Chapter 1 General Introduction

1.1 Introduction – monitoring incipient stress

Coral reefs are under threat from a variety of natural and anthropogenic sources of stress, and are perceived to be in decline worldwide (Hughes *et al.* 2003). Some of these stressors, particularly the anthropogenic impacts, might be reduced or mitigated if sufficient warning of their effects is known and acted upon. Even the "best practice" versions of current monitoring techniques are structured in ways that such knowledge too often becomes available only after a lethal stress event has begun. "Recovery" from the impact in such cases is usually more aptly described as "succession". Measurement of sublethal stress in corals has proven difficult because there are currently no simple techniques to quantify low- to medium-level stress in corals (Gates, 1990; Lesser *et al.*, 1990; Gates *et al.*, 1992; Al-Sofyani and Davies, 1993; Coles, 1993; Jokiel *et al.*, 1993; Brown, 1997, Jones, 1997).

Changes in the colour or integrity of coral tissues, including bleaching, tissue sloughing and patchy tissue necrosis have been used as indicators of stress in corals. Such responses, however, tend to occur only after high levels of stress are experienced (Gates, 1990; Jokiel *et al.*, 1993; Allison *et al.*, 1996; Jones *et al.*, 2000; Marshall & Baird 2000) and are typically followed quickly by death of the colony (Gates, 1990). Bleaching is one expression of stress in corals which is detectable without extensive lab work. Much research has been undertaken to understand the processes and

causes of coral bleaching; only to discover that it is a post-hoc symptom of extreme stress, rather than a syndrome related to a particular type of stress (Coles & Brown, 2003). Bleaching simply tells us that some stress has occurred which has caused the coral loss of symbionts, rather than identifying the cause or giving warning that damage is occurring. Partial mortality assessment suffers from an inability to record the causation of the event, and the fact that it records mortality, rather than measuring incipient stress. More subtle reactions to stress, such as decreased fecundity integrate the impact of stressors over longer periods of time, but have limited value as indicators because breeding seasons are generally short, and because they depend on the availability of prior or comparative fecundity measurements (Kojis and Quinn, 1981; Michalek-Wagner & Willis, 2001). Such measures are further complicated by taxonomic confusion, because many species within genera such as *Porites* or *Acropora* are hard to distinguish, yet have quite different reproductive patterns (Wolstenholme, 2004).

Thus, the problem with these commonly used indicators of stress in corals and coral communities is that they do not allow for rapid assessment and often indicate that corals are dying (Jokiel et al., 1993) rather than suffering incipient stress. Some technically brilliant concepts, such as PAM (Pulse Amplitude Modulated) Fluorometry were initially seen as a solution to this problem, yet address only a small part of it. PAM fluorometry has been used to measure the degree of non-photochemical quenching within the chloroplasts of zooxanthellae in hospite within corals

(Jones et al., 1998). The measurement is interpreted as the quantum photosynthetic yield (F_v/F_m) of the zooxanthellae; a quantity which is reported to be a fine predictor of damage to the xanthophyll cycling mechanism within the chloroplasts (Fitt et al. 2001, Warner et al. 1999). While PAM Fluorometry is, in fact, non-invasive and simple to operate and interpret within a restricted domain, it measures only a single parameter (photosynthetic potential) of the symbiotic zooxanthellae within the host coral, not the coral itself. It therefore addresses only a small subset of the problems that may afflict corals.

Similarly, monitoring of coral responses to environmental changes via measurements of skeletal growth parameters from coral skeletal density banding (Lough & Barnes, 1997) does not allow quick or frequent checks of the condition of massive corals. Variations in skeletal density or linear extension integrate over long periods of time to show departures from average growth during exceptional periods. The precision of skeletal density measurements is prone to fluctuate due to small variations in skeletal architecture and orientation; the sensitivity of such skeletal techniques is therefore limited. Moreover, the extraction of such records is laborious and destroys that part of the colony being examined; repeated measurements therefore risk harming the colony being assessed.

Monitoring of reef communities in order to detect subtle community changes, which may indicate environmental stress (e.g. Warwick & Clark, 1991, Risk et al., 2001) is a laborious process, which requires that a

baseline first be established against which unnatural change can be assessed. Systemic knowledge of the natural range of monitored parameters – including levels of incipient stress- is a necessary first step in developing this baseline. Detailed knowledge of reef community components is rare, however. Knowledge of the tolerance of the various components to equally diverse stressors is rarer still. Detecting stress prior to mortality in a single organism is not easy; the response of a community is more difficult to quantify, and can really only be estimated.

