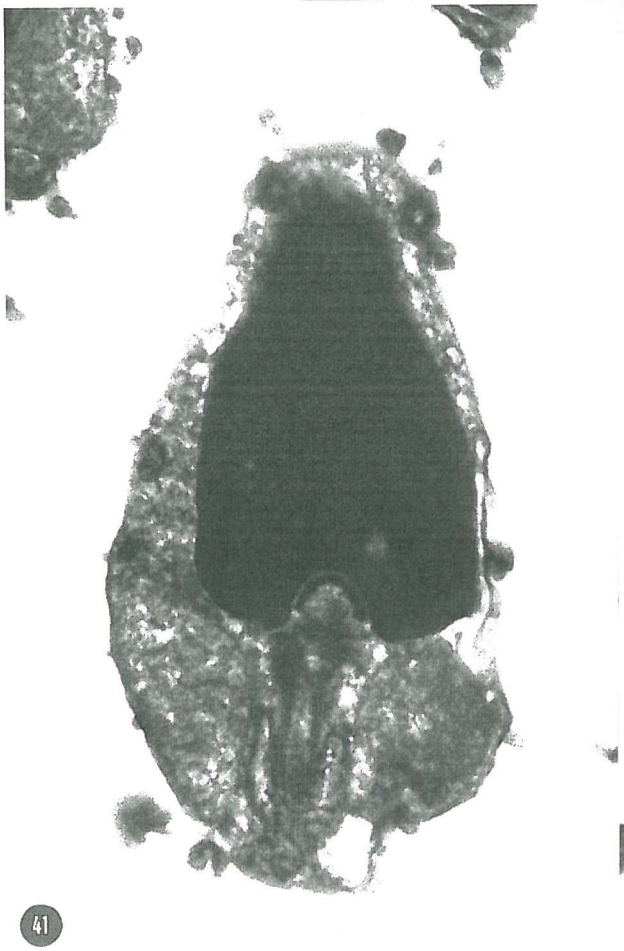


38

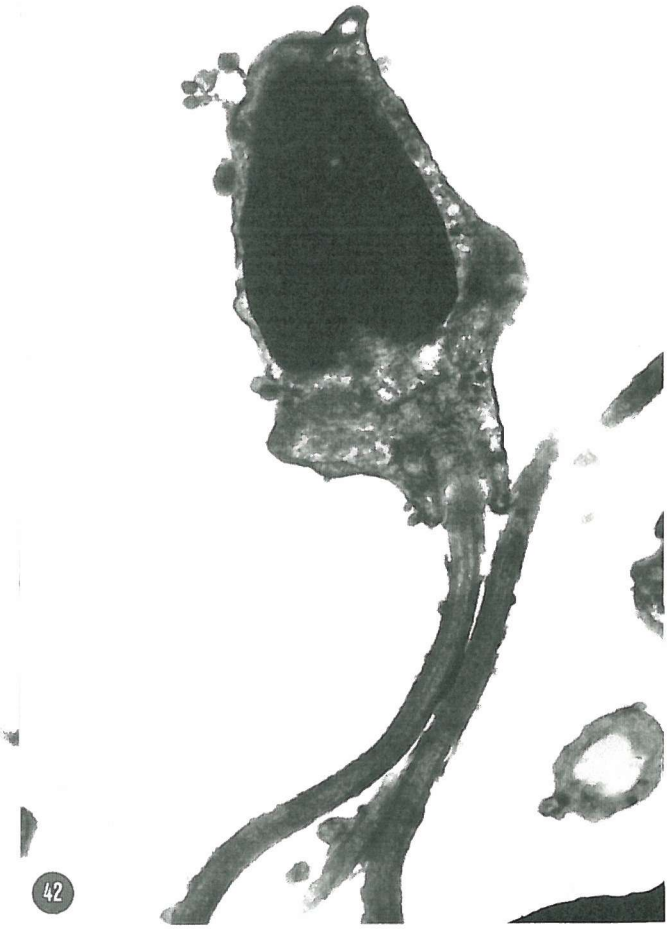
39



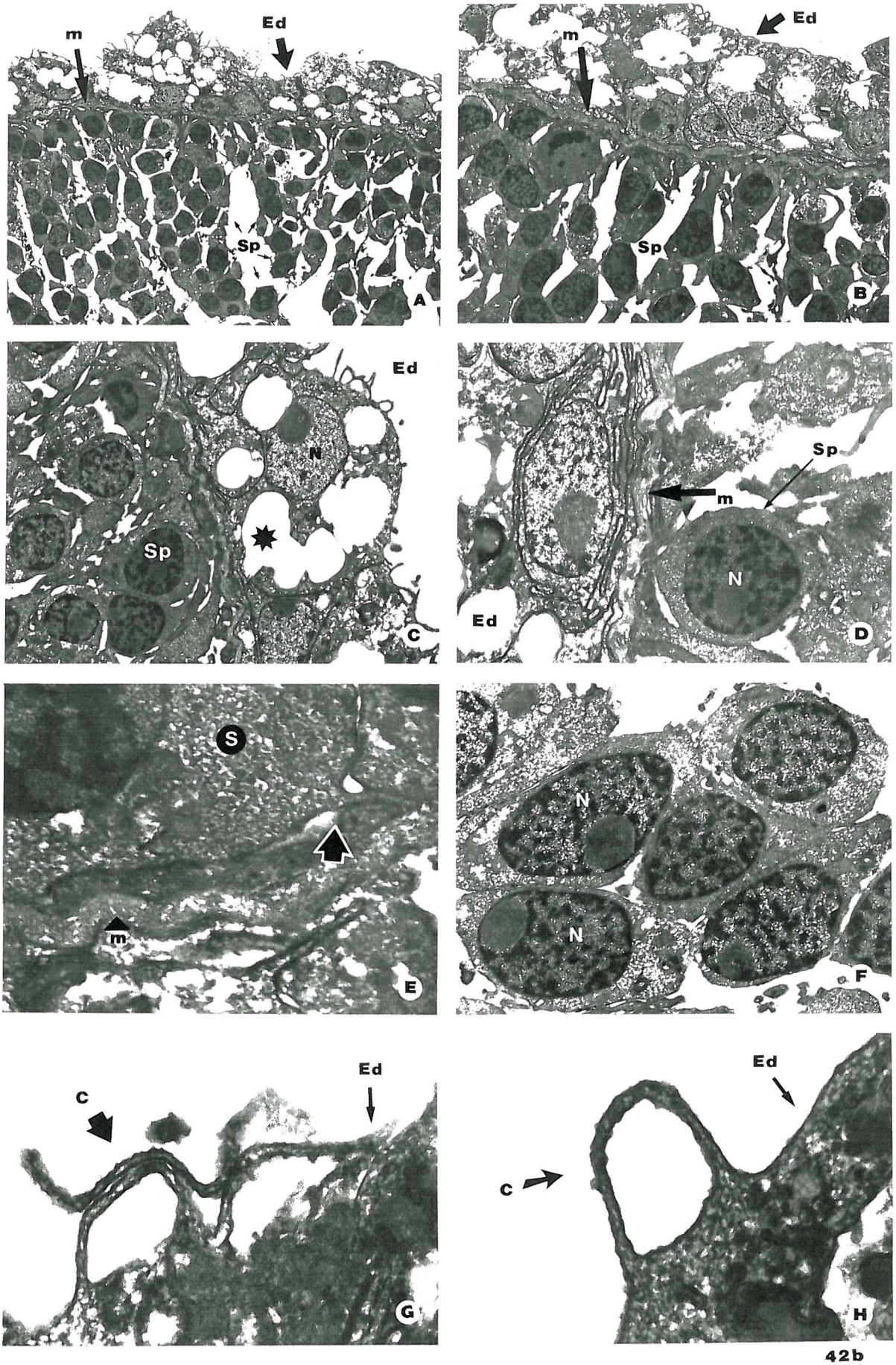
40



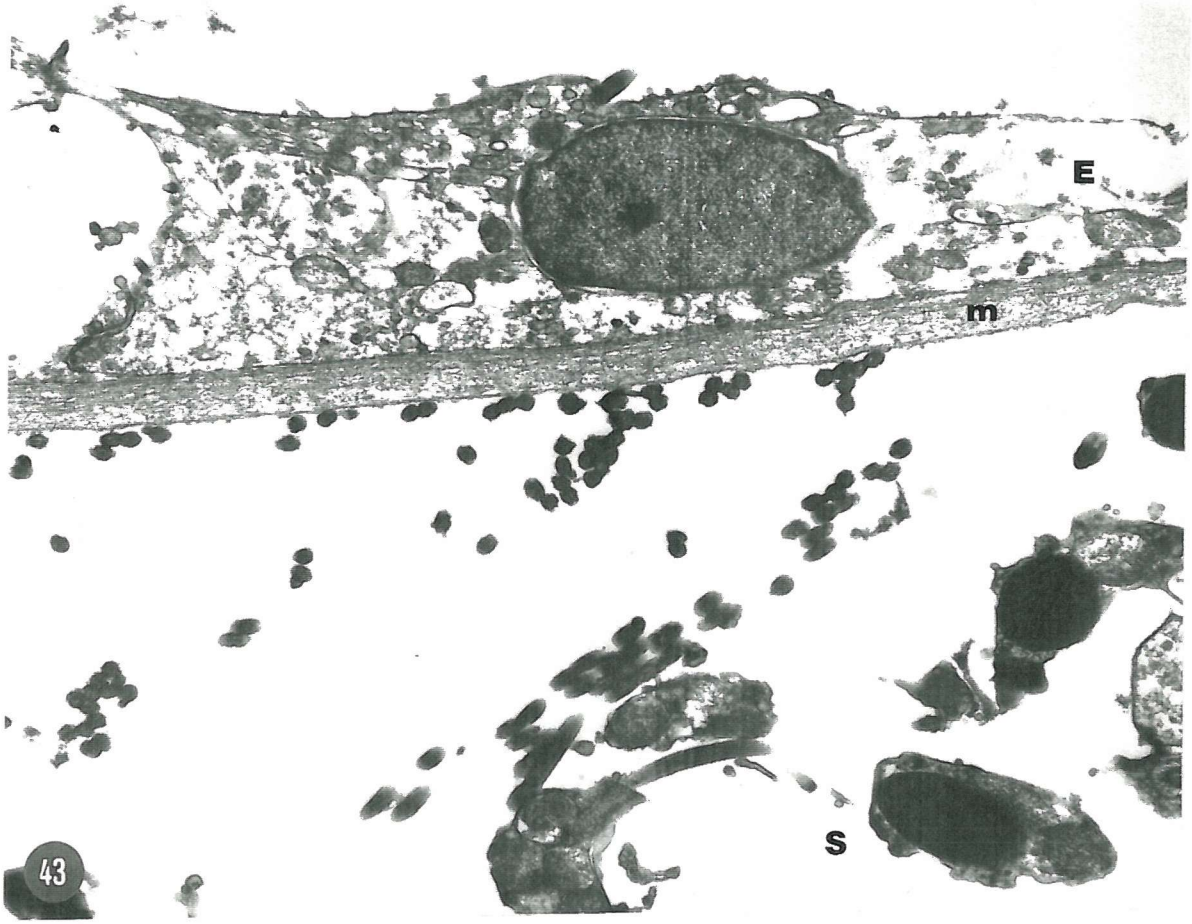
41

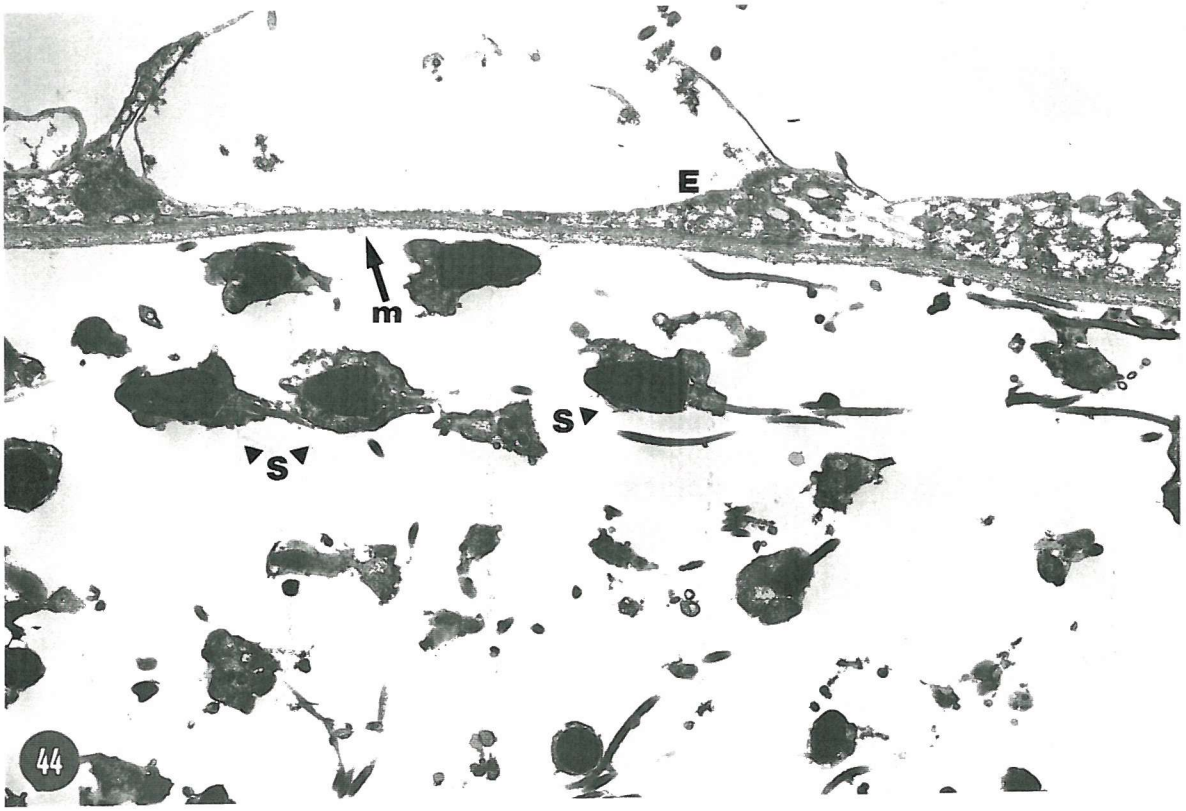


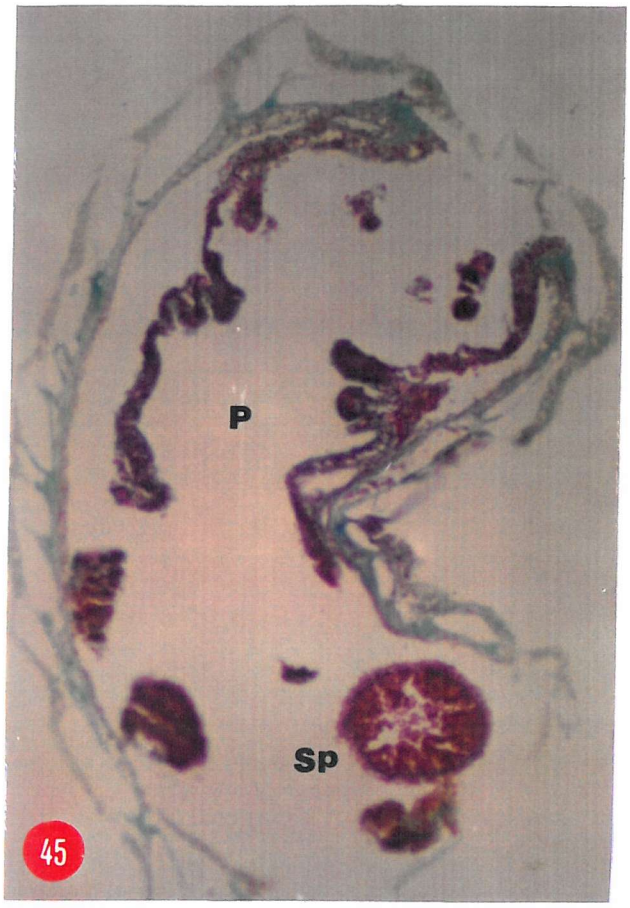
42

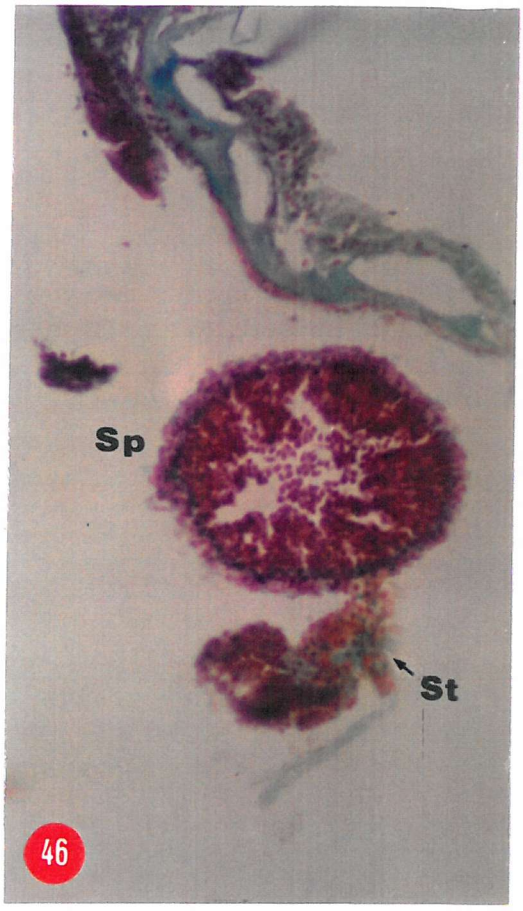


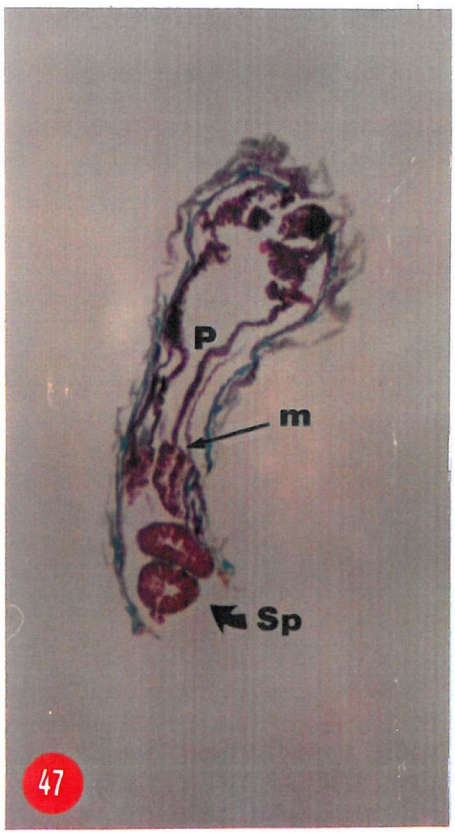
42b

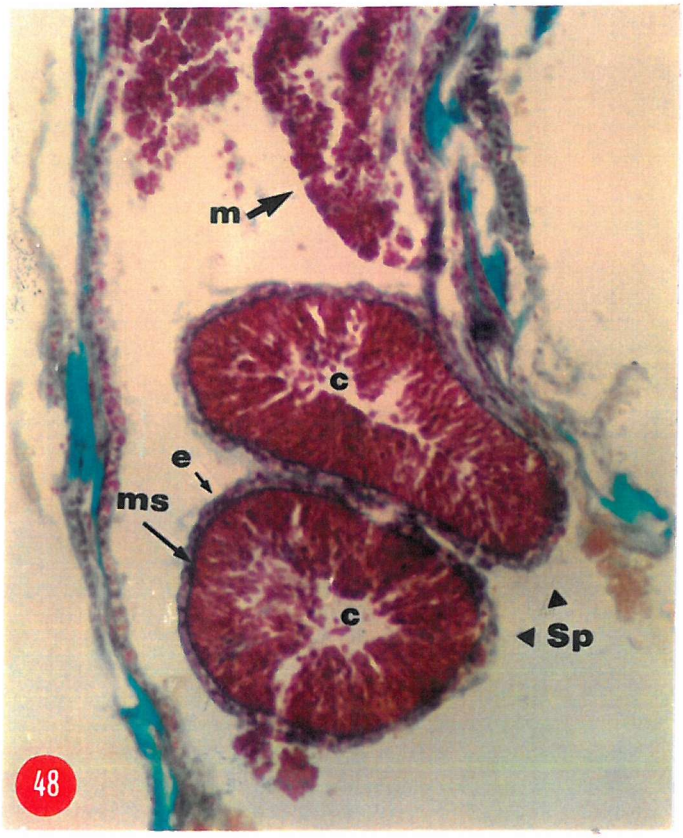

E
m
S
43

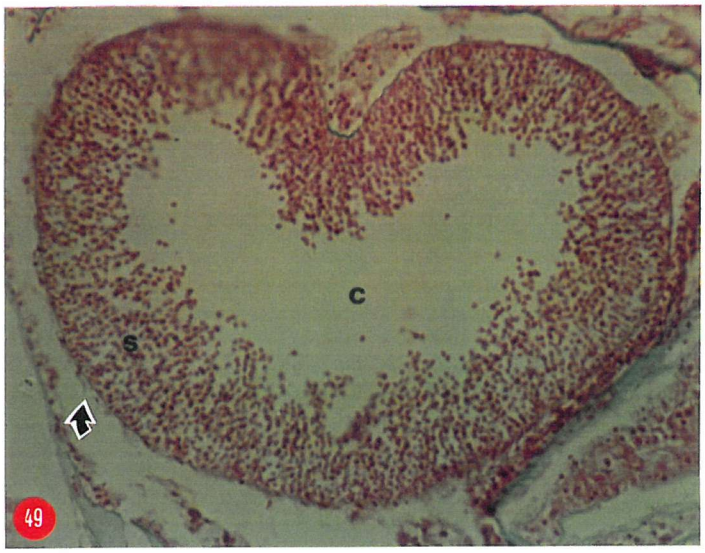

E
m
S
S
44

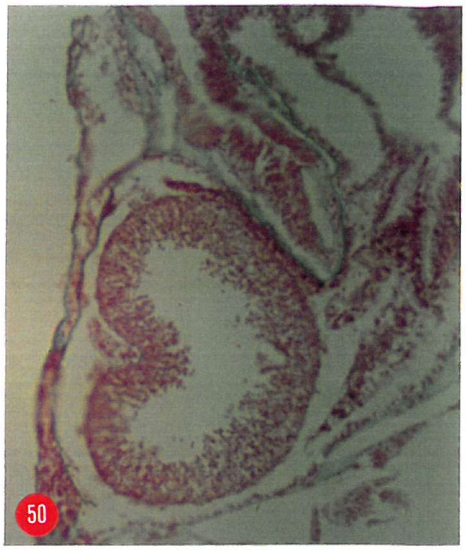

P
Sp
45

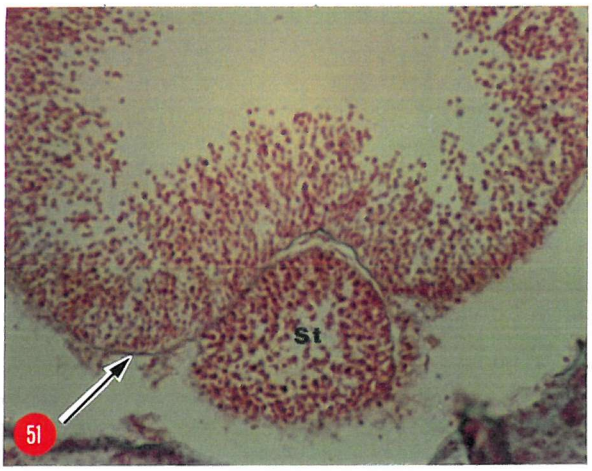

Sp
St
46

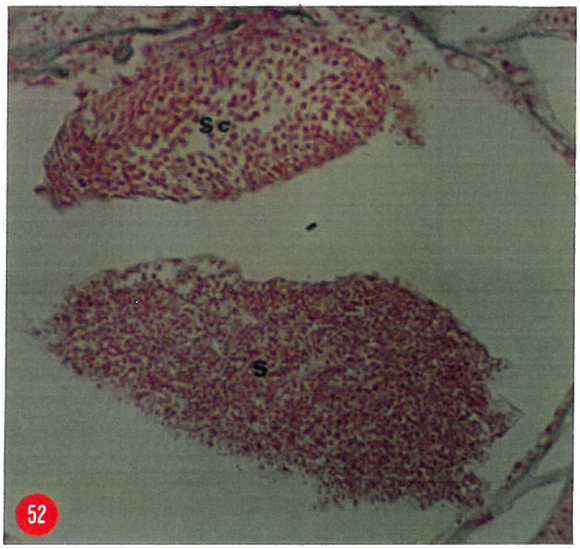

P
m
Sp
47

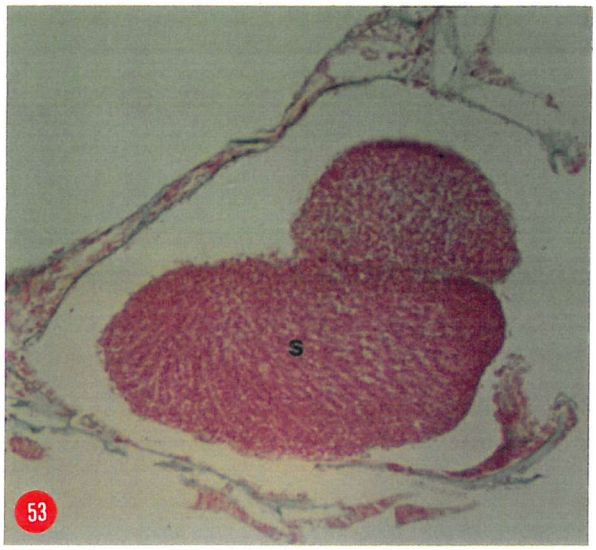

m
c
e
ms
c
Sp
48

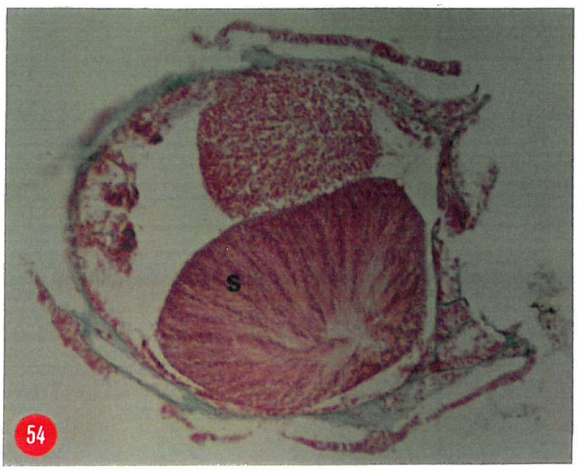

C
S
49

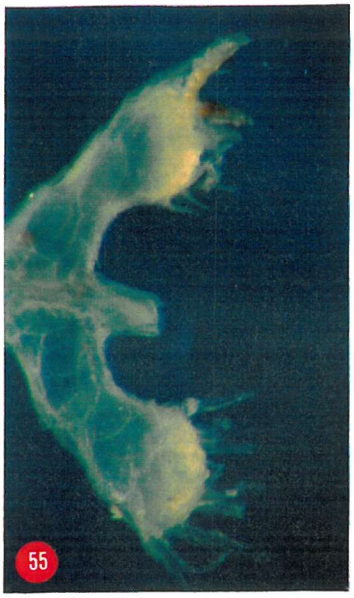

50

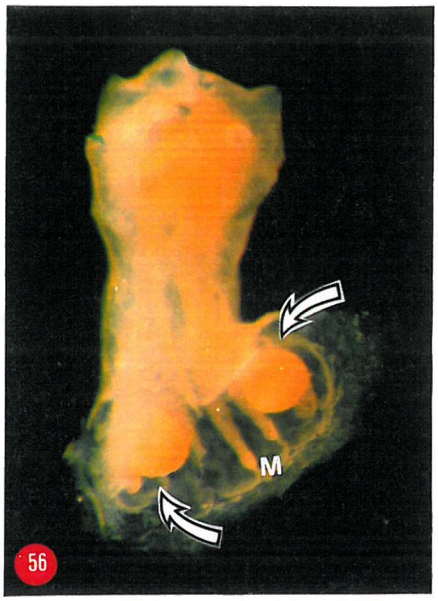

St
51


Sc
S
52

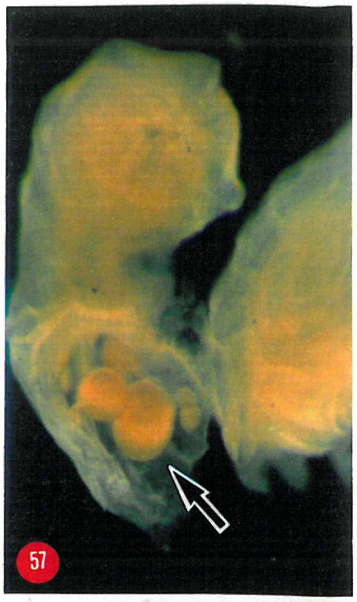

S
53

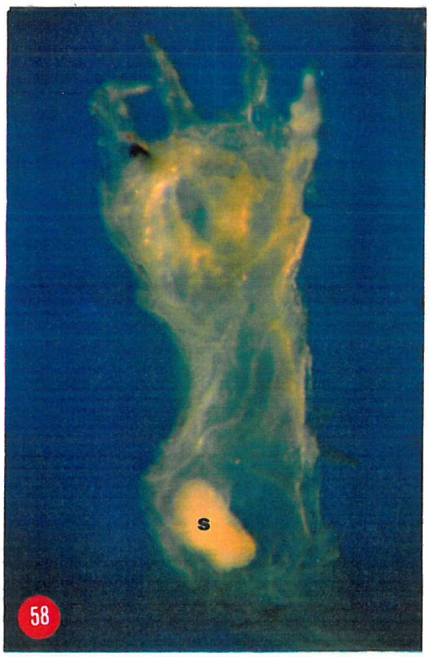

S
54


55

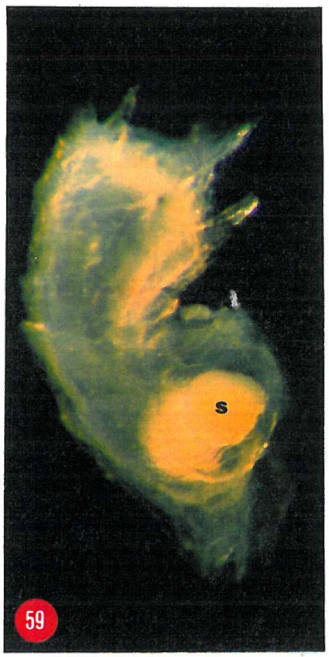


56

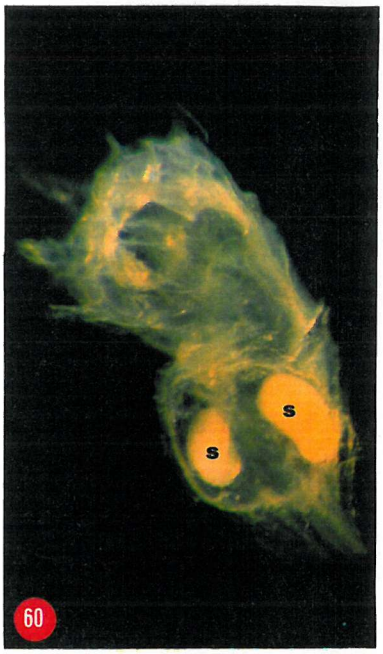

57



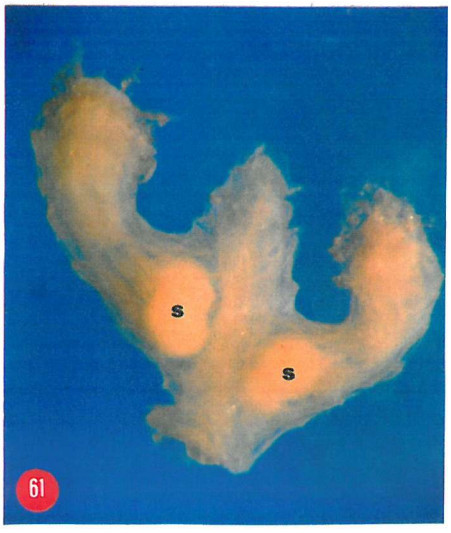
58



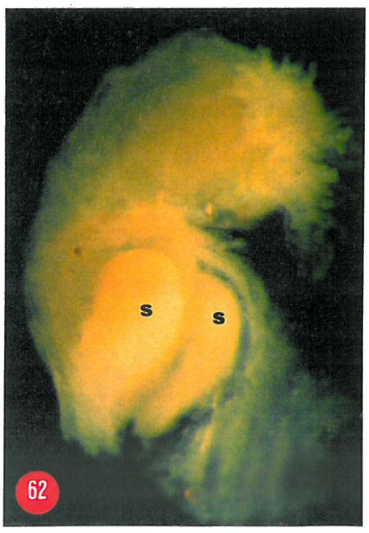
59



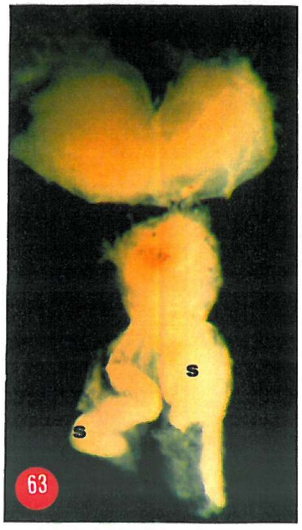
60



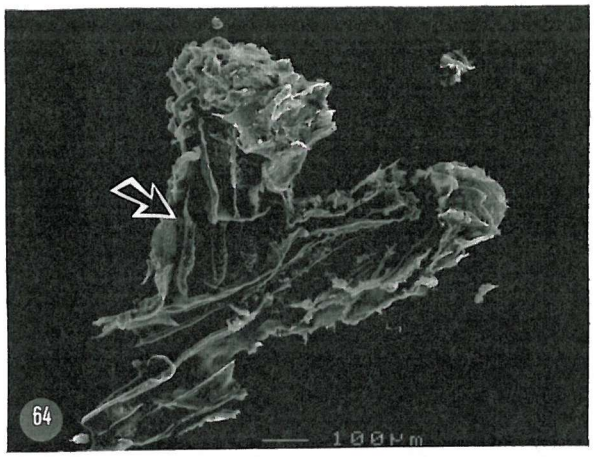
61

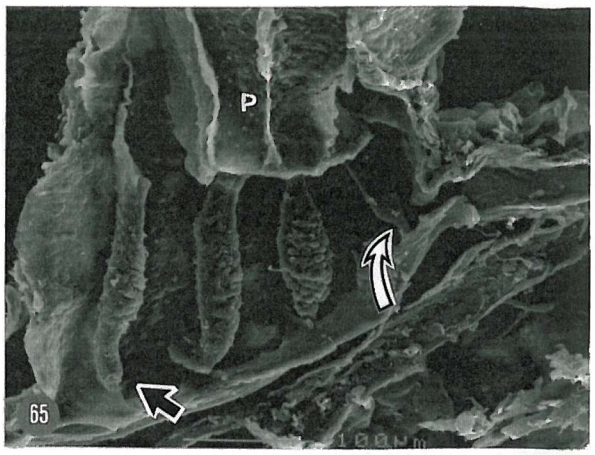


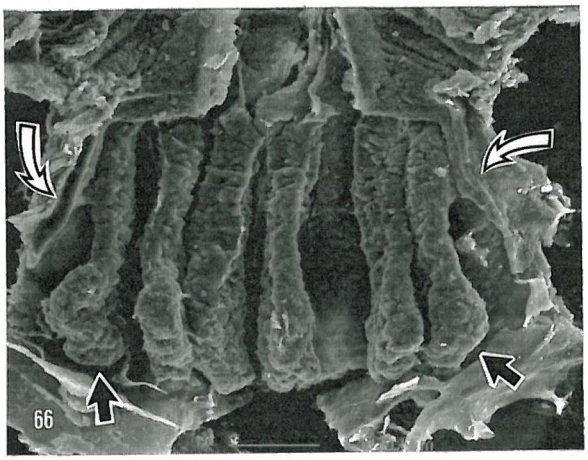
62

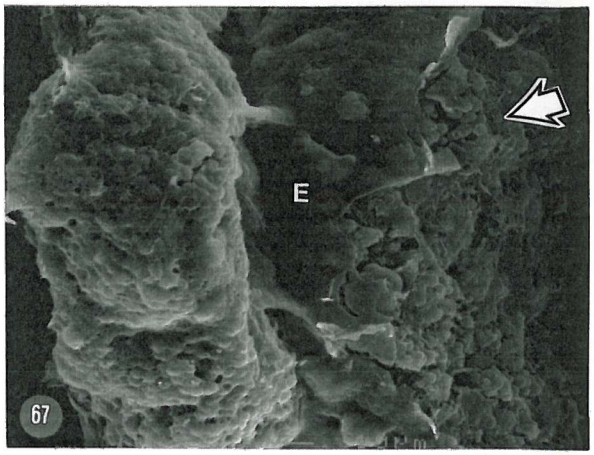


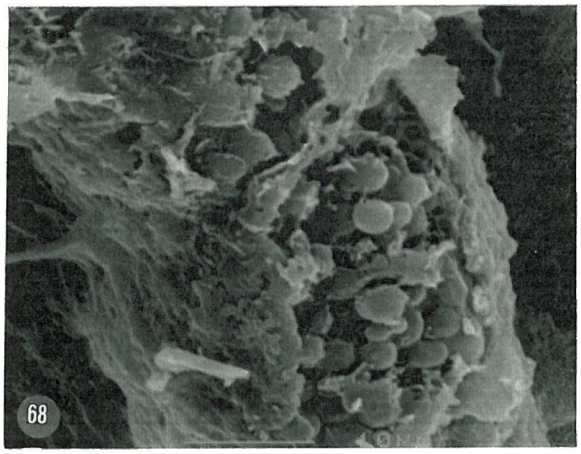
63

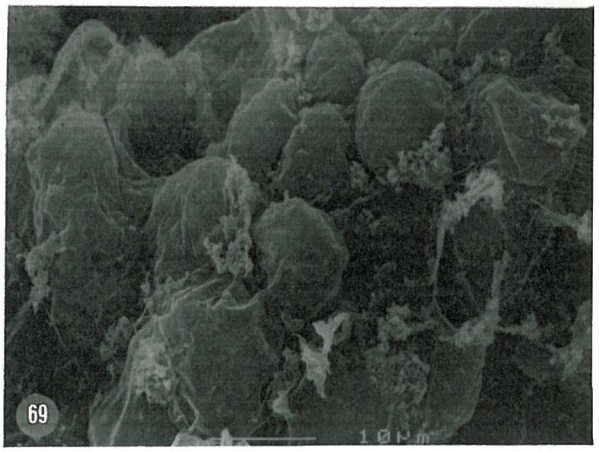

64
100µm

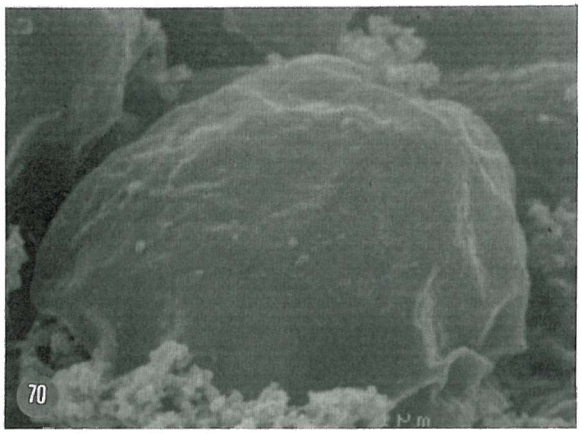

P
65
100µm

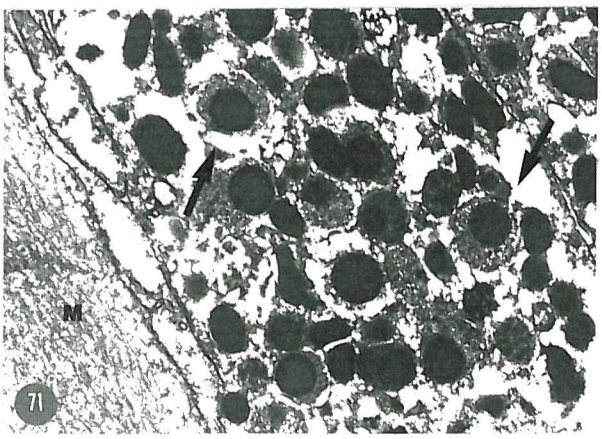

66

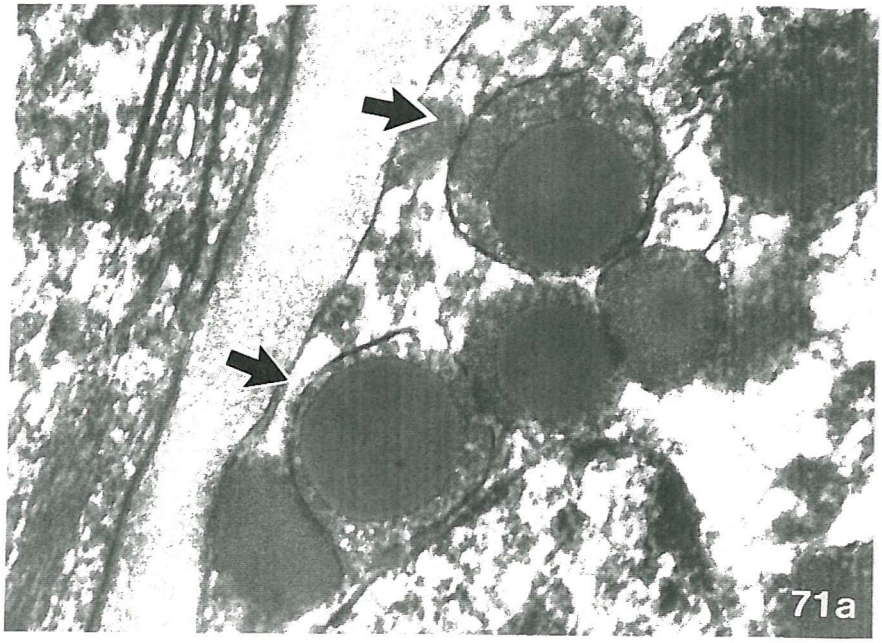

E
67

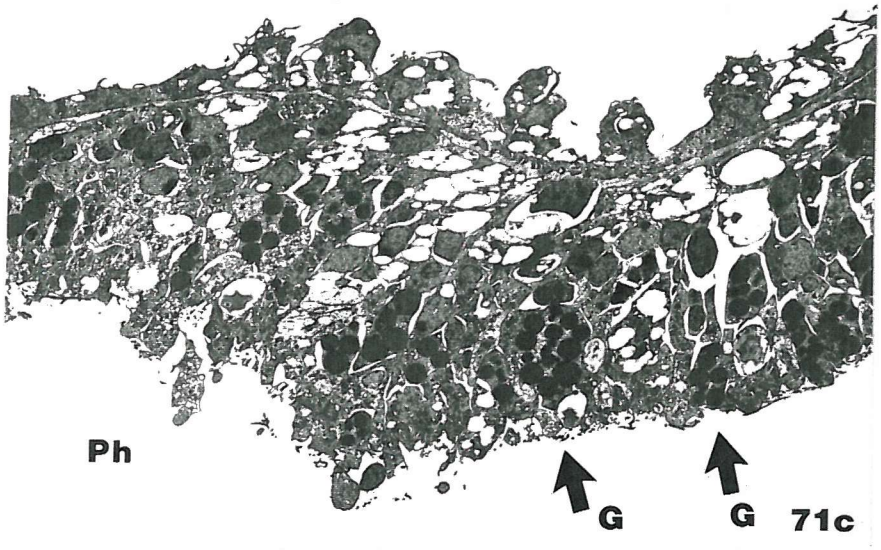

68

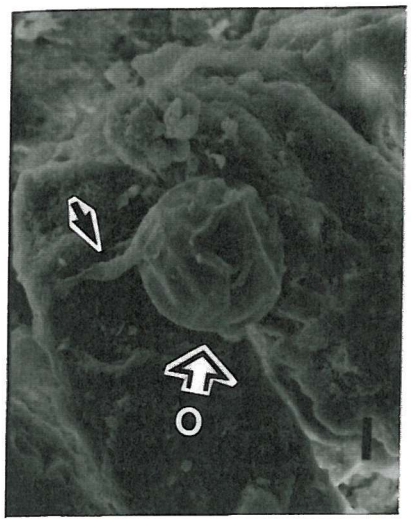

69
10µm

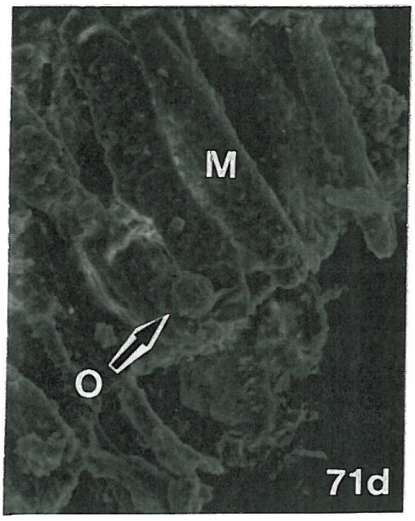

70

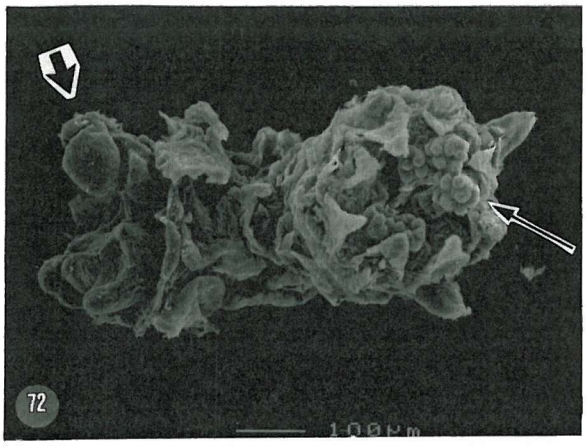

M
71

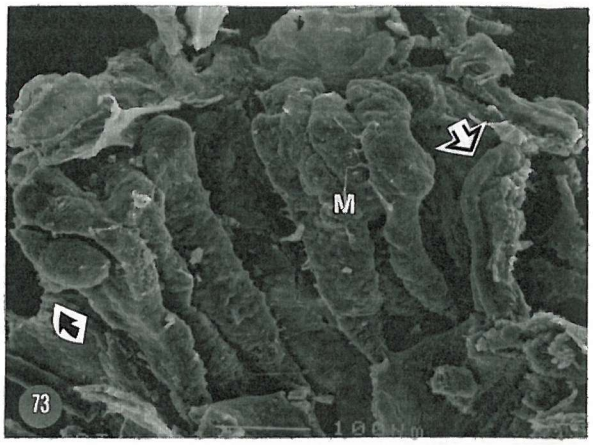

71a