1.1.1 Massive *Porites* corals as indicators of community stress levels

The most useful tool for measuring stress in corals would detect changes in coral tissues before overall health of the colony is compromised. Since stress generally represents an additional cost to the energy budget of an organism, it follows that energy reserves are depleted during stress events. A truly useful monitoring tool would therefore measure some reflection of this energy depletion prior to a stage where the survivorship of the monitored coral is in doubt. A report by Barnes and Lough (1993) described systematic variations in the thickness of the tissue layer of massive *Porites* species at various locations on the Great Barrier Reef, Australia. They attributed these variations to differences in the quality of the habitat in terms of providing nutrition to the corals in the different locations. Further, they suggested that this characteristic might be used as the basis for the development of a reactive monitoring technique because of the wide distribution of massive *Porites*.

The tissue layer of *Porites* comprises a thin band at the outer edge of the colony. It is maintained as a thin band by periodic uplift of the lower margin of the tissue zone (Barnes & Lough, 1992). Preliminary studies (Barnes & Lough, 1992, True, 1995) indicated that variation in the thickness of the tissue layer in massive *Porites* occurs over several spatial scales, from within-reef to within-region. True (1995) also reported that tissue thickness decreases with water depth and that patterns of decrease are consistent within a species of *Porites*. Moreover, this study indicated that tissue thickness appeared to have a seasonal component. Barnes & Lough (1993) suggested that corals with the greatest tissue thickness possessed the highest "nutritional status". Moreover, they speculated that the thickness of this layer might change during stress events. However, these workers left open the question of seasonal changes and whether or not these putative changes might be recorded in the skeletons of corals.

1.1.2 Proxy environmental records from coral skeletons

Retrospective detection of stress events or environmental change is an increasingly important tool for understanding climate and environmental patterns beyond the range of instrumental data. Dendrochronologists realised that variations in the width of tree rings provide good environmental records where growth of trees is dominated by one out of a suite of environmental factors. For example, tree growth is dominated by the availability of water in trees growing at the edge of a desert (Fritts, 1976). Hence, variations in the width of tree-rings reflect variations in the

availability of water. Similarly, growth of trees close to the tree line on mountains is determined largely by temperature during the summer growing season. Thus, the width of the rings in such trees reflects variations in temperature between growing seasons. Signs left by stress events, such as frost or fire scars, are frequently used to cross-match records from different sources or localities, to broaden the scale of the record or increase its length. By investigating the geographical and temporal scale of historical disturbances, paleo-climatologists can estimate the impact and severity of environmental perturbations and cycles.

X-radiography of sections cut from a growth axis of skeletons of massive corals reveals alternating bands of dense and less-dense skeleton, with general agreement that each couplet represents one year's growth (e.g., Knutson *et al.,* 1972; Hudson, 1981; Wellington and Glynn, 1983). Density bands in coral skeletons were quickly regarded as a much-needed marine analogue to tree rings.

In fact, coral skeletons offer the potential to be much more valuable proxy records of environmental history than tree rings because of the nature of the tissue layer. The information in tree rings is essentially tied up in variations in density and width of the rings. Coral density bands record information in the same way, except that, unlike tree rings, coral skeletal bands are "dead" and lie "outside" the animal. Consequently, materials co-precipitated or trapped inside the coral skeleton during growth are locked away and *in stasis* and can be dated using the annual density bands.

As well, apparent anomalies in the skeletal record become explicable when potential growth-altering factors, such as trophic dependencies or shifts (e.g. Grottoli, 2002), or stress events (e.g. Suzuki *et al.* 2003) become known.

The early promise of detailed, reliable, sub-annual scale environmental proxy records derived from coral skeletons has not been met, although many published reconstructions exist (Barnes & Lough 1993; Taylor et al, 1993, 1995; review in Lough, 2004). Several problems with the extraction of coral proxy records emerged as the techniques became more advanced. The integrity of the skeletal record, for instance, is not permanent. Because the skeleton of the colony already evacuated by tissue is not sealed, environmental water can percolate through the skeleton, carrying with it dissolved minerals and the spores of fungi and sponges, which may change the chemistry of the skeleton (see Shönberg, 2000). Moreover, the infiltration of water into pore spaces causes a certain amount of dissolution of the (metastable) aragonite skeleton and re-precipitation as calcite (a more stable form of calcium carbonate). Some techniques are available to ameliorate the effects of this process on a skeletal proxy record, but diagenesis is a serious problem for the extraction of proxy records, since calcite has different physical properties and the re-precipitation process changes the isotopic ratios within the material (McGregor & Gagan, 2003).

The nature and causes of density banding have been hotly disputed. Many studies have focussed on the biochemical processes surrounding

calcification, both *in vivo* and *in vitro* (see Barnes & Chalker, 1990; Cohen & McConnaughey, 2003 for reviews). Despite considerable research effort, however, no universally accepted model of coral calcification has emerged. Following the work of T.F. Goreau (e.g. Goreau, 1959; 1961), it was universally accepted that light, acting through endosymbiotic algae in coral tissues, was the primary control on coral calcification, although this was first reported by Kawaguti & Sakumoto (1948). Barnes & Chalker (1990) showed that, in shallow waters, the zooxanthellae of corals are light saturated for the entire year. Lough and Barnes (2000) showed that coral calcification is very sensitive to temperature, and that seasonal changes in temperature drive the calcification mechanism in shallow water corals to produce the annual density banding pattern. The existence of annual density bands in corals from localities with restricted or non-seasonal patterns of seawater temperature fluctuation suggests, however, that this is not the only factor involved in density band formation.

The amount of skeletal extension, variations in density, differences in the ratios of included stable isotopes of oxygen and carbon, as well as trace inclusions of various elements of the coral skeleton within these density bands are interpreted as recording variations in ambient conditions (Klein *et al.*, 1992; Scoffin *et al.*, 1992; Swart *et al.*, 1996, Gagan *et al.* 1996; McCulloch *et al.* 2003). Corals also include suspended (or re-suspended) particulate and dissolved materials in their skeletons during calcification, allowing interpretation of skeletal records to include events such as coastal runoff, upwelling and monsoonal wind shifts (Alibert *et al.*, 2003; Fallon

et al., 2003). Therefore, detailed knowledge of parameters controlling coral growth must give insight into the way the coral record changes in their environment. While simple models can capture many aspects of particulate and dissolved trace element inclusions within coral skeletons (e.g. Taylor *et al.* 1995), there is still some way to go to achieve mechanistic understanding of calcification (Cohen & McConnaughey, 2003). An understanding of the way isotopic records and trace inclusions are deposited in the skeleton is a necessary step to improve the way proxy records are recovered and interpreted.

Early proxy record extraction from corals assumed that skeletons of corals were simple analogue recorders of environmental variations without taking into account the role of the animal in the deposition of the skeleton (Buddemeier & Kinzie, 1976). It should be noted that the techniques used to extract these proxy records, however elegant, are correlative at best, although recently researchers are taking great strides in the investigation of cycles of skeletal density (e.g. Taylor *et al.* 1993; Barnes & Lough, 1993) and mechanisms of skeletal deposition (e.g. see Adkins *et al.* 2003; Cohen & McConnaughey, 2003; McConnaughey, 2003). The environmental information extracted from tree rings is supported by a thorough knowledge of the physiology of the trees from which they were obtained and by cross-matching records of geographically-separated trees (Fritts, 1976). The environmental information extracted from coral skeletons is not. Until recently, few studies had been published examining variations in the skeletal records between colonies (see Lough, 2004 for a review). Still

fewer were investigations into the effects of changing environments on coral skeletal records (but see Grottoli & Wellington, 1999, Grottoli, 1999, 2002).

Moreover, much of the literature addressing environmental effects on corals is the product of widely differing interpretations of proxy environmental data (see e.g. Abrams *et al.* 2003; van Woesik 2004; Abrams *et al.* 2004, Maier *et al.* 2003). In the past few years, an increasing focus of proxy-record researchers has been the ground-truthing of skeletal growth parameters and elucidation of factors controlling the chemistry of the skeleton (e.g. Grottoli, 2002, Felis *et al.* 2003, McConnaughey 2003, Rollion-Bard *et al.* 2003; Cohen & McConnaughey 2003). A picture is emerging of organisms which are dynamic and adaptable, that adjust themselves to environmental change in a number of ways, and which are much more difficult to understand than previously thought. There is a demonstrable need for the techniques of proxy record extraction to be proven against real conditions and stress events.

1.2 Structure of *Porites*

Corals of the genus *Porites* Link, 1807 are ubiquitous members of coral reef communities throughout the tropics. While neither fast growing nor competitively dominant, *Porites,* as a group, remains one of the most persistent scleractinian corals in Indo-Pacific tropical assemblages (Done, 1982).

1.2.1 Skeletal architecture

Porites are members of the order Scleractinia, family Poritidae (Veron & Pichon, 1982). As such, they are colonial and comprised of interconnected modules. Each module consists of a single polyp lying in a skeletal cup (calix). All scleractinian corals have external skeletons (Wells, 1956). The tissue of imperforate corals, such as the Faviidae and the Mussidae, lies entirely outside the skeleton; adjacent polyps communicate only via the coenosarc lying over the intervening space (Figure 1.1). Although the skeletons of perforate corals - such as the Poritidae and Acroporidae - are technically external to the tissue, the tissues are inter-connected via canaliculae that penetrate through holes within the skeleton, as well as over the surface of the skeleton.