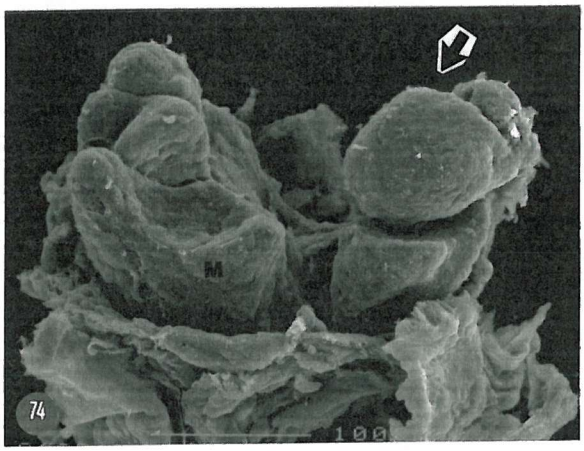

Ph
G
G
71c

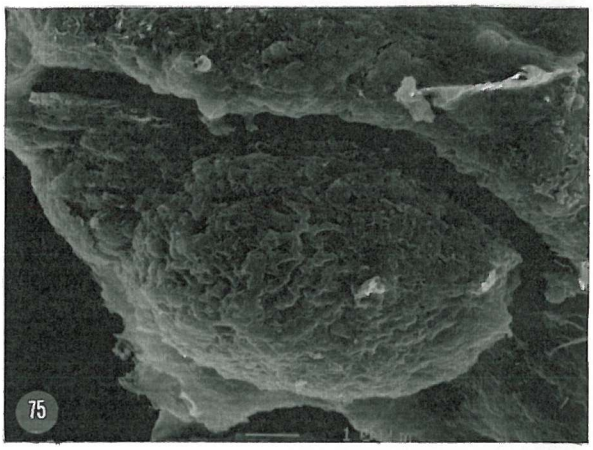

O
I

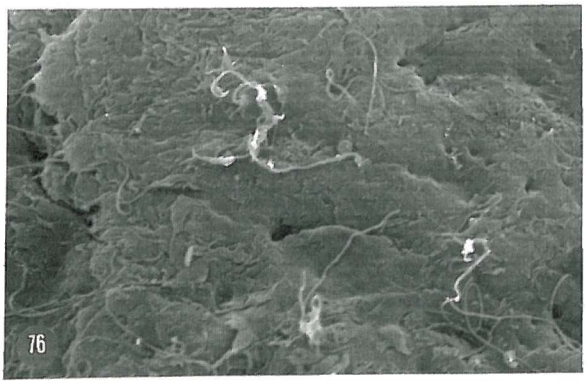

M
O
71d

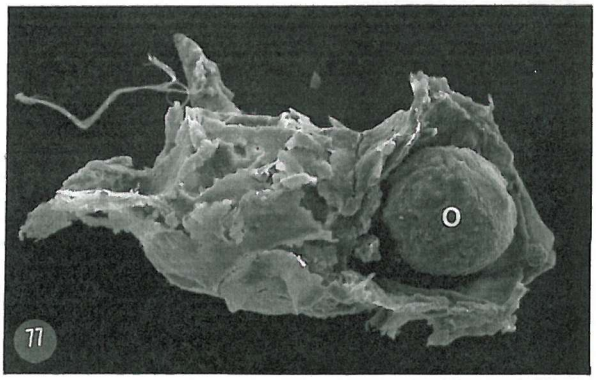

72
100µm

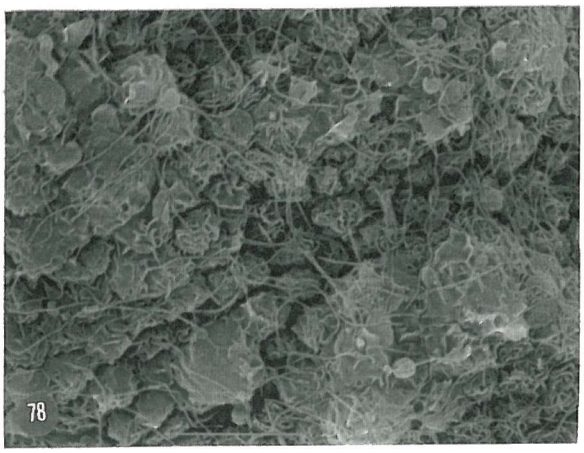

M
73
100µm

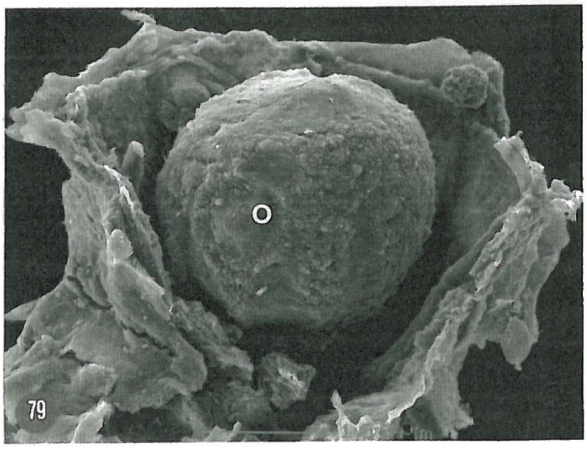

M
74
100

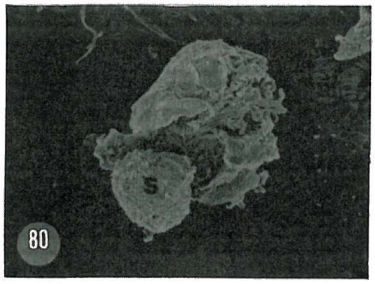

75


76

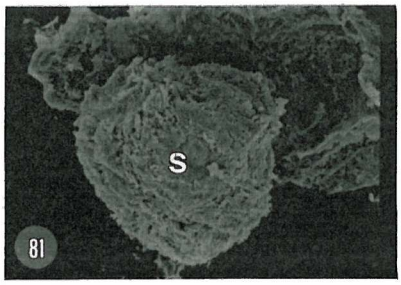

O
77


78


O
79

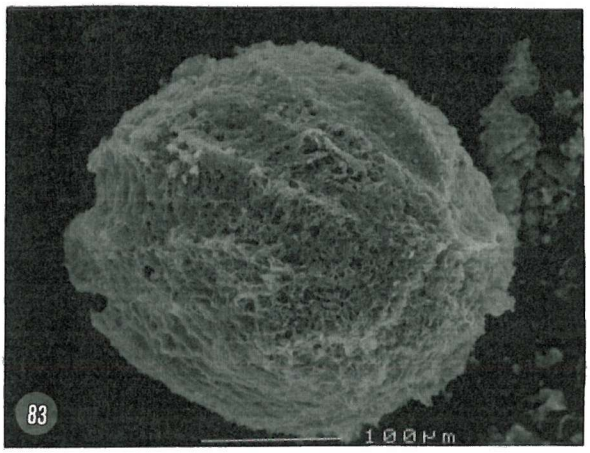


80



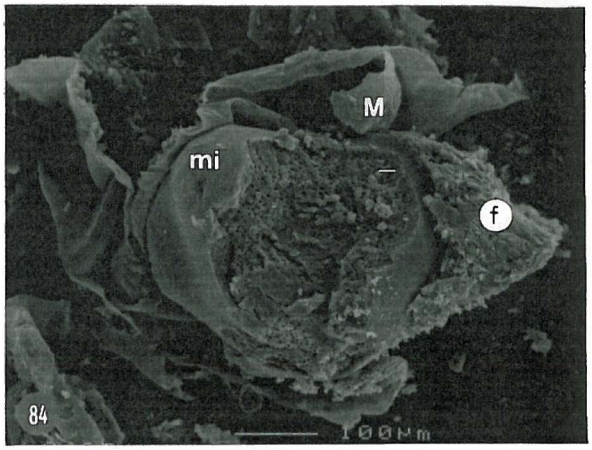
81

82



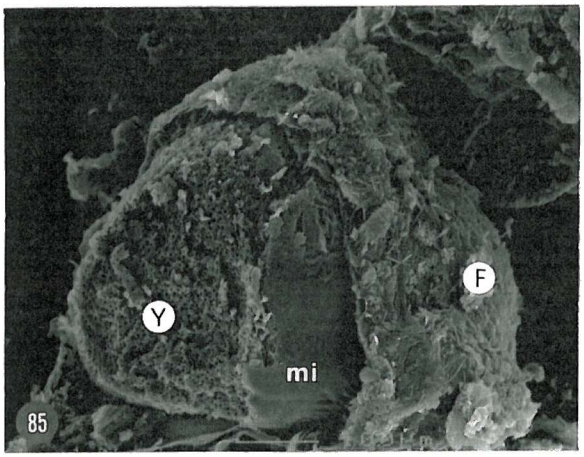
83

100μm

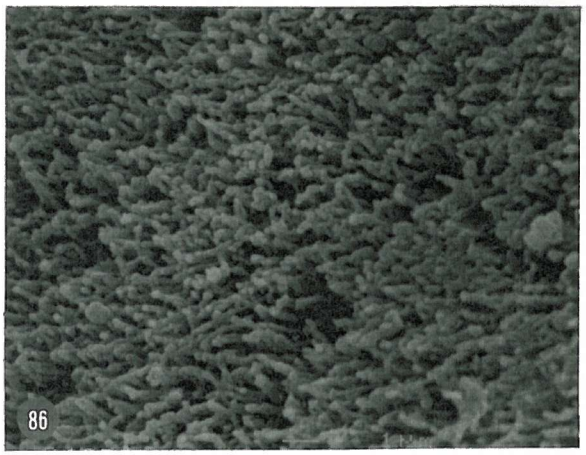


84

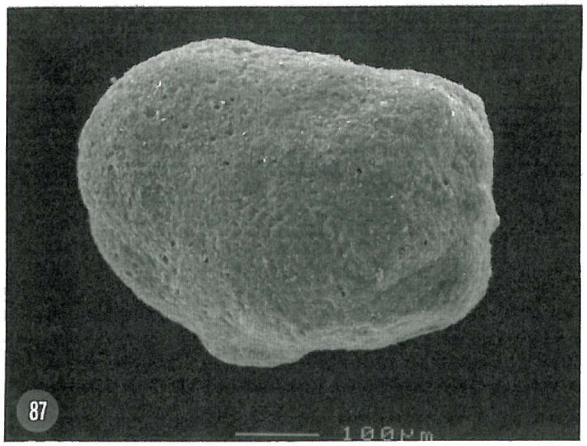
100μm



85

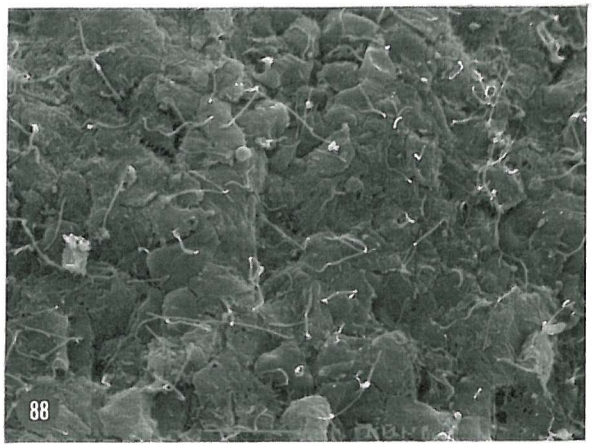


86

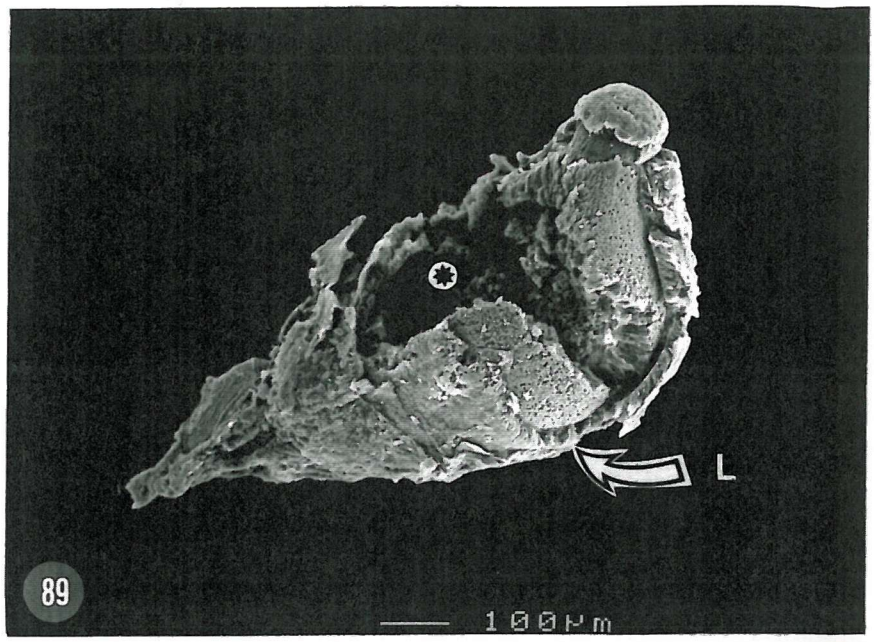


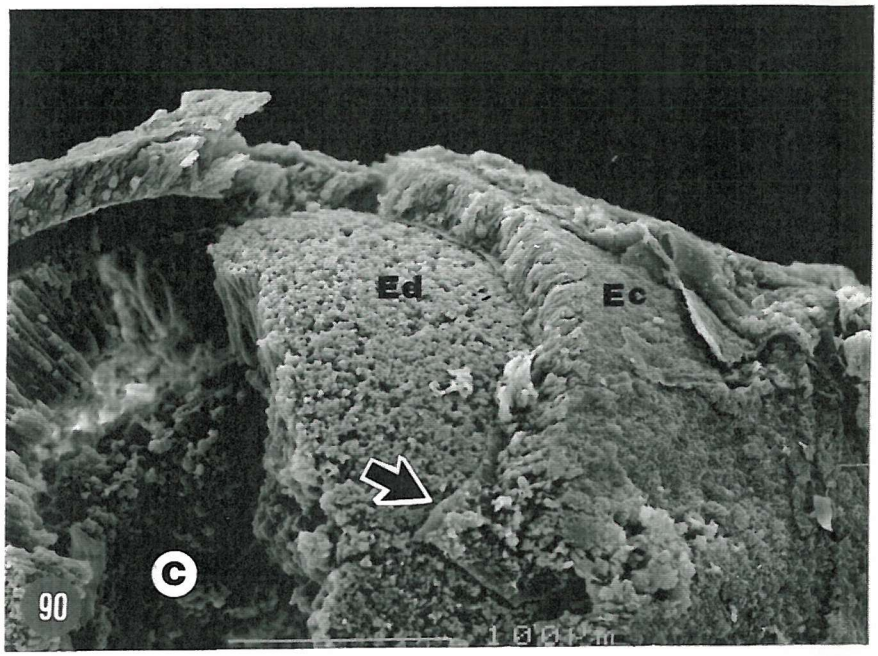
87

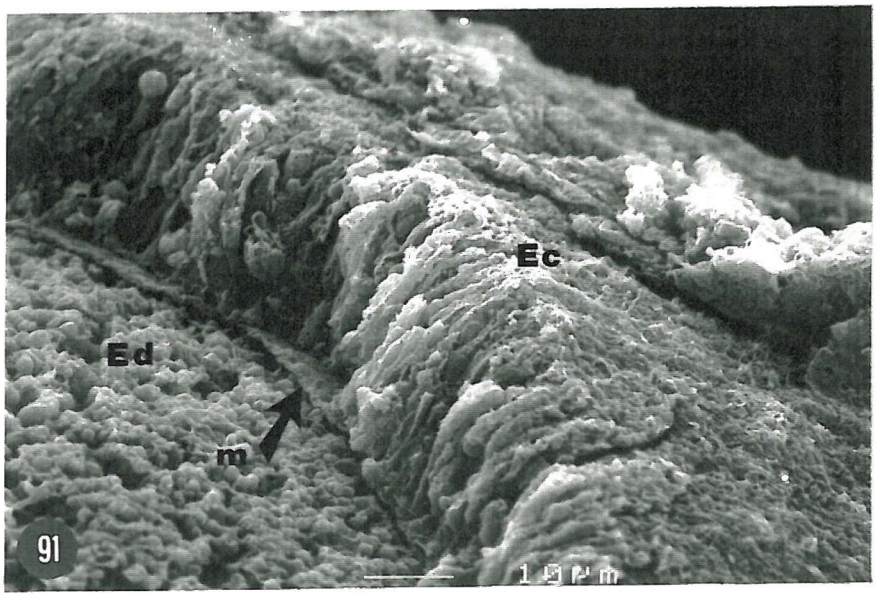
100μm

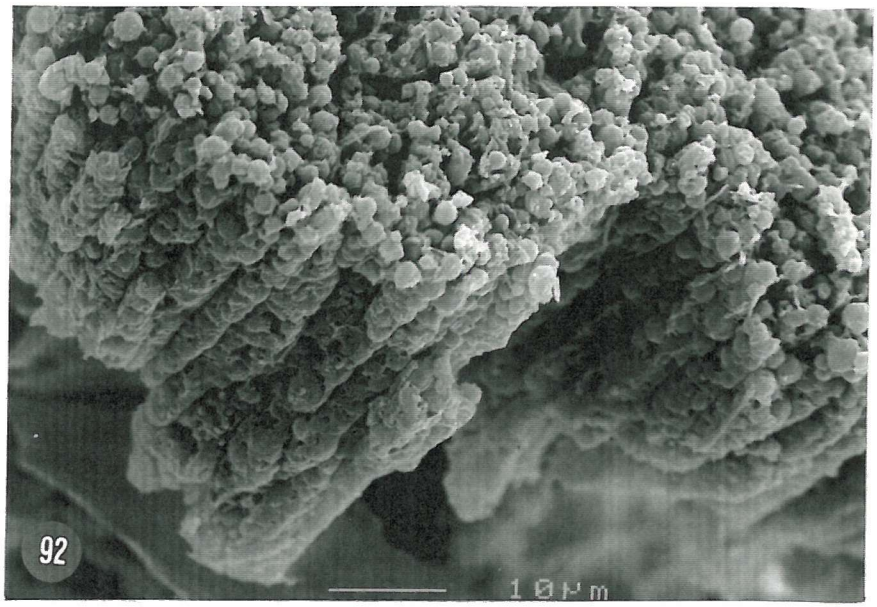


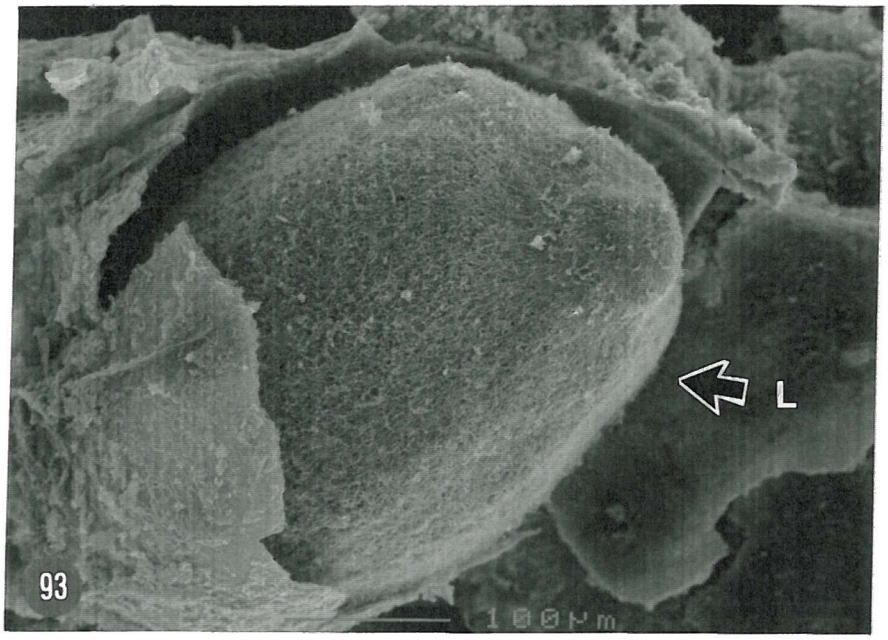
88

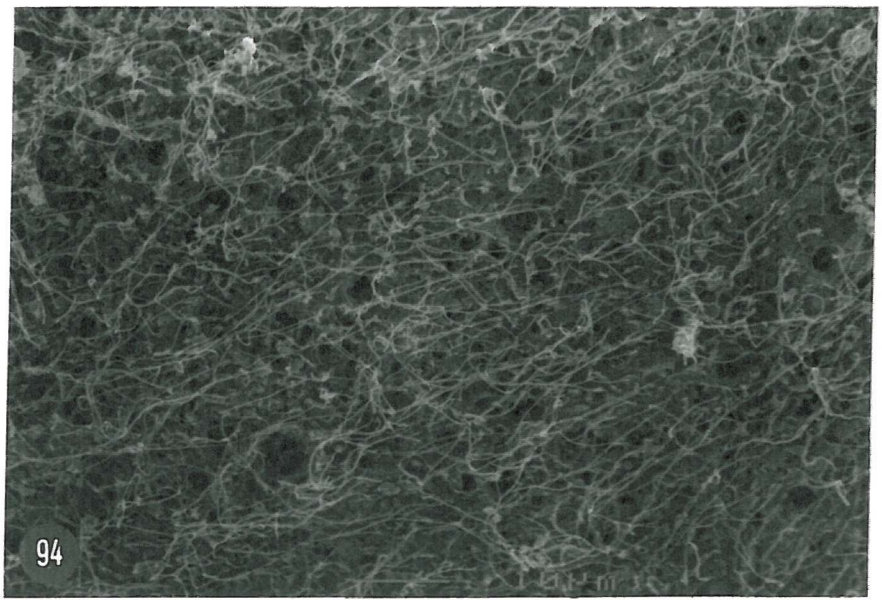

L
89
100µm

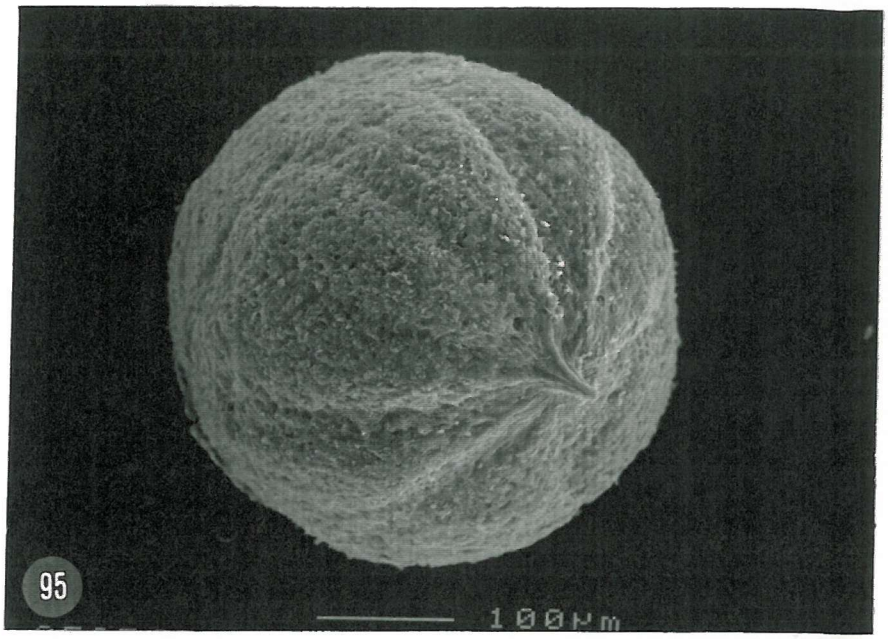

Ed
Ec
C
90
100µm

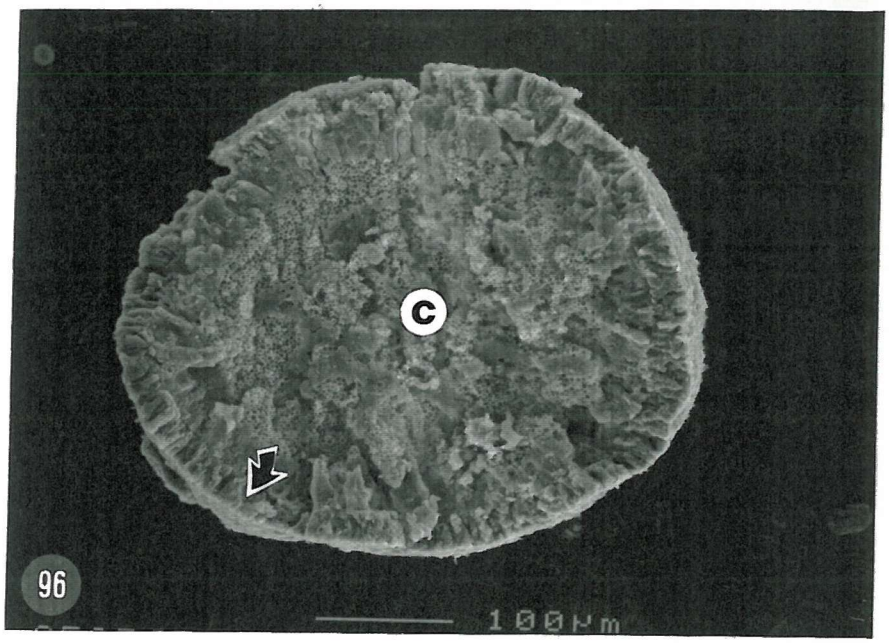

Ec
Ed
m
91
10µm


92
10µm

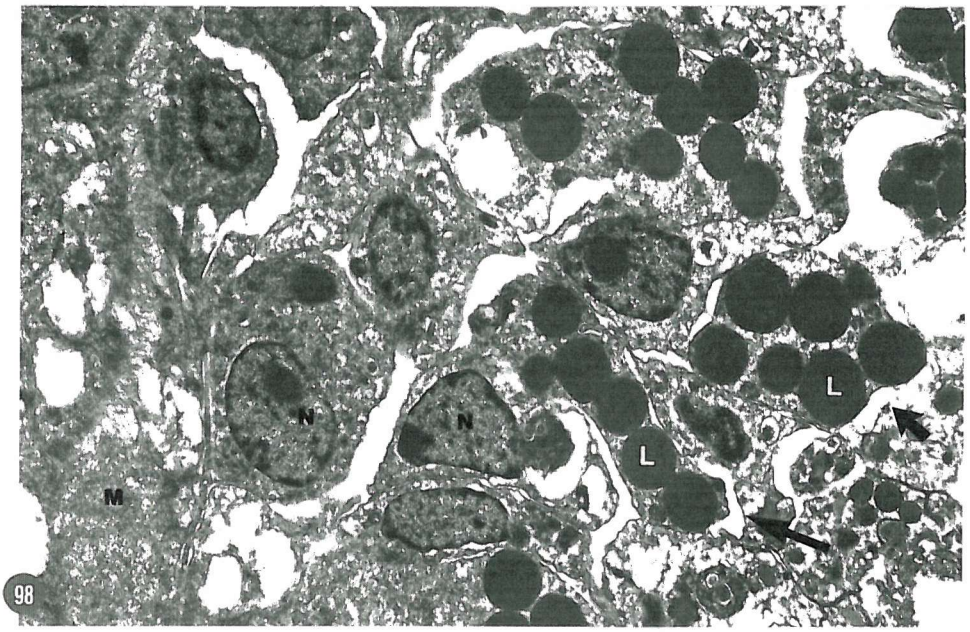

L
93
100µm


94

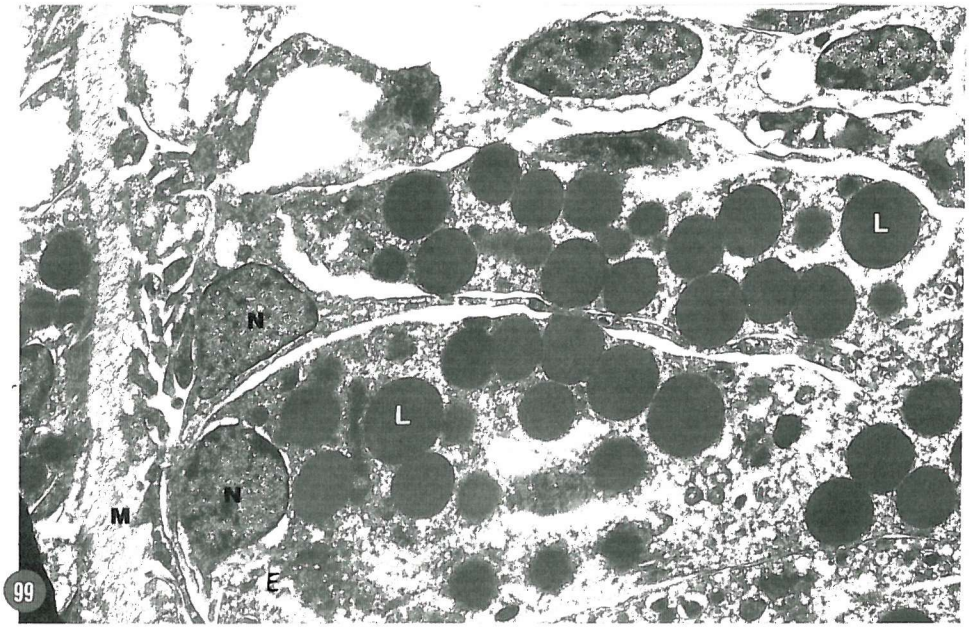

95
100µm


C
96
100µm

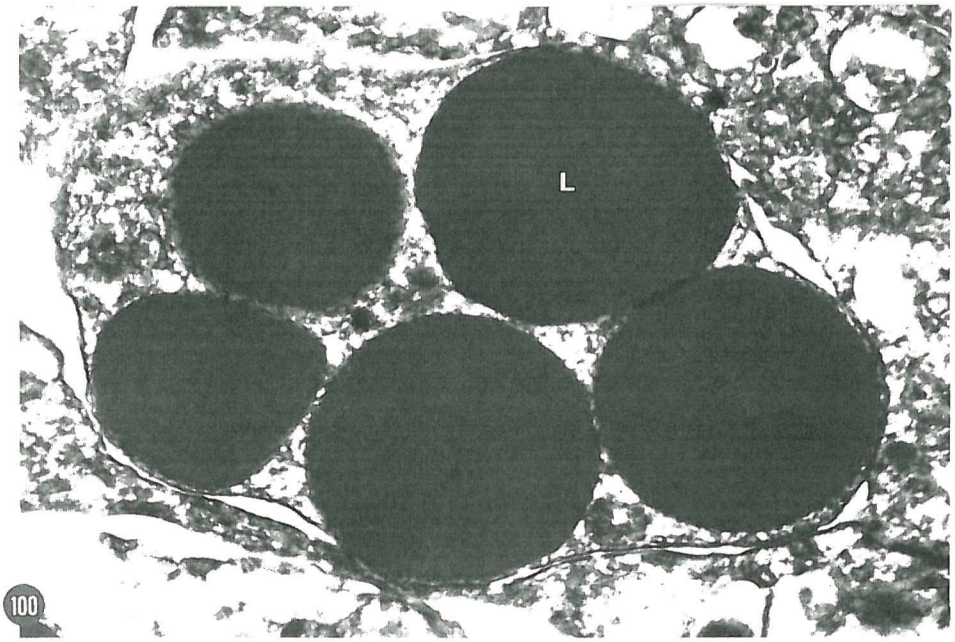

97



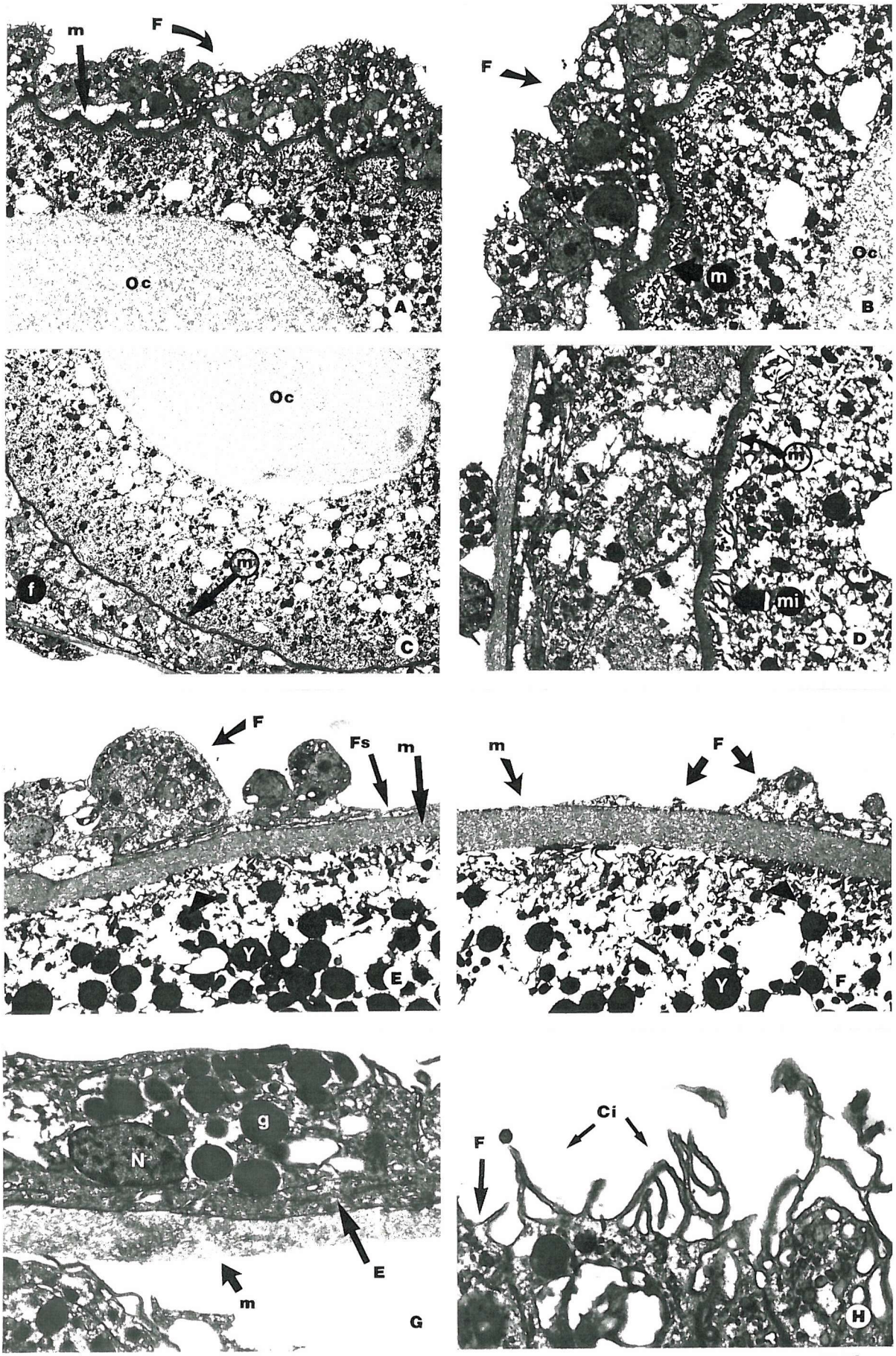
98



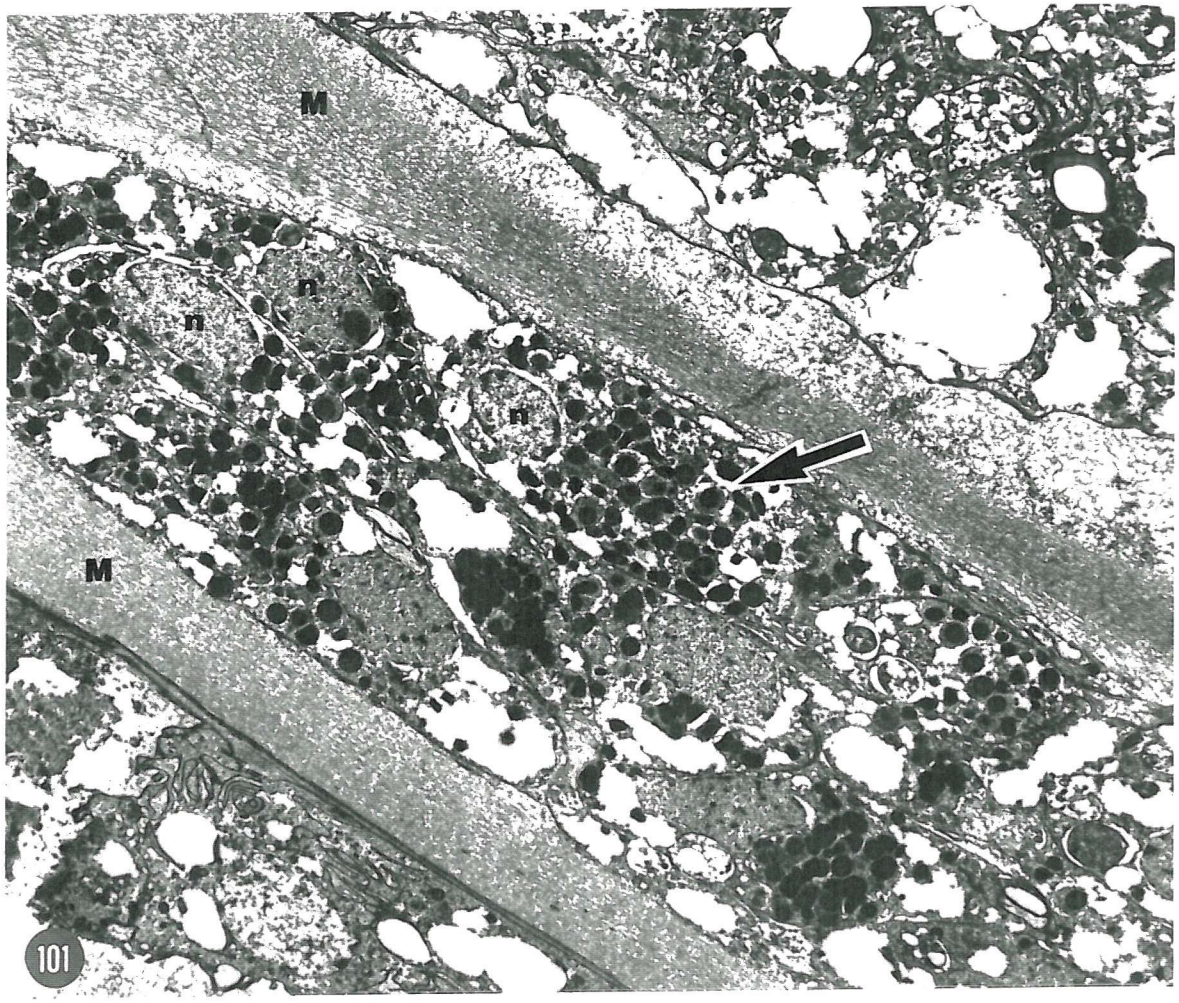
99

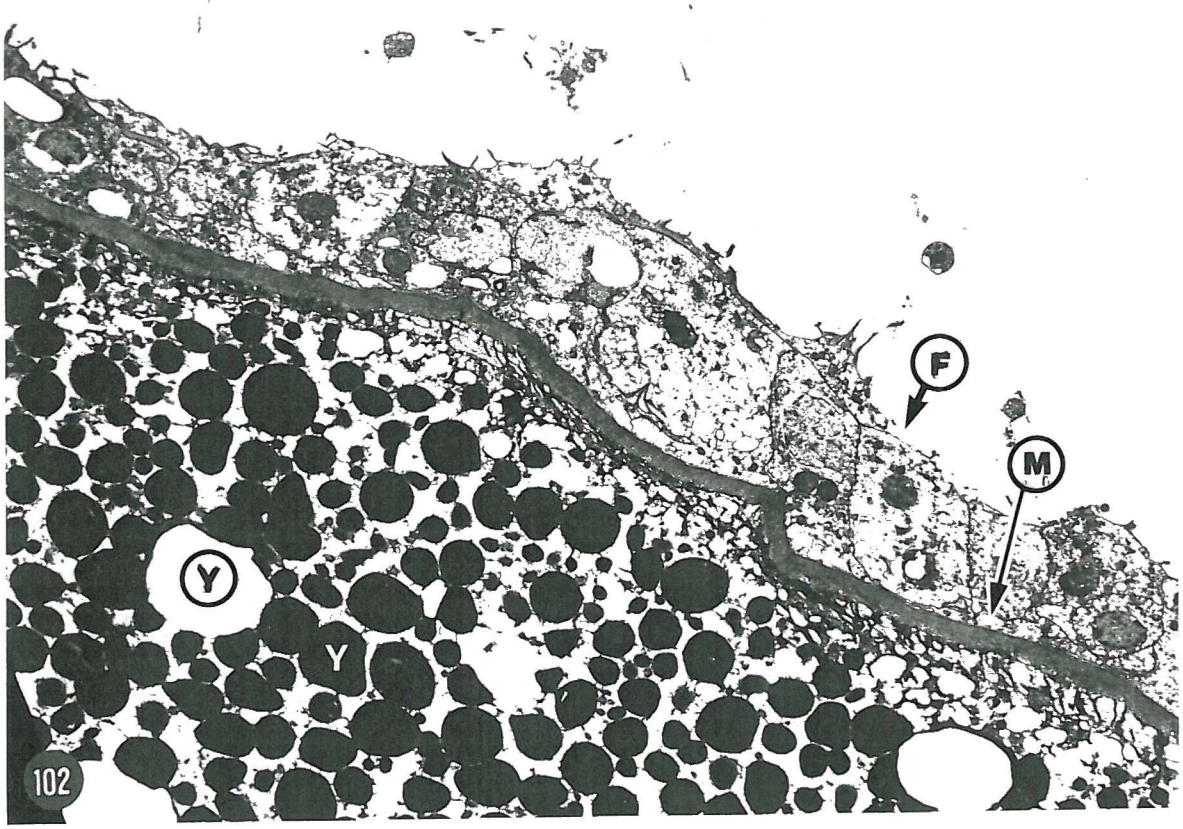


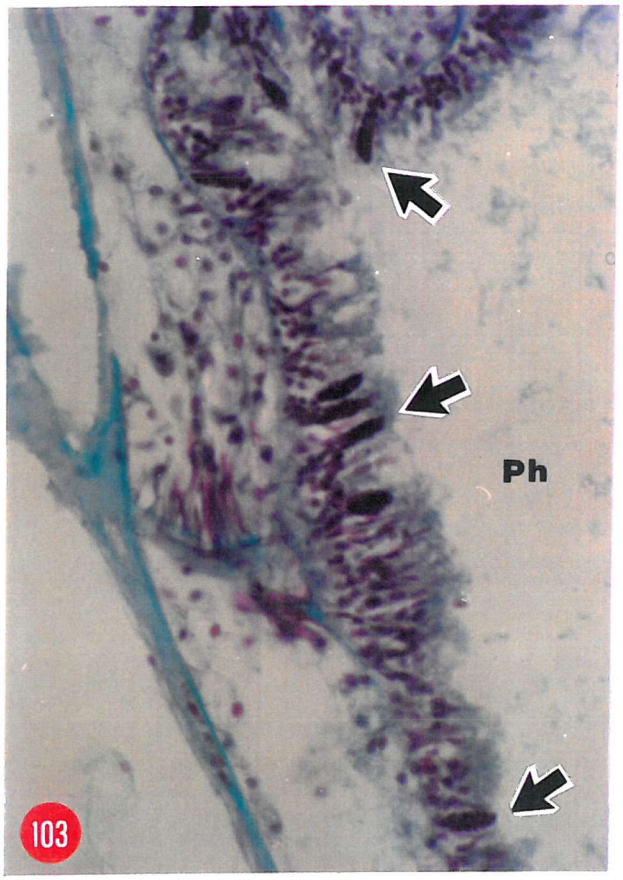
100

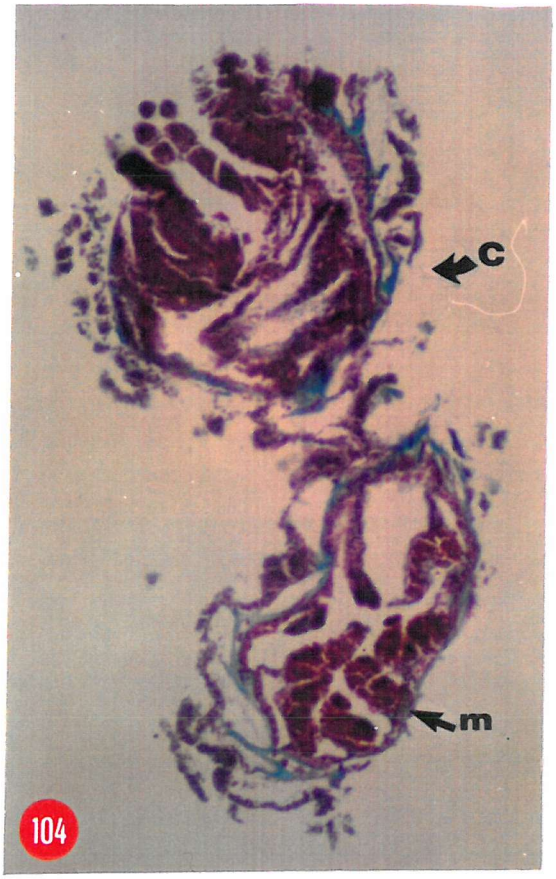


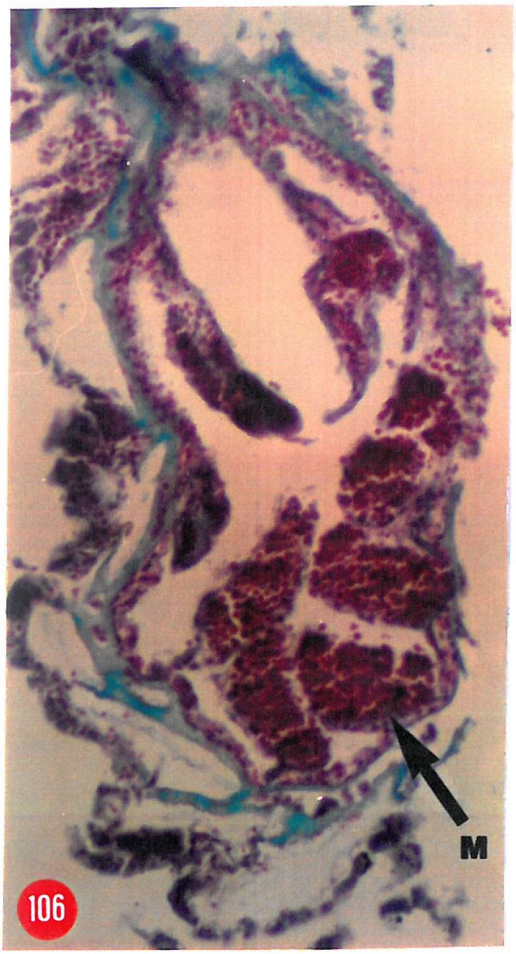
100 b

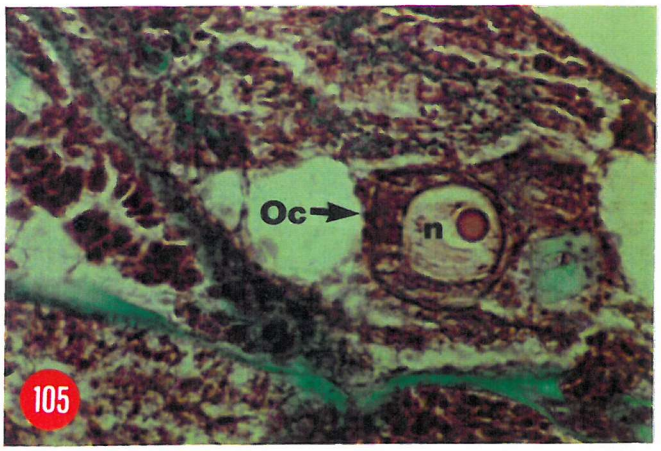

M
n
n
n
M
101

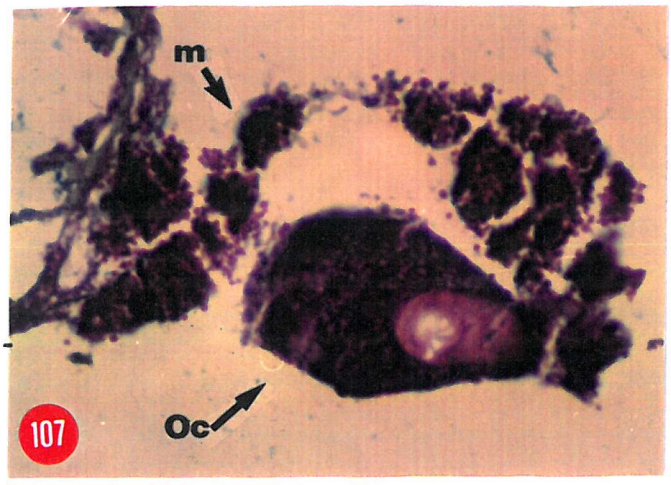

F
M
Y
Y
102

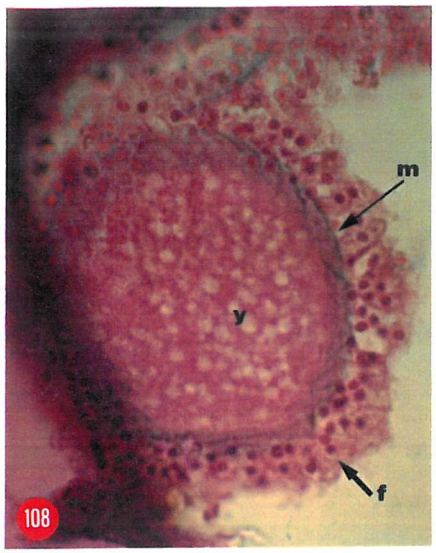

Ph
103

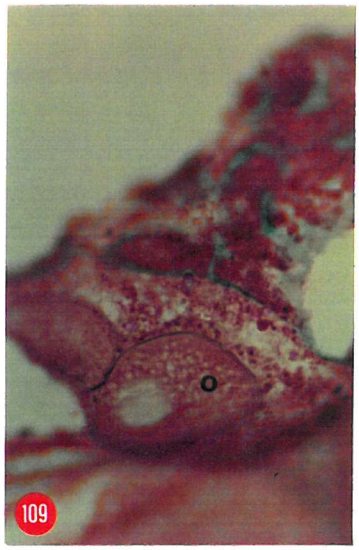

C
m
104

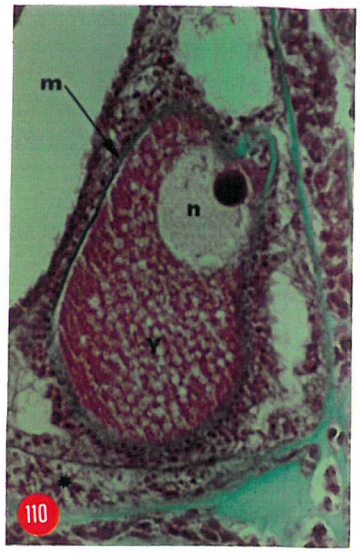

M
106