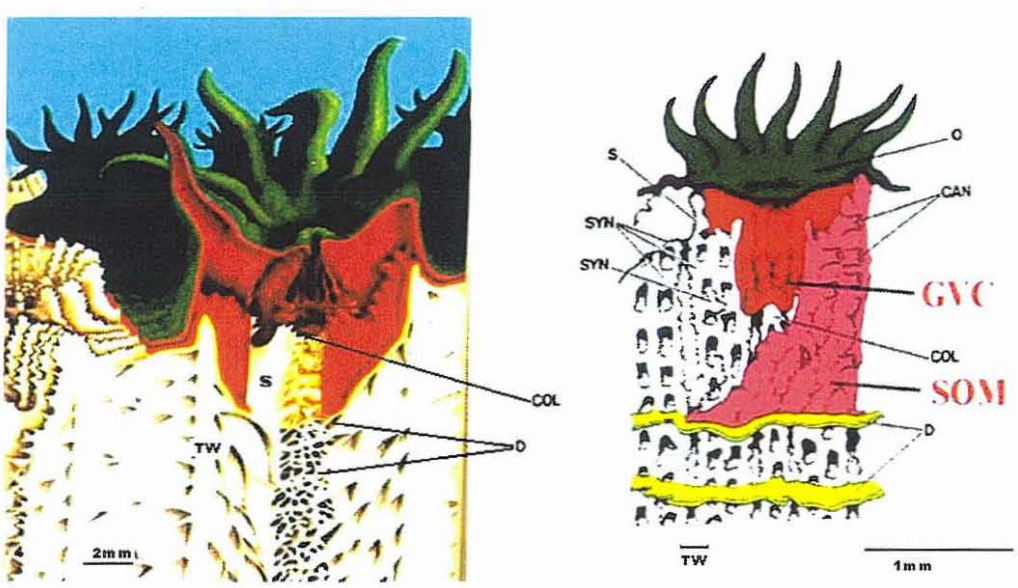

Figure 1.1. Perforate versus imperforate architecture in scleractinia. At left, imperforate skeletons, typified by Faviid corals (image from Veron, 1986); at right, stylized image of perforate architecture, typified by Poritid corals. Note that the tissue of the perforate coral ramifies extensively through the skeleton. Adjacent polyps are connected by tubules (CAN); SYN=Synapticulae, TW=Thecal Wall, S = Septum; P=Pali, COL=Columnella, D=Dissepiment; GVC =Gastro Vascular Cavity; SOM =Somatic tissue, O= Oral disk

There are only 5 species of massive Porites in the Indo-Pacific that grow to heights greater than 200 mm (Veron, 2002), however these are used indiscriminately for proxy record research, primarily because of the difficulty of distinguishing between them taxonomically. Taxonomic identification of *Porites* relies on characteristics of calix structure such as size of mature calices, calix depth, septal dentition and secondary structural elements such as radii (Veron & Pichon, 1982; Veron, 1986, Veron, 2002). Below the calix (which I define here as that part of the skeleton from the upper surface down to that part in which the septa coalesce to form the columella: Figure 1.2), massive *Porites* species are practically identical. Identification characters tend to be quite plastic and vary considerably both within a colony and within species. Skeletal architecture is heavily influenced by the habitat of the colony as well as species (Pichon, pers. com.). Partly because of this morphological inconsistency, identification of colonies used for research is often a bit dubious, and more often than not remains at the generic level.

In most colonial corals, each corallite is delimited by a vertical, circular wall called the theca. In the Poritidae, the theca is a thin, fenestrate wall, which is common to adjacent corallites (Wells, 1956). The skeletons of the Poritidae are not continuous, solid walls, but tend to be formed from a few radially arranged clusters of vertical rods (trabeculae), which coalesce arbitrarily to form a palisade (Wells, 1956). The trabeculae, in turn, are formed of fan-shaped aggregations of crystals called sclerodermites, formed around migrating centres of calcification (Cohen &

McConnaughey, 2003). The skeleton of *Porites* may be considered simply as a tangle of vertical rods arranged into radial patterns (corallites) by the organization of the secreting tissues; i.e. a sheet of adjacent polyps (Barnes & Lough, 1993). The corallite contains no extra-thecal skeleton in *Porites* since a polyp shares parts of its theca with adjacent polyps.

Associated with each polyp, and projecting from the theca towards the coelenteral cavity of the polyp are radially disposed vertical calcicular walls called septa. The thecal wall links the outer regions of the septa. Some recent reports citing evidence from electron-microscopic studies suggested that the microstructure of the crystal palisade comprising the sclerodermites might be less random than previously thought (e.g. Cuif & Dauphin 1998; Perrin, 2003); the apparent randomness may well be an artefact of the coral's growing conditions. Skeletal elements might then be composed of taxon–specific biocrystal laminates (Perrin, 2003). This hopeful prospect is yet to be rigorously examined.