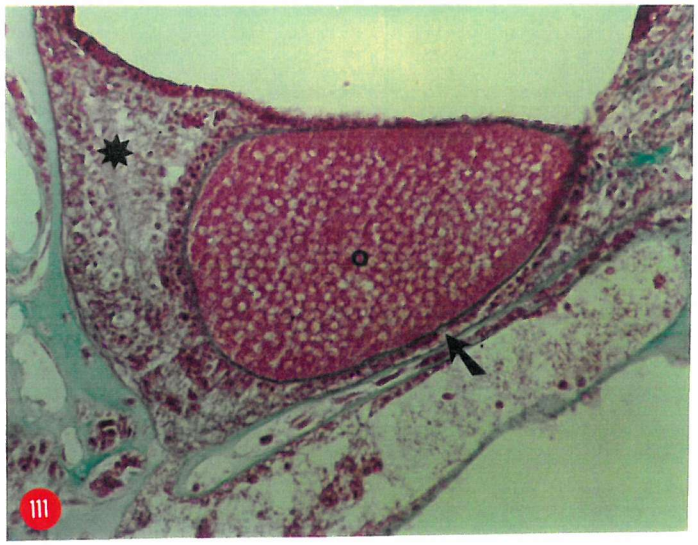

Oc
n
105

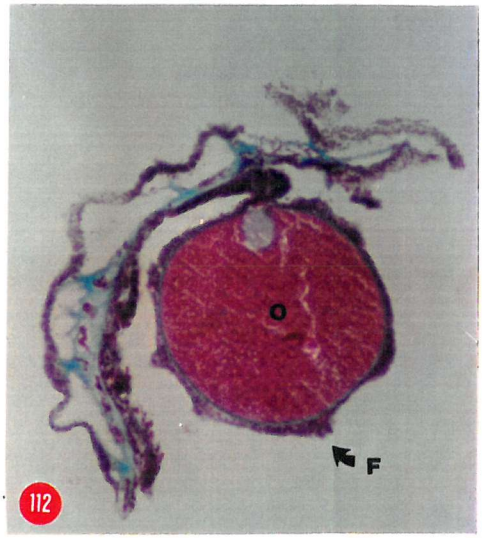

m
Oc
107

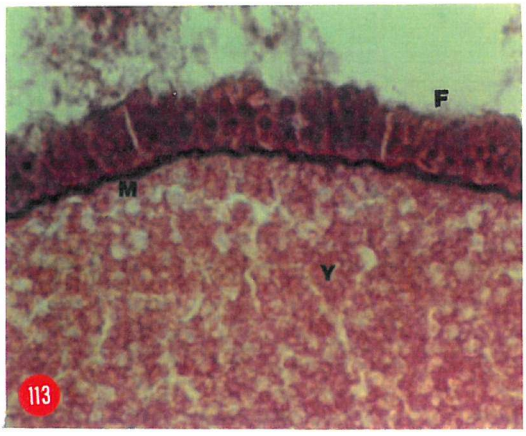

m
y
f
108

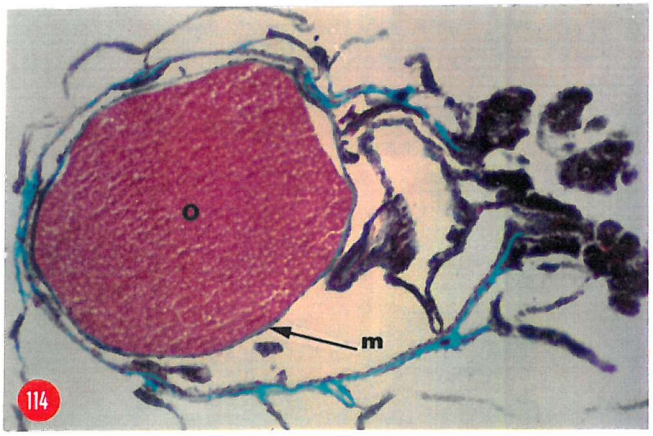

O
109

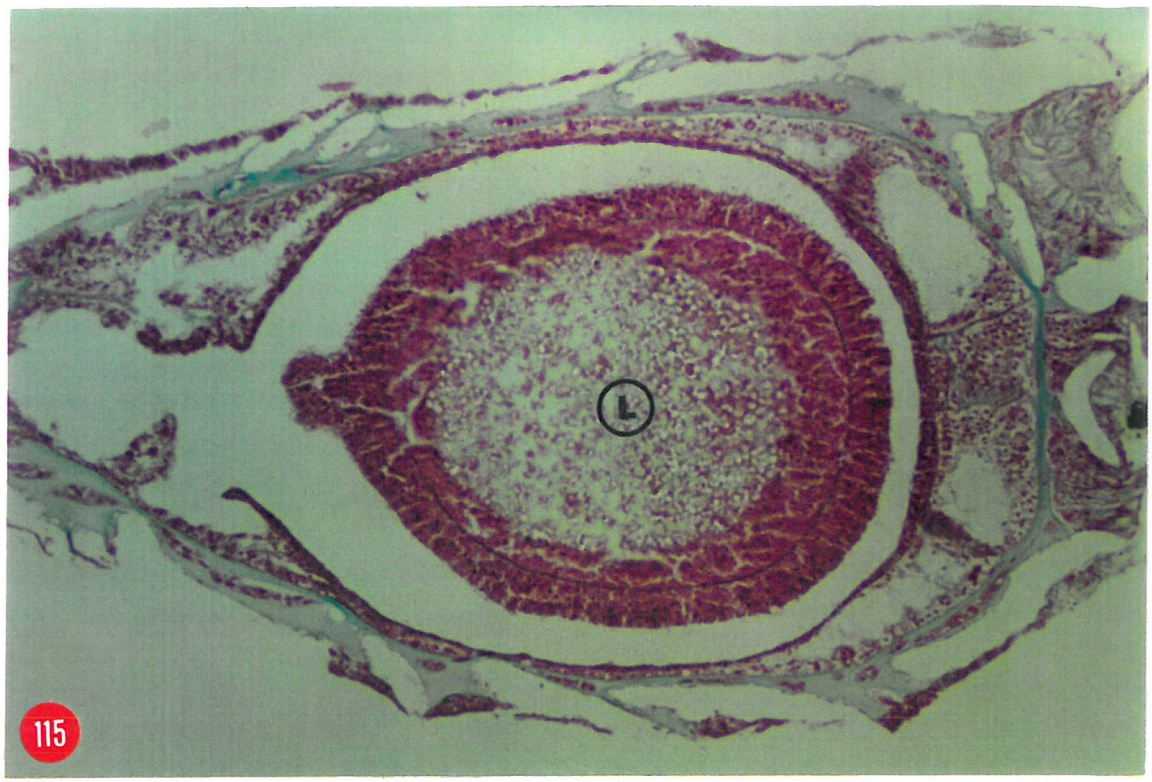

m
n
Y
110


O
111

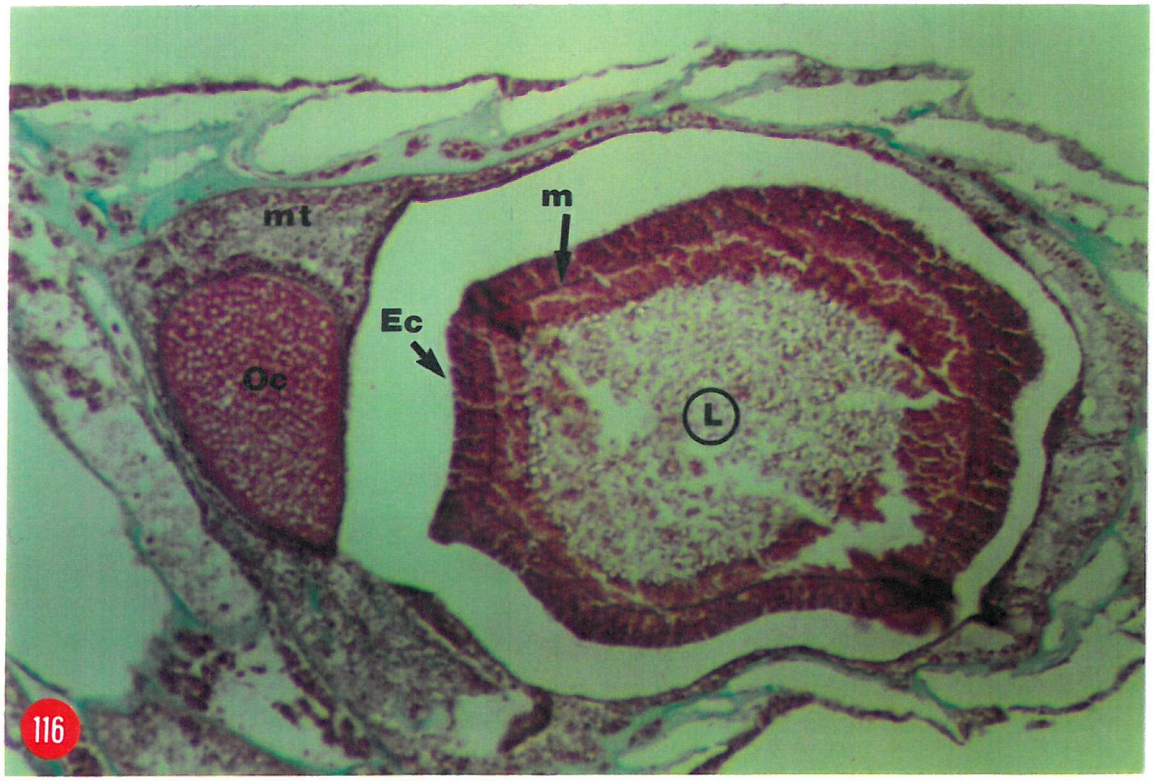

O
F
112


F
M
Y
113

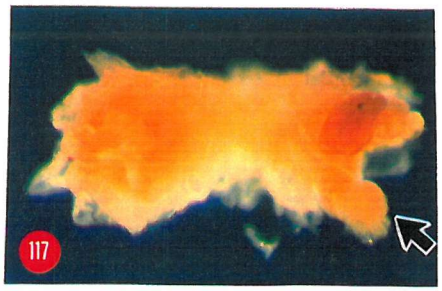

O
m
114

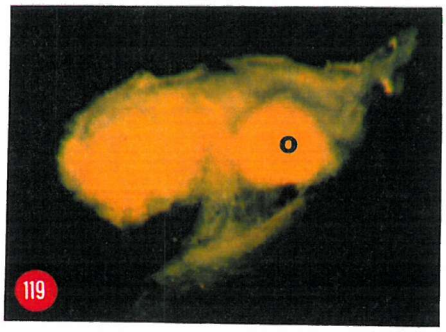


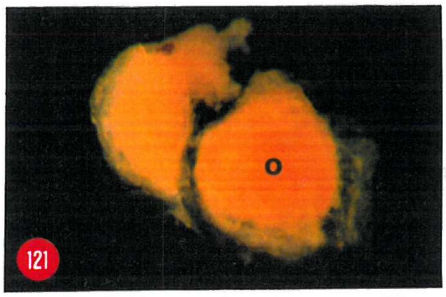
115

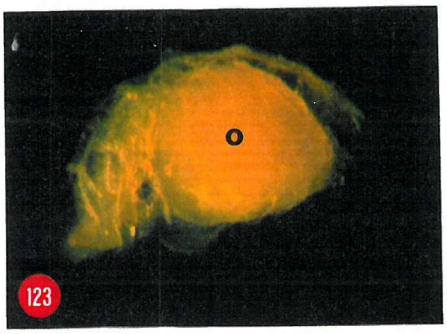


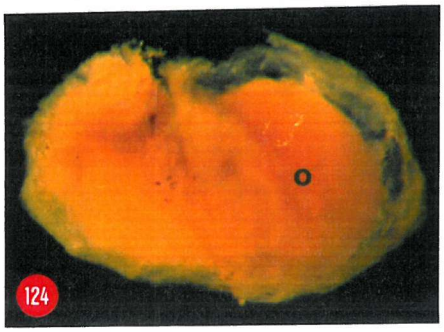
116

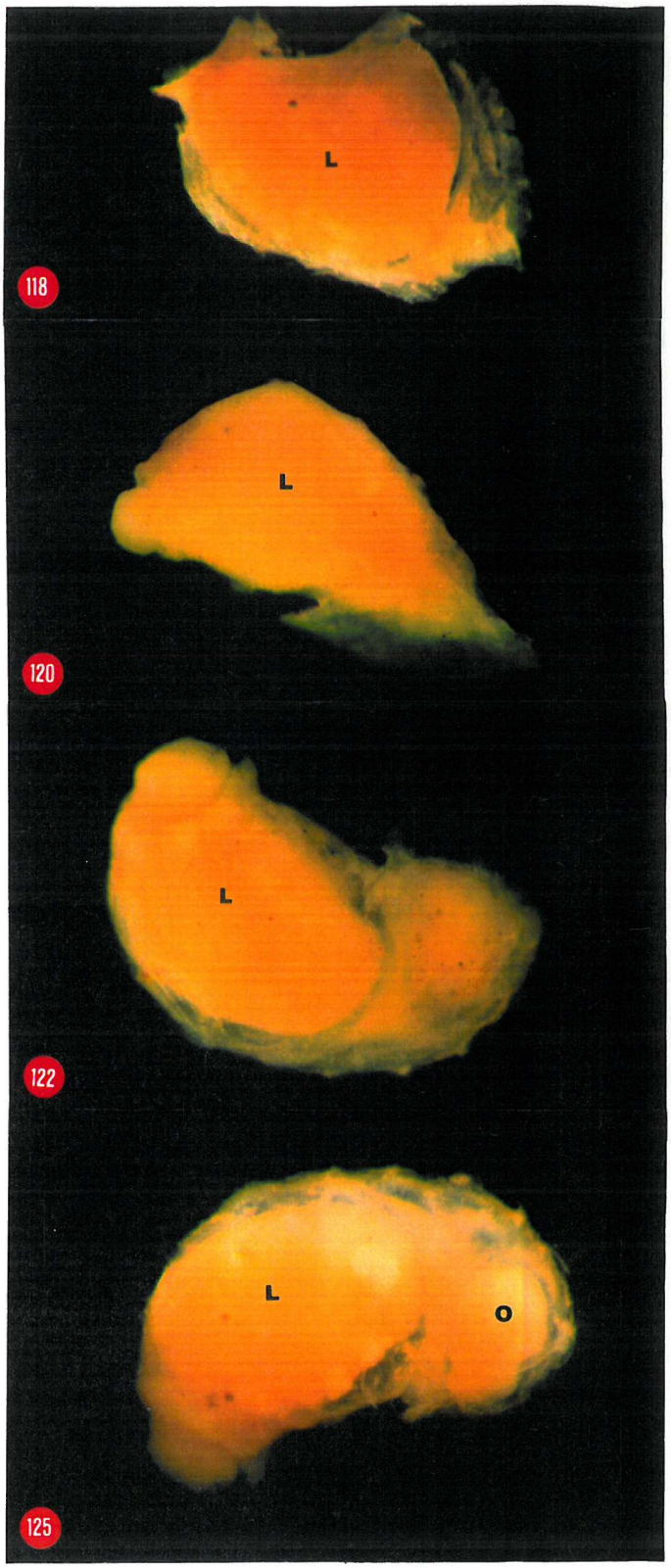

117

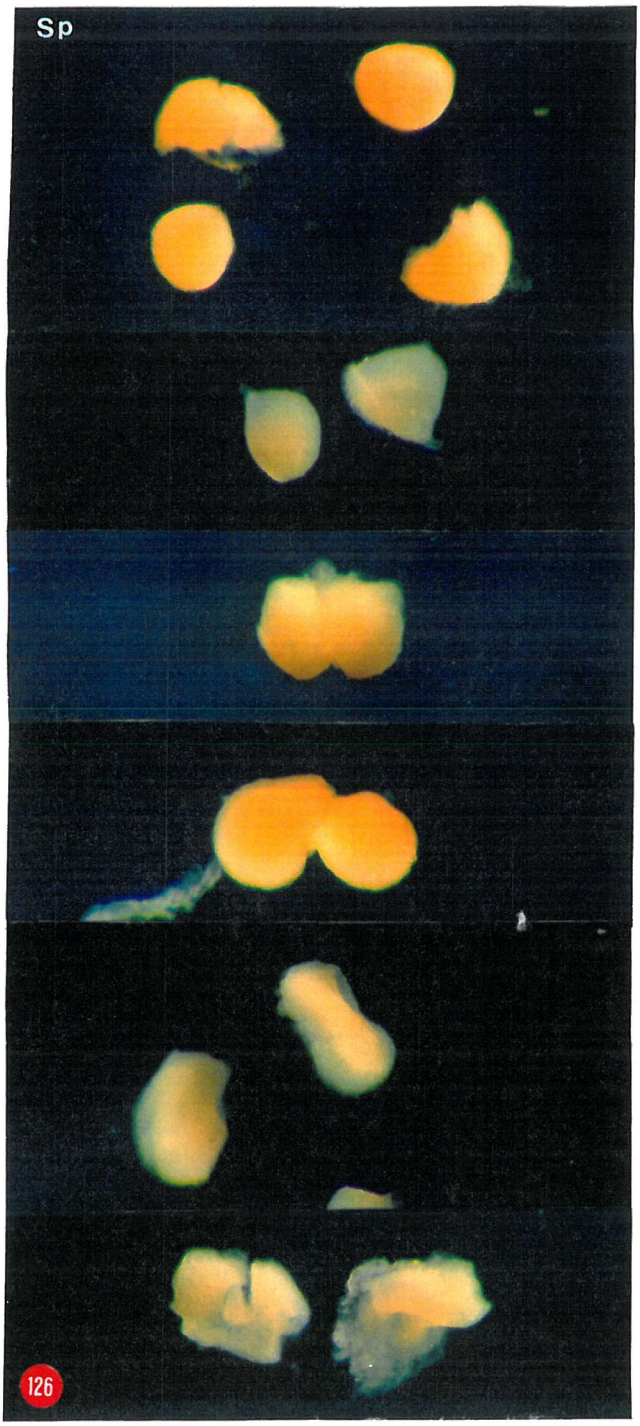

O
119

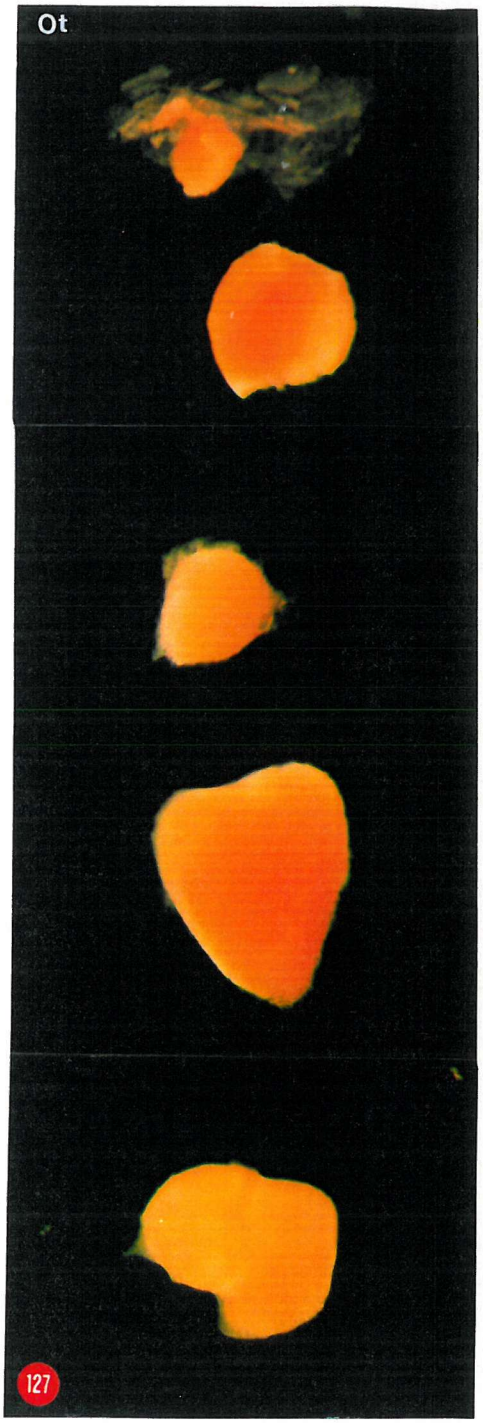

O
121

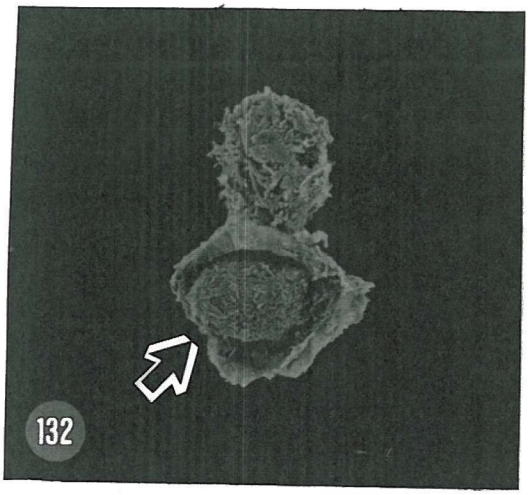

O
123

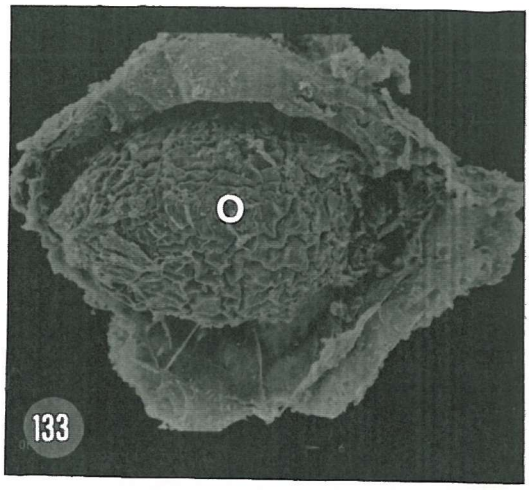

O
124

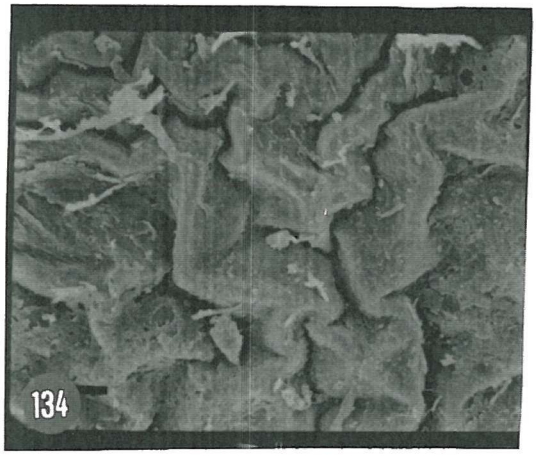

L
118
L
120
L
122
L
O
125

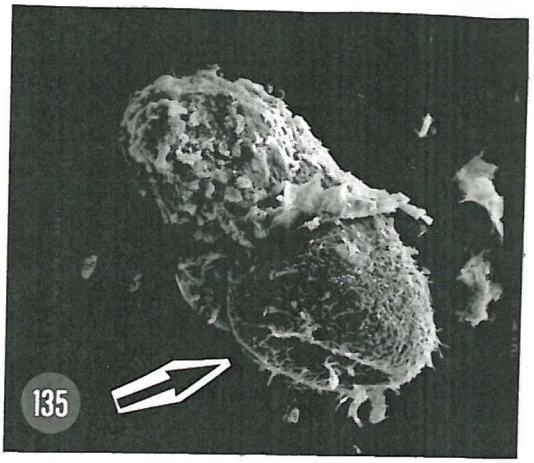

Sp
126

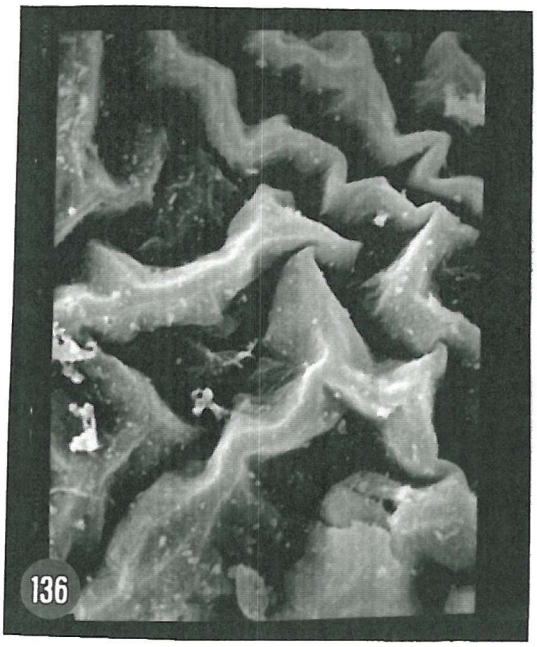

Ot
127

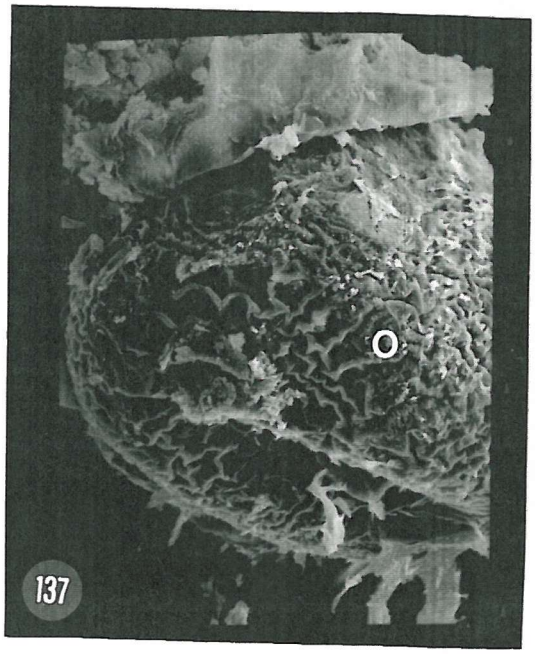

132

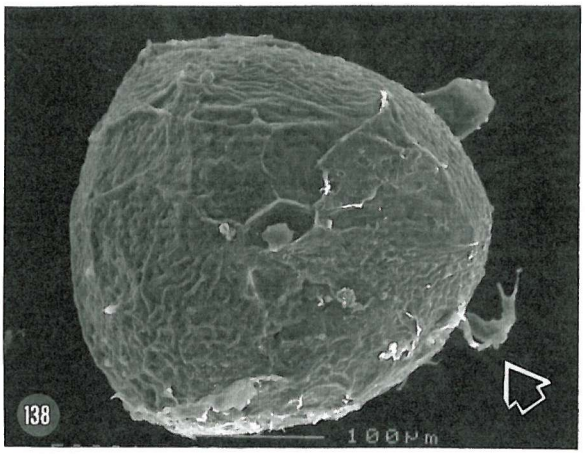

133

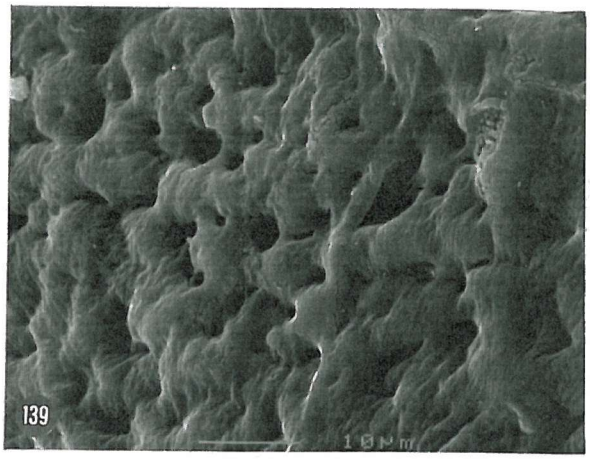

134

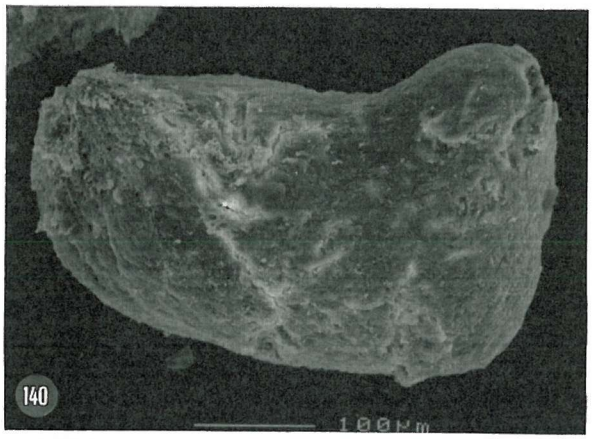

135

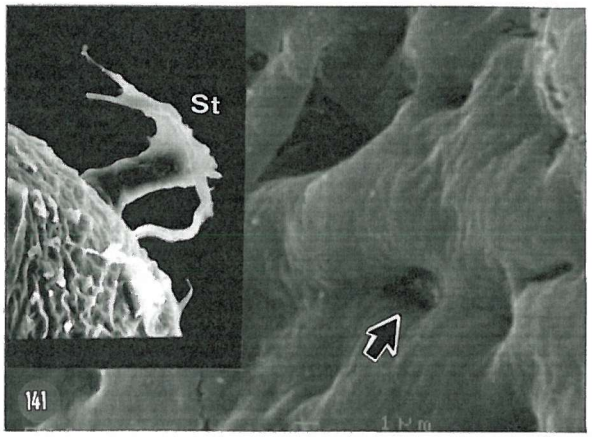

136

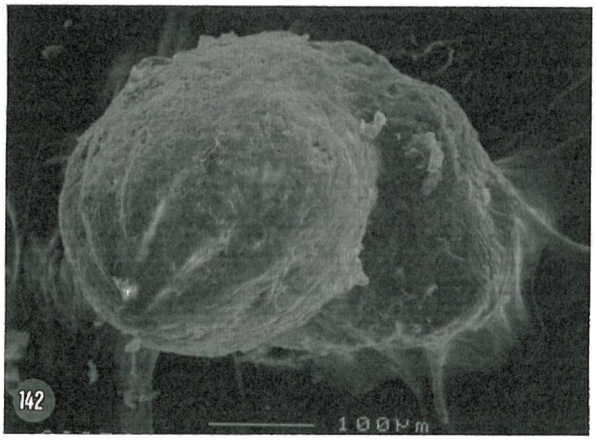

137

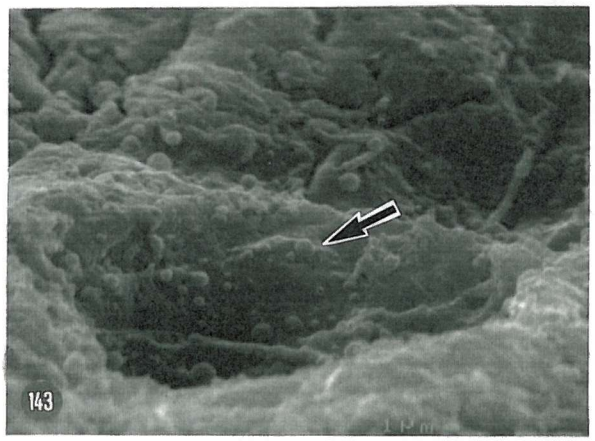

138
100µm

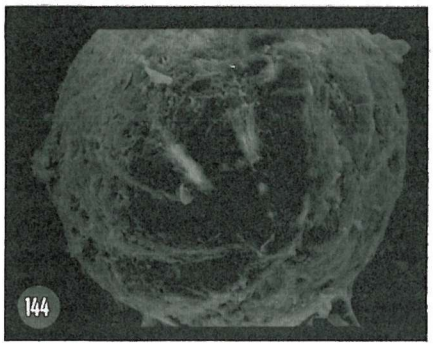

139
10µm

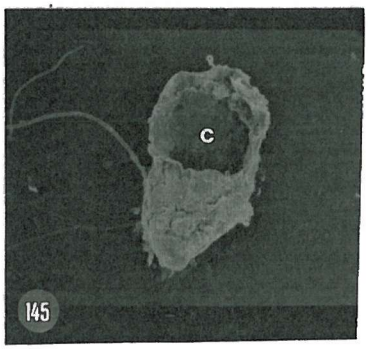

140
100µm

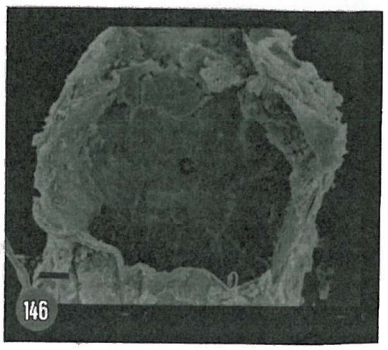

St
141
1µm


142
100µm


143
1µm


144


c
145


c
146

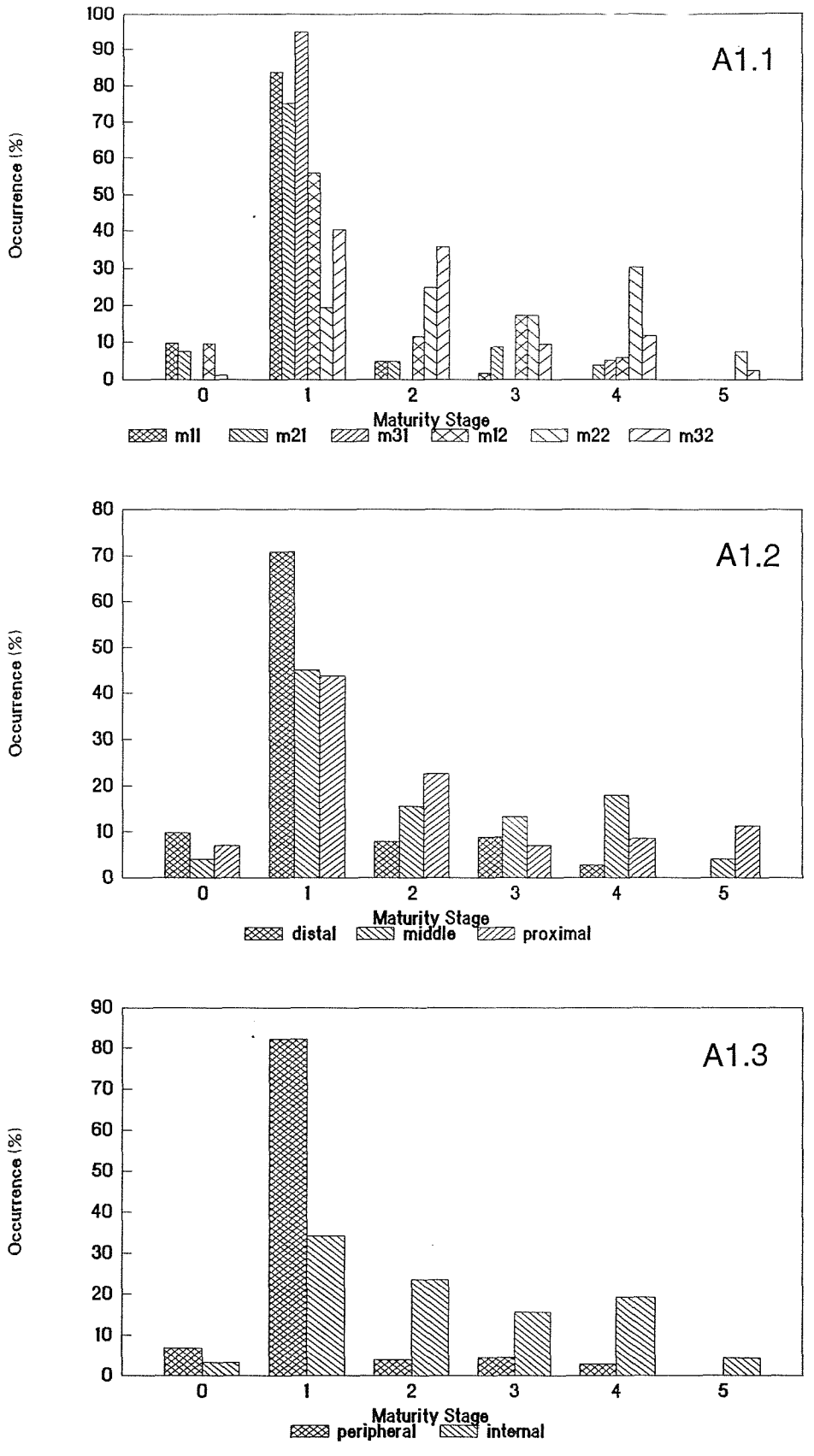


Figure A1 Distribution of maturity throughout male colonies

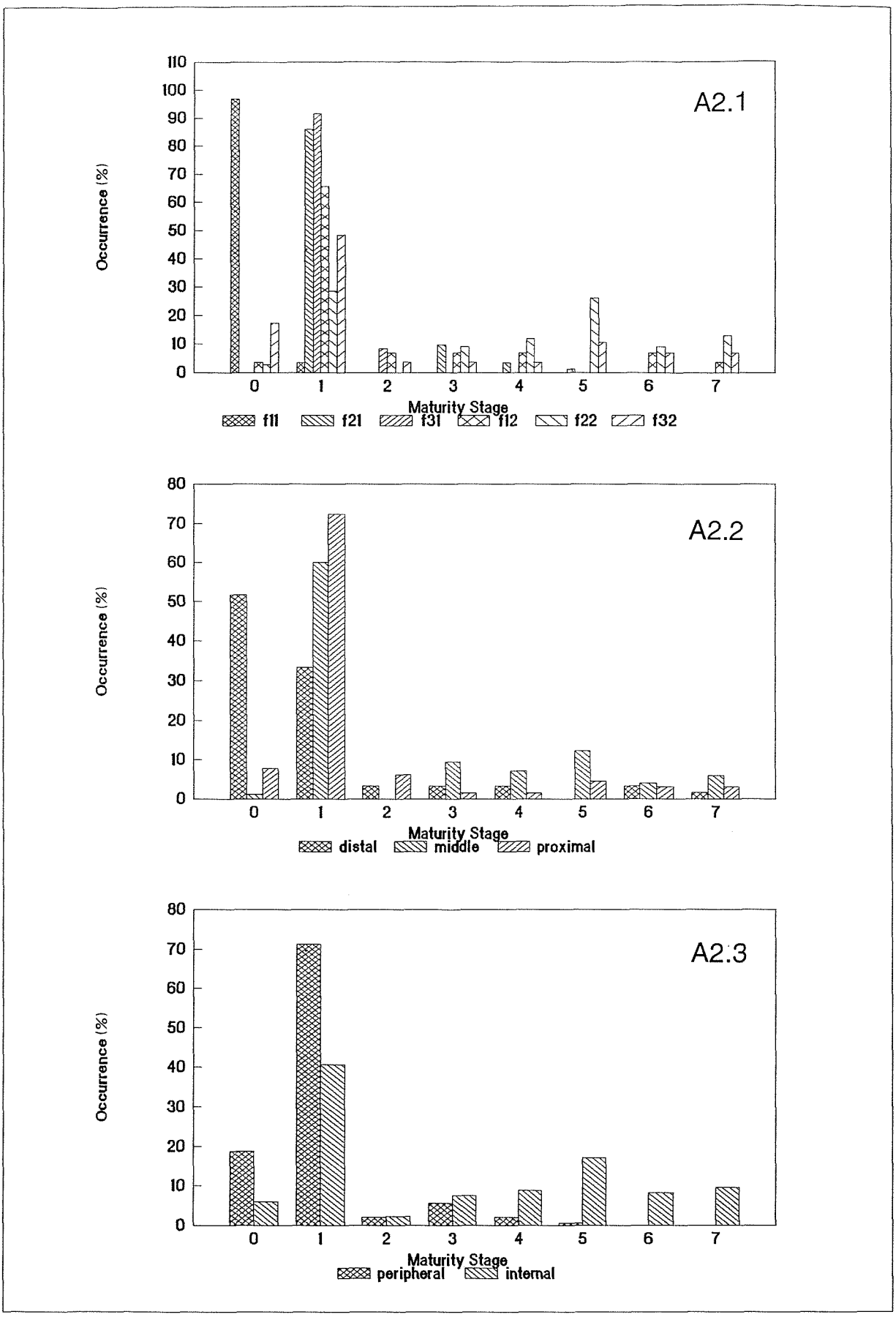


Figure A2 Distribution of maturity throughout female colonies

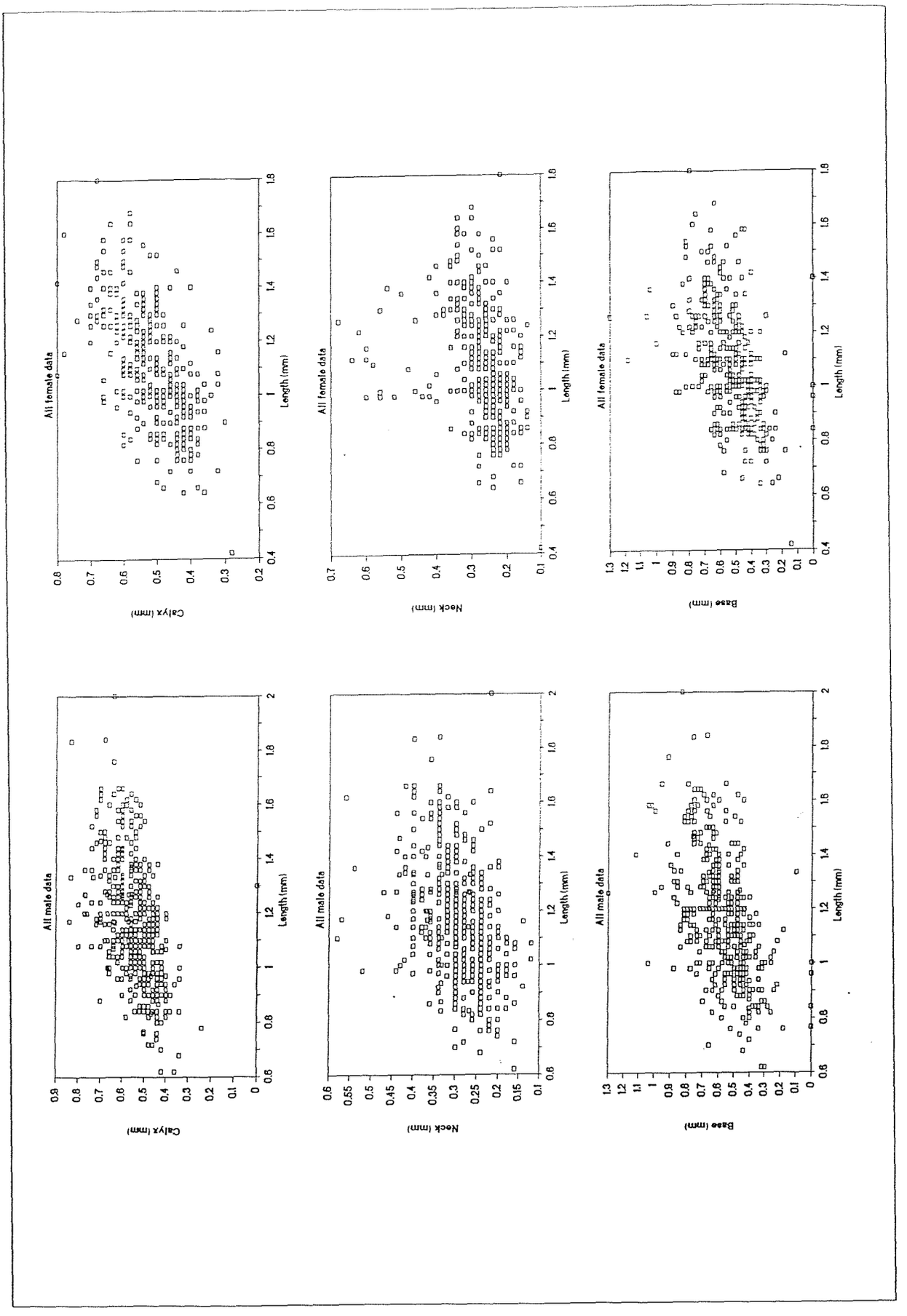


Figure A3 Comparison of polyp external dimensions male and female, all stages of maturity