Most Poritids possess some form of columella, which arises as a spire-like tangle of vertical spines from the centre of the calix. The columellae of *Porites* are generally weak, but may be spire shaped or keel-shaped. In some species, horizontal protrusions radiate from the columella (radii), which may be linked by lateral structures. The possession of radii is often associated with a less robust- appearing calicular architecture and tall, spire-like columella, and is diagnostic of certain species.

The calix, housing the principal identifying elements of *Porites* species, is relatively variable in diameter and shape within and between colonies, but tends to be relatively fixed in depth within the species (i.e. the depth of the calix is taxonomically diagnostic). In *P. lutea*, for instance, the calix floor (distinct from the dissepiment & marked by a convergence of skeletal elements to form a kind of porous platform) occurs approximately 1.5mm below the tops of the calix walls (Figure 1.2). This perforated platform represents the skeletal floor of the gastrovascular cavity. Below the calix proper, the skeleton forms a radially-arranged lattice extending through the corallum for a distance corresponding to the lifespan of the polyp which created it. The elements of the lower skeleton have the same form and arrangement as the thecae and septa forming the calix, since these are the same elements, albeit more heavily cross-linked and calcified.

Corallites of *Porites* are less distinct below the level of the calix, which extends between 0.5 and 1.5 mm below the outer surface, depending on species. Below this level, the columella and surrounding pali form synapticular bridges across to adjacent septa, creating a three-dimensional lattice. The boundaries (thecae) of adjacent corallites are difficult to distinguish from sawn specimens (most often used for density analyses), although specimens fractured along the growth axis retain some of the radial symmetry of the skeletal lattice and individual corallites may be identified by a practiced eye (Figure 1.2b).

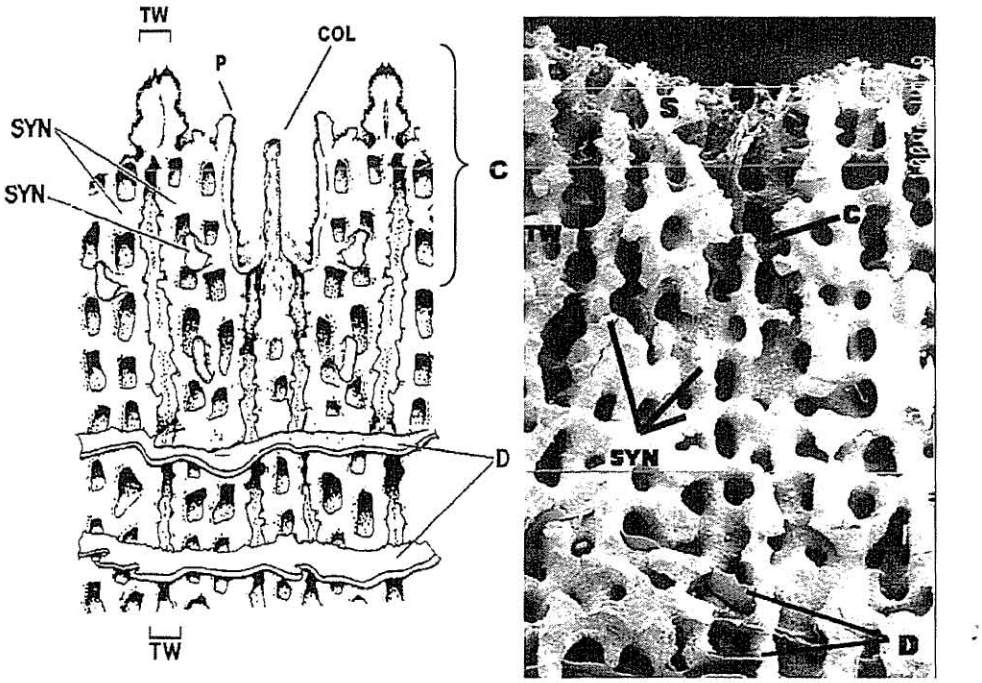

Figure 1.2a. Skeletal anatomy of stylized *Porites* spp. SYN=Synapticulae, TW=Thecal Wall, P=Palus, S= Septum, COL=Columella, D=Dissepiment, C= extent of Calix (as defined in text).
b. Composite SEM of *P. solida* skeleton through the centre of a corallite. The corallite is approximately 1mm in diameter. *Image supplied by D.Barnes, AIMS*

In massive scleractinian corals, the tissue occupies only the first few millimetres of the skeleton. The base of the tissue layer is marked by very thin, essentially horizontal, skeletal bulkheads known as dissepiments. Skeletal elements are extended at the outer surface of the tissue layer. Thickening of these elements is commonly acknowledged to occur throughout the tissue layer (Barnes & Lough, 1993), although some minor dispute exists (see e.g. Wellington *et al.* 1996; Linsley *et al.*, 1999). Dissepiments of faviid corals are secreted by the lower margin of the tissue layer in the space of a few days (Barnes, 1971); it is likely that poritid dissepiments are laid down as rapidly.