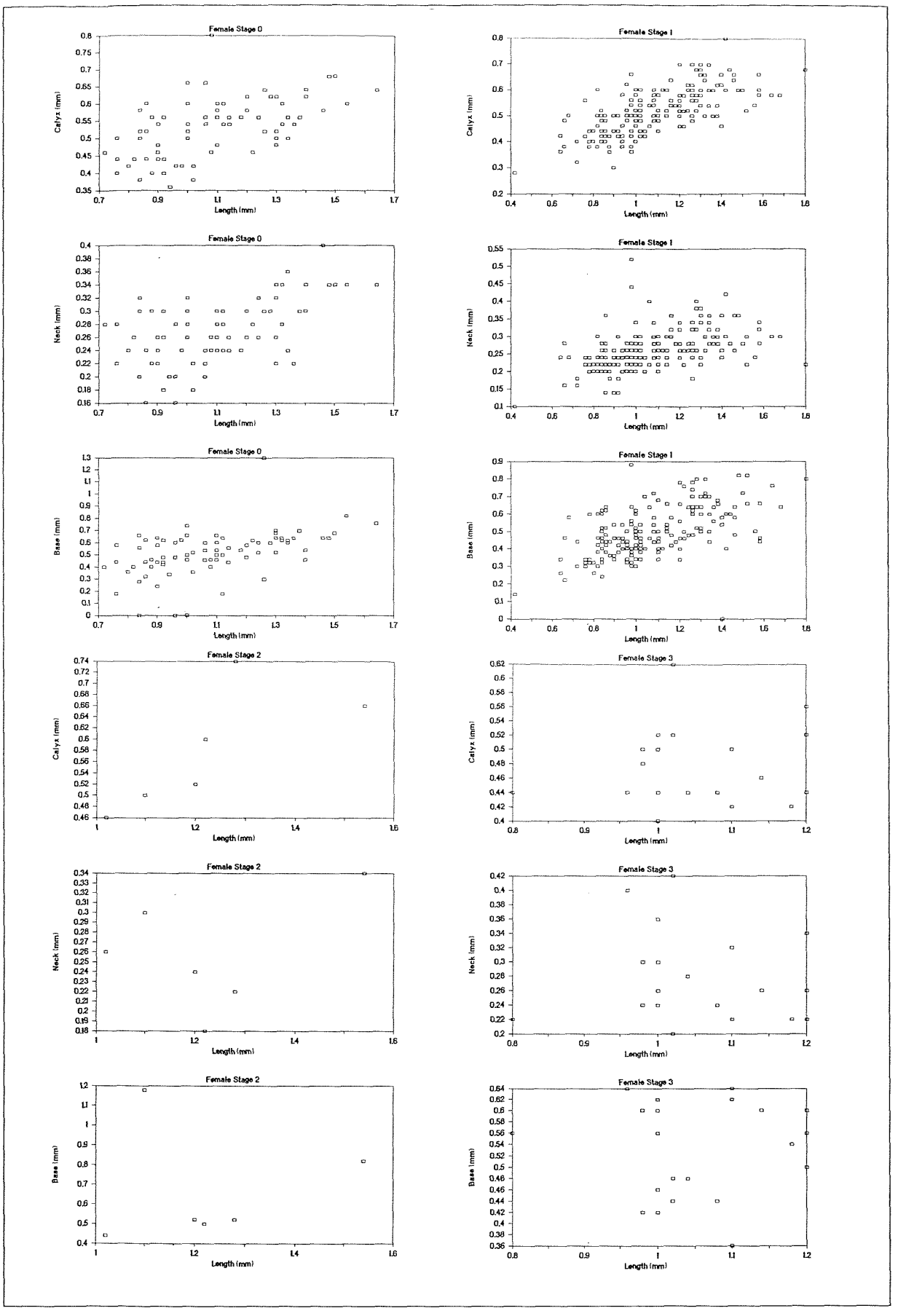


Figure A4 Comparison of polyp external dimensions female, stages 0-3

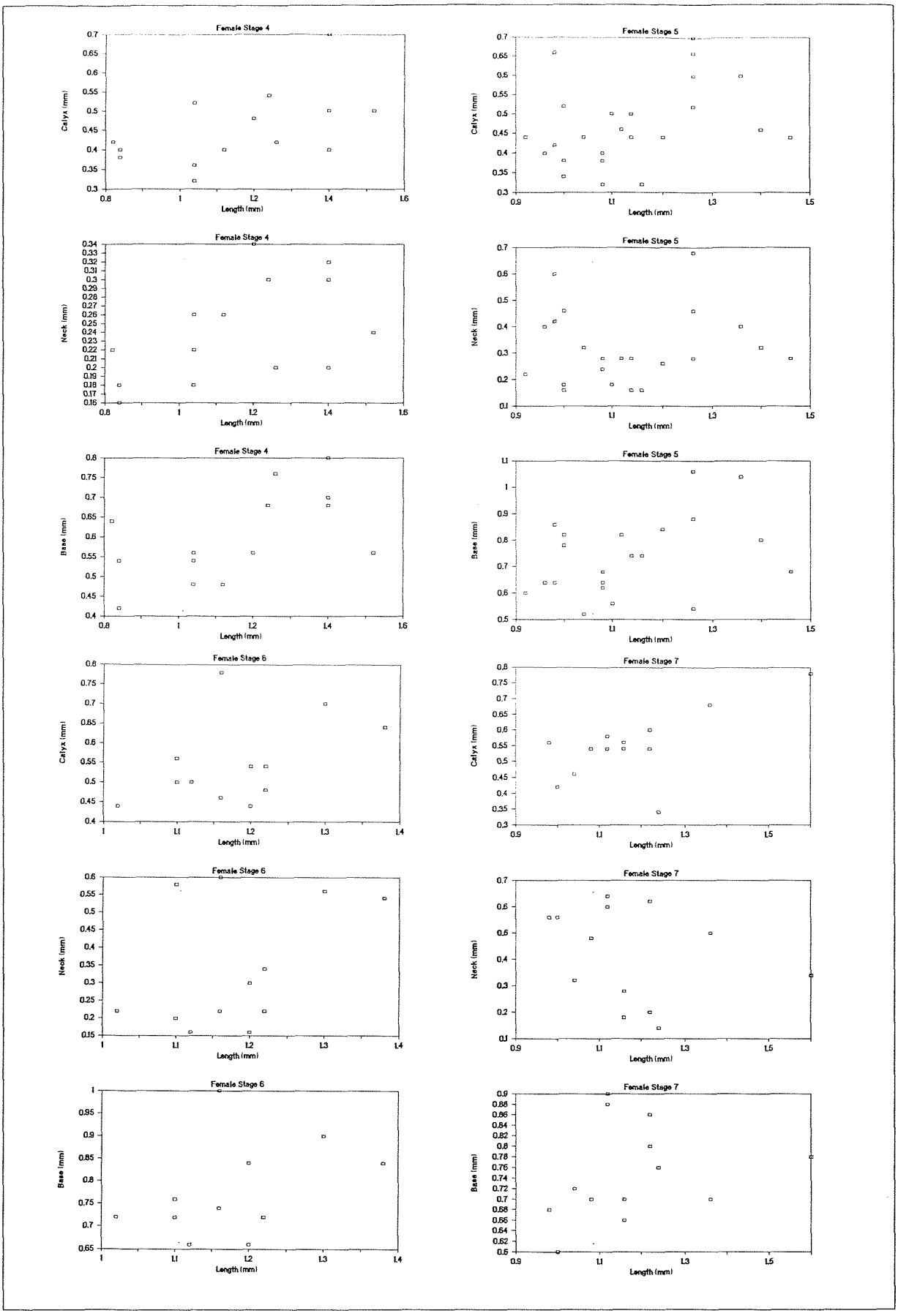


Figure A5 Comparison of polyp external dimensions female, stages 4-7

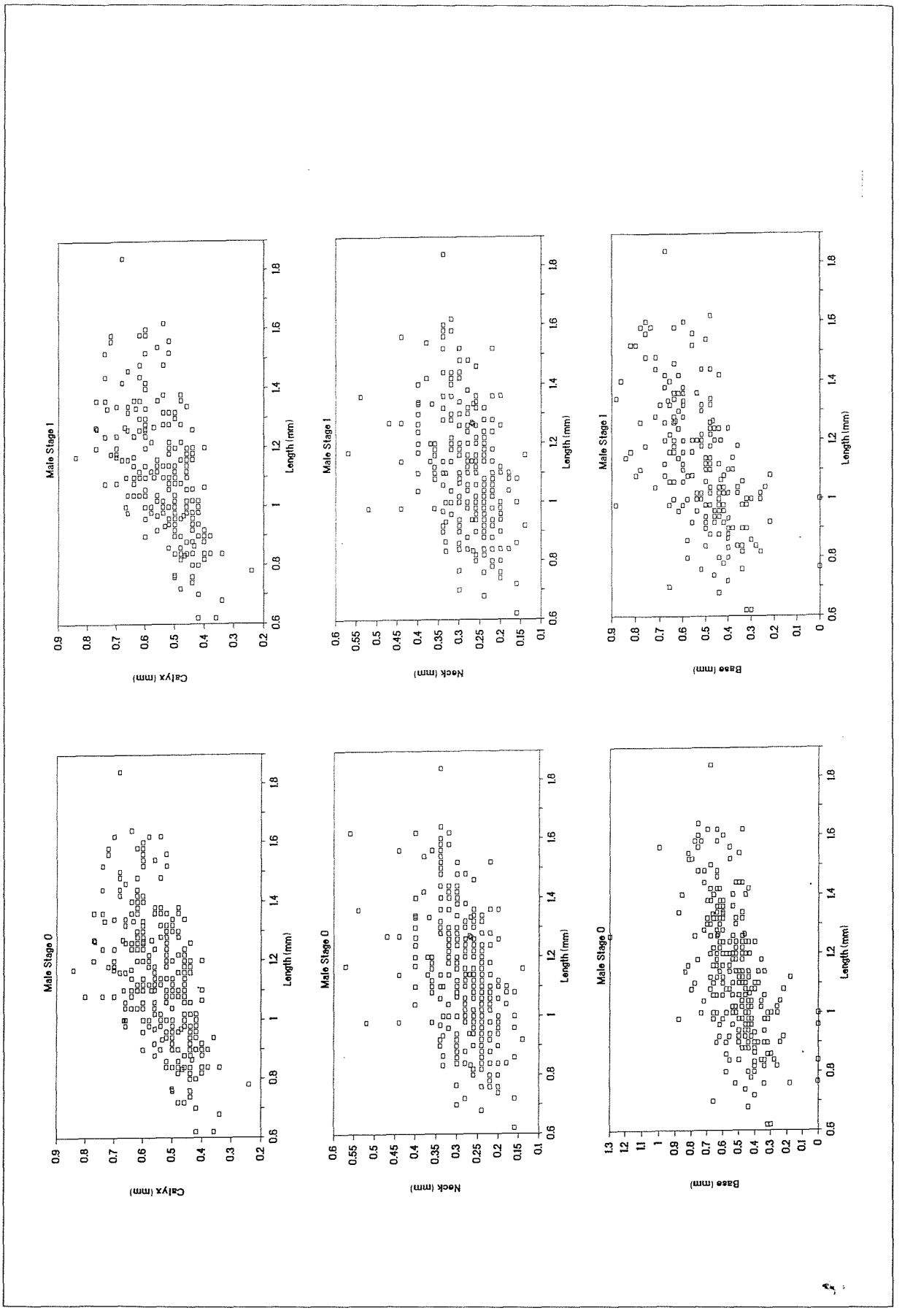


Figure A6 Comparison of polyp external dimensions male, stages 0 and 1

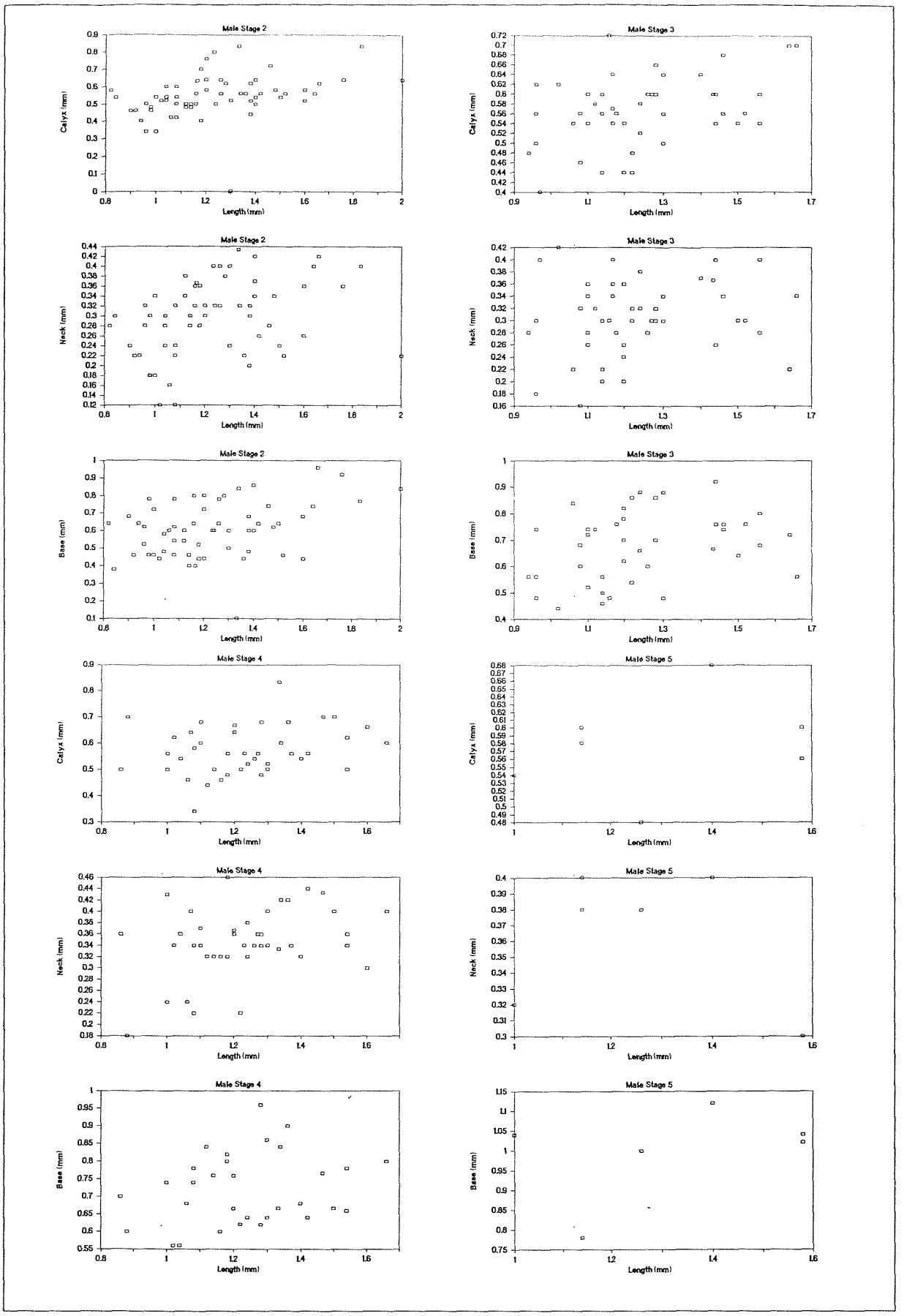


Figure A7 Comparison of polyp external dimensions male, stages 2-5

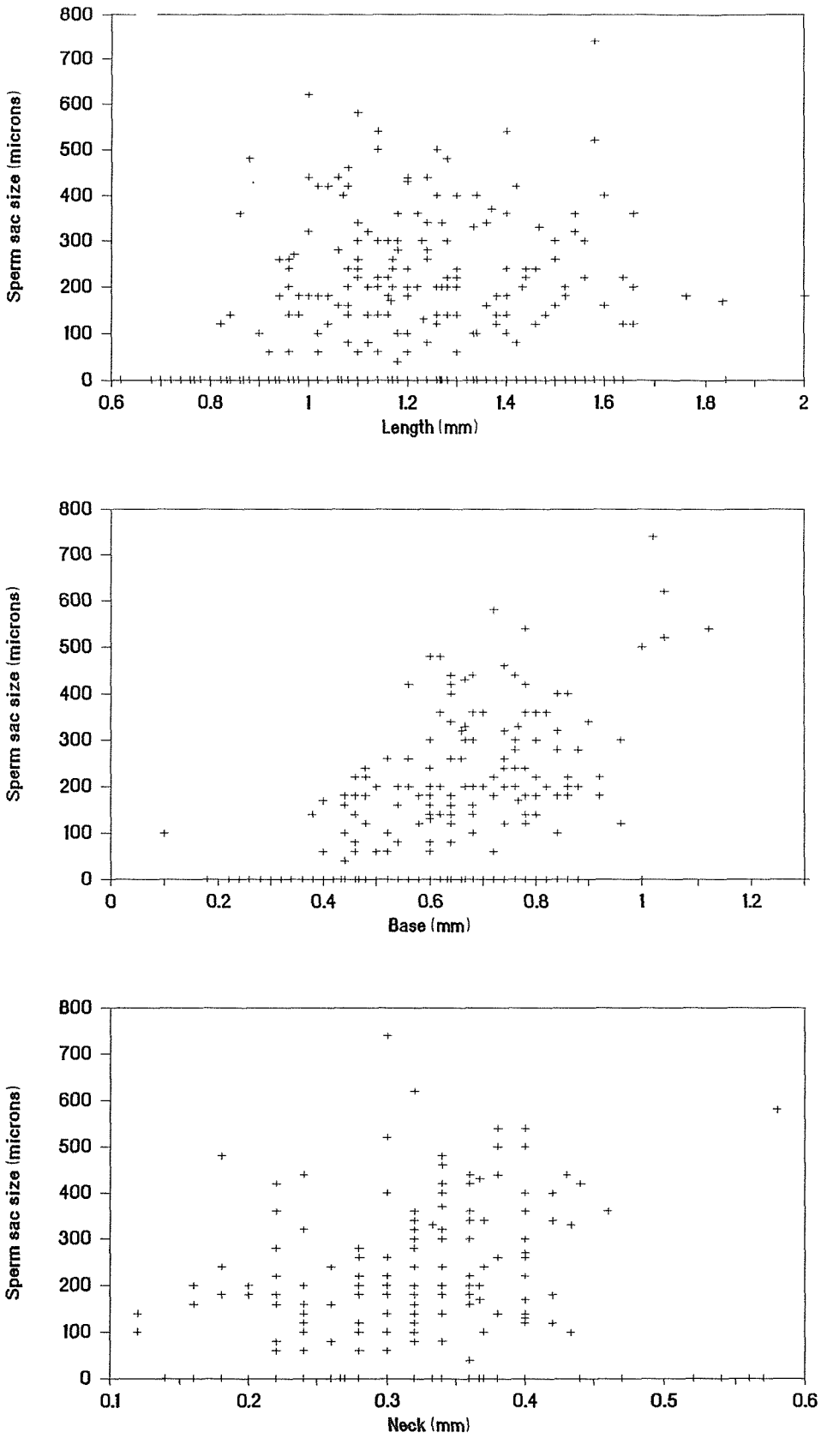


Figure A8 Comparison of sperm sac size and polyp dimensions

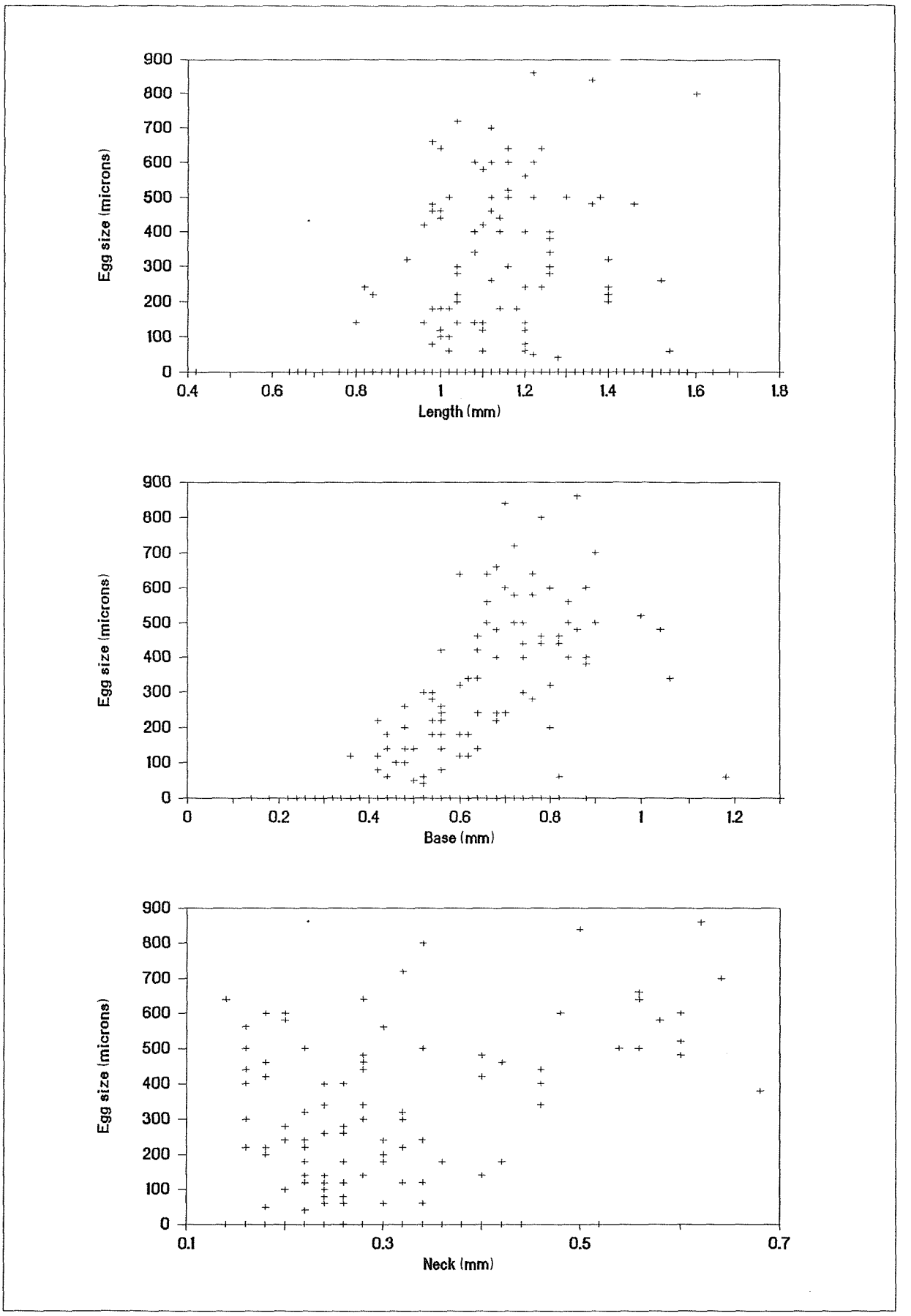
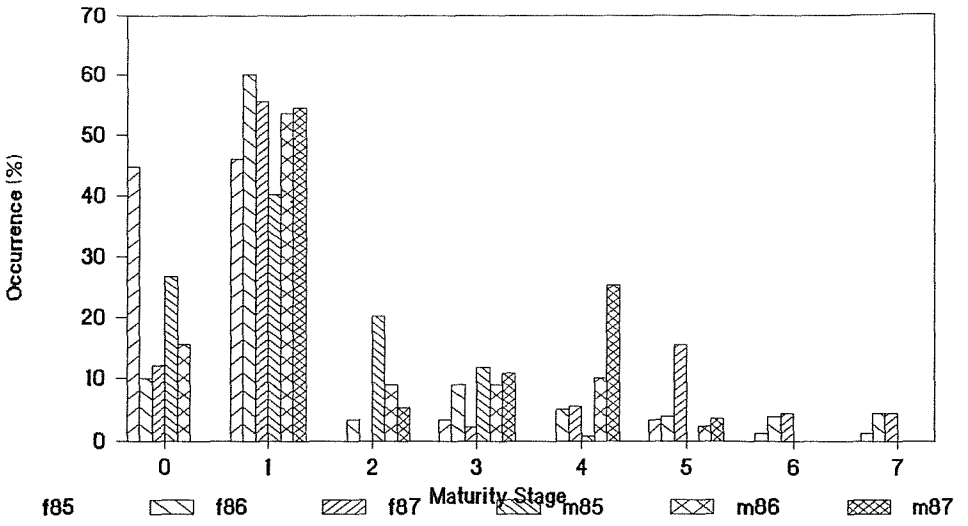
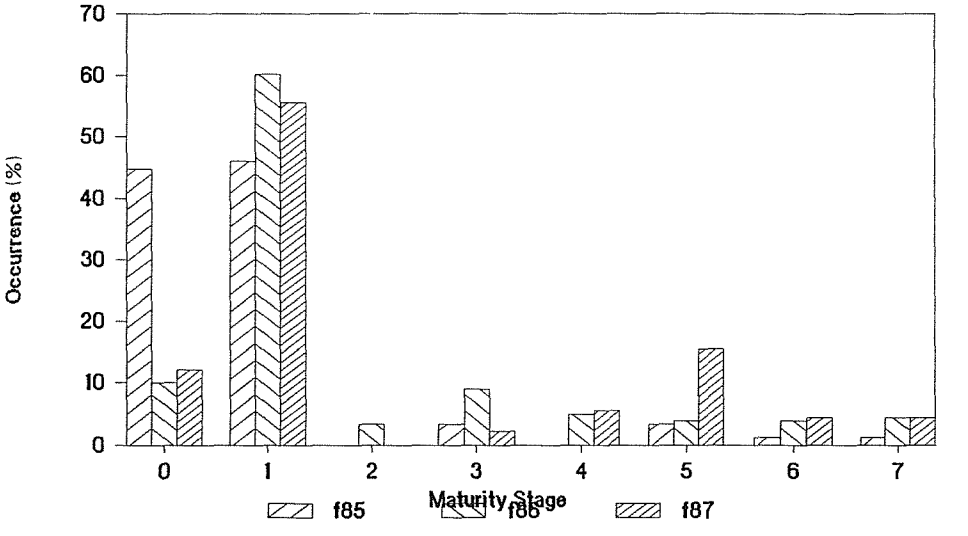


Figure A9 Comparison of egg size and polyp dimensions





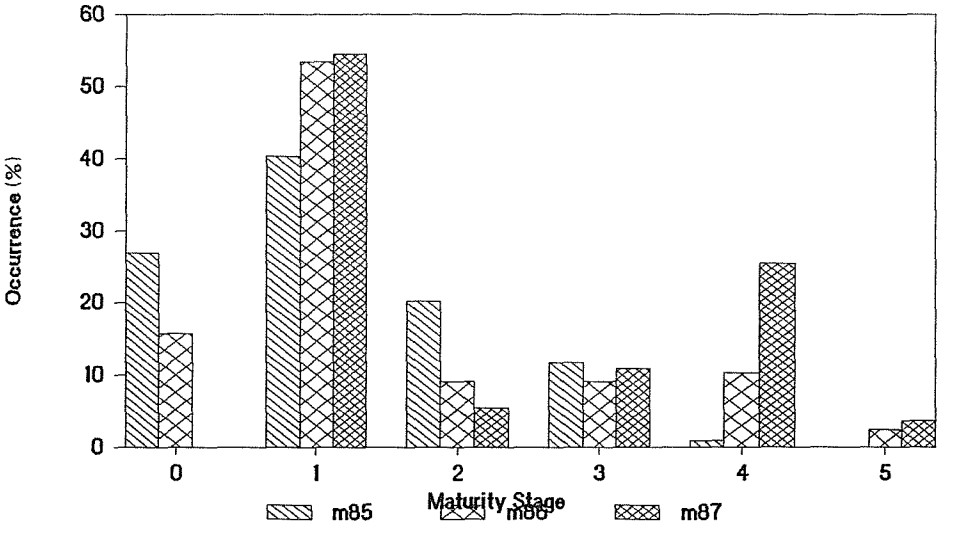


Figure A10 Distribution of maturity in sampling years 1985, 1986 and 1987 (early, mid and late summers)

CHAPTER 5
EVALUATION OF THE TAXONOMIC STATUS OF *Thouarella variabilis*

Among the primnoids collected by the Brazilian Antarctic Expeditions, the genus *Thouarella* is the most abundant and *Thouarella variabilis,* the dominant species. *Th. variabilis* shows a notable range of variation of characters. Some of the characters which have been applied in diagnosing this species are inconsistent and should be used with caution. An evaluation of the validity of these characters as reliable descriptors for this species was carried out. The description based upon the examined specimens was compared to the existing literature, to the type specimens and to the other species referred to the same genus. The description of the species presented in this chapter is a summary of the more detailed account given in Chapter 2.

Thouarella variabilis Wright & Studer, 1889

Thouarella variabilis Wright & Studer, 1889:68, pl. 21, fig. 1. - Menneking, 1905:260-262, l.9, figs 9-10, 21-22. - Verluys, 1906:37. - Gravier, 1913:589-590; 1914:56-61, pl. 1, fig. 6, pl.3, fig. 13-14. - Thomson, 1927:33, pl. 2, fig. 10. - Molander, 1929:74-75.

Thouarella variabilis var. *brevespinosa* Wright & Studer, 1889:69. - Kükenthal, 1924:298. Molander, 1929:74-75.

Thouarella variabilis var. *gracilis* Wright & Studer, 1889:70. - Kükenthal, 1924:298.

Thouarella aff. *variabilis* Kükenthal, 1912:305, pl. 20, figs 2-3.

Thouarella (Parathouarella) variabilis Kükenthal, 1919:428. - Kükenthal, 1924:297-298. - Thomson & Rennet, 1931:27-30, pl. 9, figs 4-5, pl. 12, fig. 3. Broch, 1965:30-31, p l6, figs 17-19.

TYPE SPECIMENS AND TYPE LOCALITY.- *Thouarella variabilis* var.a (referred as *the type*) Wright & Studer, 1889. *Challenger* Expedition, st. 145a, off Prince Edward Island, 310 fathoms. Present location: B.M. 89.5.27.53, N.H.M. London, UK. - *Thouarella variabilis* var. b. *brevispinosa* Wright & Studer, 1889. *Challenger* Expedition, st. 145a, off Prince Edward Island, 310 fathoms. Present location: B.M. 89.5.27.54, N.H.M. London, UK. - *Thouarella variabilis* var. c *gracilis* Wright & Studer, 1889. *Challenger* Expedition, st. 150, off Heard Island. Present location: B.M. 89.5.27.54, N.H.M. London, UK; st. 145, 140 fathoms, 27/12/1873 - B.M. 89.5.27.56, London, UK.

DIAGNOSIS. - Colony shape typically bottlebrush, occasionally biseriate. Branches highly ramified into branchlets. Polyps placed singly on the branches, sometimes in pairs, never in whorls. Polyp scales placed in less than eight longitudinal rows proximally and arranged in five to six diagonal abaxial and lateral rows and two longitudinal adaxial rows; Marginal scales conspicuously thorny; five to six scales on each abaxial longitudinal row.

DESCRIPTION.- Branching, one of two patterns: - bottlebrush colonies with ramified branches arising in opposite pairs, alternating in four directions, occasionally in three directions; - biseriate colonies with branches bent laterally, usually helically twisted, attaining a seemingly bottlebrush appearance. Branches perpendicularly or slightly upwardly directed; branch length varied from 0.65 mm to 2.23 mm long (up to 3.2 mm wide); longest branches positioned in the middle of the colony decreasing in length distally and proximally; average distance between branches is 6 mm, decreasing distally. Branches ramified dichotomously into branchlets; highly ramified branches are not perfectly dichotomous. With continuous growth, the colonies might develop side stems. Main stem horny with smooth surface or slightly grooved longitudinally; colour varied from dark brown to yellow; axis of branches slender, fragile with usually pale yellow colour. Holdfast cone-shaped and calcareous.

Polyps biserially distributed on the branches or arising irregularly in all directions; they alternate with an average distance of 1.8 mm between them, decreasing distally; average polyp number is 10 polyps per 10 mm of branch, reducing to 5-6 in middle branch regions and increasing to 20-30 in terminal branch regions. Polyps club-shaped with narrow neck and broadened calyx, directly outward or slightly upward; average length 1.08 $\pm$ 0.02 mm and maximum length 2.00 mm; average calyx diameter 0.60 mm. Scale-like sclerites thin, fragile and complexly ornamented; at the distalmost part of the polyp, eight triangular scales form an operculum and the eight thorny marginal scales surrounding the operculum are disposed in two alternating rings of four scales. Opercular scales narrow, triangular, concave on the outer surface and terminating in a pointed or blunt distal portion; up to 0.3 mm high. Marginal scales with the enlarged body well ornamented and bearing a long thorn supported in the inner surface by a strong keel; up to 0.83 mm high. Submarginal scales rounded and slightly thorny; up to 0.33 mm high/0.30 mm wide; lower body scales large, quadrangular and curved; 0.72 mm wide/0.44 mm high; adaxial scales small and plate-like; 0.19 mm in diameter. Coenenchyme scales similar to the polyp scales but smaller and less curved. Inner surface of scales ornamented by tubercles and compound warts and dorsal surface by tubercles, spines, crests, ridges or wrinkles, with free edges serrated and proximal portion indented.

MATERIAL EXAMINED.- *Antarctica, Bransfield Strait.* Off *Joinville Island*, 62°30'S, 54°16'W; 412 m; 28/01/1985; beam-trawl; PROANTAR III, st. 4743; four specimens. - Joinville Island, 62°53'S, 56°27'W; 194 m; 03/02/1986; beam-trawl; PROANTAR IV, st. 4866, three specimens. *South Shetland Islands. Livingston Island Group* - Snow Island; 80 m; 08/02/1985; PROANTAR III, one specimen.- 62°26.5'S, 59°16.0'W; 212 m; 24/02/1987; beam-trawl; PROANTAR V, st. 5052, two specimens.- Livingston Island, 62°40.0'S, 59°33.0'W; 270 m; 08/03/1987; otter-trawl; PROANTAR V, one specimen. *Elephant Island Group* - 61°08'S, 54°34'W; 240 m; 01/02/1986; beam-trawl; PROANTAR IV, st. 4862, four specimens.- 61°17'S, 54°53'W; 180 m; 02/02/1986; beam-trawl; PROANTAR IV, st. 4863, one specimen.- Off Smith and Low Islands, 63°25'S, 62°05'W; 66 m; 13/02/1986; beam-trawl; PROANTAR IV, st. 4873, one specimen.- 63°25'S, 62°19'W; 135 m; 14/02/1986; otter-trawl; PROANTAR IV, st. 4874, seven specimens.- 63°17'S, 62°30'W; 157 m; 14/02/1986; beam-trawl; PROANTAR IV, st. 4875, one specimen.- 62°39'S, 58°51'W; 170 m; 23/02/1987; otter-trawl; PROANTAR V, st. 5051, two specimens.- 63°16'S, 59°55'W; 264 m; 08/02/1986; beam-trawl; PROANTAR IV, st. 4871, one specimen. *King George Island group* - Admiralty Bay, Martel Inlet, 62°04.9'S, 58°22.1'W; 53 m; 13/02/1987; van veen; clay; PROANTAR V, st 5026, one specimen.

MATERIAL EXAMINED FOR COMPARISON. - *Thouarella variabilis* var.a (referred as *the type*; *spinosa* ? - probably mistakenly named), *Challenger* Expedition, st. 145a, off Prince Edward Island, 310 fathoms: SYNTYPES. B.M. 89.5.27.53, N.H.M. London, UK. - *Thouarella variabilis* var. b. *brevispinosa*, *Challenger* Expedition, st. 145a, off Prince Edward Island, Antarctica, 310 ftms, 27/12/1873: HOLOTYPE. B.M. 89.5.27.54, N.H.M., London. - *Th. variabilis* var. c. *gracilis*, *Challenger* Expedition, st. 150, off Heard Island, Antarctica, 150 ftms, 02/02/1874: B.M. 89.5.27.55, N.H.M. London; *Challenger* Expedition, st. 145, 140 ftms, 27/12/1873: SYNTYPES. B.M. 89.5.27.56, N.H.M. London.

EXPEDITIONS WHERE THIS SPECIES WAS COLLECTED AND WHOSE RESULTS WERE PUBLISHED - *Challenger* Expedition (1872-76); Campagnes Scientifiques du Prince Albert Ier de Monaco (1886-1888), *Hirondelle*; Expedition *Belgica* (1897-99); Deutschen Tiefsee-Expedition (1898-99), *Valdivia*; Siboga Expedition (1901-03); Deutche Südpolar-Expedition (1901-03); Swedish Antarctic Expedition (1901-03); Seconde Expédition Antarctique Francaise (1908-1910), *Pourquoi pas ?*; Australasian Antarctic Expedition (1911-14); Brategg-Expedition (1947-48), *Norvegia*.

KIND OF SUBSTRATA RECORDED. - The larvae of *Thouarella variabilis* appear to settle on most available solid substrata. Amongst the examined specimens, the colonies, which were intact, were observed mainly adhered to small stones or shells. In literature, colonies

of *Th. variabilis* have been recorded on substrata which were a mixture of sand and gravel (Versluys, 1906; Molander, 1929); rock and pebble, sand and pebble (Gravier, 1914); gravel and cobble; mixed gravel and clay (Molander, 1929).

Table 5.1 - Geographic and bathymetric distribution of *Th. variabilis*

SITE	POSITION	DEPTH (m)	AUTHOR
Prinz Edward Island, South Indic	-	567 270/558	Wright & Studer, 1889 Versluys, 1906
Heard Island, South Indic	-	270/558 570/575	Versluys, 1906 Kükenthal, 1912/1924
Gauss Station	-	350-385	Kükenthal, 1912
Baie Marguerite, between l'Ile Jenny and Terre Adélaide	67°45'S, 68°33W	254	Gravier, 1913/14
Baie Marguerite	67°45'S, 68°33'W	176	Gravier, 1913/14
Port-Lockroy, Chenal de Roosen	64°49'S, 63°30'W	50	Gravier, 1913/14
Margin of an ice bank	70°10'S, 78°30'W	460	Gravier, 1913/14
Graham Land Region, Paulet Island	63°36'S, 55°48'W	100-150	Molander, 1929
Graham Region, Seymour Island	64°20'S, 56°38'W	150	Molander, 1929
Graham Region	65°19'S, 56°48'W	400	Molander, 1929
Between Falkland Island and South Georgia, off Shag Rock-Bank	53°34'S, 43°23'W	160	Molander, 1929
-	71°26'S, 12°00'W	220	Broch, 1965
Commonwealth Bay, Adelie Land	-	82-92 100-109	Thomson & Rennet, 1931
Off the Crozets	-	1006	Thomson & Rennet, 1931
Australasian Antarctic Expedition, st.1	-	648	Thomson & Rennet, 1931
st.2	-	582	
st.7	-	110	
st.8	-	220	
st.9	-	439	
st.10	-	595	
Hirondelle, French-Expedition st. 1223	-	1642	Thomson, 1927
st. 1349 -	-	1250	Thomson, 1927

GEOGRAPHIC AND BATHYMETRIC DISTRIBUTION. - The considerable distance between the sites listed in Table 5.1. suggests that *Thouarella variabilis* has a wide geographic distribution around Antarctica and Subantarctica Regions. As early as 1914, Gravier proposed that this species might be circumpolar.

REMARKS. - The original description of *Thouarella variabilis* by Wright & Studer (1889) is not sufficiently clear concerning the consistency of the characters used by these authors to characterize the species. Moreover, the figure of the colony referred in the text does not belong to *Th. variabilis* but mistakenly to another species and the lack of illustration of the whole colony makes it even more difficult to understand the description.

Wright & Studer had difficulties in separating the individual forms specifically owing to the high degree of variation of characters. To accommodate the varied forms, Wright & Studer proposed three varieties - *Th. variabilis,* the typical form; *Th. variabilis* var. *brevispinosa*; and *Th. variabilis* var. *gracilis* - on the basis of polyp length, development of thorns and colony form.

The typical form of *Th. variabilis* differs from the other varieties by having 2 mm long polyps arranged in spirals of three, considerably long marginal thorns and branches ramified up to two to three orders. The branch divides dichotomously into branchlets still very close to the base and the last order branchlets are markedly long, giving the branch a peculiar appearance.

In the variety *Th. v. brevispinosa* the branches are sparsely distributed and poorly ramified, being mostly simple or bifid. The polyps are large (2.5 to 3.0 mm long), distant apart and not arranged in spirals. The marginal scales have short thorns when compared to the typical form of *Th. variabilis.* This variety is very similar to *Th. striata* Kükenthal (1907), differing from the latter in the size of the polyps (1.5 mm long in *Th. striata*) and on the ornamentation of the scales. *Th. striata* is highly striate.

The variety *Th. v. gracilis* has a more finely ramified form, with a flexible and thin stem and branches highly ramified into branchlets . Considering the measurements of colony length and stem thickness presented by the authors for the variety *Th. v. gracilis* and for the typical form, it is clear that the specimen referred to the *Th. v. gracilis* is much younger than the typical form of *Th. variabilis.* The polyps of *Th. v. gracilis* are placed on the branches in spirals of threes and are 2.0 mm long. The thorns of the marginal scales are longer than in the variety *Th. v. brevispinosa* but shorter than in the typical form of *Th. variabilis.*

The most consistent characters used to diagnose this species appear to be the branching pattern and the arrangement and form of sclerites. The latter is inconsistent only in the length of the thorn of the marginal scales which differs in the three varieties. The length of polyps and the branching pattern appear to be the most variable characters between the three varieties. The distribution of polyps is the same in the varieties *Th. variabilis* typica and *Th. v. gracilis*, but differs in *Th. v. brevispinosa*. Concerning the size of polyps and the branching pattern, the typical form shows intermediate characters between *Th. v. gracilis* and *Th. v. brevispinosa*. There seems to be, therefore, an intergradation between the three varieties and perhaps the validity of these varieties should be reconsidered.

The specimens examined for the present work agree in the main with the original description of the *Th. variabilis* var. *gracilis*, and partly to the typical form. With the latter, there is a discrepancy in the size of polyps and in the branching pattern. The subsequent descriptions of specimens referred to *Th. variabilis* by other authors appear to be mostly with reference to *Th. v.* var. *gracilis* but are named simply as *Th. variabilis*.

Menneking's description (1905) appears to agree with the original description of *Th. variabilis* in most aspects. Apart from longitudinal rows of body scales, Menneking also described five disorderly diagonal rows as an alternative. He provided a detailed description of the ornamentation of the scales, which agrees with the ornamentation shown by the specimens studied for this work.

Versluys (1906) commented on the deficiency of illustration in the description of Wright & Studer and on the difficulty in dealing with the great variation of characters. He added to the description of the species the information that the polyps on the proximal portion of the branches were disposed less closely spaced than on the other portions of the branches. Versluys also introduced the knowledge about the presence of a short thorn on the submarginal scales and he found it impossible to define longitudinal rows of body scales as described by Wright & Studer. About the variety *Th. variabilis brevispinosa*, he mentioned that the number of sclerites is higher and their disposition in longitudinal rows better visualized than in the typical form.

Kükenthal published several major papers where he describes this species (1907, 1912, 1919 and 1924). In his descriptions he mentioned the variation of colony form from poorly ramified to highly ramified branches. The branching pattern described for his specimens was similar to the original description, although some specimens showed a biseriate arrangement, with the branches being longer and more flexible in this case. Wright & Studer had observed that

some specimens had their stem twisted around their axis, but did not recognized a biseriate arrangement of a colony which grew helically. The polyps on Kükenthal's specimens ranged from 1.5 to 2.0 mm long, were singly placed and more aggregated on the periphery of the branches. He defined longitudinal rows of scales but did not mention how many. His descriptions and drawings of sclerites agree in all respects to the studied specimens in the present work. He also recognized the degree of variation on the length of the thorns of the marginal scales, which ranged from short to markedly long.

Kükenthal characterized briefly the varieties introduced by Wright & Studer for *Th. variabilis*. For the variety *Th. v. brevispinosa,* he considered as attributable characters the polyp size, the number of polyps and the length of the thorn of the marginal scales. For *Th. variabilis gracilis*, he considered the fragile aspect of the colony and the length of the thorns of the marginal scales. However, he regarded the latter character as inconsistent owing to its natural variation. Kükenthal concluded that on the basis of the architecture of the polyps, especially spiculation, he could probably refer his specimens to *Th. variabilis*, and he would do so without hesitation if it was not for the lack of a better description and of illustration of the distribution and architecture of the polyps. He did not, however, accommodate his specimens in varieties.

Gravier (1913, 1914) was the first author to associate the variation in the distribution of polyps, sometimes aggregated at the end branches, sometimes sparsely distributed, to environmental conditions. The description of the studied specimens are identical to Gravier's description concerning branching pattern, distribution of polyps and spiculation. Gravier attributed, with reticence, his specimens to *Th. variabilis* because of the insufficient characterization of the species given by Wright & Studer. He considered that the sclerites shown in Wright & Studer's drawings differed considerably in form from the sclerites of his specimens although similar in ornamentation. The sclerites shown in the drawings of the original description are similar to the sclerites of the class size three of the studied specimens, therefore, they were probably undeveloped. Gravier's description agreed more with the descriptions of Kükenthal than with the description of Wright & Studer.

Gravier mentioned the three varieties erected by Wright & Studer and attributed some of his specimens to the typical form only on the ground of colony form. Two of the specimens referred by Gravier to *Th. variabilis* differed markedly in colony form, one colony having shorter and more rigid branches, which he referred to the typical form, and the other longer with more flexible branches. On the grounds of colony form, Gravier felt tempted towards

separating the two forms specifically. However, the specimens were very similar in polyp architecture and spiculation which Gravier considered a more consistent character and preferred to maintain the two forms in the same species.

Thomson (1927) assigned two specimens to *Th. variabilis* on the basis of the distribution of polyps (which occurred singly, never in pairs or in whorls), size of polyp (1 mm long) and spiculation. Like the studied specimens, his description contradicts the original description concerning the size of the polyps.

Molander (1929) assigned his specimens (fragments) to the variety *Th. variabilis brevispinosa*, with which they agreed in spiculation but differed in size (1.0-1.2 mm long). Molander gave much importance to the presence of radial ridges on the surface of the scales, suggesting that this ornamentation was more evident in the *Th. variabilis brevispinosa* than in the *Th. variabilis typica.* He also proposed that the variety *Th. variabilis brevispinosa* was closely related to another species described by Kükenthal (1907), *Th. stricta*, although he probably meant *Th. striata*, because a species named *Th. stricta* had not been established.

Thomson & Rennet (1931) re-emphasized and summarized the variation of characters displayed by the specimens referred to *Thouarella variabilis.* They listed the variable characters as follows: branching pattern (bottlebrush, partly biseriate, bearing secondary stems); density of polyps (aggregate to far apart) and length of the thorns of the marginal scales. Based on these characters, Thomson & Rennet tried to group the specimens into two sets: - colonies with aggregated polyps, with looser and more irregular branching pattern and simple branches; and - colonies with polyps distributed more sparsely, regular bottlebrush appearance, and ramified branches. However, they did not neglect the fact that: - sparse and distant polyps were often smaller than crowded polyps; - the length of the thorns of the marginal scales varied within the same colony; and that - young colonies were observed bearing simple branches and polyps more distant apart. Thomson & Rennet preferred, therefore, to consider colonial form, density of polyps and length of marginal thorns as unimportant specific characters. As constant features they considered: - the presence of a keeled spine in the marginal scales; - the presence of only four scales in an abaxial row, including the marginal; - the narrow and triangular opercular scales; the absence of radial striations on the cortical sclerites; and the single disposition of the polyps, which are never in whorls, though may be densely crowded.

The studied specimens are most similar in every respect to the description by Broch (1965). This author provided a more detailed description of the sclerites. He also emphasized the importance of the brittle and rigid aspect of the basal portion of the main stem, considering it as a characteristic specific character in *Thouarella variabilis*. Broch pointed out the similarity between *Th. variabilis* and *Th. striata* (Kükenthal, 1912).

On examining the full descriptions of the aforementioned authors, the type specimens, and the studied specimens, it was possible to verify that the most constant characters applied to describe *Th. variabilis* are: the polyp shape, the arrangement of opercular and marginal sclerites, the number of sclerites per longitudinal abaxial row, and the general aspects of spiculation (apart from the length of the marginal thorns).

The most varied characters are: the colonial form, the branching pattern of the branches, the number of polyps, the length of the marginal thorns and the disposition of the abaxial rows of scales. The potential explanation for the variation occurring in each of these characters has already been discussed in Chapter 2. The appearance of the colony and the branching pattern of the branches could be a result of the intensity and direction of hydrodynamic forces, of the developmental stage of the colony and also of the presence of commensals. The distribution and number of polyps could be determined by the direction of currents or availability of food. The length of the thorns of the marginal scales was observed to vary in the different size class of polyps within the same colony as demonstrated in Chapter 2. The definition of longitudinal, transverse or diagonal rows of abaxial scales appears to be a question of approach of different authors about exactly the same thing. The abaxial scales can be disposed in 3 to 4 abaxial-lateral longitudinal rows, 5 to 6 diagonal rows or 5 transverse rows. When the polyp is bearing eggs and is consequently expanded, more longitudinal rows can be recognized. None of the authors above mentioned the reproductive stage of the polyp, therefore, it is difficult to judge the variation of this character in the descriptions. For these characters to have some taxonomic value, a range of variation for each of them has to be delineated.

Another character which should be analyzed with caution is the size of the polyps. The polyps present on the studied specimens never went beyond 2 mm in length, with the average length being about 1 mm. A polyp length of 3 mm is a considerable difference since the whole aspect of the colony changes. It must be emphasized that the studied colonies are of small size (maximum: 13 cm). Whether the size of the polyps carries on increasing with the increase of the colony size or within different environments, is not known. However, larger colonies have not always been seen having larger polyps than younger colonies.

To assume that all the variations of characters are caused by environmental influence alone could be misleading because virtually nothing is known about the environmental conditions where the specimens inhabited. It must be mentioned that some specimens of varied form come from the same station, probably ruling out the difference in environmental conditions, unless the distance of dredging and local variations are considered.

Despite the speculation about the causes of variation of characters in *Th. variabilis*, it is possible to recognize that, as suggested by Thomson and Rennet, there are numerous varied forms round the typical form of *Thouarella variabilis* and that if varieties have to be introduced to accommodate these different forms, several varieties should be proposed and not only three as originally proposed by Wright & Studer. In view of that, the introduction of varieties is of little justification and therefore the authenticity of the three varieties proposed by Wright and Studer (1889) is open to question. Further study is required to clarify the classification of this species. For that a higher number of specimens should be examined.

CHAPTER 6

TAXONOMIC REVIEW OF THE GENERA *Thouarella* and *Amphilaphis* (Family Primnoidae)

6.1 - INTRODUCTION

The family Primnoidae, comprising 25 genera, has been considered one of the best studied families of the order Gorgonacea (Bayer & Stefani, 1988), as a result of the work of Wright & Studer (1889), Versluys (1906) and especially Kükenthal (1907, 1908, 1915, 1919, 1924). The species assigned to Primnoidae, have as distinctive characters, a strongly calcified horny stem and scales- or plate-like sclerites, showing a cruciform pattern of extinction in polarized light (Bayer, 1956; Bayer & Stefani, 1988). Bayer (1982) concluded that the principal characters available for the classification and identification of primonoids are the colonial form and manner of branching; size and arrangement of polyps; number, arrangement, form and ornamentation of sclerites on the polyps and coenenchyme.

The species referred to Primnoidae were originally grouped into four subfamilies, Callozostroninae, Calyptrophorinae, Primnoidinae and Primnoinae, on the basis of presence of an operculum, aspect of sclerites and stiffness of stem (Wright & Studer, 1889). Versluys (1906) restructured the family Primnoidae into 5 subfamilies: Callozostroninae, Primnoidinae, Primnoinae, Primnoellinae and Thouarellinae. Versluys proposed the subfamily Thouarellinae to accommodate the closely related genera: *Amphilaphis, Thouarella, Plumarella* and *Stenella.* Some of the species assigned to these four genera have intermediary characters and have often been misplaced. Based upon their affinities, there appears to be some justification in Versluys' decision of grouping these four genera separately. However, Versluys' subfamilies were not accepted and Wright & Studer's original subfamilies were followed by subsequent authors.

There have been some attempts in grouping the species of the genus *Thouarella.* Versluys (1906) proposed three groups: *antarctica*-group, *hilgendorfi*-group and *koellikeri*-group, on the grounds of branching pattern and polyp distribution. Kinoshita (1908) established the subgenus, *Diplocalyptra*, based also upon branching pattern, polyp distribution, adding aspect of the operculum and spiculation. Kükenthal (1915) incorporated to *Thouarella* the genera *Amphilaphis* (Wright & Studer, 1889), *Rhopalonella* (Roule, 1908) and *Primnodendrum*

(Nutting, 1912). He invalidated Versluys' groups and Kinoshita's subgenus and introduced four new subgenera: *Amphilaphis, Euthouarella, Parathouarella* and *Epithouarella*, on the basis of number of longitudinal rows of polyp scales, distribution of polyps and presence of marginal thorns. Kükenthal also placed into *Thouarella* some specimens previously referred to *Plumarella* and *Stenella*. Bayer (1981d) restored the genus *Amphilaphis* on the basis of arrangement of opercular and marginal sclerites. Stibane (1987) also described the two genera *Amphilaphis* and *Thouarella* separately. Bayer did not, however, include the genus *Amphilaphis* in his key of the genera of Primnoidae in a later publication (1988). Kükenthal's classification, therefore, remains unchanged. The diagnosis defined by Kükenthal (1924) for the four subgenera of *Thouarella* are as follows:

Subgenus *Amphilaphis* - **Eight complete longitudinal rows of polyp scales.** The species are distinguished by the polyp distribution; presence of marginal thorns; and number of scales on each abaxial row.

Subgenus *Euthouarella* - **Longitudinal rows of polyp scales fewer than eight; marginal scales bearing thorn; polyps in pairs or in whorls of 3 to 4.** The species are distinguished by the branching pattern of colony and branches; number of whorls on 10 mm of branch; and polyp distribution.

Subgenus *Parathouarella* - **Longitudinal rows of polyp scales less than eight; marginal scales bearing thorn; and polyp placed singly**. The species are distinguished by the number of scales on each longitudinal abaxial row; ornamentation of body scales; and distribution of polyps.

Subgenus *Epithouarella* - **Longitudinal rows less than eight; and marginal scales without thorns**. The species are distinguished by the number of scales on each longitudinal abaxial row; and distribution of polyps.

There are 34 nominal species and 3 varieties attributable to *Thouarella.* To evaluate the taxonomic position of each of these species, the taxonomic characters applied for their classification were examined. Through the work carried out on *Th. variabilis* (Chapter 2-5), it was possible to verify the consistency of these characters since the species of this genus presented a similar range of variation of characters.

In Section 6.2, each of the taxonomic characters is discussed in detail and in Section 6.3 a review of the genus *Thouarella* and of the incorporated *Amphilaphis* is presented. Ideally the genera *Plumarella* and *Stenella* (= *Parastenella, Pterostenella, Dasystenella*) should have also been reviewed since some of the species assigned to the genus *Thouarella* have characters intermediate between these other genera; unfortunately, this was not realistic within the time available.

In Section 6.3, each of the nominal species referred to *Thouarella* is described according to the original and subsequent descriptions, supplemented, in some cases, by personal examination. It was possible to examine the type specimens of some of the species but others could not be located. The species are ordered according to their affinities. To allow more freedom of judgement they are not grouped in subgenera, since some of the characters used to diagnose the subgenera have been shown to be inconsistent.

Information contained in the original and subsequent descriptions was modified, without losing the essence, to conform the uniformity of the characters. Information which was not relevant was omitted. This included variable aspects such as dimensions of colonies, stems, calyx, and holdfast, and colour of specimen, which easily fades with preservation. The terminology was carefully chosen and adapted.

6.2 - TAXONOMIC CHARACTERS

COLONIAL FORM AND BRANCHING PATTERN - although the genus *Thouarella* is known for the bottlebrush appearance of the colonies, the majority of the species have been described as branched in one plane. Even the bottlebrush forms have often been said to be partly biseriate and in some cases a biseriate form is twisted along its axis attaining a bottlebrush appearance.

The great majority of the species have a racemose or monopodial branching pattern, where the main stem is continuous, bearing side branches or stems but not subdividing dichotomously distally. A few species have, however, a cymose branching pattern, where the main stem dichotomously subdivides into branches, disposed in one plane.

In bottlebrush forms the branches, simple or ramified, are disposed mostly on three sides of the main stem in ascending spirals. Sometimes they are placed on all sides, irregularly distributed or opposite, alternating in pairs. The biseriate growth forms often have true

pinnate branching pattern, where the stems give off only first order branches. The great majority of the species, with bottlebrush or biseriate growth forms, have simple branches. Some species have bifid branches and some others dichotomously ramified branches mostly up to second or third order branchlets. *Th. variabilis* is the only species with racemose growth form showing highly ramified branches (up to 5 orders).

The colony of most of the species is initially composed of a main stem surrounded by side branches, which might be simple or subdivided into branchlets. With continuous growth, the colony might give off side stems which will have branches of its own following the same original branching pattern. The side stem might give off in its turn substems which will also bear side branches. Therefore, the older the colony, the more complex the branching pattern. Side stems and substems are preferentially placed in one plane even giving off side branches on all sides. Hence, although the branching pattern is bottlebrush, the colony is fan-shaped as a whole. Based on this, it is not hard to appreciate the amount of confusion that the misinterpretation of this character has caused. Some authors have considered stems as branches and substems as twigs and the whole colony which was initially bottlebrush as fan-shaped. An author studying a young specimen of a certain species will probably have a different description from an author studying an older specimen. Added to this, there might also be the case of auto-epizoism, where the planula tend to settle on colonies of the same species, producing another colony which simulates a side stem. Auto-epizoism has not been described for this genus. Perhaps, a more careful examination could prove the contrary.

Some species were inadequately described and in the absence of illustration of the whole colony, it was sometimes impossible to define their branching pattern. The length of the branches varied from 10 to 50 mm, the average being 29 mm. Only the highest value was considered for the descriptions. The distance between branches was rarely mentioned but ranged from closely spaced to loosely distributed. The direction of the branches varied from disposed at nearly right angles to upwardly directed at acute angles.

ARRANGEMENT OF POLYPS - This has been considered a consistent character and has often been employed to diagnose species. However, it has already been shown to vary with the age of the colony (Gravier, 1914). Most of the species have the polyps singly placed on the branches, irregularly distributed, in ascending spirals of three or biserially arranged. Many species have the polyps placed in pairs and polyps are also commonly seen placed in whorls of 3 to 6, mostly 3. In many specimens, the polyps are singly placed proximally at the branch

and in pairs or even in whorls of three distally. Older specimens have also been shown to have more polyps on a whorl than younger specimens. In many species, the original and subsequent descriptions contradict on this character.

DISTRIBUTION (NUMBER) OF POLYPS - this character has also been used as a distinctive character but it is one of the most varied characters within the same colony. Among the species of *Thouarella,* the distribution of polyps varies from sparsely distributed (up to 4 mm apart) to very closely spaced (30 polyps/per 10 mm of branch).

POLYP SHAPE - In general, the polyps of the species of *Thouarella* have a narrow stalk or body and an enlarged calyx. The polyps have, therefore, been described as bell-shaped, cup-shaped, pear-shaped and mostly as club-shaped, apparently these descriptors all mean the same although some difference could be inferred. The species previously referred to the genus *Amphilaphis* do not have thorny marginal scales covering the opercular scales and have a rounded calyx. For these species, it seems more appropriate to define the polyp as club-shaped. However, it is difficult to make such distinctions. Some species show sturdy and more cylindrical polyps, which do look different from the club-shaped polyps.

POLYP SIZE - This appears to be a consistent character, as there is little natural variation. It is, however, subject to artificial variation. It should only be measured on adult polyps. The measurement of the length of the polyps can be made difficult if the polyp is incurved adaxially and will also depend on the limits of dimension chosen by the individual researcher. The smallest polyp (0.9 mm long) was described for *Th. hilgendorfi* and the longest (3 mm long) for *Th. grandiflora*, the average being 1.6 mm. Colonies with small polyps have a "fragile" appearance and colonies with large polyps a "strong" appearance.

POLYP ORIENTATION - Polyps have been described as directed upwards and adaxially incurved; directed outwards and rigid; directed outwards and slightly upwards, or slightly incurved abaxially. In general, the polyps are mostly upwards and incurved adaxially to different degrees. Some species have apparently rigid and directly outwards polyps, which gives a different appearance to the whole colony.

ARRANGEMENT OF SCALES - The arrangement of scales on the polyp has been one of the key characters to distinguish species and subgenera. The subgenus *Amphilaphis* is distinguished solely on the number of longitudinal rows of scales, which in the species assigned to this subgenus is eight and in the species of the other subgenera less than eight. As observed in *Th. variabilis*, the polyp, when not bearing reproductive products, has the

scales overlapping each other and longitudinal rows of scales, often observed distally on the polyp, are not recognized proximally. When expanded to accommodate the eggs or sperm sacs, the sclerites slide to a certain degree and longitudinal rows may then be recognized. In some species of the subgenus *Amphilaphis,* eight longitudinal rows of scales are not so readily recognized proximally. This character should, therefore, be used with caution. However, the species of this subgenus appear to have smaller, less overlapped and more organized scales than the other species.

The arrangement of the opercular and marginal scales was the key character employed by Bayer (1981d) to distinguish the genera *Thouarella* and *Amphilaphis.* According to this author, in *Thouarella* the marginal scales form two rings of four scales alternating in two transverse rows, the opercular scales form inner and outer rings of four scales each, alternating larger and smaller and the marginals are larger than operculars and fold inward over them. In *Amphilaphis* the marginal and opercular scales each form a ring of eight scales in a single transverse row and the marginals do not fold over operculars. The arrangement of marginal and opercular scales was not commonly applied in the past and to consider this character all the species of this genus would have to be reexamined. The arrangement of sclerites is difficult to observe without employing a destructive method, which is not possible when dealing with type specimens.

NUMBER OF SCALES ON EACH LONGITUDINAL ABAXIAL ROW - This appears to be a consistent character since the number of scales do not appear to vary. It has been used as a key character to distinguish species. The scales are sometimes difficult to count because longitudinal rows are not always recognizable. The number of scales on each abaxial row varies from 4 (*Th. longispinosa*) to 12 (*Th. regularis*).

SCLERITES - The scale-like sclerites of the species of *Thouarella* differ in shape and size but little in ornamentation. It is very difficult to understand a description of the aspects of the sclerites even if a drawing is provided. The drawings accompanying the descriptions of most species were inaccurate and misleading. A drawing of the ornamentation of sclerites cannot possibly compete in reliability with the illustrations provided by modern technology. Micrographs taken with an scanning electron microscope are of high precision showing a reliable picture of the sclerites. Through the studies carried out on *Th. variabilis*, it was possible to recognize the structures described.

Most of the scales have their inner surface ornamented by irregular or radially arranged warts, and on the outer surface by ridges radially oriented or projecting teeth. The free margin of the scales ranges from finely to strongly serrated or toothed, and the proximal portion is generally indented.

OPERCULUM AND OPERCULAR SCALES - The operculum of most of the species are low and covered by the marginal scales. In some species the operculum is high and uncovered. Some low opercula are also uncovered. The opercular scales are generally triangular, concave dorsally with a median keel, pointed or blunt distally.

MARGINAL SCALES - The marginal scales will vary from indistinctive rounded or triangular broad scales without distal projections to pointed, lancet-shaped or distinctive bearing pronounced sharp long thorns. It usually has an enlarged body covered on the inner side by radially or irregularly arranged warts and on the outer side by short prominences. The thorny marginal scales usually cover the opercular scales. Sometimes it is difficult to interpret what an author refers to as long or short thorns, without examining the specimen personally.

BODY SCALES - The body scales are the scales covering the body excluding the opercular scales. Owing to the range of variations shown by the marginal scales, these scales are treated separately in the descriptions of the species and are, therefore, not included in the body scales. The submarginal scales commonly have a short median thorn or spine being distinct from the lower scales, and when they do so, these scales are also mentioned separately and the other body scales are then treated as lower body scales. The adaxial scales are usually plate-like and smaller than the abaxials and laterals and if information is available about these scales, they are also mentioned separately.

COENENCHYME SCALES - The branches are commonly covered by two layers of coenenchyme scales. The upper layer is composed of flat, large scales, similar to the body scales of the polyps. These scales might be rounded, triangular, quadrangular, polygonal or irregular in shape. The inner layer has small, rounded, triangular or irregular ornamented scales. The coenenchyme scales covering the stem are small, plate-like and strongly sculptured.

MAIN STEM - The main stem is, in general, brittle, rigid and horny, longitudinally grooved and mostly oval in transverse section and flattened. Some species have the main stem circular in transverse section.

6.3 - REVIEW OF THE GENERA *Amphilaphis* and *Thouarella*

Family PRIMNOIDAE (after Bayer, 1956; Bayer & Stefani, 1988) - Strongly calcified horny stem. Scales- or plate-like sclerites, showing a cruciform pattern of extinction in polarized light.

Subfamily PRIMNOINAE (after Bayer, 1956) - Polyps with well-differentiated operculum, body scales in eight complete or incomplete rows.

Amphilaphis Wright & Studer, 1889

Amphilaphis Wright & Studer, 1889:70-71. - Versluys, 1906:20-22. - Nutting, 1908:573. - Kinoshita, 1908:49. - Bayer, 1981:936 (key).

Thouarella (Amphilaphis) Kükenthal, 1915:206-207 (key). - Kükenthal, 1919:409. - Kükenthal, 1924:289.

TYPE SPECIMEN & TYPE LOCALITY. - *Amphilaphis regularis* Wright & Studer, 1889. St. 135a, Challenger Expedition, off Inaccessible Island, Tristan da Cunha, 75 fathoms; St. 135c, off Nightingale Island, 100-150 fathoms. SYNTYPE - B.M. 1889.6.27.60, N.H.M. London, UK. (SYNTYPE - 32.12.8.7).

DIAGNOSIS. - Colony form pinnate. Main stem gives off branches on two sides in alternating series, arising at angles of about 45°. Branches simple or giving off first order branchlets of their own. Branches and branchlets parallel to one another. Polyps club-shaped, singly placed and irregularly distributed all round the branches distally and in pairs proximally. Polyps with eight longitudinal rows of scales distally; rows less defined proximally, where the polyp is narrower; 10-12 scales on each abaxial row; 7-8 scales on each adaxial row. Operculum cone-shaped formed of eight triangular scales, sometimes long and lancet-shaped. Marginal scales not prominent. Body and coenenchyme scales stout and strongly sculptured with prominences merged into radial ridges, running outwards from a nucleus projecting beyond the edge as spines. Thin coenenchyme, with two layers of scales.

REMARKS. - *Amphilaphis* means "dividing into branches" in Greek. From Wright & Studer's drawing of the type specimen, *Amphilaphis regularis*, it is possible to observe the branches

are all initially simple but when they reach a certain size, they start giving off branchlets in the same manner as the main stem. Unlike the species of *Thouarella,* the branches do not divide dichotomously into branchlets. There is a main axis with branchlets on two sides of it. The ramified branches have the axes as thick as the simple branches, suggesting that they are not as the side stems observed in the species of *Thouarella.* This genus differs from *Thouarella* for its mode of ramification and from *Plumarella* by the polyp distribution. Wright & Studer (1889) considered it to represent a transition between *Thouarella* and *Plumarella.* The main feature that *Amphilaphis* shares with *Plumarella* is the pinnate branching pattern, although the branches are not so often opposite as in *Plumarella.* The latter also has a more regular branching pattern. The polyps of *Plumarella,* unlike *Amphilaphis,* are more cylindrical and have an undeveloped operculum. The opercular scales are also considerably different in the two genus. The opercular scales of *Amphilaphis* resemble the scales of *Thouarella hilgendorfi* and *Th. moseleyi. Amphilaphis* differs from *Thouarella* in the higher number of sclerites on each longitudinal row in *Amphilaphis;* in the branching pattern and in the length of the branches. Kükenthal (1919; 1924) made *Amphilaphis* a subgenus of *Thouarella,* to which he added some other species. He then modified the diagnosis slightly to accommodate these other species. To the diagnosis was added: polyps placed partly singly all around the branch, and partly in pairs; marginal scales bearing a median rib with only a reduced spine; opercular scales concave bearing a median keel; upper coenenchyme scales large, polygonal or of irregular outline and lower scales smaller and plate-like. Bayer (1981d) revalidated the genus *Amphilaphis* in his key to the genera of Octocorallia, which he diagnosed as: "Marginal and opercular scales each forming a circle of 8 scales in a single transverse row; marginals not folding over operculars. Polyps in pairs on proximal part of twigs, irregularly scattered on distal part; inner face of opercular scales with prominent apical keel". *Amphilaphis* was also describe by Stibane (1987). Bayer (1988), however, excluded this genus from his more recent key of 1988.

The nominal species which had been attributed to *Amphilaphis* are:

Amphilaphis regularis Wright & Studer, 1889
Amphilaphis abietina Studer, 1894
Amphilaphis biserialis Nutting, 1908
Amphilaphis plumacea Thomson & Mackinnon, 1911

These species will be described in this Chapter under the name *Thouarella regularis, Th. abietina, Th. biserialis* and *Th. plumacea* as these are their current names.

Thouarella Gray, 1870

Primnoa Valenciennes, 1846, pl. 12, fig. 2. - Gray, 1859:483.

Thouarella Gray, 1870:45. - Studer, 1878:649. - Studer, 1887:50. - Wright & Studer, 1889:59-61. - May, 1899:14. - Versluys, 1906:22-24. - Thomson & Henderson, 1906:38-41 (comparative tables). - Roule, 1908:1. - Kinoshita, 1908:20-21. - Kinoshita, 1908:49-52. - Kükenthal, 1908:10-11. - Kükenthal, 1912:292-296. - Nutting, 1912:66. - Kükenthal, 1915:149 (key). - Kükenthal, 1919:405. - Thomson, 1921:77. - Kükenthal, 1924:287-288. - Broch, 1965:24. - Bayer, 1956:220. - Bayer, 1981d:936, fig. 68 (key). - Stibane, 1987:22, pl. 1, fig. 4; pl. 2, fig. 4 (comparative table). - Bayer, 1988:455 (key).

TYPE SPECIES. - *Primnoa antarctica* Valenciennes, 1846. Voyage sur la "Vénus", Falkland Islands. Present location of specimen unknown.

DIAGNOSIS. - Colony form bottlebrush, with branches distributed all around the stem, mostly in three directions, or biserially rearranged as the branches bend laterally forming a frontal and versal side; true planar growth forms, with branches arising on only two sides, pinnate or dichotomously branched, are also found; branching pattern mostly racemose, seldom cymose. Main stem, generally simple, occasionally divided into side stems, which then lie in one plane. Branches more slender than the stems and orientated, in most cases, at nearly right angles, at times upwards in a more acute angle; either remaining simple or giving off branchlets, often dichotomously branched.

Polyps singly placed on the branches, in short ascending spirals of mostly three; in pairs or in whorls of 3 to 4 or sometimes even more; if present on the stem, they are arranged irregularly. Polyps orientated outwards and upwards in relation to the branches, in some cases at almost right angle, in others adaxially incurved; polyps differ in size, ranging from 1 to 3 mm long. Adaxial side of the polyp usually shorter than abaxial side. Polyp cup-, bell-, pear- or club-shaped, with the lower portion narrow and the calyx enlarged; at times cylindrical or sturdy, with calyx, neck and bases of similar dimensions.

Opercular scales eight and covered on their bases by eight marginal scales, from which eight longitudinal rows of abaxial, lateral and adaxial body scales depart; these longitudinal rows are often recognizable distally on the polyp but not so clear proximally. Longitudinal rows mostly only two adaxially; each longitudinal abaxial-lateral row with 4 to 12, mostly 5 to 6 imbricate scales overlapping each other and with strong convex and serrated upper margins; on the inner surface numerous warts radiate outwards from a nucleus; on the outer surface

prominences or short spines often merge to form ridges, terminating in projecting teeth beyond the edge of the scale.

Operculum formed of eight pointed triangular scales, with a blunt or squared distal portion; outer surface of opercular scale with a deep longitudinal furrow and in the inner side with a median keel. When the operculum is closed the scales are overlying each other forming inner and outer rings of four scales each, alternating larger and smaller. Operculum mostly low and covered by the marginal scales, sometimes high and uncovered.

Marginal scales placed in two rings of 4 scales alternating in 2 transverse rows below operculum; triangular-shaped to varied degree, sometimes leaf-like; often bearing a prominent middle tooth, at times forming a long thorn; sometimes the thorn is lacking and the marginal scale does not differ much from the other body scales. Submarginal scales showing a reduced thorn, being a transition between the thorny marginal and the roundish lower body scales. Adaxial scales mostly small, delicate, plate-like; rarely they are similar to the abaxials. Coenenchyme thin covered by an upper layer of flat imbricate overlapping scales, with free margins strongly serrated and a lower layer of small triangular or polygonal scales. Coenenchyme of stem smaller and more sculptured than the coenenchyme of the branches.

REMARKS. - Gray (1870) introduced the genus *Thouarella* to accommodate a specimen, referred as *Primnoa antarctica*, from which he had very little information, merely a drawing of a branch showing the arrangement of polyps, with no information of the colony form. Later he received a specimen that he could refer to *Thouarella antarctica*. The original diagnosis for the genus *Thouarella* was, therefore, based on a drawing of a fragment and on a further single specimen. The diagnosis described a bottlebrush colony, whose stem gave off long, simple and filiform branches on all sides; polyps bell-shaped, covered by 4 to 5 series of large imbricate scales. Wright & Studer (1889), studying the specimens of the "Challenger" collection, observed a series of nearly related forms and modified the diagnosis of the genus, increasing the range of variation of characters. Wright & Studer (1889) considered *Thouarella* most closely related to *Stenella*. Wright Studer stated the following about the genus *Stenella* and *Thouarella*: "..it is possible that a more extended knowledge of the species will result in the two being merged into one". The main difference between the two genera as discussed by these authors was the distribution of polyps on the branches, being in pairs or in whorls in *Stenella* whereas in *Thouarella* they were singly placed. The diagnosis of Wright & Studer for *Thouarella* was, however, limited at the time. This variation of character was subsequently disregarded as some species referred posteriorly to *Thouarella* had the polyps disposed like

the ones in *Stenella*. Versluys (1906) considered the main difference between the two genera, the number of marginal scales, which is smaller in *Stenella*.

The nominal species which have been attributed to *Thouarella* are listed below according to their affinities:

1 - *Th. (Parathouarella) antarctica* Valenciennes, 1846
2 - *Th. (Parathouarella) variabilis* Wright & Studer, 1889
**Th. (P.) variabilis* var. *brevispinosa* Wright & Studer, 1889
**Th. (P.) variabilis* var. *gracilis* Wright & Studer, 1889
3 - *Th. (Parathourella) striata* Kükenthal, 1907
4 - *Th. (Parathouarella) versluysi* Kükenthal, 1907
5 - *Th. (Parathouarella) clavata* Kükenthal, 1907
6 - *Th. (Parathouarella) koellikeri* Wright & Studer, 1889
7 - *Th. (Euthouarella) longispinosa* Kükenthal, 1919
8 - *Th. (Euthouarella) hilgendorfi* Studer, 1878
9 - *Th. (Euthouarella) abeis* Broch, 1965
10 - *Th. (Euthouarella) typica* Kinoshita, 1907
11 - *Th. (Euthouarella) moseleyi* Wright & Studer, 1889
**Th. (E.) moseleyi* var. *spicata* Wright & Studer, 1889
12 - *Th. (Euthourella) laxa* Versluys, 1906
13 - *Th. (Euthouarella) tenuisquamis* Kükenthal, 1907
14 - *Th. (Euthouarella) carinata* Kükenthal, 1908
15 - *Th. (Euthouarella) flabellata* Kükenthal, 1907
16 - *Th. (Euthouarella) tydemani* Gray, 1870
17 - *Th. (Euthouarella) coronata* Kinoshita, 1908
18 - *Th. (Epithouarella) crenelata* Kükenthal, 1907
19 - *Th. (Epithouarella) affinis* Wright & Studer, 1889
20 - *Th. (Epithouarella) chilensis* Kükenthal, 1912
21 - *Th. (Amphilaphis) regularis* Wright & Studer, 1889
22 - *Th. (Amphilaphis) parva* Kinoshita, 1908
23 - *Th. (Amphilaphis) abietina* Studer, 1894
24 - *Th. (Amphilaphis) dispersa* Kükenthal, 1912
25 - *Th. (Amphilaphis) superba* Nutting, 1912
26 - *Th. (Amphilaphis) grandiflora* Kükenthal, 1912
27 - *Th. (Amphilaphis) plumacea* Thomson & Mackinnon, 1911
28 - *Th. (Thouarella) biserialis* Nutting, 1908
29 - *Th. (Thouarella) alternata* Nutting, 1912
30 - *Th. (Thouarella) recta* Nutting, 1912
31 - *Th. (Thouarella) pendulina* Roule, 1908
32 - *Th. (Thouarella) brucei* Thomson & Ritchie, 1906
33 - *Th. (Thouarella) hicksonii* Thomson, 1911
34 - *Th. (Thouarella) acanthina* Wright & Studer, 1889

SPECIES 1 - *Thouarella antarctica*

Thouarella antarctica (Valenciennes, 1846)

Primnoa antarctica Valenciennes, 1846, pl. 12, fig. 2. - Milne-Edwards, 1857:140. - Gray, 1857:286; - Gray, 1859:483. - Kölliker, 1865:135.

Thouarella antarctica: Gray, 1870:45. - Gray, 1872:482. - Studer, 1878:649. - Wright & Studer, 1889:65, pl. 21, fig. 6. - Versluys, 1906:35-36. - Hickson, 1907:9-10, pl. 2, figs 19, 24. - Gravier, 1913:460-466. - Gravier, 1914:48-56. - Molander, 1929:75.

Thouarella aff. *antarctica:* Kükenthal, 1907:203-204.

Thouarella (Parathouarella) antarctica: Kükenthal, 1915:150; - Kükenthal, 1919:433. - Kükenthal, 1924:299.

Thouarella (Euthouarella) antarctica: Broch, 1965:24-26, pl. 1, fig.2.

TYPE SPECIMEN. - *Primnoa antarctica* Valenciennes, 1846. Voyage sur la "Vénus", Iles Malouines, Falkland Islands. Present location of specimen unknown.

DIAGNOSIS. - Colony form bottlebrush; branches distributed on all sides of the main stem. Polyps singly placed. Longitudinal rows of scales fewer than eight proximally. Nine to ten transverse abaxial rows. Marginal scales bearing from foliate processes projecting as a thorn.

DESCRIPTION. - Bottlebrush colony, with branches arising on all sides of the main stem at nearly right angles. Main stem horny, calcareous, brittle, oval in transverse section and twisted in a long spiral. Branches mostly simple, 20 mm long, densely disposed. Polyps club-shaped, 2 mm long, singly placed; directed upwards and densely distributed distally at the branches. Longitudinal rows of abaxial scales not well defined proximally; nine to ten transverse abaxial rows. Opercular scales leaf-shaped, elongated, narrow and pointed, with a median furrow on the outer surface and a salient keel on the inner surface; free margins serrated; 0.67 mm long/0.35 mm wide. Marginal scales eight, middle portion enlarged and gradually tapering distally, where it bears a median thorn; inner surface with a median longitudinal keel extended into lateral platforms; enlarged body with numerous warts radially distributed; free margin serrated; higher than broad, 0.8 mm long/0.6 mm wide; thorn 0.56 mm long/0.23 mm wide. Submarginal scales bearing a projecting median cusp; indented and undulated base and strongly serrated free margins; inner surface with numerous warts radially arranged and departing from a marked nucleus. Lower body scales large, flat, with free margin strongly wrinkled, serrated and ornamented with radial ridges; broader than high, 0.6

mm long/0.65 mm wide. Coenenchyme scales vary in form and dimension, with free margin serrated and base indented; on their inner surface numerous small warts coalesce radially towards a marked nucleus.

REMARKS. - The original description of *Primnoa antarctica* by Valenciennes (1846) has no text but two inaccurate drawings. In these drawings, the branches appear poorly ramified, the polyps alternate and the marginal scales are short and do not cover the opercular scales. Subsequent descriptions are brief and do not add any information. Kölliker (1865) gave only information about general sclerite size (0.18-0.66 mm), without specifying which sclerites he measured.

On the grounds of branching pattern, Gray (1872) introduced a new genus, *Thouarella*, to accommodate the bottlebrush forms of *Thouarella antarctica* originally attributed to *Primnoa*. His description of *Thouarella antarctica* is so general that it could be the description of many other species of the genus *Thouarella*. Wright & Studer (1889) gave a much better diagnosis of the species, considering most of the available characters, particularly spiculation and giving details of all the scales and a better definition of the abaxial rows of scales. Hickson (1907) referred his specimens to *Th. antarctica* on the grounds of branching pattern, although the description of some of his specimens differed from the original description in having a few ramified branches and smaller polyps (1.5 mm). His description of the marginal scales lead Gravier (1914) to associate it to *Th. variabilis* rather than to *Th. antarctica*. Gravier (1914) appeared to be the only author to have seen the type specimen and could thus give a more reliable account on the description of the specimen. Kükenthal (1915) assigned this species to the subgenus *Parathouarella* which comprises species which have polyps singly placed, longitudinal rows fewer than eight proximally and the marginal scales bearing foliate processes. Based on Gravier's pictures (1914), Broch (1965) judged Kükenthal's decision wrong since the polyps appeared seemingly disposed in whorls rather than singly and placed this species in the subgenus *Euthouarella*. Examining the pictures, it is possible to observe that Broch was mistaken and Kükenthal was correct. The polyps are so crowded at the extremities of the branches that they give the impression of being in whorls but it is clearly seen that they are not perfectly disposed in whorls but just irregularly agglomerated.

SPECIES 2 - *Thouarella variabilis* - see Chapter 5.

SPECIES 3 - ***Thouarella striata***

Thouarella striata Kükenthal, 1907

Thouarella striata Kükenthal, 1907:204-205. - Nutting, 1912:69, pl. 10, figs 2, 2a.
Thouarella (Parathouarella) striata Kükenthal, 1915:150. - Kükenthal, 1919:426, pl. 42, fig. 67. - Kükenthal, 1924:297. - Thomson & Rennet, 1931:27. - Broch, 1965:31, pl. 7, figs 20, 21.

TYPE SPECIMEN & TYPE LOCALITY. - *Thouarella striata* Kükenthal, 1907. St. 131, Deutschen Tiefsee-Expedition, East Coast of Bouvet Island, 5°10'S, 23°2'E, 475 m. Present location: Z.M.B. 6086, Museum für Naturkunde, Humboldt-Universität, Berlin, Germany.

DIAGNOSIS. - Bottlebrush colony with branches on all sides. Polyps singly placed. Longitudinal rows of polyp scales fewer than eight proximally. Four to five scales on a longitudinal abaxial row. Marginal scales bearing foliate processes projecting as a thorn. Polyp scales strongly radially ridged.

DESCRIPTION. - Colony form bottlebrush with a cylindrical outline. Main stem gives off on all sides numerous branches at almost a right angle. Branches, 25 mm long, closely spaced in a notably dense arrangement. Extremities of the branches are in some parts turned to one side of the colony which attains an apparent biseriate arrangement. Branches subdivided dichotomously into branchlets once or twice; mostly simple distally on the colony. End branchlets from branches of the middle portion are longer than end branchlets of basal branches. Club-shaped polyps, 1.5 to 2.0 mm long, singly placed on all sides of the branches, rarely in pairs, never in whorls; directed outwards, slightly upwards in relation to the axis. They are crowdedly arranged on the branches, as close as they can get. Polyps mostly absent from the stem, apart from the very distal portion, where they are found to be very small. Longitudinal abaxial rows with four to five scales, although longitudinal rows are not easily recognized. Opercular scales triangular, pointed and bearing a middle keel on their inner surface; 0.48 mm high. Marginal scales broad, 0.6 mm high, with an enlarged body tapering into a winged spine, which is supported by a weak keel; outer surface with a median longitudinal furrow adorned with longitudinal ridges on its lower portion; free margins finely serrated. Body scales 0.6 mm wide/0.45 mm high, with finely serrated margins. Submarginal scales have a reduced thorn. For all polyp scales, the prominence of radial ridges and ribs is characteristic. Upper coenenchyme scales of the branches elongated, 0.3 mm long; lower scales small, numerous, roundish or oval in shape; around 0.1 mm in diameter. Coenenchyme scale of the stem with strong radial ridges and indented margins.

REMARKS. - The type specimen is deformed because of the formation of a worm tube, this being the reason for the apparently biseriate arrangement of branches in some parts of the colony, where the branches are bent towards the tube forming a frontal and versal side of the colony. The part of the colony which is not affected by the worm has a perfect bottlebrush arrangement, with the branches arising in all directions. The biseriate arrangement would probably not happen if there was no worm inhabiting the colony. On the distal portion of the colony, the branches are all bent towards the end of the tube that makes the tip descend. On the distal branches and on the extremity of the middle and proximal branches, the polyps are less aggregated making these regions less rigid and more loose than the lower order branchlets. On the polychaete tube, the arrangement of polyps is altered since the polyps are abnormally facing against the tube. The polyps are mostly in reproductive stage and therefore much expanded, which might explain their apparently aggregated condition, especially far from the tips. The polyps on the extremities are probably non-reproducing and consequently do not have their bases expanded being more sparsely distributed and this would probably be the aspect of the whole colony if most of the polyps were not reproducing. It should be emphasized that the reproductive state of the polyps change the whole aspect of the colony, making it denser and more rigid than normal. In normal conditions, this specimen could easily be referred to *Thouarella variabilis*, if it was not for its branches being poorly ramified. Kükenthal (1907) also pointed out the similarity of these two species and in his view they differed mainly on details of spiculation, the radial ridges existing on the scale surface of *Th. striata* are said by Kükenthal to be absent in *Th. variabilis*. Radial ridges have been, however, clearly shown in the specimens of *Th. variabilis* examined in the present study, and are shown in the figures of Chapter 2. There is no reason, therefore, to introduce a species on the grounds of variation of this character.

Nutting (1912) referred to this species an old specimen (30 cm high), highly dichotomously branched (up to 5 orders) and perfectly fan-shaped. In his description, the polyps are placed in whorls of 5, 6 or even 10 and there are 10 polyps per cm of branch; the abaxial scales are reduced to 2 or 3 and only the marginal scales have ribs. His description contradicts a great deal with the original description and could not sensibly be referred to *Th. striata*.

Broch (1965) stated the following about the two species: "The main stem of *Thouarella striata* is much more branching than that of *Thouarella variabilis*, the colonies more robust and exuberant because its twigs are more branching". It is hard to interpret what Broch (loc. cit.) meant by "branching" because if he means "more divided into branchlets", his words contradict the original description of *Th. variabilis*. The type specimen of *Th. variabilis* is more ramified than the type specimen of *Th. striata*. If he means, however, that it has a higher

number of branches, which are closely spaced, he could be mislead by the reproductive stage of the polyps giving a more dense aspect to the colony. Broch also noted that the marginal scales of *Th. striata* are constructed like those of *Th. variabilis* but that they are differently moulded, the former being more convex dorsally and having a pointed thorn and the latter being straight, having a blunt thorn. In Chapter 2, it was shown that the aspect of the scales varies considerably within the same colony. Marginal scales were observed to be slightly flat or more concave and having blunt or pointed thorns. These variations could, therefore, be misleading.

SPECIES 4 - *Thouarella versluysi*

Thouarella versluysi Kükenthal, 1907

Thouarella versluysi Kükenthal, 1907:202-203

Thouarella (Parathouarella) versluysi Kükenthal, 1915:150. - Kükenthal, 1919:428, pl. 43, fig. 68. - Kükenthal, 1924:298.

TYPE SPECIMEN & TYPE LOCALITY. - *Thouarella versluysi* Kükenthal, 1907. St. 103, Deutschen Tiefsee-Expedition, Agulhasstrom, 35°10'S, 23°2'E, 500 m. Present location: Z.M.B. 6089, Museum für Naturkunde, Humboldt-Universität, Berlin, Germany.

DIAGNOSIS. - Bottlebrush colony. Polyps singly placed and sparsely distributed. Polyp scales placed in fewer than eight longitudinal rows proximally; six scales on a longitudinal abaxial row. Marginal scales bearing foliate processes projecting as a thorn.

DESCRIPTION. - Colony form roughly bottlebrush, partly biseriate. Main stem strongly bent, giving off a well developed side stem. Branches arise on all sides of the main and side stems, mostly alternating in three directions; partly bent to the laterals forming a frontal and versal side of the colony. Branches simple or dichotomously divided into branchlets, mostly up to second order branchlets, rarely divided into higher orders; unbranched distally on the colony; average branch length 20 mm. Polyps sturdy, 1 to 2 mm long and placed singly on the branches and branchlets, never in whorls; closely spaced distally on the branches and sparsely distributed proximally; four to five polyps on 10 mm of branch, absent on the stems. Polyps usually placed in one plane, often in all directions and slightly incurved adaxially. Longitudinal abaxial rows of scales fewer than eight proximally; six scales on a longitudinal

abaxial row; four adaxially. Opercular scales triangular, pointed, projecting into a thorn distally, supported by a long median keel; with winged lateral margins. Marginal scales also triangular and pointed, bearing a long thorn, which is supported by a strong inner keel; 0.6 mm high. Body scales flattened with free margin smooth. Coenenchyme scales of the branches plate-like or with irregular shape, 0.2 mm - 0.25 mm in diameter; on the stems they are smaller, 0.12 mm in diameter.

REMARKS. - The type specimen has a fragile appearance with main and side stems slender and loose. The polyps look large when compared to the thickness of the stems. One side of the main stem has almost all the branches broken. The side stem is nearly as well developed as the main stem, It probably started its development when the colony was very young because the colony is still apparently young (less than 10 cm high). Kükenthal (1924) made a printing mistake concerning the size of the polyp, what he described as 10 mm instead of 1 mm. The mistake is a bit obvious, although might mislead those who are not familiar with the group because 10 mm polyps exist in octocorals such as in *Anthomastus*. Kükenthal found *Th. versluysi* similar to *Th. brucei* concerning the arrangement of branches, disposed on all sides of the stem, and the distribution of the polyps, which are singly placed. This specimen could, therefore, be a young form of *Th. brucei*.

SPECIES 5 - *Thouarella clavata*

Thouarella clavata (Kükenthal, 1907)

Thouarella aff. *antarctica* Kükenthal, 1907:203-204.
Thouarella clavata: Kükenthal, 1908:11.
Thouarella (Parathouarella) clavata: Kükenthal, 1915:150. Kükenthal, 1919:430, pl. 43, fig.69. - Kükenthal, 1924:298.

TYPE SPECIMEN & TYPE LOCALITY. - *Thouarella clavata* Kükenthal, 1907. St. 103, Deutschen Tiefsee-Expedition, Agulhasstrom, 35°10'5S, 23°2'0E, 500 m. Present location: Z.M.B. 6080, Museum für Naturkunde, Humboldt-Universität, Berlin, Germany.

DIAGNOSIS. - Bottlebrush colony. Polyps singly placed and sparsely distributed, crowdedly aggregate in some sites. Polyp scales placed in fewer than eight longitudinal rows proximally; five to six scales on a longitudinal abaxial row. Marginal scales bearing foliate processes projecting as thorn.

DESCRIPTION. - Colony form bottlebrush. Main stem highly twisted and irregularly giving off side stems. Main and side stems surrounded by branches oriented at almost right angles. If the colony is untwisted, it appears biserially arranged, with the branches bent towards one side of the colony forming a frontal and versal side. On the frontal side, the branches are bent inwards and are longer than the branches on the versal side. Branches 30 to 35 mm long and mostly dichotomously branched. Polyps club-shaped, sometimes sturdy, 1.5 to 2.0 mm long, singly placed and sparsely distributed on the branches; absent on the stems. Each longitudinal abaxial row with five to six scales. Opercular scales triangular with a narrow median keel. Marginal scales broad, triangular, with a short thorn, a narrow keel and ornamented with radial ribs and serrated margins. Body scales 0.5 mm wide with flattened edges. Coenenchyme scales of the branches narrow and 0.3 to 0.6 mm long; scales of the stem smaller, plate-like and adorned with warts; 0.24 mm in diameter.

REMARKS. - In some parts of the colony, there are some clumps of polyps with their bases apparently embedded in a calcareous mass. The polyps of these aggregates are short, wide and sturdy. They have more thorny and stronger marginal scales than the other polyps of the colony. Their opercular scales also have a deeper median furrow than the opercular scales of the other polyps . They look as if they belonged to another species. The clumps were also observed on another species, *Th. grandiflora*, where they were formed close to the path of an annelid polychaete. In his key for the species of the subgenus *Parathouarella*, Kükenthal (1915) used these aggregations of polyps as a valid character to separate *Th. clavata* from the other species assigned to this subgenus. These aggregations are occasional and probably caused by a stressing situation, such as the presence of a commensal. This character should be considered with caution and probably disregarded.

SPECIES 6 - ***Thouarella koellikeri***

Thouarella koellikeri Wright & Studer, 1889

Thouarella koellikeri Wright & Studer, 1889:64-65, pl.21, fig. 5. - May, 1899:14. - Versluys, 1906:35.

Thouarella (Parathourella) koellikeri Kükenthal, 1915:150. - Kükenthal, 1919:435. - Kükenthal, 1924:299.

TYPE SPECIMEN AND TYPE LOCALITY. - *Thouarella koellikeri* Wright & Studer, 1889. St. 308, Challenger Expedition, off Tom Bay, Patagonia, 175 fathoms. SYNTYPE - B.M. 1889.5.27.41, N.H.M London, UK; St. 310, Challenger Expedition, off Sarmiento Channel, Patagonia, 400 fathoms, 10/01/1876. SYNTYPE - B.M. 89.5.27.42, N.H.M. London, UK.

DIAGNOSIS. - Branches in three directions but biserially arranged. Polyps singly placed. Polyps placed in fewer than eight longitudinal rows proximally; eight scales on a longitudinal abaxial row. Marginal scales bearing foliate processes.

DESCRIPTION. - Branches coming off the main stem in three directions but attaining a biseriate or slightly secund arrangement rather than a bottlebrush form as they bend in one direction, defining a frontal and a versal side of the stem. Frontal branches simple, 10 to 25 mm long. Side stems as strong as the main stem, 80 to 100 mm long, giving off branches of their own in a similar pattern as described above. Branches occasionally ramified. Main stem horny, oval in transverse section and twisting along its axis; base calcareous and brittle. Polyps pear-shaped, arising singly in three directions, surrounding the branches in short spirals. Polyp scales placed in five abaxial-lateral and two adaxial longitudinal rows and in eight transverse rows. Opercular scales triangular, concave, having unequal sides; outer surface adorned with radial ridges and a median rib projects into a pronounced spine; 0.70 mm high/0.27 mm wide. Abaxial opercular scales pointed with serrated margins, adaxial scales small and covered by the broad lateral ones. Marginal scales higher than broad, pointed, lancet-shaped, with the median rib strongly developed into a serrated keel projecting beyond the edge of the scale; margins finely toothed; 0.62 mm high/0.54 mm wide. Submarginal scales bearing a short thorn; outer surface ornamented with radial ridges, formed from a fusion of prominences which can project beyond the edge of the scale; 0.42 mm high/0.57 mm wide. Lower body scales with convex upper edge, outer surface with diverging longitudinal ribs, of which a middle one is the most developed. Coenenchyme scales of the

upper layer irregularly triangular, polygonal or quadrangular, overlapping one another; 0.41 mm high/0.26 mm wide; coenenchyme scales of the inner layer flat, triangular or irregularly polygonal, 0.18 mm high/0.12 mm wide.

REMARKS. - Wright & Studer appear to have studied a relatively old specimen which had well developed side stems but their description is very vague and, without an illustration of the whole colony, it is difficult to understand the overall branching pattern of the colony. May (1899) rewrote the description of Wright & Studer only with respect to spiculation as if the colony aspect did not matter for its classification. Versluys (1906) suggested that this species was similar in spiculation and branching pattern to the species *Th. moseleyi*, especially because of the high position of the operculum in relation to the marginal scales which do not have prominent thorns but a short spine. Versluys observed eight abaxial longitudinal rows of scales distally on the polyp whereas in the basal part the rows were not so well defined. Wright & Studer described the polyps as large but did not give any measurements. The polyps of Versluys' specimens were 2 to 2.5 mm long.

Kükenthal (1919, 1924) defined the branching pattern of this species as being partly bottlebrush and partly biseriate and each abaxial longitudinal row had 8 scales. He placed this species into the subgenus *Parathourella* on the grounds of the polyps being singly placed on the branches and the marginal scales being leaf-shaped and having a short thorn. Examining the type specimens, it was possible to observe that on the versal side of the colony the branches are mostly broken, contributing to the biseriate aspect of the colony. There are many well developed side stems and the polyps are as club-shaped as the polyps of the species of the subgenus *Amphilaphis*.

SPECIES 7 - *Thouarella longispinosa*

Thouarella longispinosa Kükenthal, 1912

Thouarella longispinosa Kükenthal, 1912:299-301, pl. 20, fig. 1. - Gravier, 1914:61-63, pl. 7, fig. 35-36. - Thomson & Rennet, 1931:24-26, pl.9, figs 1-3; pl. 12, fig. 1.

Thouarella (Euthouarella) longispinosa Kükenthal, 1915:149. - Kükenthal, 1919:415. - Kükenthal, 1924:292. - Broch, 1965:26-27, pl. 3, figs 8-10.

Thouarella) longispinosa Utinomi, 1964:11-12, text fig. 6; pl. 2, fig. 6-7.

TYPE SPECIMEN & TYPE LOCALITY. - *Thouarella longispinosa* Kükenthal, 1912. Gauss-Station (Antarctica), Deutsche Südpolar-Expedition, 385 m, 20/10/1902. Present location: Z.M.B. 5473, Museum für Naturkunde, Humboldt-Universität, Berlin, Germany.

DIAGNOSIS. - Bottlebrush form giving off simple branches. Polyps in pairs or in whorls of 3 to 4; five whorls on 10 mm of branch. Polyp scales placed in fewer than eight longitudinal rows proximally; only four scales on an abaxial row. Marginal scales bearing a median thorn.

DESCRIPTION. - Bottlebrush colony, apparently biserially arranged. Simple branches arise on all sides of the main stem; branches from one side of the colony are strongly bent towards the other, creating a frontal and versal side. Branches 22 mm long, shorter distally on the colony. Polyps placed in whorls of 3 or 4, occasionally in pairs; singly distributed on the main stem; 5 whorls in 1 cm of branch. Polyps bell-shaped, 1.5 mm long, slightly bent adaxially. Polyp scales placed in fewer than eight longitudinal rows proximally; only four scales on an abaxial row. Opercular scales triangular and pointed; 0.32 mm long. Marginal scales long, bearing a pronounced long thorn and a strong median keel on their inner surface; up to 0.7 mm long. Body scales with rounded margins; the basal scales convex distally and jagged proximally; 0.45 mm wide; adaxial scales smaller. Coenenchyme scales flat, roughly plate-like; 0.25 mm in diameter.

REMARKS. - The biseriate arrangement of the branches was probably caused by the presence of an annelid polychaete inhabiting the colony. Perhaps in the absence of the worm the colony would have a true bottlebrush form. The main stem is twisted along its axis giving the biseriate colony a bottlebrush appearance. If untwisted, it is biseriate. In short branches, the polyps are placed in pairs whereas in longer branches they are placed in whorls of three or four. The marginal scales are markedly thorny, sometimes only four scales are thorny, especially the two lateral and abaxial scales, whilst the others are less pronounced. The figured specimen for the original description of Kükenthal (1912) is a small specimen (57 mm long). The branches are long when compared with the size of the colony, although its length (22 mm long) is just below average when compared to other species. Kükenthal (1912) suggested that *Th. longispinosa* was similar to *Th. laxa* and *Th. tydemani* because of their similarly long marginal thorn. *Th. laxa* has, however, polyps distributed preferentially in pairs whereas the polyps in *Th. longispinosa* are mostly in whorls of 3 or 4. *Th. tydemani* has the branches arising on two sides of the main stem and the number of abaxial scales is higher than in *Th. longispinosa*. Kükenthal named this species *longispinosa* because of its long marginal thorns.

The specimen studied by Gravier (1914) was also small and fragile, but it was older than Kükenthal's specimen, being 105 mm long and had longer branches, 30 mm long. This colony has a less clear bottlebrush appearance because the branches are more sparsely distributed around the main stem.

Thomson and Rennet (1931) examined several specimens that could be referred to this species and described two distinct types of branching pattern: 1 - loosely branched form bearing short simple branches (30 mm long) on all sides of the main stem. There are 2-4 polyps in a whorl. 2 - "While the branches arise on all sides of the axis, there has been a compression, so that a bilateral colony has resulted. In consequence there is a superficial differentiation of what might be called, as in pennatulids, the dorsal and ventral surfaces; and another consequence is that the branches come to lie in two or three parallel planes"; average length of the branch is 40 mm and there are 4-6 polyps in a whorl. Thomson & Rennet described the marginal scales as being 1.6 mm long. Thomson & Rennet suggested that *Th. longispinosa* is markedly similar to *Stenella acanthina* concerning details in spiculation, distribution of polyps, number of scales abaxially and branching pattern. What identifies them apart is the difference in the position of the polyps towards the branches and the polyps of *S. acanthina* being more rigid than the polyps in *Th. longispinosa*.

Utinomi (1964) studied a very young colony, whose main stem gives off a few branches. The polyps are not placed in whorls but singly or in pairs. The spiculation described by this author agrees more with *Th. variabilis* than with *Th. longispinosa*. His specimen probably belongs to another species other than to *Th. longispinosa*.

The specimens studied by Broch (1965) agreed in most aspects with the original description. They were higher than Kükenthal specimens (150 mm long) and had longer branches (30 to 40 mm long). The whorls consisted mostly of 4 polyps and seldom of 3, exceptionally 5. In one specimen, 6 whorls were found per cm against the 5 described in the original description. The marginal scales are flattened, whilst in the original description they are convex, and do not have such long thorns.

SPECIES 8 - ***Thouarella hilgendorfi***

Thouarella hilgendorfi (Studer, 1878)

Plumarella hilgendorfi Studer, 1878:648-649, pl. 2, fig. 15 a-e.

Thouarella hilgendorfi: Wright & Studer, 1889:62, pl. 21, fig. 4. - Versluys, 1906:24-29, text fig. 17-25, pl. 1, fig. 4; pl. 2, fig. 7. - Kükenthal, 1907:206. - Kinoshita, 1908:21- 22, pl.5, fig. 42. - Nutting, 1912:66-67. - Thomson, 1927:33-34, pl. 1, fig. 23; pl. 6, figs 4,5.

Thouarella (Euthourella) hilgendorfi: Kükenthal, 1915:150. - Kükenthal, 1919:415. - Kükenthal, 1924:293.

TYPE SPECIMEN AND TYPE LOCALITY. - *Thouarella hilgendorfi* Studer, 1878. St. 192, Challenger Expedition, off the Ki Islands, South Papua, 140 fathoms, 26/09/1874. Present location: M.B. 89.5.27.40, N.H.M. London, UK.

DIAGNOSIS. - Bottlebrush form giving off simple branches. Polyps mostly in pairs, occasionally singly placed or in whorls. Six whorls on 10 mm of branch. Polyp scales placed in fewer than eight longitudinal rows proximally; six to eight sclerites on an abaxial row. Marginal scales bearing a long median thorn.

DESCRIPTION. - Young colony bottlebrush, partly biseriate; old colony fan-shaped with main stem giving off at its lower portion side stems, sometimes in almost opposite pairs, disposed in obtuse angles in relation to the main stem; from the main and side stems, fine branches arise in three directions at almost right angles. Main stem horny, calcareous, rigid, oval in transverse section and longitudinally grooved. Branches simple, rarely divided, 30 to 40 mm long. Polyps club-shaped, 0.9 mm long, absent on the main stem; singly placed on the branches, although seemingly in pairs; occasionally in whorls. Polyp scales disposed in six to eight transverse rows and five longitudinal rows. Opercular scales eight, pointed, lancet-shaped and toothed on their free margin; 0.38 mm long. Marginal scales with strong projecting thorn extending beyond the opercular scales. Submarginal scales with convex upper margin, bearing a rough spine. Lower body scales nearly round, 0.2 mm in diameter, with flat and serrated margins; with a few blunt warts. Coenenchyme scales elongated, oval, three cornered or four-cornered plates and overlapping each other.

REMARKS - One of the specimens described by Wright and Studer (1889) bifurcated dichotomously into two equal side stems, already 2 mm above the holdfast. The side stems appear always to lie in one plane. The main stem is oval in transverse section and its longer axis is parallel to the direction of the side stems. The drawings of Studer (1878) show a pinnate arrangement of branches. Together with Wright (1889), he described the branches arising from three sides of the main stem nearly at right angles. The latter description also contradicts the original description concerning the distribution of polyps. In the original description, the polyps are shown in pairs and in Wright & Studer's description they are singly placed, although Wright and Studer claimed to have had a better examination. The polyps are placed in spirals of three around the branch giving the impression of being opposite.