Consequently, the dissepiments of faster- growing *Porites* seem to appear as sheets common across many corallites more often than in slower-growing corals. The dissepiments of slower-growing corals often appear as if the lower margins of the tissue layer were "bubbling up" at different rates in non-contiguous corallites, and seldom appear to extend as a sheet between many corallites (although adjacent corallites "share" a dissepiment, these are quite often not horizontal, so that corallites separated by three or four places might contain slightly different volumes of tissue). Faster-growing colonies on the Great Barrier Reef tend to deposit 12-13 dissepiments within annual density bands (Barnes & Lough, 1992), strongly suggesting lunar periodicity. This aspect is examined more thoroughly in Chapter 2.

1.2.2 Tissue

The tissue of *Porites* is a continuous sheet that occupies the skeleton down to the first dissepiment. Adjacent polyps are appressed and highly interconnected through the porous shared thecae. The tissue thus forms a continuous band at the outer margin of the colony. This is in contrast to the tissue of imperforate corals (such as the faviid *Montastrea* used in many Caribbean studies of density banding) in which the thecal wall is solid and calices are separated by extra-thecal skeleton. In these corals, only a thin band of tissue joins adjacent polyps, overlying the extra-thecal skeleton.

Morphologically, the tissue layer of *Porites* consists of two regions: the gastrovascular region and the subgastric region (Figure 3; True, 1995), which roughly correspond to the areas above and below the calix floor. In fresh and dried tissue samples, the two regions are easily discerned by colour and overall appearance. The gastrovascular cavity occupies the upper portion of the skeleton to a depth of approximately 1-2.5mm, depending on species and time of year. This region of tissue, encapsulated within the skeletal calix, contains the basic components common to all hexacorals – glandular endoderm, mesenteries and involuted tentacles. It is here that the basic trophic and reproductive functions of the polyp are concentrated. While zooxanthellae are found in the lower regions, the density of algal symbionts in the upper section of tissue is vastly greater than in the lower part (True, 1995, unpubl.).

In the sub-gastric region below the level of the calix, the tissue layer is comprised of a 3-dimensional lattice of tubules, enveloping and permeating the skeletal matrix (Figure 3). In a similar way to the skeleton, individual polyps are difficult to distinguish in lateral view of an unbroken sheet of decalcified tissue, although separation of polyps is easily performed using needles. The polyps do not taper from top to bottom; while the volume of tissue: skeleton diminishes with depth within the tissue layer as the skeletal elements thicken, the corallite does not encroach on the gastrovascular cavity in the same way as that of imperforate massive corals such as faviids. This narrowing of polyps of imperforate corals towards their base is due to the formation of dissepiments that slope

downwards and inwards from the theca. The dissepiments in *Porites* are horizontal sheets, thus the constriction of the polyp does not occur.

The lower region of the tissue layer appears quite homogeneous, and individual polyps are extremely difficult to distinguish in lateral views of cut (rather than dissected) tissue. Cross-linking between septa superimposes a three-dimensional skeletal lattice on the radial symmetry of the polyp. It is difficult to distinguish the vertical cavities separated by the septa below the level of the columella. Once dissected out from the sheet of tissue, the separated polyp appears as an angular, slab-sided tube, connected to adjacent polyps on each of 4-6 sides by many fine tubules.

The lowest margin of the tissue layer generally appears as a pale spongiform layer, often quite ragged in appearance. The ectodermal layer adjacent to, and secreting, the skeleton is the calicoblastic ectoderm. This ectoderm is adjacent to all skeletal elements within the tissue layer. By definition, endoderm envelops the coelenteron: skeletal elements are thus secreted by and surrounded by calicoblastic ectoderm, itself surrounded by mesoglea and endoderm. The calicoblastic ectoderm is histologically distinct from the ectodermal layer that is presented to the environment. It is assumed that all resources required for the deposition of skeleton are translocated to the calicoblastic ectoderm from elsewhere in the tissue layer, since this tissue does not contain zooxanthellae, nor is it exposed directly to the water column via the coelenteron. It is this tissue which must form the dissepiment during tissue uplift; reduction in the volume of

tissue during these episodes therefore occurs in the region above the basal calicoblastic ectoderm.