Versluys (1906) examined a considerably old colony (500 mm high) which had several side stems disposed in one plane, with side stems of their own. The branches (55 mm long) are distributed as in the original description, in a biseriate arrangement. The branches on the young stems are less densely distributed than on the old stems. Versluys contested the arrangement of polyps described by Wright & Studer and agreed with the original description by Studer (1878) where the polyps are placed in pairs or in the case of his specimens also in whorls of three, and seldom singly, with 6 whorls in 10 mm of branch. He also observed polyps on the stems. Kükenthal (1907) observed branches on all sides of the stem but in the majority they were biserially distributed with polyps placed in pairs and in one plane. He also observed single polyps on the stem. Kinoshita (1908) observed up to 4 polyps on a whorl. Nutting (1912) agrees, in his description, with Wright & Studer (1889) on the arrangement of branches arising on all sides of the stem. He described the distribution of polyps as being in whorls or in short spirals of three and 18 whorls in 2 cm of branch. Like Versluys (1906), Nutting (1912) also noted the slenderness of the polyps and the prominent marginal spines. Kükenthal (1924) redescribed the distribution of polyps as being in pairs or in whorls of three instead of in pairs or singly as he had described in 1907. Kükenthal (loc. cit.) observed 6 scales on a longitudinal row whilst Thomson (1927) observed 8. One of the specimens that Thomson studied had the polyps placed singly and another specimen had the polyps in pairs. On personally examining the colony studied by Wright & Studer (1889), which is held at the Natural History Museum, London, it was possible to verify that the branches are disposed in all directions in some portions of the colony and attain a biseriate arrangement in other portions. The colony has a fragile appearance with the polyps looking small in relation to the thickness of the axes and stems. The marginal and opercular scales do not differ much in appearance and the polyps are singly placed on the stem. There is a holotype of the same species at the Naturkunde Museum in Berlin, probably mistakenly classified as such.

SPECIES 9 - *Thouarella abeis*

Thouarella abeis Broch, 1965

Thouarella (Euthouarella) abeis Broch, 1965:29-30, pl. 5, figs 14-16; pl. 7, fig. 22.

TYPE SPECIMEN & TYPE LOCALITY. - *Thouarella abeis* Broch, 1965. Coronation Island, South Orkney, 11/11/1959, 170 - 150 fathoms. Present location: B 970, Zool. Museum, Oslo, Norway.

DIAGNOSIS. - Colony mostly bottlebrush, partly biseriate. Branches simple. Polyps in whorls of 3 to 5; 7 to 8 whorls on 10 mm of branch. Polyp scales placed in fewer than eight longitudinal rows proximally; 5 to 6 scales on each abaxial row. Marginal scales bearing a median thorn.

DESCRIPTION. - Colony form bottlebrush with the main stem giving off branches on all sides; middle branches broken and deprived of coenenchyme; one side of the colony is flattened, with the branches being in a biseriate arrangement, specially distally. Branches, simple, 35 mm long, upwardly oriented in the proximal part of the colony and at nearly right angles in the middle. Polyps cylindrical, without a narrow middle portion, 1 mm long, incurved adaxially and placed in whorls of 3 to 5, occasionally 6; there are 7 to 8 whorls on 10 mm of branch; polyps present on the stem only distally, where they are scattered distributed. Longitudinal rows of polyp scales fewer than eight; five to six scales on each abaxial row. Opercular scales covered by marginal scales. Marginal scales with a prominent, straight and acute thorn; distal part of the scale smooth or radially striped and free margins serrated; proximal part with warts and indented margin. Submarginal scales have their distal portion tapering into a short spine. Lower body scales are more rounded, adorned with radial ridges distally and with warts and spines proximally.

REMARKS. - Broch (1965) noted that the middle portion of the colony was probably damaged as a result of predation. If this is so, this is the first record of predation in *Thouarella.* Broch did not hesitate in placing this species in Kükenthal's subgenus *Euthouarella* being positioned between *Th. hilgendorfi* and *Th. typica.*

SPECIES 10 - *Thouarella typica*

Thouarella typica Kinoshita, 1907

Thouarella typica Kinoshita, 1907:230-231. - Kinoshita, 1908:23-24, pl. 2, fig. 9, 10; pl. 5, fig. 43. - Nutting, 1912:68.

Thouarella (Euthouarella) typica Kükenthal, 1915:150. - Kükenthal, 1919:417. - Kükenthal, 1924:293.

TYPE SPECIMEN & TYPE LOCALITY. - *Thouarella typica* Kinoshita, 1907. West coast of Satsuma, Japan. Present location unknown.

DIAGNOSIS. - Branches on all side, but attaining a biseriate arrangement. Polyps in pairs or in whorls of 3 to 4. Ten to eleven whorls on 10 mm of branch. Polyp scales placed in fewer than eight longitudinal rows proximally; five to six sclerites on an abaxial row. Marginal scales bearing a median thorn.

DESCRIPTION. - Fan-shaped colony. Side stems arising in acute angles from a cylindrical main stem and dividing dichotomously into substems. Main, side and sub stems giving off slender, flexible and simple branches, mostly 25 mm long, rarely 35 mm. Branches arise on all side of the stems but bent towards two sides, disposing biserially. Polyps in pairs or in whorls of 3, rarely 4, exceptionally isolated; 10 to 11 whorls on 10 mm of branch. Polyps 1 mm long, adaxially incurved; adaxial side shorter than abaxial side. Polyp scales placed in 5 to 6 longitudinal abaxial rows; 3 to 4 scales on each adaxial row. Opercular scales alternating smaller with larger; outer surface slightly concave and bearing only a rudimental keel. Marginal scales bearing a well developed thorn. Body scales rounded or oval, with serrated free margin. Coenenchyme scales of the branches slender and similar to the body scales.

REMARKS. - Kinoshita suggested that this species is most closely related to *Th. hilgendorfi.*

SPECIES 11 - *Thouarella moseleyi*

Thouarella moseleyi Wright & Studer, 1889

Thouarella moseleyi Wright & Studer, 1889:61-62, pl. 14, figs 1, 1a; pl.21, fig. 2. - Versluys, 1906:29-30, pl. 1, fig. 6; text fig. 26, 27. - Thomson & Dean, 1931:209.

Thouarella moseleyi var. *spicata* Thomson & Henderson, 1906:42-43, pl.3, figs 2, 4.

Thouarella (Euthouarella) moseleyi Kükenthal, 1915:150. - Kükenthal, 1919:417. - Kükenthal, 1924:294.

TYPE SPECIMEN AND TYPE LOCALITY. - *Thouarella moseleyi* Wright & Studer, 1889. St. 171, Challenger Expedition, off the Kermadec Islands, 600 fathoms, hard ground, 15/07/1874. B.M. 1889.5.27.39, N.H.M. London, UK.

DIAGNOSIS. - Colony branched in one plane. Polyps in pairs; five pairs on 10 mm of branch. Polyp scales placed in fewer than eight longitudinal rows proximally. Marginal scales with a median thorn.

DESCRIPTION. - Biserially arranged colony, with branches coming off from the main stem alternating in two directions. Main stem thin, slightly calcareous, flexible and somewhat flattened. Branches thin, flexible, and mostly simple, seldom bearing lateral branchlets; 0.5 to 0.6 mm thick; 15 to 20 mm long. Side stem 35 mm long, with lateral branches of its own. Polyps club-shaped, 1.5 mm, directed outwards, homogeneously distributed, placed in pairs on the branches and at the bases singly in short spirals of three; irregularly distributed on the main stem. Polyps appear large when compared to the thickness of the branches. Scales disposed in five transverse rows and four longitudinal rows. Operculum cone-shaped. Opercular scales short, spear-shaped, with unequal lateral sides; grooved in the middle line, expanded in the base and finely toothed in the lower portion; 0.38 mm high/ 0.20 mm wide. Body scales with a convex upper edge, slightly toothed, ornamented with radially arranged spines and a middle tooth and two lateral teeth, especially developed on the marginal scales; 0.23 mm high/0.27 mm wide. Adaxial scales small and covered by the lateral scales. Coenenchyme with two layers of scales, the upper layer formed of large flat four- or five-sided scales (0.36 mm high/0.15 mm wide) and the lower layer of small triangular or polygonal scales (0.12 mm high/0.10 mm wide).

REMARKS. - Wright & Studer (1889) considered the species *Th. moseleyi* as the transitional form between the genera *Stenella* and *Thouarella* for sharing characters with the species of both genera. These authors suggested that the characteristic pattern of the genus *Thouarella,* where the branches form a spiral coming off in three directions, was retained in this species even having an apparent biseriate arrangement with branches in two directions. They studied the arrangement in detail and observed that "between two twigs, which are given off on two sides at different heights, one polyp is placed on the stem in such a way that the two twigs are arranged in a short spiral with the polyp; at a few points a short twig is developed in place of the polyp". In the specimen studied by Versluys (1906) there are 5 pairs of polyps in 10 mm of branch and the polyps are described as having a cylindrical form. Based on the variation of some characters Thomson & Henderson (1906) proposed the variety *Th. moseley* var. *spicata.* This variety has a markedly ridged, cylindrical and rigid main stem; the branches do not strictly alternate; polyps are only in pairs, never in short spirals; the operculum forms a high cone; and the marginal scales have strong spines, which may be bifid. These authors observed that the branches distributed laterally, bent more to one side, as described by Wright & Studer, were induced partly by the presence of a polychaete worm on one side of the stem.

Examining the specimen, it was possible to observe that the colony is small and fragile. There were two small polychaetes inhabiting the colony and the branches had grown round them, starting to form their tube. It appears that the marginal scales have short thorns but to confirm this a better examination should be undertaken. The variety *Th. m. spicata* is very similar to the typical species. It differs from the latter by being more thorny. Many branches are broken and carefully examining them it is possible to see that they are in fact alternating. This variety should, therefore, be reassessed.

SPECIES 12 - *Thouarella laxa*

Thouarella laxa Versluys, 1906

Thouarella laxa Versluys, 1906:30-32, pl. 1, fig. 5; pl. 3, fig. 8.; text figs 28-32. - Kükenthal & Gorzawsky, 1908:36, pl. 2, fig. 13.

Thouarella (Euthouarella) laxa Kükenthal, 1915:150. - Kükenthal, 1919:417. - Kükenthal, 1924:293-294, text fig. 164.

TYPE SPECIMEN AND TYPE LOCALITY. - *Thouarella laxa* Versluys, 1906. St. 88, Siboga Expedition, 0°34'6N, 119°8'.5E, Makassar-Strasse, 1301 m. Present location unknown, possibly at the Institute for Taxonomic Zoology, Amsterdam, The Netherlands.

DIAGNOSIS. - Colony branched apparently in one plane. Polyps in pairs; four pairs on 10 mm of branch. Polyp scales placed in fewer than eight longitudinal rows proximally. Marginal scales bearing long thorns; four of the eight opercular scales considerably larger and distinct from the remaining.

DESCRIPTION. - Branches disposed pinnately distally on the main stem and arising on all sides from the older or lower portion of the stem, although bent to the laterals being apparently biserially arranged. Branch extremities bent towards one side of the colony, forming a frontal and versal side of the colony. Branches sparsely distributed on the main stem; mostly 30 mm long, occasionally 40 or even 50 mm. Polyps placed in pairs on the branches and irregularly scattered on the main and side stems; 4 pairs per 10 mm of branch; Polyps densely distributed proximally on the branches, where a worm path is formed. These polyps are smaller than normal. Polyp size (normal) 1.2 to 1.5 mm long. Scales placed on longitudinal rows, fewer than eight proximally where they overlap one another; five scales on each abaxial row. Low operculum enclosed by the marginal thorns. Opercular scales of varied size, four inner smaller scales alternate with four outer larger scales; outer opercular scales relatively strong, with concave outer surface and supported by an irregular enlarged distal margin; 0.36 mm long. Marginal scales disposed in two rings, an outer and an inner ring, and bearing long thorns; 0.75 mm long, where half the length is the thorn. Submarginal scales bearing a short thorn. Lower body scales 0.36 mm in diameter; outer surface roughly flat, inner surface ornamented with small warts. Coenenchyme scales on the branches slightly rounded, with larger but less numerous warts than the polyp scales; average diameter: 0.3 mm, maximum diameter: 0.4 mm; On the main and side stems coenenchyme scales are smaller, 0.2 mm in diameter, occasionally 0.3 mm in diameter or larger.

REMARKS. - Versluys (1906) studied an old specimen (400 mm high) which had the main stem highly ramified into secondary stems and the colony attained a fan-shaped form. Each stem gave off numerous single branches. He also described a young specimen which had a fragile appearance and a main stem surrounded by single branches. Worms inhabit both sides of the colony and affect the density and arrangement of branches forming their path. The scales are similar to the scales of *Th. hilgendorfi* although larger. Versluys observed on a damaged polyp, where a scale is missing, marked small and delicate plate-like scales. Perhaps, these scales were involved in a process of regeneration of the damaged portion

rather than normally existing as an underlying layer. The specimen studied by Kükenthal & Gorzawsky (1908), which they referred to this species, had the polyps not only placed in pairs but also in whorls of three and exceptionally four. The polyps were more densely distributed on the branch distally than proximally, differing from the original description. The presence of worm tubes or paths were not, however, mentioned by the latter authors, which might possibly explain the difference in distribution of polyps.

SPECIES 13 - *Thouarella tenuisquamis*

Thouarella tenuisquamis (Kükenthal, 1907)

Thouarella regularis Kükenthal, 1907:206-207.

Thouarella tenuisquamis: Kükenthal, 1908:11.

Thouarella (Euthouarella) tenuisquamis: Kükenthal, 1915:151. - Kükenthal, 1919:421, pl. 42, fig. 65. - Kükenthal, 1924:295.

TYPE SPECIMEN & TYPE LOCALITY. - *Thouarella tenuisquamis* Kükenthal, 1907. St. 210, Deutschen Tiefsee-Expedition, Gross-Nicobar, 4°53'1S, 93°33'5E, 752 m. Present location: Z.M.B. 6087, Museum für Naturkunde, Humboldt-Universität, Berlin, Germany.

DIAGNOSIS. - Colony branched in one plane. Polyps in pairs; seven to eight pairs on 10 mm of branch. Polyp scales placed in fewer than eight longitudinal rows proximally. Marginal scales not bearing a prominent thorn.

DESCRIPTION. - Pinnate colony giving off branches on two sides oriented at almost right angles. Branches bent towards one side of the colony creating a frontal and versal side on the colony. Branches simple, 18 mm long and sparsely distributed. Polyps 1.2 mm long, placed in pairs, regularly biserially distributed; directed outwards in relation to the axis, rarely adaxially incurved; 7-8 pairs per 10 mm of branch; polyps more densely distributed proximally on the branch. Polyp scales placed in fewer than eight longitudinal rows proximally; six scales on each abaxial longitudinal row; 0.25 mm wide. Polyp scales delicate, with serrated flattened margins. Marginal scales fragile, with a pointed distal portion, a narrow keel and triangular outline; 0.26 mm wide/0.55 mm long. Opercular scales small, notably fragile, bearing ridges projecting as a spine supported by a keel. Upper coenenchyme scales similar to the polyp scales; 0.25 wide; Lower scales plate-like and 0.15 mm in diameter.

REMARKS. - At the lower part of the colony, some branches are bent to one side and some to the other forming a polychaete tube on each side. There is only one worm inhabiting one of the tubes. At the middle and upper portion of the colony, the branches are bent only to the dorsal side, where the formation of a polychaete tube is evident. Perhaps, in the absence of the worm, the colony would have a more recognizable biseriate arrangement. At the middle of the colony there is a small side stem and one branch subdividing dichotomously.

The disturbance caused by the worm is not only seen in the branching pattern, but also in the distribution and number of polyps. As mentioned in the original description, the polyps are more closely spaced proximally at the branches. This is observed on branches which are forming the path of the worm. It is as if the polyps padded the way of the worm, where they are as aggregated as they can get. Evidence of the influence of the worm can be seen in areas where there is no tube formation. In these areas, the polyps are sparsely distributed like in the distal portion of most branches. The sclerites of the polyps on the tube are more thorny and these polyps are often in reproductive stage.

Kükenthal (1907) introduced the species *Thouarella regularis* to accommodate a specimen that in 1908 he transferred to *Th. tenuisquamis* because he had decided to incorporate the genus *Amphilaphis* into *Thouarella* and a species named *Amphilaphis regularis* already existed. This author suggested that this species was most closely related to *Th. laxa* and *Th. tydemani*. The main character used by Kükenthal to discern this species from the latter two was the distribution of polyps, 7-8 pairs of polyps per 10 mm of branch. The distribution of polyps, as mentioned above, was caused by the presence of a polychaete commensal, and therefore, this character should be evaluated with caution.

SPECIES 14 - *Thouarella carinata*

Thouarella carinata Kükenthal, 1908

Thouarella carinata Kükenthal, 1908:11, pl. 1, fig. 1.
Thouarella (Euthouarella) Carinata Kükenthal, 1915:150. - Kükenthal, 1919:423, pl. 42, fig. 66. - Kükenthal, 1924:296.

TYPE SPECIMEN & TYPE LOCALITY. - *Thouarella carinata* Kükenthal, 1907. Japan, Okinose and Urugakanal, 400 fathoms. Present location: Z.M.B. 6078, Museum für Naturkunde, Humboldt-Universität, Berlin, Germany.

DIAGNOSIS. - Colony branched in one plane. Polyps in whorls of three; seven to eight whorls on 10 mm of branch. Polyp scales placed in fewer than eight longitudinal rows proximally. Marginal scales lancet-shaped bearing a prominent median thorn.

DESCRIPTION. - Fan-shaped colony, with main stem giving off long side stems only on one side of the colony; branches arise in one plane, oriented upwards in an acute angle in relation to the main and side stems. Branches mostly simple, 18 mm long, bent towards one side of the colony forming a frontal and versal side on the colony. Polyps, 1.3 mm long and placed in whorls of three, occasionally in pairs; 7-8 whorls per 10 mm of branch; on the stem the polyps are sometimes packed together. Polyp scales placed in fewer than eight longitudinal rows proximally; six scales on each abaxial longitudinal row six; adaxially in lesser numbers. Opercular scales triangular, rounded distally; alternating 4 larger, 0.36 mm high, with 4 smaller, 0.18 mm high. Marginal scales long, lancet-shaped bearing a thorn, which is supported by a strong keel on the inner side; 0.65 mm high. Body scales 0.36 mm in diameter. Coenenchyme scales of the branches similar to the polyp scales; scales on the stem are smaller, mostly plate-like, 0.12 mm in diameter.

REMARKS. - The main stem of the type specimen is twisted and for this reason, in some parts of the colony, the branches seem to arise in three different directions. Most branches are, however, disposed in one plane. A small polychaete inhabited the colony. A few branches are curved on it as if they were to start a new tube. Kükenthal (1908) suggested that *Th. carinata* is closest related to *Th. typica* concerning the short branches and the position of the polyps on the branch.

SPECIES 15 - *Thouarella flabellata*

Thouarella flabellata Kükenthal, 1907

Thouarella flabellata Kükenthal, 1907:207-208.
Thouarella (Euthouarella) flabellata Kükenthal, 1915:150. - Kükenthal, 1919:418, pl.42, fig. 64. - Kükenthal, 1924:294-295.

TYPE SPECIMEN AND TYPE LOCALITY. - *Thouarella flabellata* Kükenthal, 1907. St. 25, Deutschen Tiefsee-Expedition, East Africa cost, 1°48.2'S, 45°42.5'E, 1644 m. Present location: Z.M.B. 6083, Museum für Naturkunde, Humboldt-Universität, Berlin, Germany.

DIAGNOSIS. - Colony branched in one plane. Polyps in pairs and in whorls of 3; five whorls on 10 mm of branch. Polyp scales placed in fewer than eight longitudinal rows proximally. Marginal scales with a median thorn.

DESCRIPTION. - Colony form roughly fan-shaped. Main stem strong, with side stems arising on both sides in an arrangement seemingly plane, although oriented in different angles. Side stems dichotomously divide into substems. Main stem, side stems and substems give off branches likewise, in one plane. Branches mostly simple, the longer lower branches occasionally branched into branchlets; average branch length 30 mm, occasionally 40 mm long. Polyps placed on the branches mostly in pairs, occasionally in whorls of three; pairs of polyps usually biserially disposed and sparsely distributed; 5 to 6 pairs on 10 mm of branch; on the stems polyps are disorderly distributed. Polyp in average 1 mm long, slender and pear-shaped; directed outwards and slightly upwards, not bent towards the branch. Each longitudinal abaxial row with five scales and adaxial with four. Opercular scales eight: four larger, pointed and bearing thorny projections on the margins and a middle keel; and four smaller, thin, triangular, concave on their outer surface and rounded distally. Marginal scales broad, wing-like and triangular, with a short blunt thorn and bearing a strong keel and middle groove. Body scales 0.3 mm high and 0.36 mm wide, semicircle-shaped or flat with smooth free margins, serrated overlapping edges and surface ornamented by fine radial ridges. Coenenchyme scales from the branches 0.3 mm in diameter and irregular in shape. Coenenchyme scales from the stems 0.1 mm in diameter, plate-like with marked serrated margins.

REMARKS. - The type specimen described by Kükenthal (1907) appears to be an old colony. Its distal part is wanting and the existing lower part is markedly ramified into side stems, which are divided into substems. The main stem and side stems are thick and strong when compared to the branches. The colony has a markedly large and strong calcareous holdfast, which spread over a polychaete tube. The long lower branches subdivided into branchlets, as mentioned by Kükenthal, look more like a side stem starting to develop since it has a racemose or monopodial branching pattern. As the branches are arranged at different angles, a frontal and versal side as seen in other fan-shaped species is not easily recognized in this species, or at least in this specimen. However, some of the side stems have their branches bent towards one direction, probably caused by environmental conditions. Younger colonies, with a few side branches, would possibly have a more recognizable fan-shaped form and very young colonies, without any side branches would possibly attain a regular pinnate form. The type specimen is highly colonized by epibionts such as solitary scleractinians, ophiuroids and polychaetes. The marginal scales do not appear notably thorny, perhaps because of their

triangular shape which soften the thorny appearance. Kükenthal (1907) found *Th. flabellata* closest related to *Th. tydemani* concerning colony form and as well the form of the polyp and variety of scales. This species is also similar to *Th. moseleyi.*

SPECIES 16 - *Thouarella tydemany*

Thouarella tydemany Gray, 1870

Hookerella pulchella Gray, 1870:45 (Dr. Hooker's drawing nº 255). (?)
Thouarella tydemany: Versluys, 1906:32-35, pl. 1, fig. 2.
Thouarella (Euthouarella) tydemany: Kükenthal, 1915:150. - Kükenthal, 1919:420. - Kükenthal, 1924:295.

TYPE SPECIMEN & TYPE LOCALITY - *Thouarella tydemany* Versluys, 1906. St. 297, Siboga Expedition, 10°39'S, 123°40'E, East of Rotti, 520 m. Present location unknown, possibly at the Institute for Taxonomic Zoology, Amsterdam, The Netherlands.

DIAGNOSIS. - Fan-shaped colony. Polyps in pairs and in whorls of 3; six whorls on 10 mm of branch. Polyp scales placed in fewer than eight longitudinal rows proximally. Marginal scales with a median thorn.

DESCRIPTION. - Colony branched in one plane, with numerous side stems disposed also in one plane. Branches simple, arising from the stems in two directions attaining a pinnate arrangement. Branches mostly bent towards the frontal side of the colony. Side stems divide dichotomously into substems, also bearing side branches biserially arranged. Branch length 15 - 20 mm, occasionally 30 mm; branches of substems shorter, 10 mm long. Polyps bell-shaped, 1.5 mm long, placed on the branches in pairs or in whorls of three; 6 pairs per 10 mm of branch; on the stems the polyps are placed irregularly somehow biserially between the bases of the branches. Each longitudinal abaxial row with five scales. Marginal scales large, bearing long thorns, 0.45 mm long, occasionally 0.5 mm. Opercular scales eight: 4 small convex inner scales bearing a blunt keel distally and 4 larger concave outer scales broad distally; outer scales have larger warts than inner scales; scales vary in size, a large scale being 0.36 mm long; width 2/3 of the length. Coenenchyme scales thick and overlapping; on

the base of the stem the coenenchyme scales are thicker, plate-like, with irregular outline ornamented with high elevations and ridges on their outer surface and numerous warts on their inner surface; 0.35 mm in diameter, occasionally much larger, 0.70 mm in diameter.

REMARKS. - The specimen studied by Versluys (1906) was not very high (200 mm) and already gave off so many side stems, although from the figure presented by Versluys it seems that part of the colony is lacking. Existing worm paths affect the branching pattern of the colony. Versluys considered the polyps of this species similar to the polyps of *Th. laxa*, to which they differ by being broader, more robust and for having longer marginal thorns and varied opercular scales. Versluys suggested that *Th. tydemany* is very similar to *Hookerella pulchella* described by Gray (1870). Gray's description is very brief and inaccurate, and it lacks reliable illustration. Examining the type specimen of *H. pulchella*, Versluys confirmed that the colony form was identical to *Th. tydemani*, however, he did not examine the polyps. In conclusion, on the possibility of these two species being the same and in the absence of a reliable description for *H. pulchella,* Versluys introduced the species *Th. tydemani* and invalidated Gray's species. It seems, from Versluys statement, that Wright & Studer also had doubts about Gray's species.

SPECIES 17 - *Thouarella coronata*

Thouarella coronata Kinoshita, 1908

Thouarella (Diplocalyptra) coronata Kinoshita, 1908:56-59, text fig. 4-6.
Thouarella coronata Kükenthal, 1915:151.
Thouarella (Euthouarella) coronata Kükenthal, 1919:425. - Kükenthal, 1924:296.

TYPE SPECIMEN & TYPE LOCALITY. - *Thouarella (Diplocalyptra) coronata* Kinoshita, 1908. Insel Udsi, Provinz Satsuma, 80 fathoms. Present location unknown.

DIAGNOSIS. - Fan-shaped colony, with the main stem subdivided distally. Polyps in pairs or whorls of three. Polyp scales placed in fewer than eight longitudinal rows proximally. Marginal scales bearing a long median thorn.

DESCRIPTION. - Fan-shaped colony with a cymose branching pattern, where the main stem dichotomously subdivides into simple branches, disposed in one plane. End branches mostly 20-25 mm long, but can reach up to 50 mm long. Main stem flattened. Polyps placed in pairs or whorls of three, rarely four, on the slender branches and on the thick main stem; when in pairs, they are biserially disposed; about 17 to 20 polyps in 30 mm of branch. Polyps 1.0 to 1.4 mm long, rigid, incurved adaxially only on the extremities of the branches; adaxial side of the polyp shorter than the abaxial side. Longitudinal rows of body scales clearly defined distally but undefined proximally, where the polyp body is narrow. Polyp scales placed in fewer than eight longitudinal rows proximally; six scales on a longitudinal abaxial row, including the opercular scale. Opercular scales triangular, finely serrated, outer surface concave, inner surface ornamented with warts and radial ridges; varied in size; 0.20 to 0.32 mm high. Marginal scales triangular, leaf-shaped, with concave outer surface, and bearing a long thorn supported by a strong inner median keel; alternating in two rings of scales; 0.60 mm high/0.32 mm wide. Coenenchyme scales slender, rounded or elliptic, with warts on the outer surface; 0.3 mm, rarely 0.4 mm, in diameter.

REMARKS. - Kinoshita's (1908) description of *Th. coronata* is clear and detailed. However, he only illustrated the polyp and sclerites. Without any illustration of the branching pattern and arrangement of polyps, it is very hard to compare his species to the other species of this genus, specially because of its peculiar cymose branching form, which seems to be uncommon among the species of *Thouarella*. Most of the species of this genus have a racemose or monopodial branching pattern, where the main stem is continuous, bearing side branches or stems but not subdividing dichotomously distally. Kinoshita identified three different sizes of opercular scales, starting from a minute size. The last type he refers as inner marginal scales of opercular type. They were probably true opercular scales rather than marginal scales. Kükenthal (1915) did not adopt the subgenus *Diplocalyptra*, introduced by Kinoshita, as he believed that *Th. coronata* could be assigned to the subgenus *Euthouarella* instead, on the grounds of branching pattern, His description was, however, too brief and without any strong argument to support the removal of this species from the original subgenus. In a later publication, Kükenthal (1924) gave a better diagnosis of this species, although he omitted essential information about details of spiculation.

SPECIES 18 - *Thouarella crenelata*

Thouarella crenelata Kükenthal, 1907

Thouarella crenelata Kükenthal, 1907:205.

Thouarella (Epithouarella) crenelata Kükenthal, 1915:151. - Kükenthal, 1919:436, pl.43, fig. 70. - Kükenthal, 1924:300-301.

TYPE SPECIMEN & TYPE LOCALITY. - *Thouarella crenelata* Kükenthal, 1907. St. 131, Deutschen Tiefsee-Expedition, East Coast of Bouvet Island, 5°10'S, 23°2'E, 475 m. Present location: Z.M.B. 6081, Museum für Naturkunde, Humboldt-Universität, Berlin, Germany.

DIAGNOSIS. - Colony densely branched with simple branches biserially arranged. Polyps singly placed. Polyp scales placed in fewer than eight longitudinal rows proximally; nine to ten scales on each abaxial row. Marginal scales not thorny; free edges of all scales strongly serrated.

DESCRIPTION. - Colony form biseriate, with the main stem giving off branches oriented at angles of 60° to 90°. Branches, 20 mm long, mostly simple, occasionally ramified in the lower part of the colony; the branches are bent to one side of the colony forming a frontal and versal side, especially in the lower part of the colony. Polyps club-shaped, directed upwards and slightly inwards, 2 mm long, singly placed, closely spaced and arising all around the branches; 10-12 polyps on 10 mm of branch; absent from the main stem. Polyp scales placed in fewer than eight longitudinal rows proximally; nine to ten scales on each abaxial row; nine adaxially. Opercular scales leaf-shaped, strongly serrated, with the outer surface ornamented with fine radial ridges; half the size of the marginal scales. Marginal scales strongly serrated, without pronounced thorn or keel. Body scales with free margins strongly serrated; 0.35 mm wide/0.25 mm high. Coenenchyme scales of the axis plate-like or oval, with serrated margins, 0.15 mm in diameter. Scales of the main stem are similar although more sculptured with ridges.

REMARKS. - The type specimen is inhabited by a polychaete, and that is probably the reason for the biseriate arrangement of the branches. They are bent to the side where the worm tube is formed. In another colony of the same species the worm tube starts on one side of the colony and continues to the opposite. Part of the branches are, therefore, bent towards one side and part towards the opposite side, creating a confusing branching pattern. On the tube, the polyps are biserially arranged against the path of the worm. Based on this

observation, Kükenthal (1907) described the arrangement of the polyps preferentially biseriate. In a later paper (1924), he corrected this information by describing them as arising on all sides of the polyp, which in normal condition is the true information. Kükenthal (1907) suggested that this species was similar to *Th. koellikeri* in colony form, although the branches do not seem to arise on all sides of the main stem as in *Th. koellikeri* but preferentially on two sides and the marginal scales are less thorny in *Th. crenelata*.

SPECIES 19 - *Thouarella affinis*

Thouarella affinis Wright & Studer, 1889

Thouarella affinis Wright & Studer, 1889:66-68, pl. 21, fig.3. - Versluys, 1906:37.
Thouarella (Epithouarella) affinis Kükenthal, 1915:151. - Kükenthal, 1919:435. - Kükenthal, 1924:300.

TYPE SPECIMEN AND TYPE LOCATION - *Thouarella affinis* Wright & Studer, 1889. St. 135d, off Inaccessible Island, Tristan da Cunha, 15/07/1874, 55-70 fathoms. Present location: B.M. 1889.5.27.44, N.H.M. London, UK.

DIAGNOSIS. - Bottlebrush colony. Polyps sparsely distributed on the branches. Polyp scales placed in fewer than eight longitudinal rows proximally; seven scales on each longitudinal abaxial row. Marginal scales not bearing thorns, being simply toothed.

DESCRIPTION. - Colony form bottlebrush, with branches arising apparently on all sides of the main stem, mostly in three directions, at nearly right angles and in ascending spirals. Main stem hard, brittle and oval in transverse section; twisting of the main stem may reach 360°. Branches simple, occasionally bifid near the base and closely spaced, 1.5 to 2 mm distant from each other; up to 50 mm long. Polyps club-shaped, 2 mm long, singly placed on the branch, in short spirals of always three or four, but never in whorls or in pairs; densely distributed at the branch periphery; the very end of the branch is occupied by a polyp. Scales on each longitudinal abaxial row in number of seven. Opercular scales triangular, lancet-shaped, pointed, concave, with two lateral teeth; 0.4 mm long/0.33 mm wide; adaxial opercular scales smaller than the abaxial. Marginal scales higher than broad, 0.42 mm high/0.33 mm wide, with a long projecting tooth distally. Submarginal scales with middlemost

tooth lengthened; highest scales nearly lancet-shaped. Lower body scales large, symmetrical, broader than long, the upper margin convex, finely serrated and with large rough warts; adaxial scales smaller, polygonal, and overlapping one another. Coenenchyme thin and composed of a single layer of flat scales, irregularly three- or four-sided; 0.3 mm height/0.47 mm wide.

REMARKS. - Wright & Studer (1989) suggested that *Th. affinis* was very similar to *Th. antarctica*, differing only in size and sculpture of the polyps. The shape of marginal scales and the absence of long thorns is probably the major difference between the two species. Wright & Studer showed contradiction when describing the coenenchyme layer. At first they mentioned that there was a single layer of sclerites and at the end of the text they described two layers of coenenchyme scales. The description of these authors is rich in details but poor in illustration, containing only drawings of sclerites. It is difficult, therefore, to ascertain the arrangement of scales on the polyps, the distribution of polyps on the branches, and the colony form. Versluys (1906) did not sufficiently clarify whether his description was based on the type specimen or on other specimens. He did not recognize the same branching pattern mentioned in the original description, where the stem is twisted in a long spiral. Versluys observed eight longitudinal rows of scales in the distal portion of the polyp but proximally they were not clear. Kükenthal (1915) placed this species in the subgenus *Epithouarella* based on absence of marginal thorns.

SPECIES 20 - *Thouarella chilensis*

Thouarella chilensis Kükenthal, 1908

Thouarella chilensis Kükenthal, 1908:11. - Kükenthal,1912:302-304, pl. 21, fig. 5.
Thouarella (Epithouarella) chilensis Kükenthal, 1915:151. - Kükenthal, 1919:436. - Kükenthal, 1924:300.

TYPE SPECIMEN & TYPE LOCALITY. - *Thouarella chilensis* Kükenthal, 1912. Iquique (Chile), Deutsche Südpolar-Expedition. Present location: Z.M.B. 6079, Museum für Naturkunde, Humboldt-Universität, Berlin, Germany (only fragments of the holotype. Location of the colony unknown, perhaps Breslaw).

DIAGNOSIS. - Bottlebrush colony. Polyps singly placed and densely distributed at the extremities of the branches. Polyp scales placed in fewer than eight longitudinal rows proximally; seven to eight scales on each longitudinal abaxial row. Marginal scales not bearing thorns, being simply toothed.

DESCRIPTION. - Colony form densely bottlebrush, with main stem giving off one large side stem. Branches simple, arising on all sides of the main and side stems, 22 mm long and shorter at the base of the colony. Polyps singly placed, 2 mm long, study, regularly thick and only slightly bent adaxially; notably aggregated distally on the branches making the branches appear club-shaped; sparsely distributed proximally; also present on the lower part of the stems. Polyp scales placed in fewer than eight longitudinal rows proximally; seven to eight scales on each longitudinal abaxial row. All polyp scales with fine, delicate and serrated free margin; 0.42 mm wide/0.25 mm high. Opercular scales narrow and leaf-shaped; 0.4 mm high. Marginal scales triangular and pointed, without thorn but with a round blunt distal tooth, strongly dentated and sculptured with longitudinal ribs; 0.6 mm high. Coenenchyme scales of the axes larger than the scales on the stems, which have deeply serrated margins and are 0.15 mm in diameter.

REMARKS. - Compared to other species of *Thouarella*, *Th. chilensis* appears to have more aggregate polyps on the extremities of the branches. The branching pattern and the distribution of the branches give a regular aspect to the colony. The side stem appears to be larger than the main stem. The main stem is markedly thick, being able to withstand the large side branch. It looks more like a case of auto-epizoism, where a larva settles on the parent colony and develops a new colony, which resembles a side stem.

SPECIES 21 - *Thouarella regularis*

Thouarella regularis (Wright & Studer, 1889)

Amphilaphis regularis Wright & Studer, 1889:71-72, pl. 15, figs 1, 1a; pl. 21, fig. 7. - Versluys, 1906:22. - Thomson & Ritchie, 1906:854. - Nutting, 1908:573-574.

Thouarella regularis: Kükenthal, 1907:206-207.

Thouarella (Amphilaphis) regularis: Kükenthal, 1915:149. - Kükenthal, 1919:409. - Kükenthal, 1924:289.

TYPE SPECIMEN & TYPE LOCALITY. - *Amphilaphis regularis* Wright & Studer, 1889. St. 135a, Challenger Expedition, off Inaccessible Island, Tristan da Cunha, 75 fathoms; St. 135c, off Nightingale Island, 100-150 fathoms. SYNTYPE - B.M. 1889.6.27.60, N.H.M. London, UK. (SYNTYPE - 32.12.8.7.).

DIAGNOSIS. - Pinnate colony. Polyps singly placed, occasionally in pairs. Polyp scales placed in eight longitudinal rows. Marginal scales bearing a thorn.

DESCRIPTION. - Colony form pinnate, with main stem giving off several long side stems on two sides; main and side stems bearing branches biserially and arranged in parallel. Main stem flattened, hard, brittle, becoming flexible and thinner distally. Branches simple, 40 to 50 mm long, arising from the stems at angles of 45°. Polyps club-shaped, 1.5 to 2.0 mm long, singly placed on all sides of the branches in irregular ascending spirals, occasionally in pairs. Polyp scales placed in seven or eight transverse rows and arranged in eight longitudinal rows, overlapping one another proximally; 10 to 12 scales on a longitudinal abaxial row and 7 to 8 on each adaxial row. Adaxial scales different and thinner than abaxial scales. Opercular scales roughly triangular, with a median longitudinal furrow on the outer surface and a convex keel on the inner side; free margins toothed and ornamented with prominent spines; 0.46 mm high/0.25 mm wide; and form a blunt low cone when the operculum is closed. Marginal scales broad, with a toothed, convex upper margin and a median keel projecting into a long pointed or toothed spine; adorned with radially arranged ridges and ribs projecting beyond the edges; 0.37 mm high/0.33 mm wide. Submarginal scales broader than high, bearing a short median rib, running outwards from a nucleus, and with projecting spines; 0.26 mm high/0.33 mm wide. Lower body scales thick, covered with large, rough prominences running out into short spines resulting in a toothed free edge. Adaxial scales thin and rounded, without prominent sculpture. Upper coenenchyme scales irregular, polygonal or triangular, with marginal nucleus and free margin toothed, from where sharp, short spines extend; 0.27 mm high/0.20 mm wide. Lower coenenchyme scales small, rounded and thin, with a more central nucleus and weaker sculpture; 0.10 mm in diameter.

REMARKS. - The type specimen of *Thouarella regularis* has a true pinnate form, with only branches of first order being produced from the main and side stems. The rigid stems and axes keep the branches straight and regularly arranged. The branches are markedly long, decreasing in length distally. Wright & Studer (1889), when describing *Amphilaphis regularis* noted that growth takes place at the end of the branches and they suggested that the same happened in *Thouarella*. In the specimens examined in the present study it was observed that polyps arise preferentially in the subterminal portions and in areas close to where the

branches bifurcate into branchlets. In species with simple branches, with or without side stems, however, the an extension of the axis is probably terminal and the polyps arise where there is extension and free space available. It is probable that Wright & Studer referred to species of *Thouarella* which had simple branches. Another specimen studied by Wright & Studer had the branches coming off partly perpendicular to the main stem and partly in one plane as a result of the main and side stems being twisted in a long spiral. If untwisted the colony is all pinnate. Thomson & Ritchie (1906) commented upon the noticeable variation among the specimens that they referred to this species, especially concerning the number and arrangement of polyps. They also complained about the lack of coherence between the text and the figures of Wright & Studer, noting the confusion over the understanding of the details of the spiculation of the polyps. In fact, Wright & Studer's drawings of sclerites are very inaccurate and unreliable. Projecting middle ridges described by Wright & Studer for the polyp scales are not mentioned in Thomson & Ritchie. The latter authors described the scales as having pronounced ridges and projecting spines beyond the whole free edge of the scale rather than specifically in the middle.

Kükenthal (1907) had already introduced a species named *Thouarella regularis* to accommodate a specimen that in 1908 he transferred to *Th. tenuisquamis* (see species 13). In 1915, Kükenthal incorporated *Amphilaphis regularis* into *Thouarella*, then named *Thouarella regularis* and placed it into the subgenus *Amphilaphis*.

SPECIES 22 - *Thouarella parva*

Thouarella parva Kinoshita, 1908

Thouarella (Diplocalyptra) parva Kinoshita, 1908:53-56, text figs 1-3.
Thouarella (Amphilaphis) parva Kükenthal, 1915:149. - Kükenthal, 1919:410. - Kükenthal, 1924:290.

TYPE SPECIMEN & TYPE LOCALITY. - *Thouarella (Diplocalyptra) parva* Kinoshita, 1908. Insel Kodzu, South of the Provinz Idzu, Japan. Present location unknown.

DIAGNOSIS. - Fan-shaped colony. Polyps in pairs and biserially arranged. Polyp scales placed in eight longitudinal rows; five scales on each longitudinal abaxial row. Marginal scales without a thorn.

DESCRIPTION. - Colony in one plane, dichotomously branched, with the branches arising at an angle of 50°. Main stem fragile. Polyps in pairs disposed biserially, 6 polyps on 10 mm of branch. Polyps rigid, rarely adaxially incurved and 1 mm long. Eight longitudinal rows of polyp scales; 5 scales on each abaxial row. Opercular scales completely covered by the marginal scales. Opercular scales small and strong, concave on the outer surface, with serrated narrow margins and adorned by warts converging into ridges, which end as projecting teeth; 0.36 mm high/0.18 mm wide. Marginal scales longer than wide; 0.4 mm long. Body scales broad transversally, ornamented by warts and with free edges serrated; 0.38 mm wide. Coenenchyme scales slender, oval, flattened; 0.2 mm in diameter, with serrated margins and ornamented by warts.

REMARKS. - There is a certain doubt about what Kinoshita (1908) refers to as opercular and marginal scales. In his description, the marginal scales are long and covering the small opercular scales. It appears, however, that the specimen described by Kinoshita has two types of opercular scales, one small, triangular and narrow, and a second long scale, square distally (which he described as marginal). The marginal could be the rounded, toothed scales below the second type of opercular described. From Kinoshita's drawing, it is difficult to recognize longitudinal rows as described by Kükenthal. The colony form was badly described, therefore, any judgement about the taxonomic position of this species can not be made until the type specimen is examined.

SPECIES 23 - *Thouarella abietina*

Thouarella abietina (Studer, 1894)

Amphilaphis abietina Studer, 1894:65. - Menneking, 1905:255-260, pl. 8, figs 7,8; pl. 9, figs 17-20. - Versluys, 1906:22.

Thouarella (Amphilaphis) abietina: Kükenthal, 1915:149. - Kükenthal, 1919:410. - Kükenthal, 1924:290.

TYPE SPECIMEN & TYPE LOCALITY. - *Amphilaphis abietina* Studer, 1894. Pacific Ocean, 1°7'N, 81°4'E, 3182 m. Present location unknown.

DIAGNOSIS. - Fan-shaped colony. Polyps in pairs and sparsely distributed. Polyp scales placed in eight longitudinal rows; six scales on each longitudinal abaxial row. Marginal scales without thorns. High and pointed operculum.

DESCRIPTION. - Colony in one plane, with main stem giving off side stems at almost right angles. Branches arising from the main and side stems also at right angles and subdivided into branchlets. Polyps in pairs, 3-4 mm distant apart on the branches and 2 mm distant on the end branchlet. Polyps 2 mm long and incurved adaxially. Polyp scales placed in 8 longitudinal rows; six scales on each longitudinal abaxial row; adaxials fewer and smaller; 0.18 mm high/0.54 mm wide; and overlapping one another. Opercular scales triangular, longitudinally ridged and strongly indented; 0.86 to 1.2 mm long; operculum notably high. Marginal scales with rounded margins and ornamented with numerous prominences. Coenenchyme scales triangular, quadrangular or polygonal, ornamented with warts but not with ridges; 0.12 to 0.32 mm long.

REMARKS. - Studer (1894) suggested that *Amphilaphis abietina* was similar to *Th. (Amphilaphis) regularis* described by Wright & Studer (1889), from which it differed mainly concerning the flabby appearance of the colony; the size and distance between polyps; markedly high operculum; and details of the opercular scales. Compared to the other species of the genus *Thouarella*, the operculum shown by *Th. abietina* is much higher and somewhat peculiar. Figures showing a branch and details of the scales (Menneking, 1905), are the only available illustration for this species. The lack of illustration of the whole colony, makes it difficult to interpret the description of the colony form.

SPECIES 24 - *Thouarella dispersa*

Thouarella dispersa Kükenthal, 1912

Thouarella dispersa Kükenthal, 1912:307-309, pl. 20, fig. 4.
Thouarella (Amphilaphis) dispersa Kükenthal, 1915:149. - Kükenthal, 1919:411, pl. 31, fig. 11. - Kükenthal, 1924:290-291.

TYPE SPECIMEN & TYPE LOCALITY. - *Thouarella dispersa* Kükenthal, 1912. Twist (Antarctica), Deutsche Südpolar-Expedition, 2450 m, 01/03/1903. Present location: Z.M.B. 5468, Museum für Naturkunde, Humboldt-Universität, Berlin, Germany.

DIAGNOSIS. - Fan-shaped colony. Polyps singly placed on the branches. Polyp scales placed in eight longitudinal rows; five to six scales on each longitudinal abaxial row. High and pointed operculum. Marginal scales with projecting spine.

DESCRIPTION. - Colony form in one plane. Main stem simple, without side stems, giving off branches on two sides, oriented upwards at an angle of 60°. Branches simple and 15 to 25 mm long. Polyps club-shaped, 1.5 to 2.0 mm long, singly placed and sparsely distributed on the branches and main stem, often biserially disposed, and slightly incurved adaxially. Polyp scales placed in eight longitudinal rows; five to six scales on each abaxial longitudinal row. Opercular scales 0.8 mm long, narrow, leaf-shaped, with a strong median keel supporting one or more projecting spines and a median deep groove on the outer side. Marginal scales triangular and pointed, not bearing a prominent thorn but with only a feeble median spine. More distal body scales narrow, leaf-shaped and pointed, with smooth margins; not more than 0.6 mm wide, majority smaller. Coenenchyme scales fragile, polygonal or rounded, with a few warts and without ridges; 0.25 mm in diameter.

REMARKS. - Kükenthal (1912) described the branches bending preferentially towards one side of the colony creating a frontal and versal side. After personal examination, it was possible to observe that the colony is twisted having the branches on the upper part bent to one side and the branches on the lower part bent to the opposite side. If untwisted it agrees with the original description. The colony looks young for it has short, loose and fragile branches and a slender main stem. The extremities of the branches are bent against the distal portion of the colony and the polyps preferentially positioned towards the distal portion of the colony, against the tips of the branches. It appears as if the branches were bent away from the active current and the polyps were turned towards it.

SPECIES 25 - *Thouarella superba*

Thouarella superba (Nutting, 1912)

Primnodendron superbum Nutting, 1912:71-72, pl. 9, figs 2, 2a; pl. 19, fig. 4.
Thouarella (Amphilaphis) superba: Kükenthal, 1915:151. - Kükenthal, 1919:412.
- Kükenthal, 1924:291.

TYPE SPECIMEN & TYPE LOCALITY. - *Primnodendrum superbum* Nutting, 1912. St. 4778, Albatross Expedition, Semisopochnoi Island, Japan, 43-33 fathoms. Present location: Cat. No. 30691, U.S.N.M., USA.

DIAGNOSIS. - Fan-shaped colony, densely branched. Polyps singly placed on the branches. Polyp scales placed in eight longitudinal rows; six to seven on each longitudinal abaxial row. Marginal scales with projecting sharp spine.

DESCRIPTION. - Colony highly branched in one plane. Main stem immediately divides into three stems, the central stem forming the main part of the colony. Each stem gives off several branches which in turn, give off numerous closely spaced branchlets. Branches arising on all sides of the stem, although apparently disposed in one plane. Polyps club-shaped, 2 mm long, singly placed in all directions around the branches, being closely distributed; adaxially incurved and upwardly oriented. Eight longitudinal rows of scales; six to seven scales on each abaxial row; 3 to 4 in the adaxials. Operculum well developed. Opercular scales pointed; smaller and not pointed adaxially. Marginal scales bearing sharp spines abaxially and laterally, adaxial scales bearing only reduced spines or none. Two or three transverse rows of scales below the marginals have similar but smaller points. Body scales with a flaring thin margin and a central spine; the spine decreases from the marginal scales towards the base of the polyp; basal scales do not have central spines. Coenenchyme scales smaller than body scales and irregular in shape.

REMARKS. - The type specimen has a peculiar branching pattern, apparently differing considerably from all the other species of *Thouarella,* perhaps deserving to be accommodated in a new genus. Kükenthal (1924), however, placed this specimen into the genus *Thouarella,* subgenus *Amphilaphis.*

SPECIES 26 - *Thouarella grandiflora*

Thouarella grandiflora Kükenthal, 1912

Thouarella grandiflora Kükenthal, 1912:309-310, pl. 21, fig. 6.
Thouarella (Amphilaphis) grandiflora Kükenthal, 1915:149. - Kükenthal, 1919:304, pl. 21, fig. 6. - Kükenthal, 1924:291.