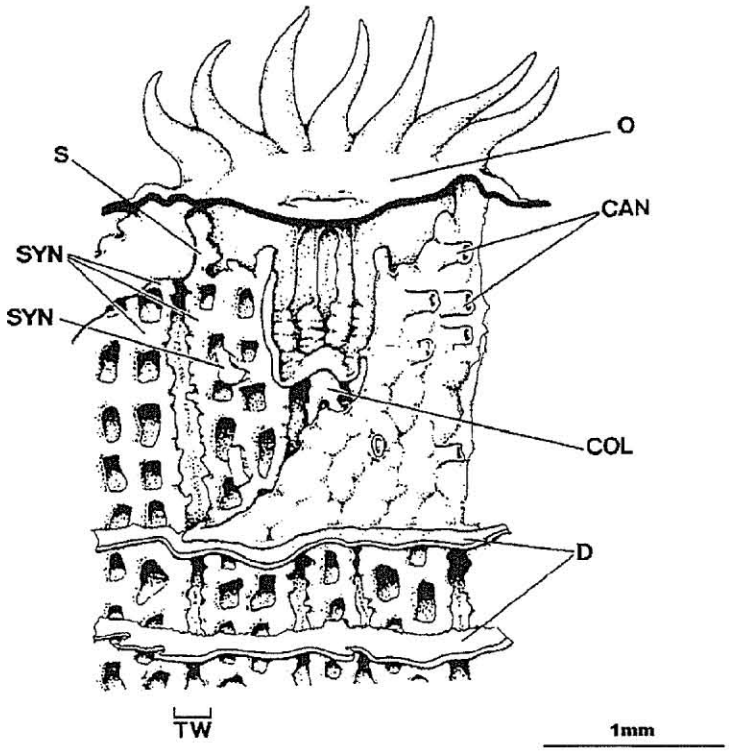

Figure 1.3 Tissue/skeleton relationship in *Porites*. SYN=Synapticulae, TW=Thecal Wall, COL=Columella, D=Dissepiment; O= Oral disk; CAN= Canaliculae linking adjacent polyps

1.3 Rationale for this project

Monitoring of the responses of coral to environmental change and linking those responses to past environmental changes is difficult and relies for the most part on correlative proxy techniques. So far, there has been no serious attempt to directly measure biologically meaningful effects of environmental changes on corals that could both explain the consequences of sublethal stress and enhance the interpretation of proxy environmental records. Taylor *et al.* (1993) showed that variations in the thickness of the

tissue layer of massive *Porites* can change the appearance of density bands. They were attempting to model coral growth in order to derive simple rules to visualise how variations in tissue thickness, calcification rate and extension rate, both inter- and intra-annually, might affect the appearance of density bands. The main use of proxy environmental data from corals has been in reconstruction of seawater temperatures. Temperatures are obtained from annual average calcification and extension rates. A mechanistic understanding of the rules governing these parameters is predicated of the effective use of such a tool.

The AIMS (or Townsville) model of density band formation constructed by Taylor *et al.* (1993) provided a simple analogue of coral growth to allow interpretation of density band variations in the context of environmental change. It provided a basis for subsequent exploration of growth variations and trace inclusions and the interpretation of variations in skeletal density. Coupled with Barnes & Lough's (1993) report that the thickness of the tissue layer might correspond to variations in the quality of the coral's growing environment, it also suggested a mechanism for detecting changes in habitat quality in the life of a colony. They reasoned that if the thickness of the tissue layer changes in response to changes in habitat quality, then this might be reflected in concomitant changes in patterns of variation in skeletal density or growth.

It was quickly noticed by proxy-record researchers that not every colony possessed the 12 or 13 dissepiments per cycle that correspond to a strict

lunar periodicity. Most colonies do not form one dissepiment every month, even though they extend their skeletons almost continuously (albeit not consistently). The colonies are thus more-or-less "choosing" the time of tissue uplift (or are constrained to it by the prevailing conditions). This behaviour would clearly be constrained by physical limitations on the amount of tissue supportable by the colony (see Darke & Barnes, 1993), however, allowing control of the degree of uplift to revert to the animal predicates an ability for the animal to respond to changing environmental conditions.

If, for example, the prevailing conditions caused a slowing of linear extension, the colony may not achieve a threshold thickness of tissue which would allow it to reduce by 15-25% (corresponding to the normal spacing between dissepiments as a fraction of average TTL), or perhaps has not exceeded the upper margin of tissue supportable (cf. Darke & Barnes, 1993), and thus does not provoke an "upward tension". Alternatively, a colony which has the energy surplus to support a larger mass of somatic tissue may "choose" not to uplift the lower margin, but to accommodate the extra tissue mass, since it is not constrained by energetic considerations (Darke & Barnes, 1993). Barnes & Lough's (1993) report suggested that this behaviour has the potential to reflect degradation of their environment, since they reasoned that TTL might be characteristic of a certain environmental quality. The possibility therefore exists that the thickness of the tissue layer can be used as a gauge of how well the colony was doing at the time of collection.

Early investigation (True 1995) showed that different habitats within a typical mid-shelf reef were characterised by colonies with fairly similar tissue thickness. As well, there were distinct trends in the variation of tissue thickness of various species according to the depth of water they occurred in.