TYPE SPECIMEN & TYPE LOCALITY. - *Thouarella grandiflora* Kükenthal, 1912. Gauss-Station (Antarctica), Deutsche Südpolar-Expedition, 385 m, 07/02/1903. Present location: Z.M.B. 5469, Museum für Naturkunde, Humboldt-Universität, Berlin, Germany.

DIAGNOSIS. - Fan-shaped colony. Polyps singly placed on the branches. Polyp scales placed in eight longitudinal rows; seven to eight on each longitudinal abaxial row. Marginal scales with only a short projecting spine.

DESCRIPTION. - Colony branched in one plane, apparently pinnate, with branches arising at an angle of 45°, and side stems being parallel to each other. Branches simple and 40 mm long. Polyps up to 3 mm long, singly placed on the branches; biserially or irregularly distributed, occasionally crowded distally on the branch; directed outwards. Polyp scales placed on eight longitudinal rows; seven to eight scales on each longitudinal abaxial row; eight adaxially. Opercular scales narrow and triangular, with a thick median keel and indented lateral margins; 0.6 mm high. Marginal scales barely different from the other body scales, bearing only short projecting spines. Body scales leaf-shaped, with the free margins serrated and overlapped edge indented; 0.6 mm wide. Upper coenenchyme scales fragile, disc-like, 0.3 mm in diameter; lower coenenchyme scales smaller with strong radial ribs.

REMARKS. - The main stem of the type specimen is peculiar for having an irregular shape, apparently heavily twisted. The more irregularly curved area coincides with the path inhabited by a polychaete. The worm is large compared to the size of the colony. Only on one side of the apparently pinnate colony, side stems are developed, making the colony asymmetric. Polyps are not always developed on one plane as stated in the original description. They can be distributed on all sides of the colony as well as biserially. In two sites of the colony, there are unusual aggregation of polyps. These clumps of polyps are seemingly embedded in a calcified mass. These aggregations are apparently located at the end of the polychaete path. Kükenthal (1912) suggested that the closest related species to *Th. grandiflora* was *Th. regularis*.

SPECIES 27 - ***Thouarella plumacea***

Thouarella plumacea (Thomson & Mackinnon, 1911)

Amphilaphis plumacea Thomson & Mackinnon, 1911:680-681, pl. 65, fig. 3; pl. 68, fig. 3; pl. 74.

Thouarella (Amphilaphis) plumacea: Kükenthal, 1915:149. - Kükenthal, 1919:414. - Kükenthal, 1924:291-292.

TYPE SPECIMEN & TYPE LOCALITY. - *Amphilaphis plumacea* Thomson & Mackinnon, 1911. St. 22, 40, 44, Thetis Expedition, Barrenjoey, Australia, 30-40 fathoms. Present location unknown.

DIAGNOSIS. - Fan-shaped colony, with cymose branching pattern. Polyps singly placed on the branches. Polyp scales placed in eight longitudinal rows; 8 to 12 on each longitudinal abaxial row. Marginal scales without thorns.

DESCRIPTION. - Fan-shaped colony with a cymose branching pattern, where the main stem dichotomously subdivides into simple branches, disposed in one plane. Branches ramify dichotomously or irregularly into branchlets. Polyps singly placed and arranged in a spiral, occasionally biserially placed. Polyps club-shaped, 1 to 1.5 mm long, orientated at the branch at an angle of 45° to 60°. Polyp scales placed in eight longitudinal rows of scales; 8 to 12 scales on each abaxial longitudinal row; less adaxially. Opercular scales triangular and elongated, concave on the outer surface, bearing a median ridge on the inner surface, ornamented by warts and with free margin covered by ridges; 0.4 mm high/0.2 mm wide; they form a high operculum. Marginal scales not differing much from the other body scales, although better developed. Body scales with free edge crisply waved, outer surfaced with strong radial ridges and inner surface with numerous warts aggregated around a nucleus; 0.31 mm wide/0.27 mm high. Coenenchyme scales rounded, with a central nucleus surrounded by warts and free margin with a few low ridges; 0.13 mm in diameter.

REMARKS. - The type specimens have a true planar form. The main stem subdivides distally like the other stems. The colony is not very large (185 mm long) and is already highly ramified. Thomson & Mackinnon (1911) separated this species from *Th. regularis* by the following criteria: slender and more highly branched; polyps in a spiral and crowded together;

smaller size of polyps and greater number of scales on the abaxial rows. From *Amphilaphis abietina*, *Th. plumacea* differ in: the close spiral arrangement of polyps; and the fact that the branches do not come off in right angles from the stem.

SPECIES 28 - *Thouarella biserialis*

Thouarella biserialis (Nutting, 1908)

Amphilaphis biserialis Nutting, 1908:573, pl. 43, fig. 3; pl. 47, fig. 4.
Thouarella biserialis: Kükenthal, 1919:438. - Kükenthal, 1924:301 (*species dubiae atque incertae sedis*)

TYPE SPECIMEN & TYPE LOCALITY. - *Amphilaphis biserialis* Nutting, 1908. St. 3982, Albatross Expedition, off Kauai, Hawaii, 40-233 fathoms. Present location: Cat. No. 22583, U.S.N.M., USA.

DIAGNOSIS. - Polyps placed in pairs on the branches; occasionally in whorls of three. Five scales on each abaxial row. Opercular scales alternating in two rings of four.

DESCRIPTION. - Main stem gives off alternate branches at intervals of about 18 mm. Polyps club-shaped, 1.5 mm long, outwardly oriented and placed in pairs on the stem, occasionally in whorls of three distally on the branches. Polyp scales placed in four longitudinal and five transverse rows. Low operculum nearly concealed by the marginal scales. Opercular scales apparently alternating in two rings of four. Marginal scales broadly triangular, ribbed and fluted. Body scales large, imbricate, with the undulating free margins toothed and the outer surface sculptured with radial ridges and furrows. Coenenchyme scales broad, lamelliform, rounded or oval in outline.

REMARKS. - The original description by Nutting (1908) was unclear and lacked important information. The drawings were inaccurate and the lack of illustration of the whole colony conveys a misleading interpretation. Nutting suggested that this species was similar to *Callogorgia gilberti*, from which it differed mainly by the polyps not being appressed against the axis and for the larger size of the coenenchyme scales. It also resembles the species of the genus *Plumarella* to which he avoided referring this species because it has the polyps disposed in pairs. Nutting assumed that the features of this species did not agree completely

with the definition of the genus *Amphilaphis,* but it is closely related to it. Kükenthal (1924) placed this specimen in the genus *Thouarella* instead, among his *species dubiae atque incertae sedis.* Without examining the type specimen, no judgement about the taxonomic position of this species can be made.

SPECIES 29 - *Thouarella alternata*

Thouarella alternata Nutting, 1912

Thouarella alternata Nutting, 1912:69-70, pl. 9, figs 1, 1a; pl. 19, fig. 3. - Kükenthal, 1919:438. - Kükenthal, 1924:301 (*species dubiae atque incertae sedis* - mistaken for *Th. attenuata*).

TYPE SPECIMEN & TYPE LOCALITY. - *Thouarella alternata* Nutting, 1912. St. 5080, Albatross Expedition, Omai Saki Light, Japan, 505 fathoms. Present location: Cat. No. 30097, U.S.N.M., USA.

DIAGNOSIS. - Bottlebrush colony with ramified branches. Polyps singly placed and alternating on the branches. Each longitudinal abaxial row with 8 to 10 scales. Low cone-shaped operculum. Scales markedly delicate. Marginal scales bearing long sharp thorns.

DESCRIPTION. - Colony form bottlebrush with branches arising on three sides of the main stem. Branches up to 32 mm long, 3 mm distant from each other, and subdividing dichotomously into branchlets. Polyps 1.5 mm long, singly placed and alternating with each other, rarely in pairs; sparsely distributed on the branches, averaging 2 mm apart; upwardly oriented and incurved adaxially. All the scales are markedly delicate, with free margins heavily toothed and surface ornamented with warts. Marginal scales bear long sharp thorns (up to 1 mm long); adaxially the thorns are short or absent; free margins finely serrated. Each longitudinal abaxial row with 8 to 9 scales; 6 to 7 in the laterals; and 4 in the adaxials. Low and conical operculum. Opercular scales flat, triangular and delicate.

REMARKS. - From Nutting's (1912) figures, it is possible to observe that the colony has a fragile appearance and the slender branches bear small polyps. The branching pattern is not dense and the colony appears to have a high porosity, since the branches are sparsely distributed. This could be the reason for the polyps not being aggregated at the terminal portion of the branches, but homogeneously distributed along the branch. If it was not for the

high number of abaxial scales and for the apparently poorly ramified branches, this specimen could easily be referred to *Th. variabilis*. Kükenthal's (1924) description of this species, to which he mistakenly refers as *Th. attenuata*, contradicts the original description concerning the branching pattern. Nutting described branches arising on three sides of the main stem, implying a bottlebrush appearance. Kükenthal probably based his description on Nutting's description and figures, and concluded that the branches were arising on two sides of the main stem, implying that the branching pattern was biseriate. From Nutting's figure, a biseriate arrangement could really be suggested as the branches appear bent forwards. However, without examining the type specimen, it is dangerous to make such judgement. The original description omits some characters such as branching pattern of the branches and details of the body and coenenchyme scales. Kükenthal (1924) placed this species among his *species dubiae atque incertae sedis*.

SPECIES 30 - *Thouarella recta*

Thouarella recta Nutting, 1912

Thouarella recta Nutting, 1912:67-68, pl. 7, figs 1, 1a; pl. 19, fig. 2. - Kükenthal, 1919:440. - Kükenthal, 1924:302.

TYPE SPECIMEN & TYPE LOCALITY. - *Thouarella recta* Nutting, 1913. St. 5079, Albatross Expedition, Omai Saki Light, Japan, 475-505 fathoms. Present location: Cat. No. 30040, U.S.N.M., USA.

DIAGNOSIS. - Bottlebrush colony. Polyps irregularly singly placed on the branches. Scales on each longitudinal abaxial row in number of 6. Low operculum covered by marginal scales. Marginal scales bearing long sharp thorns.

DESCRIPTION. - Colony form bottlebrush, with branches arranged on the main stem in spirals of three distally and irregularly proximally. Branches, up to 24 mm long, mostly simple, often bifid, occasionally giving off branchlets. Polyps 1.4 mm long, singly placed, scattered, distant apart and irregularly distributed; disposed biserially or distributed along a spiral; directed outwards, slightly upwards. Scales on each longitudinal abaxial row in number of 6. All scales with free margin toothed. Operculum low and covered by the marginal scales. Opercular scales triangular, with free margin toothed. Marginal scales bear a long thorn. Coenenchyme scales placed in two layers are rounded or irregular, with imbricated edge.

REMARKS. - The type specimen appear to be relatively young (47 mm long) and somehow damaged. Nutting (1912) noted that this species resembled *Th. laxa*, from which it differed in the arrangement of polyps, which are rarely in pairs. The whole appearance of the colony, based on Nutting's drawing, resembles the young form of *Th. alternata* Kükenthal (1919) placed this species among his *species dubiae atque incertae sedis*.

SPECIES 31 - *Thouarella pendulina*

Thouarella pendulina (Roule, 1908)

Rhopalonella pendulina Roule, 1908:4-5, pl. 1, figs 5-8. - Gravier, 1914:70-77, pl. 3, figs 15,16; pl. 5, figs 21-25; pl. 10, figs 56-59.

Thouarella pendulina: Kükenthal, 1919:440. - Kükenthal, 1924:302 (*species dubiae atque incertae sedis)*

TYPE SPECIMEN & TYPE LOCALITY. - *Rhopalonella pendulina* Roule, 1908. Expedition Antarctic Française, Booth-Wandel Island, Antarctica, "collected from cormorant's nests", probably fished by the birds not far from the coast. Present location unknown.

DIAGNOSIS. - Bottlebrush colony, with ramified branches (up to 3 orders) arising on all side of the colony. Polyps singly placed all around the stem, and closely spaced. Scales placed in diagonal rows; five to six abaxial scales. Marginal scales thorny.

DESCRIPTION. - Colony form bottlebrush and densely branched, with main stem bearing on all sides, at nearly right angles, closely spaced branches; 1 to 3 mm distant apart. Main stem flexible and finely grooved longitudinally. Branches proximally short and rigid and middle branches flexible and long, 40 to 42 mm long; occasionally biserially disposed; distal and proximal branches simple and middle branches ramified dichotomously into branchlets (up to 3 orders). Polyp 1 to 1.5 mm long, incurved adaxially, singly placed all around the branch and mostly disposed close together; polyp numbers vary from sparsely distributed to markedly packed together, up to 70 polyps on 10 mm of a terminal region of a branch; also aggregated at the base of the branches. Polyp scales not placed in longitudinal or transverse rows, but preferentially diagonal; with five to six abaxial scales. Marginal scales six, convex dorsally, heavily indented proximally and with serrated margins; four abaxial scales narrow and triangular, bearing a thorn, an inner keel and having their outer surface smooth and the inner

surface ornamented with warts; two adaxial scales similar to the abaxial although broader, less triangular and more leaf-shaped; abaxial: 0.55 mm high/0.30 mm wide; adaxial: 0.48 mm high/0.40 mm wide. Opercular scales six or eight, triangular and narrow, indented proximally, bearing an inner keel, smooth on the outer surface, ornamented with warts on the inner surface and with serrated free edges; 0.35 mm high/0.20 mm wide. Body scales broader than higher, ornamented with warts sometimes arranged radially on their inner surface, indented proximally and with heavily serrated free margins. Adaxial scales much smaller, 0.05 to 0.06 mm long/0.035 mm wide, of irregular outline and covered with warts on the inner surface. Coenenchyme scales small, varied in form, heavily covered by prominences on the inner surface.

REMARKS. - Etymology of *Rhopalonella* (greek): club. Despite the specimen studied by Roule (1908) being already almost 350 mm long, it did not give off any side stems, common in old colonies of other species. He noted that the colony showed in its lower portion spines similar to the spines present in antipatharians. However, these spines could be broken lower branches, which is often seen in other species of *Thouarella*. The aggregation of polyps proximally at the branches, shown by Roule (1908) and Gravier (1914), might have been caused by the presence of a commensal since the same sort of polyp distribution has been observed in other colonies inhabited by a polychaete. Observing their figures, one has the impression that some parts of the colony, which are biseriate are flattened as if they were forming a path. However, the presence of a polychaete was not mentioned by any of the authors. Gravier (1914) mentioned that the basal polyps were, in the great part, reproducing and had enlarged bases. This could explain some of the aggregation of polyps. The polyps are so aggregated that sometimes they are artificially placed in whorls, but in general they are placed singly. Gravier (1913) suggested that because of the aspect of the operculum and the arrangement of polyps, which are singly placed, the species *Rhopalonella* could be referred to the subfamily Primnoinae. Other characters such as branches arising all around the main stem and singly placed polyps would include this species into the *Thouarella antarctica*-group. It would, however, differ in the aspect of the polyp armature and spiculation, specially concerning the marginal scales. Kükenthal (1919) placed this species into the genus *Thouarella* among his *species dubiae atque incertae sedis*. He did not give any argument for doing so. Apart from the high number of polyps, the other characters shown by this species are similar to the ones shown by *Th. variabilis*.

SPECIES 32 - *Thouarella brucei*

Thouarella brucei Thomson & Ritchie, 1906

Thouarella brucei Thomson & Ritchie, 1906:852-854, pl. 1, fig. 1; pl. 2, fig. 1. - Kükenthal, 1919:439. - Kükenthal, 1924:301 (*species dubiae atque incertae sedis*). *Thouarella (Euthouarella) brucei* Broch, 1965:27-28, pl. 4, figs 11-13.

TYPE SPECIMEN AND TYPE LOCALITY. - *Thouarella brucei* Thomson & Ritchie, 1906. Burdwood bank, 54°43'S, 60°14'W, 61 fathoms, hard bottom, prawn-trawl and dredge, 09/03/1948. Present location: holotype - B.M. 1912.11.9.2, N.H.M. London, UK. lectotype (topotype) - B 969 Zool. Museum, Oslo, Norway (?).

DIAGNOSIS. - Bottlebrush colony, with branches arising in three directions; sclerites highly ornamented. Marginal scales with foliate processes.

DESCRIPTION. - (Thomson & Ritchie, 1907; type specimes ?, N.H.M., London) - Colony form bottlebrush, upright branched and with branches arising from the main stem in at least three directions at irregular intervals; larger branches tend to curve inwards towards the main stem; side stems bearing branches of their own. Main stem stout and rigid and composed of horny and calcareous material. Polyps pear-shaped, 1 mm long, adaxially incurved, distributed closely spaced, arising in all directions without definite arrangement. Polyp scales placed usually in seven longitudinal rows and 5 transverse rows. Scales similar in size and structure, with convex upper margin, often quadrangular-shaped and thickly tuberculated; occasional ridged tubercles run outwards from a nucleus; proximal margin more ragged than the distal free margin. Seven opercular scales with a long projecting ridge and a narrow leaf-like wing.

(Broch, 1965 & personal observation; lectotype ?; Zool. Museum, Oslo) - Colony form bottlebrush, with closely spaced branches arising on all sides of the main stem, occasionally perfectly alternating, making a markedly regular branching pattern. On one side of the colony a slightly biseriate arrangement can be noted, especially distally. Branches simple, rarely dichotomously branched. Polyps club-shaped, 1 - 1.5 mm long, placed in whorls mostly of 3, occasionally 4 or even of 5 distally on the branches; proximally in pairs or sometimes singly placed; on the extremities of the mostdistal branches they might be disposed in pairs; absent on the stem; on the biseriate flat side the polyps are placed in one plane; 4 - 5 whorls on 10 mm. Scales similar in size and structure. Opercular scales leaf-shaped. Marginal scales

triangular, lancet-shaped with a median ridge on their inner side, and armed with two or one slender spines distally; they do not cover the opercularis. Body scales irregularly rounded with a serrate free margin and indented base. Marginal and submarginal scales adorned with radiating veins and small warts.

REMARKS - The existing lectotype is the specimen described by Broch (1965) and his description contradicts the original description of Thomson & Ritchie (1906). There is, however, a type specimen at the N.H.M., London. This specimen does not agree with the original description either. Because there is doubt about the actual type specimen, it was considered appropriate to present both descriptions.

The colony appears to be just a side branch from the main branch which is lacking. Thomson & Ritchie certainly did not describe the type specimen held by the Museum in Oslo because their description of the branching pattern of the colony does not apply to that specimen. They probably described an old colony which had several main side stems. What they named twigs or branchlets are in fact simple branches. Thomson and Ritchie mentioned that the number of transverse rows varied, five being the common number. Kükenthal (1924) placed this species among his *Species dubiae atque incertae sedis*. Broch (1965) questioned the original description concerning the long projecting ridges of the marginal scales shown in Pl. 2, Fig. 1 (in: Thomson & Ritchie, 1906). Broch noted that the three spicules with long and folded apical projections belonged to the type of marginal scales found in *Thouarella longispinosa, Th. variabilis* and *Th. striata*. He assumed that Thomson & Ritchie had contaminated samples and that they probably observed sclerites of other species. Broch placed this species in the subgenus *Euthouarella* and added: "On the other hand the numerous and comparatively small scales of the polyps and their arrangement show near affinity both with the subgenus *Amphilaphis* and with *Th. (Euthouarella) chilensis*". *Th. chilensis* is in fact assigned to the subgenus *Epithouarella*. With the absence of marginal thorns, this species would in fact be better placed in the subgenus *Epithouarella* rather than in *Euthouarella*. Perhaps, that is what he meant.

SPECIES 33 - *Thouarella hicksoni*

Thouarella hicksoni Thomson, 1911

Thouarella hicksoni Thomson, 1911:886, pl. 44, fig. 3a,b; pl.45, fig. 1. - Kükenthal, 1919:439. - Kükenthal, 1924:301 (*Species dubiae atque incertae sedis*).

TYPE SPECIMEN & TYPE LOCALITY - *Thouarella hicksonii*, Thomson 1911. South-west of Cape St. Francis, Cape Colony (South Africa), 74 fathoms. Present location: B.M. 1962.7.20.36, N.H.M. London.

DIAGNOSIS. - Bottlebrush colony with mostly simple branches. Polyps singly placed. Marginal scales bearing thorns.

DESCRIPTION. - Colony form bottlebrush, with branches at the lower portion of the colony longer than the terminal branches. Branches mostly simple, occasionally ramified. Polyps pear-shaped, 1.94 mm long, singly placed on the branches and rarely present on the main stem. Polyp scales placed in 4 to 5 longitudinal rows, with free margins partly or completely serrated. Marginal scales bearing thorns and 0.56 mm long. Coenenchyme scales vary in size, from 0.068 to 0.22 mm in diameter.

REMARKS. - Examining the type specimen held at the N.H.M. London, it was possible to observe that the branches are often subdivided into branchlets, contradicting the original description. The polyps are singly placed and marginal, and opercular scales do not differ a great deal giving the uppermost portion of the calyx a rounded resemblance. The longitudinal rows mentioned by Thomson were probably only the abaxial because of the small number (4 to 5). Kükenthal (1924) did not place this species into any of the proposed subgenera and preferred to refer it to his *species dubiae atque incertae sedis*. Examining the type specimen it was possible to confirm that it has really a bottlebrush growth form.

SPECIES 34 - *Thouarella acanthina*

Thouarella acanthina (Wright & Studer, 1889)

Stenella acanthina Wright & Studer, 1889:59, pl. 14, fig. 3; pl. 20, fig. 10.
Stenella (Dasystenella) acanthina Versluys, 1906:48.
Thouarella acanthina: Kükenthal, 1915:151. - Kükenthal, 1919:441. - Kükenthal, 1924:302 (*species dubiae atque incertae sedis*)

TYPE SPECIMEN AND TYPE LOCALITY. - *Stenella acanthina* Wright & Studer, 1889. St. 320, Challenger Expedition, off Rio de la Plata, 600 fathoms, bottom green sand. Present location: SYNTYPE - B.M. 1889.5.27.48, 48a, N.H.M. London, UK.

DIAGNOSIS. - Bottlebrush colony. Polyps in whorls of 3 to 4. Five scales on each longitudinal abaxial row. Marginal scales fewer than eight and bearing long thorns.

DESCRIPTION. - Colony form dense with main stem giving off branches on all sides arranged in an incomplete spiral. Main stem hard, consisting of numerous concentric rings of fibrous substance, with calcareous particles. Polyps 2 mm, in whorls of 3 to 4. Scales on each abaxial rows in number of five. Opercular scales 0.7 mm high/0.3 mm wide. Marginal scales fewer than eight and bearing long thorns; 1.5 mm high/0.7 mm wide. Body scales 0.8 mm/0.6 mm. Adaxial scales markedly smaller than abaxials. Coenenchyme scales oval disc-like, often densely packed; 0.3 mm/0.2 mm.

REMARKS. - The original description of the branching pattern is very brief and inaccurate, making it difficult to understand the growth form of the colony. Versluys (1906) considered the branching pattern similar to *Thouarella variabilis*. *Th. acanthina* could not, however, be included in the *antarctica*-group because the polyps are whorls rather than singly placed. He also noted that the marginal scales are not eight in number but six or even five or four, which would be characteristic of *Stenella* (= *Pterostenella, Dasystenella*). Kükenthal (1915) considered this species similar to *Th. longispinosa*.

CHAPTER 7
SUMMARY

This chapter summarizes the knowledge obtained about the genus *Thouarella*, especially about the species *Th. variabilis*, through the work presented in the previous chapters. As already stated in this thesis, prior to this study much was known about the taxonomy but virtually nothing was known about the biological aspects of this group. The work presented herein adds much information to so poorly a known group. By complementing the information obtained in this study with other examples in the literature, it has been possible, in the following section, to attempt to outline the life history of the species *Th. variabilis.* In this chapter the taxonomic status of the species of the genus *Thouarella* is also discussed.

7.1 - LIFE HISTORY OF *Thouarella variabilis*

Thouarella variabilis produces a non-feeding, non-pelagic lecithotrophic planula larva. The swimming planula most likely settles soon after release in a site not far from the parent colony, probably resulting in the patchy distribution of the species.

The planula is relatively large (860 µm long) and composed of an inner epithelium, the endoderm, a connective layer, the mesoglea, and an outer epithelium, the ectoderm. The ectodermic cells are abundantly ciliated in a fully developed planula. The yolk content of the larva is already partly consumed and a coelenteron begins to form whilst the planula is still inside the polyp.

Once settled, the planula retracts the cilia, develops the mesenteries, outgrows the eight tentacles and the oral surface invaginates forming the mouth and the pharynx which leads to a gastric cavity. Posteriorly the pinnules develop on the tentacles. The sclerites might have already started developing at this stage. The epidermis is formed of ectodermal cells, and so is the pharynx and the two asulcal mesenteries. The body wall and the remaining six mesenteries are lined with endodermal cells. On one side of the pharynx a strongly ciliated longitudinal tract, the siphonoglyph, is formed. Together with the siphonoglyph, the asulcal mesenteries, which are heavily ciliated, are responsible for maintaining the water circulation inside the colony. The tentacles and pinnules are highly contractile and are abundantly supplied with nematocysts.

The scleroblasts, present in the mesoglea, will be responsible for the production of the calcareous sclerites, which will surround the soft parts of the polyp as a calcareous armature. The sclerites, which were investigated, showed that the calcium carbonate is deposited as calcite and not as aragonite, as previously thought for the family Primnoidae (Bayer, 1981c). The uptake and deposition of calcium decreases with the decrease of temperature (Alemmand & Grillo, 1992) and perhaps this is the reason for the fragile consistency of the sclerites of the cold water Antarctic and deep-sea primnoids. The rate of calcification might be incremented with the increase in water temperature during the Antarctic summer.

The very young polyp (up to 0.3 mm long) is composed of an aggregate of blunt scales without any proper form. With continuous growth, the scales are moulded in a more defined shape. When about 0.8 mm long, the polyp resembles the adult polyp but the scales are still underdeveloped. The polyp attains an average adult size when around 1 mm long, and a reinforced calcified armature, by heavier calcification and fortification of spines and thorns, when over 1.2 mm long. The maximum polyp size observed was just less than 2 mm.

An operculum, enclosing the soft parts of the polyp, is formed by eight small triangular scales at the distalmost portion of the polyp. The operculum is surrounded by eight marginal scales, bearing sometimes markedly long and sharp thorns. The submarginal scales are also provided with a shorter thorn. The lower body scales are larger and curved to conform to the contours of the body of the polyp. The very basal body scales are shaped so as to fit in with the coenenchyme scales of the axis. The polyp is capable of inclining towards the axis because the adaxial marginal scales do not bear long thorns and the body adaxial scales are small and plate-like, and often covered by the laterals. When the polyp expands by bearing reproductive products, these small scales move apart enabling the expansion of the base of the polyp. The abaxial and lateral body scales overlap one another and are placed in five diagonal rows. When the polyp expands these scales slide apart and are disposed in longitudinal rows.

The base of the scales are indented and heavily tuberculated keeping them anchored in the mesoglea. The inner surface of the scales are provided with compound warts, radially or irregularly disposed and their outer surface with short spines. The warts on the inner surface of the overlying scale engage the short spines and tubercles of the underlying scale keeping them interlocked avoiding excessive extensions of the scales but giving a certain flexibility to the polyp. The calcareous armature surrounding the soft parts provides the polyp with skeletal support and protection against predators.

The primary polyp will give rise to a second polyp by asexual reproduction, budding. The lower portion of the gastric cavity of the polyps are interconnected by a small canal lined with endoderm, the solenia, which is covered by a thin coenenchyme. The new buds will derive from outgrowth of the solenia. In the sub-tropical species *Xenia macrospiculata* (Benayahu *et al.*, 1989), a young colony of 4 polyps is formed in 5 to 6 months. This information is not available for *Th. variabilis*.

A common horny axial skeleton is formed. It originates below the base of the primary polyp, being secreted to the outside of the ectodermal cells (Chapman, G., 1974). The axial skeleton consists of gorgonin, a proteinaceous matrix in which flexible collagen fibres are embedded. The gorgonin is stiffened by sclerotization and by mineralization since it is permeated by calcareous components. The layer of coenenchyme scales covering the axis also contributes to the stiffness of the axial skeleton.
With the development of the colony, the axial skeleton grows in extension and in density. The deposition of skeletal material is discontinuous because of tidal and seasonal variations. During the eutrophic summer months in the Antarctic, deposition is probably high, decreasing in the oligotrophic winter months. This variation in deposition results in the formation of growth layers, which can be detected in cross sections of the main stem. In young colonies (up to 5 or 6 years old), the growth layers are large and well delineated, and therefore, the estimation of age is possible. In colonies up to 10 years old, the growth layers are not so distinguishable but they are still possible to detect. In older colonies, the outer growth layers are thin and apparently compressed against each other. The layers are not easily detectable, and the determination of age by counting the growth layers is, for this reason, unreliable.

With the extension of the axis new buds arise and an erect colony takes shape. The colony is always attached to a solid substrate by means of a calcareous enlargement of the base of the main stem. The main stem will give off side branches mostly perpendicularly directed, slightly upwards. A young colony of *Th. variabilis*, such as seen in Figure 2, will show a regular branching pattern with branches arising from the main stem in opposite or slightly offset pairs, in four directions. The colony will maintain its regular branching pattern on the condition that there is no dominant current direction and the current flows within the boundary layer, where the young colony has developed, and the main stream are similar and that there is no polychaete commensal inhabiting the colony.

If the current in the sheltered boundary layer is subject to a different flow than that in the main stream, the colony will grow helically twisted. Once the colony is passively redirected and

held in that position during periods of skeletal growth, progressive changes in orientation occur in the direction of the water movement.

If the colony is subject to unidirectional or bidirectional current-generated water movements, the branches of the originally bottlebrush colonial form will tend to passively bend perpendicular to the current. The branches will be, therefore, positioned on the sides of the colony, which will attain a biseriate arrangement forming a frontal and versal side. By alteration in the direction of the flow, the biseriate colony will twist along its axis attaining a deceptive bottlebrush appearance.

The presence of polychaete commensals inhabiting the colony will induce deformations of the branches and consequent alteration of the branching pattern. The polychaete might settle on the octocoral when the colony is still very young. As the branches grow they bend towards the worm, by an unknown process, and a tube is formed. On doing that, the branches alter their position and a biseriate arrangement of the colony is seen. The understanding of the branching pattern of the colony becomes even more complicated when two polychaete tubes are formed or when the tube is confined to certain portions of the colony and the regular bottlebrush branching pattern is maintained in other portions.

Combining the alterations caused by hydrodynamic forces and the deformations caused by polychaete commensals, a great variation of branching pattern is to be expected.

Large colonies might give off side stems which will be disposed in one plane and will bear branches of their own. The maximum colony height recorded for *Th. variabilis* is 340 mm (Thomson & Rennet, 1931). The size limit might be determined by the maximum force the holdfast can resist before it detaches from the solid substrate and the colony dies by abrasion.

The branches are divided into branchlets, which are dichotomously branched in one plane early in development. The highly ramified branches are complex and pseudo-dichotomous. The number of branch orders can vary. According to Velimirov (1976), areas of high availability of food favours specimens with few ramifications, long end branchlets and numerous polyps and in areas poor in energetic resources the specimens with highly ramified narrow branches, bearing fewer polyps are in advantage.

The polyps arise on the branches singly and alternating one another, sometimes in short spirals of threes. The polyps are directly upwards and inclined towards the axis. On the

proximal and middle regions of the branches, they are sparsely distributed, whereas on the distal region they are aggregated together.

As mentioned above, the availability of food may influence the number of polyps on the branches. In the bottlebrush forms of *Th. variabilis*, the closely spaced highly ramified branches will result in low colony porosity. Consequently, the most appropriate feeding position is on the terminal portion of the branches, where the polyps are often found in higher number. The middle and proximal portions of the branches are probably less adequately nourished and a lower number of polyps is found. On the exposed distalmost part of the colony, where the branches are mostly simple, the polyps are also in high numbers. In colonies which have more sparsely distributed branches and high porosity, the polyps are more regularly distributed. In bottlebrush colonial forms which modify the current flow, eddy currents can form which supply the polyps disposed on all sides of the axes with food.

The arrangement of the polyps may be affected by their position in relation to the food bearing current (Wainwright & Koehl, 1976; Leversee, 1976; Jeyasuria & Lewis, 1987). In biseriate stiff colonies, the polyps may tend to dispose biserially on the side of the branch which is in the most favourable feeding position. If the stems and branches are flexible enough to turn to the current, the polyps will not be in disadvantage if disposed all around the branch.

The feeding habits of *Th. variabilis* could not be defined since no particulate food was observed on the tentacles or in the body cavities of the polyps. Nematocysts were, however, observed in abundance on the tentacles. Perhaps, these organisms rely on absorption of dissolved organic material (DOM). For some species such as *Heteroxenia fuscescens* (Schlichter, 1982), DOM is the most important nutritional supply. The absorption of DOM is suggested to be an advantage for animals inhabiting an energy poor environment (Lawson, 1990).

During the first reproductive stages, fat granules are present in massive concentrations in the cells from the base of the tentacles to the lower part of the pharynx. They are even more abundant in the endodermic cells of the mesenteries in the gastric cavity.

The polyps which are positioned in a most favourable feeding position, such as the terminal portion of the branches and the distalmost part of the colony, do not usually reproduce. These polyps probably carry on feeding and distributing the dissolved compounds to the rest of the colony via the solenia, whereas the middle and proximally positioned polyps do not feed because of being blocked with reproductive products.

The colonies of *Th. variabilis* are gonochoric. In both, male and female polyps, the gonads develop preferentially on the two lateral mesenteries close to the two asulcal mesenteries on the adaxial side of the polyp. The polyp fecundity is low, perhaps as a result of the polyp having a short gastric cavity compared to the relatively large oocytes and sperm sacs produced. The female polyps produce only one oocyte and the male polyps produce in most cases only two sperm sacs at a time.

In the first stage of the development of male gonads, the germ cells present in the mesenterial endoderm migrate to the mesenterial mesoglea. The spermatocytes accumulates at the lower portion of the mesenteries, searching for space, bulge into the gastric cavity drawing around themselves the mesoglea in which they are embedded and the outer layer of mesenterial endoderm. The two developing sperm sacs will be surrounding by heavily ciliated and cuboidal endodermal cells which become elongated as the sperm sacs grow in size. The spermatozoa are fully developed and the sperm sac ready to spawn when over 500 µm. At this stage the endoderm is fragile and breakable. The sperm sacs are expelled through the mouth. Other small sperm sacs might be in development after the former gonads have been released.

Although, in the female polyps only one oocyte will be preferentially developed to maturation at a time, oocytes in different stages of development might also be present. The oocytes will develop preferentially on the lowermost portion or the lateral mesenteries close to the two asulcal mesenteries. The mature oocyte reaches a length of 660 µm and a width of around 500 µm. It detaches from the mesentery and is loose in the gastric cavity. It occupies 60% of the volume of the polyp and it is ready to be fertilized.

The presence of oocytes in different stages of development within the same polyp suggests a two year cycle of oogenesis or continuous gametogenesis with release of larvae occurring throughout the year. There is, however, a general trend of development of oocyte and sperm sacs during summer with the late stages of development being mostly observed at late summer.

The energetic reserves, clearly seen during the first reproductive stages, decrease considerably at the later stages, suggesting that the production of a single egg is very costly. To have continuous gametogenesis the polyp would have to continuously build up energetic reserves.

Fertilization is internal and the embryo will move to the upper part of the polyp cavity where the larva planula will be developed. The larva will be brooded inside the polyp cavities until

maturation is reached. This reproductive mode provides protection for the embryo, enhancing early survival and compensating for the low fecundity of the polyp.

7.2 - TAXONOMIC STATUS OF THE SPECIES OF THE GENUS *Thouarella*

From the information obtained about some aspects of the life history of *Th. variabilis*, it was possible to understand some of the causes of the variation of the characters applied to classify and identify the species of the genus *Thouarella.*

The characters that have been used to describe each of the 34 nominal species attributable to *Thouarella* are:

Colonial form/branching pattern
Orientation/length and distance between branches
Number of branch orders
Aspect of the main stem
Arrangement/orientation/number and distance between polyps
Polyp shape/size and orientation
Aspect of the operculum
Arrangement of body scales
Arrangement of opercular and marginal scales
Number of scales on each longitudinal abaxial row
Shape/size/ornamentation of:
    opercular/marginal (length of thorn)/submarginal/
    lower body/adaxial/coenenchyme scales

The key characters applied to distinguish the species of this genus are shown in Table 7.1. These characters were employed by the mentioned authors in an attempt to group the species of the genus *Thouarella* in groups or subgenera and for the elaboration of identification keys.

Most of these characters show some degree of variation caused by extrinsic aspects such as: intensity and direction of currents, water temperature, availability of food, presence of commensals; and intrinsic aspects such as: growth and reproductive stage of the polyps, stiffness of the axes and developmental stage and porosity of the colony.

Table 7.1 - Characters employed for the diagnosis of the species of the genus *Thouarella*.

CHARACTER	AUTHOR				
	Versluys, 1906	Roule, 1908	Kinoshita, 1909	Nutting, 1912	Kükenthal, 1924
Branching pattern	X		X	X	X
Number of branch orders					X
Polyp arrangement	X	X	X	X	X
Polyp number		X			X
Polyp size		X			
Polyp orientation			X		
Aspects of the operculum			X		
Arrangement of scales					X
Number of scales on each abaxial row					X
Shape and ornamentation of scales			X		X

For these characters to have some taxonomic value, a range of variation for each of them within every species has to be delineated. Understanding some of the causes of variation, this delineation may be possible. However, it is important to re-emphasize that to rely on most variations of characters on environmental influence alone could be misleading because virtually nothing is known about the environmental conditions the specimens inhabited and sometimes specimens of varied forms come from the same station. Some considerations about the key characters are presented as follows.

COLONIAL FORM AND BRANCHING PATTERN - Variations could be caused by intensity and direction of currents, presence of commensal and developmental stage of the colony. This character could be consistent if side stems are not considered for the diagnosis of the branching pattern since their existence varies with the age; if the influence of the presence of commensals is evaluated; and if limits are drawn between:

- RACEMOSE BRANCHING PATTERN, BOTTLEBRUSH OR BISERIATE;
- RACEMOSE BRANCHING PATTERN, TRUE PINNATE;
- CYMOSE BRANCHING PATTERN.

NUMBER OF BRANCH ORDER - Variation caused by availability of food, direction of currents and developmental stage of the colony. This character requires further examination but perhaps could be used, although with some caution, as:

- SIMPLE BRANCHES
- RAMIFIED BRANCHES (FROM BIFID TO FIVE ORDERS)

POLYP ARRANGEMENT - This is one of the most consistent characters, however, the number of polyps on a whorl could vary with developmental stage of the colony. This character should be further examined. The categories could be:

- PLACED SINGLY
- PLACED IN PAIRS
- PLACED IN WHORLS (FROM 3 TO SIX)

POLYP NUMBER - This is one of the most inconsistent characters. The number of polyps can vary considerably within the same colony. Variations might be caused by availability of food, length of branchlets and presence of commensal. This character certainly deserves better examination.

POLYP SIZE - It appears to be a constant character. However, little is known about the limit of size within each species and, therefore, this character should be used with caution.

POLYP ORIENTATION - This is a constant character and could have two categories:

- DIRECTED OUTWARDS
- ADAXIALLY INCURVED

ASPECT OF THE OPERCULUM - This is apparently a consistent character and could have two categories:

- HIGH OPERCULUM, NOT COVERED BY THE MARGINAL SCALES
- COVERED BY THE MARGINAL SCALES

ARRANGEMENT OF SCALES - It can be one of the most consistent characters but should be much better evaluated. Polyps should be observed under a scanning electron microscope for a better understanding of the arrangement of the scales.

NUMBER OF SCALES ON EACH LONGITUDINAL ABAXIAL ROW - This appears to be a consistent character. The number of scales varies from 4 to 12.

SHAPE AND ORNAMENTATION OF SCALES - The shape of opercular and marginal scales appear to be a distinctive character. The length of the marginal thorn is variable and should be used with caution. The shape and ornamentation of scales require much better investigation.

Kükenthal (1915) elaborated and updated (1924) an identification key for the species and subgenera of the genus *Thouarella* applying the characters shown in Table 7.1. This is the only existing key for this genus. In order to evaluate the reliability of this key, it is reproduced in Table 7.2.

The species that Kükenthal could not place in the key, he placed amongst his *Species dubiae atque incertae sedis*: this includes *Th. alternata (*mistaken by *attenuata*); *Th. biserialis; Th. brucei; Th. hicksoni; Th. pendulina; Th. recta; Th. acanthina.*

The key has its validity, however, many of the characters used by Kükenthal in his key were demonstrated earlier in this section, to be inconsistent. The existence of eight longitudinal rows of abaxial scales in the species of the subgenus *Amphilaphis* is not really clear since in the description of some of these species, Kükenthal mentioned that longitudinal rows were not so clear proximally in the polyp. Kükenthal uses marginal thorns to separate some species and the length of the marginal thorn was shown to vary within the same colony.

Kükenthal also often uses the number of polyps per cm of branch as a specific character. The species *Th. longispinosa, Th. hilgendorfi, and Th. typica*, for example, are separated into different species solely on the number of whorls in one cm of the branches. The type specimen of the species *Th. tenuisquamis* appears to be a young specimen of the type specimen of *Th. grandiflora.*

Kükenthal introduced the species *Th. striata* to accommodate a specimen which had different ornamentation of the scales. *Th. striata* is very similar to *Th. variabilis* var. *brevispinosa.* It would have been referred to it was it not for its striate polyp scales. In Chapter 2, it was shown that the ornamentation of the scales vary within the same colony. Perhaps, *Th. striata* is in fact different in this aspect but to confirm that the scales of this species should be better examined.

The species *Th. clavata* was introduced to accommodate a specimen which showed sites of agglomeration of polyps. Through personal observation, it was possible to verify that these agglomerations were caused by the presence of a polychaete commensal.

Table 7.2 - KEY FOR THE SUBGENERA AND SPECIES OF THE GENUS *Thouarella* - (after Kükenthal, 1924)

1 - Body scales in eight longitudinal rows .....Subgenus *Amphilaphis*
Body scales in fewer than eight longitudinal rows.....2
2 - Marginal scales bearing thorn.....3
Marginal scales without thorn .....Subgenus *Epithouarella*
3 - Polyps placed in pairs or in whorls of up to 4 .....Subgenus *Euthouarella*
Polyps placed singly .....Subgenus *Parathouarella*

Subgenus *Amphilaphis*

1 - Polyps placed in pairs or in whorls 2
Polyps placed singly 4
2 - Marginal scales bearing thorn .....*Th.(A.) regularis*
Marginal scales without thorn 3
3 - 5 scales in each longitudinal abaxial row .....*Th.(A.) parva*
6 scales in each longitudinal abaxial row .....*Th.(A.) abietina*
4 - Marginal scales bearing thorn 5
Marginal scales without thorn 6
5 - Polyps sparsely distributed .....*Th.(A.) dispersa*
Polyps densely distributed .....*Th.(A.) superba*
6 - 7 to 8 scales on each longitudinal abaxial row .....*Th.(A.) grandiflora*
8 to 12 scales on each longitudinal abaxial row .....*Th.(A.) plumacea*

Subgenus *Epithouarella*

1 - 7 to 8 scales on each longitudinal abaxial row 2
9 to 10 scales on each longitudinal abaxial row .....*Th.(E.) crenelata*
2 - Polyps regularly distributed on the branches .....*Th.(E.) affinis*
Polyps densely distributed on the branches which attains a cylindrical shape .....*Th.(E.) chillensis*

Table 7.2 Contd

Subgenus *Euthouarella*

1 - With side branches 2
Without side branches .....*Th.(E.) coronata*
2 - Side branches surrounding the stem 3
Side branches placed in one plane 4
3 - 5 whorls on 1 cm of branch .....*Th.(E.) longispinosa*
6 whorls on 1 cm of branch .....*Th.(E.) hilgendorfi*
10 to 11 whorls on 1 cm of branch .....*Th.(E.) typica*
4 - 4 whorls on 1 cm of branch .....*Th.(E.) laxa*
5 whorls on 1 cm of branch 5
6 whorls on 1 cm of branch .....*Th.(E.) tydemany*
7 to 8 whorls on 1 cm of branch 6
5 - Polyps placed in pairs .....*Th.(E.) moseleyi*
Polyps placed in pairs or in whorls of 3 .....*Th.(E.) flabellata*
6 - Polyps in pairs .....*Th.(E.) tenuisquamis*
Polyps in whorls of 3 .....*Th.(E.) carinata*

Subgenus *Parathouarella*

1 - 4 to 6 scales on each longitudinal abaxial row 2
8 scales on each longitudinal abaxial row .....*Th.(P.) koellikeri*
9 to 10 scales on each longitudinal abaxial row .....*Th.(P.) antarctica*
2 - Polyp scales radially striated .....*Th.(P.) striata*
Polyp scales not radially striated 3
3 - Polyps regularly distributed on the branches 4
Polyp often densely distributed and forming clumps .....*Th.(P.) clavata*
4 - 4 scales on each longitudinal abaxial row .....*Th.(P.) variabilis*
6 scales on each longitudinal abaxial row .....*Th.(P.) versluysi*

Based on these observations, it is clear that Kükenthal's key should be restructured. Perhaps, with the information obtained in this study a more reliable key could be introduced. However, a reliable work could only be done when enough specimens of each of the species have been examined. Without it, only suggestions can be made. It is outside the scope of this thesis to present a definite classification of the genus *Thouarella*. It was the aim of this work to evaluate the taxonomic status of the genus and to provide sufficient knowledge, based on which a major work could be done. Most natural history museums holding Antarctic or deep-sea collections, have a great amount of unidentified specimens of primnoids that could be referred to the genus *Thouarella*. If these specimens are examined and identified, much can be done for the adequate characterization of the species of this genus.

REFERENCES

ACHITUV, Y. & Y. Benayahu. 1990. Polyp dimorphism and functional, sequential hermaphroditism in the soft coral *Heteroxenia fuscescens* (Octocorallia). *Mar. Ecol. Prog. Ser.*, **64**: 263-369.

ALLEMAND, D. & M-C. Grillo. 1992. Biocalcification mechanism in Gorgonians: ^{45}Ca uptake and deposition by the Mediterranean red coral *Corallium rubrum*. *Jnl. Exp. Zool.*, **262**: 237-246.

ARNAUD, P.M. 1977. Adaptations within Antarctic benthic ecosystems. In: *Adaptations within Antarctic ecosystems*, edited by Llano, G.A., Gulf Publ. Co., Houston, pp. 135-157.

ARNAUD, P.M., K. Jazdzewski, P. Presler & J. Sicinski. 1986. Preliminary survey of benthic invertebrates collected by Polish Antarctic Expeditions in Admiralty Bay, King George Island, South Shetland Islands, Antarctica. *Pol. Polar. Res.*, **7**: 7-24.

ARNTZ, W.E., T. Brey & V.A. Gallardo. Antarctic Zoobenthos. *Alfred Wegener Institute for Polar and Marine Research, Bremerhaven*. (In press)

ASHWORTH, J.H. 1899. The structure of *Xenia hicksoni*, nov. sp., with some observations on *Heteroxenia elizabethane*, Kölliker. *Quarterly Journ. Microsc. Sci.*, (new series) **42** (3): 245-304, Pls 23-27.

ASHWORTH, J.H. 1900. Report on the Xeniidae collected by Dr Willey. *Zoological results based on material from new Britain, New Guinea, Loyalty Islands and elsewhere, collected during the years 1895, 1896 and 1897, by Arthur Willey*, part 4: pp 509-530, Pls. 52-53.

BABCOCK, R. 1990. Reproduction and development of the blue coral *Heliopora coerulae* (Alcyonaria: Coenothecalia). *Marine Biology*, **104**: 475-481.

BAKUS, G. 1981. Chemical defense mechanisms on the Great Barrier Reef, Australia. *Science*, **211**: 497-498.

BAKUS, G. & M. Thun. 1979. Bioassays on the toxicity of Caribbean sponges. *Collog. Int. C.N.R.S.*, **291**: 417-442.

BAYER, F.M. 1955. Contributions to the nomenclature, systematics, and morphology of Octocorallia. *Proc. U.S. Natnl. Mus.* **105**: 207-220, Pls 1-8.

BAYER, F.M. 1956. Octocorallia. In: Moore, R.C. (ed.) *Treatise on Invertebrate Paleontology. Part F. Coelenterata. Geol. Soc. Amer.* and Univ. Kansas Press, 163-231, 134-162 Figs.

BAYER, F.M. 1973. Colonial organization in octocorals. In: Boardman, R.S. *et al.* (eds.) *Animal Colonies; their development and function through time*. Stroudsburg, Pennsylvania, 69-93, Figs. 1-23.

BAYER, F.M. 1974. Studies on the anatomy and histology of *Plexaura homomalla* in Florida. In: Bayer, F.M. & A.J. Weinheimer (eds.). *Idem. Stud. Trop. Oceanogr.*, **12**: 62-100, 1-62 Figs.

BAYER, F.M. 1981a. Status of knowledge of octocorals of world seas. In: *Seminarios de Biologia Marinha*, Academia Brasileira de Ciencias, Rio de Janeiro, 3-12, 1 Fig.

BAYER, F.M. 1981b. Utilization and conservation of Plexaura homomalla. In: *Seminarios de Biologia Marinha*, Academia Brasileira de Ciencias, Rio de Janeiro, 13-17.

BAYER, F.M. 1981c. Recent advances in research on octocorals. In: *Seminarios de Biologia Marinha*, Academia Brasileira de Ciencias, Rio de Janeiro, 19-102, Figs 1-7.

BAYER, F.M. 1981d. Key to the genera Octocorallia exclusive of Pennatulacea (Coelenterata: Anthozoa), with descriptions of new taxa. *Proc. biol. Soc. Wash.*, **94** (3): 902-947, Figs 1-80.

BAYER, F.M. 1982. Some new and old species of the primnoid genus *Callogorgia* Gray, with a revalidation of the related genus *Fanellia* Gray (Coelenterata: Anthozoa). *Proc. Biol. Soc. Wash.*, **95** (1): 116-160.

BAYER, F.M., M. Grasshoff & J. Verseveldt. 1983. Illustrated trilingual glossary of morphological and anatomical terms applied to Octocorallia. E.J. Brill & W.Backhuys (eds.), Leiden, 75pp.

BAYER, F.M. & J. Stefani. 1988. Primnoidae (Gorgonacea) de Nouvelle-Caledonie. *Bull. Mus. Nat. Hist, Nat. Paris*, **193**: 449-518.

BEAMS, H.W. & R.G., Kessel. 1983. Cnidaria. In: Adiyodi K.G. & Adiyodi R.G. (eds.). *Reproductive biology of invertebrates, Vol. I. Oogenesis, oviposition, and oosorption.* John Wiley & Sons, New York, 31-66.

BEHETI-GONZALEZ, P.A. & M. Guardiola. 1979. Ciclo reproductivo de *Plexaura homomalla* (Esper, 1792) forma *kuekenthali* Moser, 1921 (Gorgonacea). *Acad. Serie. Gencais de Cuba*, **3**: 99-104.

BENAYAHU, Y. 1989. Reproductive cycle and developmental processes during embryogenesis of *Clavularia hamra* (Cnidaria, Octocorallia). *Acta. Zoologica.*, Stockholm, **70** (1): 29-36.

BENAYAHU,Y. 1990. Reproduction and developmental pathways of Xeniidae (Octocorallia: Alcyanacea). *Hydrobiologia.*

BENAYAHU,Y. 1991. Reproduction and developmental pathways of Red Sea Xeniidae. *Hydrobiologia*, **216/217**: 125-130.

BENAYAHU, Y. & Y. Loya. 1983. Surface brooding in the Red Sea soft coral *Parerythropodium fulvum fulvum* (Forskal 1775). *Biol. Bull. mar. biol. Lab.*, Woods Hole, **165**: 353-369.

BENAYAHU, Y. & Y. Loya. 1984. Life history studies on the Red Sea soft coral *Xenia macrospiculata* Gohar, 1940. I. Annual dynamics of gonadal development. *Biol. Bull. mar. biol. Lab.*, Woods Hole, **166**: 32-43.

BENAYAHU, Y. & Y. Loya. 1985. Settlement and recruitment of a soft coral: why is *Xenia macrospiculata* a successful colonizer? *Bull. Mar. Scie.*, **36**: 177-188.

BENAYAHU, Y. & Y. Loya. 1986. Sexual reproduction of a soft coral: synchronous and brief annual spawning of *Sarcophyton glaucum* (Quoy and Gaimard). *Biol. Bull. mar. biol. Lab.*, Woods Hole, **170**: 32-42.

BENAYAHU, Y., Y. Achituv & T. Berner. 1988. Embryogenesis and acquisition of algal symbionts by planulae of *Xenia umbellata* (Octocorallia: Alcyonacea). *Marine Biology*, **100**: 93-101.

BENAYAHU, Y., T. Berner & Y. Achituv. 1989. Development of planulae within a mesogleal coat in the soft coral *Heteroxenia fuscescens*. *Marine Biology*, **100**: 203-210.

BERKMAN, P.A., D.S. Marks & G.P. Shreve. 1986. Winter sediment resuspension in McMurdo Sound, Antarctica, and its ecological implications. *Polar Biology*, **6**: 1-3.

BIRKELAND, C. 1969. Consequences of differing reproductive and feeding strategies for the dynamics and structure of an association based on the single prey species, *Ptilosarcus guerneyi* (Gray), Ph.D. Thesis, University of Washington.

BODUNGEN, B. 1986. Phytoplankton growth and krill grazing during spring in the Bransfield Strait, Antarctica -implications from sediment trap collections. *Polar Biology*, **6**: 153-160.

BRAZEAU, D.A. & H.R. Lasker. 1988. Inter- and intraspecific variation in gorgonian colony morphology: quantifying branching patterns in arborescent animals. *Coral Reefs*, **7**: 139-143.

BRAZEAU, D.A. & H.R. Lasker. 1989. The reproductive cycle and spawning in a Caribbean gorgonian. *Biol. Bull.*, **176**: 1-7.

BRAZEAU, D.A. & H.R. Lasker. 1990. Sexual reproduction and external brooding by the Caribbean gorgonian *Briareum asbestinium*. *Marine Biology*, **104**: 465-474.

BROCH, H. 1965. Some octocorals from antarctic waters. Scientific Results of the "Brategg" Expedition, 1947-48. *Chr. Christensens Hvalfangstmuseum i Sandefjord, Pub.* **26**: 19-38, Pls 1-7.

BUDDEMEIER, R.W., R.C. Schneider & S.V. Smith. 1981. The alkaline earth chemistry of corals. In: *Proc. 4th Int. Coral Reef Symp., Manila*, **2**: 81-85.

BULLIVANT, J.S. & J.H. Dearborn (eds). 1967. The fauna of the Ross Sea. *New Zealand Department of Scientific and Industrial Research Bulletin*, **176**: 1-76.

CARLISLE, J.G., C.H. Turner & E.E. Ebert. 1964. Artificial habitat in the marine environment. *Calif. Fish Game, Fish Bull.*, **124**: p.66.

CARY, L.R. 1914. Observations upon the growth and ecology of gorgonians. *Carnegie Inst. Wash., Pap. Tortugas Lab.*, **5**: 81-90.

CHAMBERLAIN, J.A. & R.R. Graus. 1975a. Water flow and hydromechanical adaptations of branched reef corals. *Bull. Mar. Sci.*, **25** (1): 112-125.

CHAPMAN, G. 1974. The skeletal system. In: *Coelenterate Biology: Reviews and new perspectives*, Academic Press, London, 93-128.

CHAVES, K.E. 1954. Aspects on the biogeochemistry of magnesium. 1. Calcareous marine organisms. *J. Geol.*, **62:** 266-283.

CHIA, F.S. & M.A Rostron. 1970. Some aspects of the reproductive biology of *Actina equina* (Cnidaria: Anthozoa). *J. mar. biol. Ass. U.K.*, **50**: 253-264.

CHIA, F.S. & B.J. Crawford. 1973. Some observations on gametogenesis, larval development and substratum selection of the sea pen *Ptilosarcus guerneyi*. *Marine Biology*, **23**: 73-82.

CLARK, G.R. 1974. Growth lines in invertebrate skeletons. *A. Rev. Earth Planet. Sci.* **2**: 77-99.

CLARK, F.W. & W.C. Wheeler. 1922. The inorganic constituents of marine invertebrates. *Prof. Paper U.S. Geol. Surv.*, 124: 1-62.

CLARKE, A. 1984. Lipid composition of two species of *Serolis* (Crustacea, Isopoda) from Antarctica. *Br. Antarct. Survey. Bull.*, **64**: 37-53.

CLARKE, A. 1988. Seasonality in the Antarctic marine environment. *Comparitive Biochemistry and Physiology*, **90B**: 461-473.

CLARKE, A. 1990. Temperature and evolution: Southern Ocean cooling and the Antarctic marine fauna. In: *Antarctic Ecosystems: ecological change and conservation*, Kerry K.R. & Hempel G. (Eds.), Springer-Verlag, Berlin, 9-22.

CLARKE, A. 1992. Reproduction in the cold: Thorson revisited. *Intern. J. Invert. Reprod.*, **22** (1-3): 175-184.

CLARKE, A. & J.A. Crame. 1989. The origin of the Southern Ocean marine fauna. In: *Origins and evolution of the Antarctic biota*, ed. Crame, J.A., *Geological Society Special Publication*, **47**: 253-268.

COLL, J.C., S. La Barre, P.W. Sammarco, W.T. Williams & G.J. Bakus. 1982a. Chemical defences in soft corals (Coelenterata: Octocorallia) of the Great Barrier Reef: A study of comparative toxicities. *Mar. Ecol. Prog. Ser.*, **8**: 271-278.

COLL, J.C., B.F. Bowden, D.M. Tapiolas & W.C. Dunlap. 1982b. *In situ* isolation of allelochemicals released from soft corals (Coelenterata: Octocorallia): A totally submersible sampling apparatus. *J. Exp. Mar. Biol. Ecol.*, **60**: 293-299.

DALYELL, J.G. 1838. On the reproduction of the *Virgularia* or *Pennatula mirabilis*. *Edinburgh New Philos. Journ.*, **27**: 379-381.

DAWSON, R., W. Schramm & M. Bolter. 1985. Factors influencing the production, decomposition and distribution of organic and inorganic matter in Admiralty Bay, King George Island. In: Siegfried, W.R., *et al.* (eds.), *Antarctic nutrient cycles and food webs*, Springer-Verlag, Berlin, 109-114.

DAYTON, P.K. 1990. Polar benthos. In: *Polar oceanography, Par B: Chemistry, biology and geology*, edited by Smith, W.O., Academic Press, London, pp. 631-685.

DEER, Howie & Zussman, 1966. An Introduction to the rock forming minerals. Commonwealth Printing Press Ltd., 528pp.

DINESEN, Z.D. 1985. Aspects of the life history of a stolon bearing species of *Efflatuonaria* (Octocorallia: Xeniidae). In: Gabrie, C. *et al.* (eds.) *Antenne Museum-EPHE*, Proc. 5th Int. Coral Reef Congr., **6**: 89-94.

DRUFFEL, E.R.M., L.L. King, R.A. Belastock & K.O. Buesseler. 1990. Growth rate of a deep-sea coral using $_{210}$Pb and other isotopes. *Geoch. Cosmoch. Acta*, **54**: 1493-1500.

DUNBAR, M.J. 1968. Ecological development in polar regions: a study in evolution. Prentice-Hall Inc., Englewood Cliffs, New Jersey, 1-113.

DUSTAN, P. 1975. Growth and form in the reef-building coral *Montastrea annularis. Marine Biology,* **33**: 101-107.

ECKELBARGER, K.J. & R.L. Larson. 1988. Ovarian morphology and oogenesis in *Aurelia aurita* (Scyphozoa: Semaeostomae): ultrastructural evidence of heterosynthetic yolk formation in a primitive metazoan. *Mar. Biol.*, **100**: 103-115.

EMLET, R.B., L.R. McEdward & R.R Strathmann. 1987. Echinoderm larval ecology viewed from the egg. In: Jangoux, M. & Lawrence, J,M, (eds.), *Echinoderm studies*, **2**: 55-136, A.A. Balakema, Rotterdam.

EREZ, J. 1978. Vital effect on stable-isotope composition seen in Foraminifera and coral skeletons. *Nature*, **273**: 199-202.

ESFORD, L.E. & J.C. Lewis. 1990. Stiffness of Caribbean gorgonians (Coelenterata, Octocorallia) and Ca/Mg content of their axes. *Mar. Ecol. Prog. Ser.* **67** (2): 189-200.

FADALALLAH, Y.H. 1983. Sexual reproduction, development and larval biology in scleractinian corals. *Coral Reefs*, **2**: 129-150.

FADALALLAH, Y.H. & J.S. Pearse. 1982a. Sexual reproduction in solitary corals: overlapping oogenic and brooding cycles, and benthic planulae in *Balanophyllia elegans. Marine Biology*, **71**: 223-231.

FADALALLAH, Y.H. & J.S. Pearse. 1982b. Sexual reproduction in solitary corals: synchronous gametogenesis and broadcast spawning in *Paracyathus stearnsii. Marine Biology*, **71**: 233-239.

FARRANT, P. 1985. Reproduction in the temperate Australian soft coral *Capnella gaboensis.* In: *Proceedings of the 5th Int. Coral Reef Congr. Tahiti*, **4**: 319-324.

FAUTIN, D.G., & R.N. Mariscal. 1991. Cnidaria: Anthozoa. In: *Microscopic anatomy of invertebrates, Volume 2: Placozoa, Porifera, Cnidaria and Ctenophora*, Wiley-Liss, Inc., 267-358.

FISHELSON, L. 1973. Ecological and biological phenomena influencing coral species composition on the reef tables at Eilat (Gulf of Aqaba, Red Sea). *Mar. Biol.*, **19**: 183-196.

GILI, J.M. & I.A. Garcia. 1985. Biologia de *Paramuricea Clavata* (Anthozoa: Octocorallia) a les costes Catalanes. I Creixement i caracteristiques generals. *Bullt. Inst. Cat. Hist. Nat.*, (Sec. Zool.), **52** (6): 25-32.

GOHAR, H.A.F. 1940. Studies on the Xeniidae of the Red Sea. *Pub. Mar. Biol. St. Ghardaqa (Red Sea)*, **2**: 27-118.

GOHAR, H.A.F. 1948. A description and some biological studies of a new alcyonarian species *Clavularia hamra* Gohar. *Pub. Marine Biol. Sta. Ghardaqa (Red Sea)*, **6**: 3-33, Pls 1-3.

GOHAR, H.A.F. & H.M. Roushdy. 1961. On the embryology of the Xeniidae (Alcyonaria)(with notes on the extrusion of larvae). *Pub. Marine Biol. Sta. Ghardaqa (Red Sea)*, **11**: 43-70, Pl. 1-3.

GOLDBERG, W.M. 1974. Evidence of a sclerotized collagen from the skeleton of a gorgonian coral. *Comp. Biochem. Physiol.* **49B**: 525-529.

GOLDBERG, W.M., 1976. A comparative study of the chemistry and structure of gorgonian and antipatharian coral skeletons. *Mar. Biol.* **35**: 253-267.

GOLDBERG, W.M. 1978. Chemical changes accompanying maturation of the connective tissue skeletons of gorgonian and antipatharian corals. *Mar. Biol.* **49**: 203-210.

GOLDBERG, W.M. 1991. Chemistry and structure of skeletal growth rings in the black coral *Antipathes fiordensis* (Cnidaria, Antipatharia). *Hydrobiologia*, **216**: 403-409.

GOLDBERG, W.M. & R.D. Hamilton. 1974. The sexual cycle in *Plexaura homomalla. Stud. Trop. Oceanogr. Miami*, **12**: 58-61.

GOREAU, T.J. 1977. Carbon metabolism in calcifying and photosynthetic organisms: theoretical models based on stable isotope data. *Proc. 3rd. Int. Coral Reef Symp.*, **2**: 395-401.

GOSLINE, J.M. 1971. Connective tissue mechanics of *Metridium senile, J. exp. Biol.*, **55**: 775-795.

GRANGE, K.R. 1985. Distribution, standing crop, population structure, and growth rates of black coral in the southern fiords of New Zealand. *NZ J. Mar. Freshw. Res.* **19**: 467-475.

GRANT, R.E. 1826. Observations on the spontaneous motions of the ova of the *Campanularia dichotoma, Gorgonia verrucosa, Caryophyllaea calycularis* etc. *Edingburgh New Philos. Journ.* **1**: 150-156.

GRANT, R.E. 1827. Notice regarding the structure and mode of generation of the *Virgularia* and the *Pennatula phosphorea. Edingburgh Journ. Sci.* **7**: 330-335. (Also in *Frorieps Notizen* **19**, 337-341)

GRANT, R.E. 1829. Further observations on the generation of the *Virgularia mirabilis. Edinburgh Journ. Sci.* **10**: 350-351. (Also in *Frorieps Notizen* **24**: 247)

GRAVIER, C. 1913. Seconde expedition antartique francaise (1908-1910). Alcyonaires (ire note preliminaire). *Bull. Mus. Nat. Hist. Nat. Paris*, **19**: 451-455.

GRAVIER, C. 1914. Alcyonaires. Deuxieme expedition antartique francaise (1908-1910). *Sciences naturelles: Documents scientifiques*, 1-118, Pls. 1-10.

GRAY, J.E. 1857. Description of the new genera of Gorgonidae. *Proc. Zool. Soc. London.*, 1857: 158-159, 8 Pls.

GRAY, J.E. 1859. Description of some new genera of lithophytes or stony zoophytes. *Proc. Zool. Soc. London*, 1859: 479-486.

GRAY, J.E. 1870. Catalogue of the lithophytes or stony corals in the collection of the British Museum. London, British Museum. [i-iv] + Pls 1-51.

GRAY, J.E. 1872. *Thouarella antarctica* from the Falkland Islands. *Ann. Mag. Nat. Hist.*, **9** (4): 481.

GRIGG, R.W. 1970. Ecology and population dynamics of the gorgonians, *Muricea californica* and *Muricea fruticosa.* Ph.D. Thesis, Univ. Calif., San Diego. pp. 261.

GRIGG, R.W. 1972. Orientation and growth form of sea fans. *Limnology and Oceanography,* **17** (2): 185-192.

GRIGG, R.W. 1974. Growth rings: annual periodicity in two gorgonian corals. *Ecology,* **55** (4): 876-881.

GRIGG, R.M. 1975. The commercial potential of precious corals in the western Caroline Islands, Micronesia. *Univ. Hawaii Sea Grant Tech. Rep. UNIHI-Sea Grant AR-75-03*, i-v + 1-14, Figs 1-2.

GRIGG, R.M. 1976. Fishery management of precious and stony corals in Hawaii. *Univ. Hawaii Sea Grant Tech. Rep. UNIHI-Sea Grant TR-77-03*: i-viii + 1-48, Figs 1-23.

GRIGG, R.W. 1977. Population dynamics of two gorgonian corals. *Ecology*, **55** (4): 876-881.

GRIGG, R.W. 1979. Reproductive ecology of two species of gorgonian corals: relation to vertical and geographical distribution. In: Stancyk, S.E.(ed.), *Reproductive ecology of marine invertebrates*, Univ. South Carolina Press, Columbia, 41-59.

HART, T.J. 1942. Phytoplankton periodicity in Antarctic surface waters. *'Discovery' Rep.* **8**: 329-396.

HARTNOLL, R.G. 1975. The annual cycle of Alcyonium digitatum. *Est. Coastal Mar. Sci.*, **3**: 71-78.

HARTNOLL, R.G. 1977. Reproductive strategy in two British species of *Alcyonium.* In: Keegan, P.O. *et al.* (eds.), *Biology of benthic organisms*, Pergamon Press, Oxford, 321-328.

HEMPEL, G. 1985. Introductory theme paper: On the biology of polar seas, particularly the southern ocean. In: *Marine Biology of polar regions and effects of stress on marine organisms*, edited by J.S. Gray & Christiansen, M.E., Wiley, London, pp. 3-33.

HICKSON, S.J. 1895. The anatomy of *Alcyonium digitatum. Quart. Journ. Microsc. Sci.*, **37 (4)**: 343-388, Pls. 36-69.

HICKSON, S.J. 1907. Coelenterata I. Alyconaria. *National Antarctic (Discovery) Expedition, Nat. Hist.*, **3**: 1-15, Pls. 2. British Museum.

HICKSON, S.J. 1940. The species of the genus *Acabaria* in the Red Sea. *Publ. Marine Biol. Sta. Ghardaqa (Red Sea)*, **2**: 3-22, Pls 1-2 (English with Arabic summary)

HILL, M.D., 1906. Notes on the maturation of the ovum, of *Alcyonium digitatum. Quarterly Journ. Microsc. Sci.* (new series), **49** (3): 493-505.

HORNE, A., G. Fogg & D. Eagle. 1969. Studies *in situ* of the primary production of an inshore Antarctic sea. *J.Mar. Biol. Assoc. U.K.*, **49**: 393-405.

HUNTLEY, M., D.M. Karl, P. Niiler & O. Holm-Hansen. 1991. Research on Antarctic coastal ecosystem rates (RACER): an interdisciplinary field experiment. *Deep-Sea Research*, **38** (8/9): 911-941.

HYMAN, L.H. 1940. *The invertebrates. Volume I. Protozoa through Ctenophora.* New York, McGraw-Hill Book Co. pp i-x + 1-716, Figs 1-221.

JABLONSKI, D. & R.A. Lutz. 1983. Larval ecology of marine benthic invertebrates: paleobiological implications. *Biol. Rev.*, **58**: 21-89.

JAZDZEWSKI, K., W. Jurasz, W. Kittel, E. Presler, P. Presler & J. Sicinski. 1986. Abundance and biomass estimates of the benthic fauna in Admiralty Bay, King George Island, South Shetland Islands. *Polar Biology*, **6**: 5-16.

JEYASURIA, P. & J.C. Lewis. 1987. Mechanical properties of the axial skeletons on gorgonians. *Coral Reefs,* **5**: 213-219.

JUNGERSEN, H,F.E. 1888. Uber bau und entwicklung der kolonie von *Pennatula phosphorea*. L. Zeitschr. Wissenschaftl. Zool., **47**: 626-649, Pl. 39.

KIM, K., W.M. Goldberg & G.T. Taylor. 1992. Architectural and mechanical properties of the black coral skeleton (Coelenterata: Antipatharia): A comparison of two species. *Biol. Bull.*, **182**: 195-209.

KINGSLEY, R.J. & N. Watabe. 1984. Synthesis and transport of the organic matrix of the spicules in the gorgonian *Leptogorgia virgulata* (Lamark) (Coelenterata: Gorgonacea). *Cell Tissue Res.*, **235**: 533-538.

KINOSHITA, K. 1908. *Diplocalyptra*, eine neue Untergattung von *Thouarella* (Primnoidae). *Annot. Zool. Japan.* **7** (1): 49-60.

KINOSHITA, K. 1910. Ueber die Postembryonale Entwicklung von *Anthoplexaura dimorpha* Kükenthal. *Jnl. Coll. Sci. Univ. Tokyo*, **23**: Art 14, pp 14.

KINZIE, R.A. 1973. The zonation of West Indian Gorgonians. *Bull. Marine Sci.*, **23** (1): 93-155, Figs 1-34.

KITANO, Y., N. Kanamori & T. Oomori. 1971a. Measurements of distribution coefficients of strontium and barium between carbonate precipitate and solution - abnormally high values of distribution coefficients measured at early stages of carbonate formation. *Geochem. J.*, **4**: 183-206.

KITANO, Y., N. Kanamori, T. Oomori, S. Yoshioka & A. Tokuyama. 1971b. Factors controlling the trace element contents of calcareous marine skeletons. *J. Geol. Soc. Japan*, **77**: 535-545 (In Japanese).

KITANO, Y., M. Okumura & M. Idogaki. 1975. Incorporation of sodium, chloride and sulfate with calcium carbonate. *Geochem. J.*, **9**: 75-84.

KOEHL, M.A.R. 1984. How do benthic organisms withstand moving water? *Am. Zool.*, **24**: 57-70.

KÖLLIKER, R.A.V. 1865. Icones histologicae. oder Atlas der vergleichenden Gewebelehre. Zweite Abtheilung. Der feinere Bau der hoheren Thiere. Erstes Heft. Die Bindesubstanz der Coelenteraten. (reviewed by Verrill, 1866). Verlag von Wilhelm Englemann, Leipzig, 87-181, Pls. 10-19, 13 Figs.

KONISHI, K. 1981. Alcyonarian spiculite: Limestone on soft corals. In: *Proc. 4th Int. Coral Reef Symp., Manila*, **1**: 643-649.

KOWALEVSKY, A.O. & A.F. MARION. 1882. Sur le developpement des Alcyonaires. *C.R. Acad. Sci. Paris*, **95**: 562-565.

KOWALEVSKY, A.O. & A.F. MARION. 1883. Documents pour l'histoire embryonogenique des alcyonaires. *Ann. Mus. Hist. Nat. Marseille.* Zoologie 1 (Memoire No.4), 1-50, Pls.1-5.

KÜKENTHAL, W. 1907. Gorgoniden der Deutschen Tiefsee-Expedition. *Zool. Anzeiger.*, **31** (7): 202-212.

KÜKENTHAL, W. 1908. Diagnosen neuer Gorgoniden (4. Mitteilung). *Zool. Anzeiger.*, **33** (1): 9-20.

KÜKENTHAL, W. 1912. Die Alcyonaria der deuschen Sudpolar-Expedition 1901-1903. In: E. von Drgalski (ed.) *Deutsche Sudpolar-Expedition 1901-1903. 13 Band Zoologie*, **5** (3): 289-349, 20-23 Pls.

KÜKENTHAL, W. 1915. System und Stammesgeschichte der Primnoidae. *Zool. Anzeiger.*, **46** (5): 142-158.

KÜKENTHAL, W. 1919. Gorgonaria. *Wiss. Ergebn. deutschen tiefsee-Exped. "Valdivia"*, **13** (2):1-946, 30-89 Pls.

KÜKENTHAL, W. 1924. Gorgonaria. *Das Tierreich*, Walter de Gruyter & Co., Berlin, **47**: 1-478, 209 Figs.

KÜKENTHAL, W. & H. Gorzawsky. 1908. Japanische Gorgoniden. 1. Teil: Die familien der Primnoiden, Muriceiden und Acanthogorgidiiden. In: Doflien, F.(ed.), Beitrage zur Naturgeschichte Ostasiens. *Abhandl. math.-phys. Klasse K. Bayer. Akad. Wissensch., Suppl.-Bd.* **1** (3): 1-71, Pls. 1-4.

LABOREL, J. 1960. Contribution a l'etude directe des peuplement benthiques sciaphiles sur substrat rocheux en Mediterranee. *Rec. Trav. Stat. Mar. Endoume*, **33**: 117-173.

LACAZE-DUTHIERS, H. 1887. Sur le developpement des Pennatules (*Pennatula grisea*) et les bonnes conditions biologiques que presente le laboratoire Arago pour les etudes zoologiques. *C.R. Acad. Sci. Paris*, **104** (8): 463-469.

LANG DA SILVEIRA, F. & T. Van't Hof. 1977. Regeneration in the gorgonian *Plexaura flexuosa* (Cnidaria, Octocorallia). *Bijdr. Dierk.*, **47** (1): 98-108.

LASKER, H.R. 1981. A comparison of the particle feeding abilities of three species of gorgonian soft coral. *Mar. Ecol. Prog. Ser.*, **5**: 61-67.

LAWSON, G.S. 1990. The Ecology of *Acanella arbuscula* from 2000m in the North East Atlantic Ocean. MSc Thesis, University of Southampton, England.

LE JEHAN, S. & P. Treguer. 1985. The distribution of inorganic nitrogen, phosphorus, silicon and dissolved organic matter in surface and deep waters of the Southern Ocean. In: Siefried, W.R. *et al.* (eds.) *Antarctic Nutrient Cycles and Food Webs*, Springer-Verlag, Berlin, 22-29.

LEVERSEE, G.J. 1976. Flow and feeding in fan-shaped colonies of the gorgonian coral *Leptogorgia. Biol. Bull.*, **151**: 344-356.

LEWIS, J.B. 1981. Estimates of secondary production of coral reefs. In: *Proc. 4th Int. Coral Reef Symp., Manila*, **2**: 369-373.

LEWIS, J.C. & E. Von Wallis. 1991. The function of surface sclerites in gorgonians (Coelenterata, Octocorallia). *Biol. Bull.*, **181** (2): 275-288.

LEWIS, J.C., T.F. Barnowski & G.J. Telesnicki. 1992. Characteristics of carbonates of gorgonian axes (Coelenterata, Octocorallia). *Biol. Bull.*, **183** (2): 278-296.

LIPPS, J.H. & C.S. Hickman. 1982. Origin, age, and evolution of Antarctic and deep-sea faunas. In: *The environment of the deep sea*, edited by Ernst, W.G. & Morin, J.G., Prentice-Hall, Eaglewood Cliffs, New Jersey, pp. 324-356.

LOWENSTAM, H.A. 1964. Coexisting calcites and aragonites from skeletal carbonates of marine organisms and their strontium and magnesium contents. *In:* Miyake, Y. & T. Koyama (eds.) *Recent researches in the fields of hydrosphere. Atmosphere and nuclear geochemistry.* Maruzen Co. Ltd., Tokyo.

MATTHEWS, A. 1916. The development of *Alcyonium digitatum*, with some notes on the early colony formation. *Quarterly Journ. Microsc. Sci.* (new series), **62**: 42-94, 51 Figs., Pls 3-5.

MAY, W. 1899. Alcyonarien. *Hamburger Mag. Samm.*, L. Friederichsen & Co., Hamburg, 1-22.

McCLINTOCK, J.B. & J.S. Pearse. 1986. Organic and energetic content of eggs and juveniles of Antarctic echinoids and asteroids with licithotrophic development. *Comp. Biochem. Physiol.*, **85A**: 341-345.

MEIGEN, W. 1903. In: *Naturf. Gessel. Freiburg Berlin*, **13**: p 13.

MENNEKING, F. 1905. Uber die Anordnung des Schuppen und das Kanalystem dei *Stachyodes ambigua* (Stud.), *Caligorgia flabellum* (Ehrb.), *Calyptrophora agassizii* (Stud.), *Amphilaphis abientina* (Stud.) und *Thouarella variabilis* (Stud.). *Arch. Naturgesch.*, **71** (3)1: 245-266, 8-9 Pls.

MILNE-EDWARDS, H. & J. Haime. 1857. Histoire naturelle des coralliares ou polypes proprement dits. al a *Libraire Encyclopedique de Roret*, Paris, **1**: 1-326, 8 Pls.

MOLANDER, A.R. 1929. Die Octactiniarien. *Further Zool. Res. Swedish Antarctic Exped. 1901-3*, **2** (2): 1-86, 1-5 Pls.

MOSELEY, H.W. 1876. On the structural relations of *Heliopora*. *Philos. Trans.*, **166**: 91.

MUZIK, K. & S.A. Wainwright. 1977. Morphology and habitat of five Fijian sea fans. *Bull. Mar. Sci.*, **27** (2): 308-337.

MÜLLER, R. 1910. Uber die Eireifung bei den Alcyonacean. *Ach. ges. Physiol.*, **136**: 141-146, 4 Figs.

MYRCHA *et al.* 1985. In: Siegfried, W.R. *et al.* (eds.) *Antarctic Nutrient Cycles and Food Webs*, Springer-Verlag, Berlin.

NEVILLE, A.C. & B.M. Luke. 1969. A two-system model for chitin-protein complexes in insect cuticles. *Tissue & Cell*, **1**: 689-707.

NONATO, E.F., T.A.S. Brito, P.C. Paiva & M.A.V. Petti. 1992. Programa Antartico Brasileiro: Projeto "Bionomia da fauna bentonica Antarctica". Atividades subaquaticas realizadas na Baia do Almirantado a partir da VI Expedicao (1988). *Relat. int. Inst. oceanogr. Univ. S Paulo*, **(33)**: 1-12.

NONATO, E.F., M.A.V. Petti, P.C. Paiva & T.A.S. Brito. 1992. Programa Antartico Brasileiro: amostragem de organismos bentonicos realizadas nas seis primeiras expedicoes (1982 a 1988), com participacao do N/Oc. "Prof. W. Besnard". *Relat. int. Inst. oceanogr. Univ. S Paulo*, **(32)**: 1-12.

NUTTING, C.C. 1908. Descriptions of the Alcyonaria collected by the U.S. Bureau of Fisheries Steamer "Albatross" in the vicinity of the Hawaiian Islands in 1902. *Proc. U.S. Natl. Mus.*, **34**: 543-601, 41-51 Pls.

NUTTING, C.C. 1912. Descriptions of the Alcyonaria collected by the U.S. Bureau of Fisheries Steamer "Albatross", mainly in Japanese waters during 1906. *Proc. U.S. Natl. Mus.*, **43**: 1-104.

OHDE, S. & Y. Kitano, Y. 1981. Behaviour of minor elements in the transformation of coral Aragonite to Calcite. In: *Proc. 4th Int. Coral Reef Symp., Manila*, **2**: 91-94.

ORDER, 1985. In: Siegfried, W.R. etal (eds.) *Antarctic Nutrient Cycles and Food Webs*, Springer-Verlag, Berlin.

PALMISANO, A.C. & C.W. Sullivan. 1985. Physiological responses of microalgae in the ice-platelet layer to low-light conditions. In: Siegfried, W.R. *et al.* (eds.) *Antarctic Nutrient Cycles and Food Webs*, Springer-Verlag, Berlin, 84-88.

PEARSE, J.S., J.B. McClintock & I. Bosch. 1991. Reproduction of Antarctic benthic marine invertebrates: tempos, modes, and timing. *American Zoologist*, **31**: 65-80.

PEARSON, T.H. & R. Rosenberg. 1987. Feast and famine: structuring factors in marine benthic communities. In: *Organization of communities in past and present*, edited by Gee, J.H.R. & Giller, P.S., Blackwell, Oxford, pp. 373-395.

PECHERZEWSKI, K. 1980. Distribution and quantity of suspended matter in Admiralty Bay, King George Island, South Shetland Islands. *Polish Polar Res.*, **1**: 75-82.

PIKE, J.E. 1974. The prostaglandins: their history, development and potential. *Stud. Trop. Oceanogr. Miami*, **12**: 9-16, 1-7 Figs.

PRESLER, P., 1980. Phenological and physiographical observations carried out during the first wintering at the Artowski station in 1977. *Pol. Arch. Hydrobiol.*, **27**: 245-252.

PRUSZAK, Z. 1980. Current circulation in the water of Admiralty Bay (region of Artowski Station on King George Island). *Polish Polar Res.*, **1**: 55-74.

RAKUZA-SUSZCZEWSKI, S. 1980. The role of near-shore research in gaining an understanding of the functioning of Antarctic ecosystems. *Pol. Arch. Hydrobiol.*, **27**: 229-233.

READ, 1970. Rutley's Elements of Mineralogy. Thomas Murby & Co. 560 pp.

REES, J.T. 1972. The effect of current on growth form in an octocoral. *J. Exp. Mar. Biol. Ecol.*, **10**: 115-124.

RICE, A.L., P.A. Tyler & G.J.L. Paterson. 1992. The Pennatulid *Kophobelemnon Stelliferum* (Cnidaria: Octocorallia) in the Porcupine Seabight (North-east Atlantic Ocean). *J. mar. biol. Ass. U.K.*, **72**: 417-434.

RIEDL, R. 1966. *Biologie der Meereshölen.* Verlag Paul Parey, Hamburg, 636 pp.

RIEDL, R. 1971. Water movement: animals. *In:* O. Kinne (ed.) *Marine ecology.* Wiley-Interscience, London. **1** (2): 1123-1150.

RINKEVICH, B. & Y. Loya. 1979. The reproduction of the Red Sea coral *Stylophora pistillata*. I. Gonads and planulae. *Mar. Ecol. Prog. Ser.* **1**: 133-144.

RINKEVICH, B. & Y. Loya. 1984. Does light enhance calcification in hermatypic corals? *Mar. Biol.*, **80**: 1-16.

ROULE, L. 1908. Alcyonaires. *Exped. Antarct. Franc. (1903-1905). Sciences Naturelles: Documentes Scientifiques*, **15**: 1-6. Pl 1.

ROULE, L. 1932. Observations sur l'ontogenese du Veretille (*Veretillum cynomorium* Pall.). *Arch. Zool. exp.gen. Paris*, **74**: 233-247.

SAKSHANG, E. & O. Holm-Hansen. 1984. Factors governing pelagic production in polar oceans. In: *Marine Phytoplankton and Productivity*, Holm-Hansen, O. *et al.* (eds.), Lecture Notes on Coastal and Estuarine Studies, Springer, Berlin, **8**: 1-16.

SAMMARCO, P.W. & J.C. Coll. 1988. The chemical ecology of alyconarian corals. *Bioorganic Marine Chemistry*, Springer-Verlag, New York, **2**: 87-116.

SAMMARCO, P.W., J.C. Coll, S. La Barre & B. Willis. 1983. Competitive strategies of soft corals (Coelenterata: Octocorallia): Allelopathic effects on selected Scleractinian corals. *Coral Reefs*, **1**: 173-178.

SAMP, R. 1980. Selected environmental factors in the waters of Admiralty Bay (King George Island, South Shetland Islands) December, 1978 - February, 1979. *Polish Polar Res.*, **1** (4): 53-66.

SCHWARTZMAN, G. & D.M. Opresko. 1992. Infrared spectrum of the skeletal axis of antipatharian corals (Cnidaria: Anthozoa). *Bull. Mar. Sci.*, **50** (2): 352-356.

SCHLICHTER, D. 1982. Epidermal nutrition of the Alcyonarian *Heteroxenia fuscescens* (Ehrb.): Absorption of dissolved organic material and lost endogeneous photosynthates. *Oecologia* (Berl.), **53**: 40-49.

SEBENS, K.P. 1983. The larval and juvenile ecology of the temperate octocoral *Alcyonium siderium* verril. I. substratum selection by benthic larvae. *Jnl. Exp. Mar. Biol. Ecol.*, **71**: 73-89.

SEBENS, K.P. 1984. Water flow and coral colony size: interhabitat comparison of the octocoral *Alcyonium siderium. Proc. Nat. Acad. Sci. USA*, **81**: 5473-5477.

SEBENS, K.P. & J.S. Miles. 1988. Sweeper tentacles in a gorgonian octocoral: morphological modifications for interference competition. *Biol. Bull.*, **175**: 378-387.

SPINDLER, M. & G.S. Dieckmann. 1991. Das Meereis als Lebensraum. *Spektrum der Wissenschaft*, **2**/1991, 48-57.

STIBANE, F.A. 1987. Primnoide Oktokorallen (Coelenterata: Anthozoa, Order Gorgonacea Lamouroux, 1816) aus dem Sudatlantik. *Mitt. hamb. zool. Mus. Inst., Hamburg*, **84** (10): 17-26.

STIMSON, J.S. 1978. Mode and timing of reproduction in some common hermatypic corals from Hawaii and Enewetak. *Marine Biology*, **48**: 173-184.

STUDER, T. 1878. Ubersicht der Anthozoa Alyconaria, welche wahrend der Reisse S.M.S Gazelle um die Erde gesammelt wurden. *Monatsber. K. preuss. Akad. Wiss. Berlin*, 1878: 632-688, Pls.1-5.

STUDER, T. 1887. Versuch eines Systemes der Alyconaria. *Arch. Naturgesch.*, **53** (1): 1-74, Pl. 1.

STUDER, T. 1894. Note preliminaire sur les alcyonaires. Reports on the dredging operations...carried on by the U.S. Fish Commission steamer "Albatross", during 1891...X. *Bull. Mus. Comp. Zool. Harvard*, **25** (5): 53-69.

SUZUKI, H. 1971. Notes on *Cornularia* (Stolonifera, Alcyonacea) found in the vicinity of Manazaru Marine Biological Laboratory. *Sci. Rep. Yokohama Natn. Univ.*, **II** (18): 1-6.

SVOBODA, A. 1970. Simulation of oscillating water movement in the laboratory for cultivation of shallow water sedentary organisms. *Helgoländer wiss. Meersunters.*, **20**: 676-684.)

SVOBODA, A. 1973. Beitrag zur Okologie, Biometrie und systematik der Mediterranean *Aglaophenia* Arten. (Hydroidea). Ph.D. dissertation, Vienna.

SVOBODA, A. 1976. The orientation of *Aglaophenia* fans to current in laboratory conditions (Hydrozoa, Coelenterata). *In:* G.O.Mackie (ed.) *Coelenterate Ecology and Behavior,* Plenum Press, New York. 41-48 pp.

SWART, P.K. 1981. The Carbon isotope composition of organic material in coral skeletons and its effect on early diagenesis. In: *Proc 4th Int. Coral Reef Symp., Manila*, **2**: 87-90.

SZFRANSKI, Z. & M. Lipski. 1982. Characteristics of water temperature and salinity at Admiralty Bay (King George Island, South Shetland Islands, Antarctica) during the austral summer 1978/79. *Pol. Polar Res.*, **3** (1-2): 7-24.

SZMANT, A.M. 1986. Reproductive ecology of Caribbean reef corals. *Coral Reefs*, **5**: 43-53.

SZMANT-FROELICH, A., P. Yevich & M.E.Q. Pilson. 1980. Gametogenesis and early development of the temperate coral *Astrangia danae* (Anthozoa: Scleractinia). *Biol. Bull. (Woods Hole, Mass.)*, **158**: 257-269.

TANNER, A.C. 1985. The role of bacteria in the cycling of nutrients within the maritime antarctic environment. In: Siegfried, W.R. etal (eds.) *Antarctic Nutrient Cycles and Food Webs*, Springer-Verlag, Berlin, 123-127.

THÉODOR, J. 1963. Contribution à l'étude des Gorgones. III. Trois formes adaptives d'*Eunicella stricta* en fonction de la turbulence et du courant. *Vie et Milieu,* **14**: 815-818.

THÉODOR, J. 1967. Contribution a l'etude desgorgones (VII): Ecologie et comportment de la planula. *Vie et Milieu*, **18** (2-A): 291-301, 1-8 Figs, 8 Pls.

THÉODOR, J., & M. Denizot. 1965. Contribution à l'étude des Gorgones. I. A propos de l'orientation d'organismes marins fixés végétaux et animaux en fonction du courant. *Vie et Milieu,* **16**: 237-241.

THOMSON, J.A. & W.D. Henderson. 1906. *An account of the alcyonarians collected by the Royal Indian Marine Survey Ship Investigator in the Indian Ocean. Part I. The Alcyonarians of the deep sea.* Calcutta, Indian Museum. pp i-xvi + 1-132, Pls. 1-10.

THOMSON, J.A. & J. Ritchie. 1906. The Alcyonarians of the Scottish National Antarctic Expedition. *Trans. Roy. Soc. Edinburgh*, **41** (3): 851-860, Pls 1-2. (January 18).

THOMSON, J.A. & D.L. Mackinnon. 1911. The alcyonarians of the "Thetis" Expedition. *Australian Mus. Mem.*, **4**: 661-695, Pls. 41-82. (January 27)

THOMSON, J.A. 1907. Note on *Primnoa reseda* from the Faeroe Channel, and on its embryos. *Proc. Roy. Phys. Soc. Edinburgh*, **17** (2): 65-72, Pls 1-2.

THOMSON, J.A. 1927. Alcyonaires provenant des campagnes scientifiques dy Prince Albert Ier de Monaco. *Res. Camp. Sci. Monaco*, **73**: 1-77, 1-6 Pls.

THOMSON, J.A. & N.I. Rennet. 1931. Alyconaria, Madreporaria, and Antipatharia. *Australasian Antarctic Exped. Sci. Rept.*, **9:**, 1-46, 8-14 Pls.

THOMSON, J.S. 1911. The Alcyonaria of the Cape of Good Hope and Natal. Gorgonacea. Proc. Zool. Soc. London, 1911: 870-893, Pls. 43-45. (December).

THOMSON, J.S. 1921. South African Alcyonacea. *Trans. Roy. Soc. South Africa*, **9** (2): 149-175, Pls. 5-6.

THORSON, G. 1950. Reproduction and larval ecology of marine bottom invertebrates. *Biological Review*, **25**: 1-45.

TILER, 1985. In: Siegfried, W.R. etal (eds.) *Antarctic Nutrient Cycles and Food Webs*, Springer-Verlag, Berlin.

TURSCH, B., J.C. Braekman, D. Daloze & M. Kaisin. 1978. Terpenoids from coelenterates. In: Scheur,P.J. (ed.) *Marine natural products, chemical and biological perspectives, Vol II.* Academic Press, New York, 247-296.

UEHARA, T, M. Sato & K. Yamazato. 1987. General description of developmental stages in a soft coral *Lobophytum Crassum* Marenzeller. *Galaxea*, **6**: 185-193.

UTINOMI, H. 1964. Some octocorals from the antarctic waters off Prince Harold Coast, *Jap. Antarctic Res. Exped. Sci. Rept.* (E), **23**: 1-14, Figs. 1-6, Pls. 1-2.

VALENCIENNES, A. 1846. Zoophytes., In: Dupetit-Thouars, A. *Voyage autour du monde sur la fregate "la Venus" pendant les annees 1836-1839*, Atlas de Zoologie, 1-15 Pls.

VAN ALSTYNE, K. & V.J. Paul. 1992. Chemical and structural defenses in the sea fan *Gorgonia ventalina*: effects against generalist and specialist predators. *Coral Reefs*, **11**: 155-159.

VAN ALSTYNE, K., C.R. Wylie, V.J. Paul & K. Meyer. 1992. Antipredator defenses in tropical pacific soft corals (Coelenterata: Alcyonacea). I. Sclerites as defenses against generalist carnivorous fishes. *Biol. Bull.*, **182** (2): 231-240.

VELIMIROV, B. 1973. Orientation in the sea fan *Eunicella cavolinii* related to water movement. *Helgoländer wiss. Meeresunters.* **24**: 163-173.

VELIMIROV, B. 1975. Wachstum und altersbestimmung der gorgonie *Eunicella cavolinii* (Growth and age determination in the sea fan *Eunicella cavolinii*). *Oecologia* **19**: 259-272.

VELIMIROV, B. 1976. Variations in growth forms of *Eunicella cavolinii* Koch (Octocorallia) related to intensity of water movement. *J. Exp. Mar. Biol. Ecol.*, **21**: 109-117.

VELIMIROV, B. 1980. Growth aspects and magnesium carbonate concentrations of the reef building octocoral *Heliopora coerulea* (Pallas) after transplantation. *Marine Ecology*, **1**: 155-168.

VELIMIROV, B. & E.L. Böhm. 1976. Calcium and magnesium carbonate concentrations in different growth regions of gorgonians. *Marine Biology*, **35**: 269-275.

VELIMIROV, B. & J. King. 1979. Calcium uptake and net calcification rates in the octocoral *Eunicella papillosa. Marine Biology*, **50**: 349-358.

VERSLUYS, J. 1906. Die Gorgoniden der Siboga Expedition II. Die Primnoidae. *Siboga-Exped. Monogr.*, **13A**: 1-187, Figs 1-178, 1-10 Pls.

VERMEIJ, G.J. 1978. *Biogeography and adaptation: patterns of marine life*, Harvard Univ. Press, Cambridge, Mass.

VIGHI, M. 1970. Ricerche sul ciclo riproduttivo del corallo rosso (*Corallium rubrum* (L.)) del promontorio di Portofino. *Atti Accad. naz. Lincei. Memorie*, **10** (8) (IIIa, 1): 1-26

VIGHI, M. 1972. Etude sur la reproduction du *Corallium rubrum* (L.). *Vie Milieu*, Ser. A, **23**: 21-32.

WÄGELE, J.W. & T.A.S. Brito. 1990. Die sublitorale Fauna der maritimen Antarktis. Erste Unterwasserbeobachtungen in der Admiralitätsbucht. *Natur und Museum*, **120**: 269-282.

WAINWRIGHT, S.A. & J.R. Dillon. 1969. On the orientation of sea fans (genus *Gorgonia*). *Biological Bulletin,* **136** (1): 130-139.

WAINWRIGHT, S.A. & M.A.R. Koehl. 1976. The nature of flow and the reaction of benthic Cnidaria to it. *In:* G.O.Mackie (ed.) *Coelenterate Ecology and Behavior,* Plenum Press, New York. 5-21 pp.

WALKER, T.A. & G.D. Bull. 1983. A newly discovered method of reproduction in gorgonian coral. *Mar. Ecol. Prog. Ser.*, **12**: 137-143.

WEBER, J.N. & P.M.J. Woodhead. 1970. Carbon and oxygen isotope fractionation in the skeletal carbonate of reef-building corals. *Chem. Geol.*, **6**: 93-123.

WEBER, J.N. & E.W. White. 1973. Carbonate minerals as precursors of new ceramic, metal and polymer materials for biomedical applications. *Miner. Sci. Engng.*, **5** (2): 151-165.

WEINBERG, S. 1979. The light-dependent behaviour of planula larvae of *Eunicella singularis* and *Corallium rubrum* and its implication for octocorallian ecology. *Bijdragen tot de Dierkunde*, **49** (1): 16-30.

WEINBERG, S. 1986. Mediterranean octocorallia: description of *Clavularia carpedium* n.sp. and synonymy of *Clavularia crassa* and *C. ochracea* on etho-ecological grounds. *Bijdr. Dierk.*, **56**: 232-246.

WEINBERG, S. & F. Weinberg. 1979. The life cycle of a gorgonian: *Eunicella singularis* (Esper, 1794). *Bijdragen tot de Dierkunde*, **48 (2)**: 127-139.

WELLS, J.W. 1963. Coral growth and geochronometry. *Nature,* **197**: 948-950.

WHITE, M.G. 1984. Marine benthos. In: *Antarctic ecology Vol 2*, edited by Laws, R.M., Academic Press, London, pp. 421-461.

WHITE, R.A., E. Shors & E.W. White. 1981. Uniform microporous biomaterials prepared from marine skeletal precursors. In: *Proc. 4th. Int. Coral Reef Symp., Manila*, **2**: 95-98.

WILBUR, K.M. 1976. Recent studies of invertebrate mineralization. In: *The Mechanisms of mineralization in the invertebrates and plants.* Watabe, N. & Wilbur, K.M. (eds.) Univ. South Carolina Press, Columbia, 79-108.

WILSON, E.B. 1880. The early stages of *Renilla*. *Amer. Journ. Sci. Arts*, **20** (3): 446-449, Pl. 9.

WILSON, E.B. 1883. The development of *Renilla*. *John Hopkins University Circulars*. **22**: 78-79. (April)

WRIGHT, E.P. & T. Studer. 1889. Report on the Alcyonaria collected by HMS Challenger during the years 1873-1876. *Rept. Sci. Res. Challenger. Zool.*, **31**: 1-314, 43 Pls.

YAMAZATO, K., M. Sato & H. Yamashiro. 1981. Reproductive biology of an Alcyonacean coral *Lobophytum crassum* Marenzeller. In: *Proc. of the 4th Int. Coral Reef Symp., Manilla*, **2**: 671-678.