Together, these results suggested that tissue thickness might provide a gauge of coral health, if it could be assumed that habitats characterised by colonies with thinner tissue provided fewer resources for coral growth. However, the evidence, like much proxy environmental research, was still quite correlative rather than empirical. Manipulative experimentation remains the most effective tool for testing hypotheses and it was this premise that provoked the next logical stage of research described here. The overall aim of my research is to determine whether or not tissue thickness variation can be used as a simple tool to monitor coral condition.

Four basic questions underlie much of the research described in the following chapters:

- Does the thickness of the tissue layer (TTL) of massive *Porites* spp. vary systematically in space and time? Clarification of this question will provide insight into patterns of skeletal growth and indicate whether TTL might be a useful indicator of changes in environmental conditions in space and time.

- Does TTL reflect the quality of the coral's environment; i.e. is it possible to change TTL by manipulating the environment? This question addresses the major concerns of any prospective monitoring technique: is the measure sensitive to change, and how rapidly can the response be detected?
- Is it possible to predict the severity of stress events by examining changes in TTL? Answering this question will further enhance the usefulness of a prospective monitoring technique by enabling it's use as a comparative measure.
- Are stress events in the life of a coral recorded in the skeleton in ways that can be detected by techniques for extracting proxy environmental records? This question addresses a major issue in proxy environmental record research: do the records faithfully record environmental changes that affect the corals? Can events such as the 1998 mass bleaching event be detected reliably in such a way that their frequency in the past can be estimated?

Ultimately, it was envisaged that extrapolation of the results of the research proposed for this dissertation could be used as the basis for a simple reactive monitoring technique for coral reef management. Additionally, the validity of an important proxy record reconstruction technique would be assessed against a record of biological variation for the first time. Not only would this provide an opportunity to ground-truth the theory of the technique, but it would also provide much-needed insight into the interpretation of data recovered by such techniques.

1.4 Outline of the thesis

1.4.1 Chapter 2

In Chapter 2, I examine temporal and spatial patterns in tissue layer thickness variation. It is important to identify possible environmental correlates for variation in TTL in order to isolate biologically important parameters. If TTL varies seasonally, then the use of TTL as a monitoring tool is complicated by the necessity to sample consistently in time between sites. Seasonal variation, however, also means that the seasonally varying parameters (principally light and temperature) may be the most important environmental parameters to the coral. To address this question, a long-term survey of tissue thickness variation was carried out at Orpheus Island, central GBR. If TTL varies consistently in space, then causation of the variation is more easily identified, since parameters that vary along similar gradients are proximal causes of the TTL variation. To examine this question, surveys of *Porites* were carried out at widely scattered reefs both within the GBR and in parts of south East Asia.

1.4.2 Chapter 3

In Chapter 3, I examine the effects of stress on the TTL of massive *Porites* spp. Monitoring the condition of an organism requires knowledge of the effects of stress, since a basic precursor to the development of a biological monitoring technique is the knowledge that the measured parameter in fact reflects the condition of the organism. Previous work (Chapter 1) had identified light as an important correlate to TTL; therefore lack of light was targeted as a potential stress agent.

Firstly, a field experiment was conducted, in which large colonies were shaded continuously for almost two years. Colonies were repeatedly sampled over that period and the results compared to unstressed colonies described in the previous chapter. Secondly, thermal stress was used to examine the relationship between TTL and the lipid content of the tissue layer in corals under stress. Lipids are a standard storage medium in animals; they were therefore used as a proxy for the "available energy" of a colony. The relationship between tissue thickness and level of lipid within the tissue might then be interpreted as an indication of whether TTL could be used as a direct measure of the energy level of a colony under stress. This stress was compared with a natural high-impact stress event, when the reefs of the central GBR were affected by the worldwide mass-bleaching event of 1998. Recovery of colonies from the 1998 event was documented using both TTL and lipid levels. The applicability of experimental data to real-world stress events could thus be estimated.

1.4.3 Chapter 4

In Chapter 4 I investigate the ability of corals to record the effects of stress events in their skeletons. The existence of this ability is of major consequence to proxy record research, since the fidelity of proxy environmental records is determined by how the corals respond to fluctuations in their environment. In this chapter, commonly-employed skeletal isotope ratios (^{13}C:^{12}C and ^{18}O:^{16}O) are used as representative proxy records. Core samples were extracted from the central growth axes

of three large colonies (two of these colonies (of different species) were shaded during the experiment described in Chapter 3; the third colony was a control). The cores were sampled at minute intervals spanning several years' growth before and after the stress events and the ratios of skeletal stable isotopes of oxygen and carbon were analysed by mass spectrometry. The use and fidelity of these high-resolution proxy records for identification of stress events is analysed and the differences and similarities between different colonies and species is discussed. The effects of both the shading stress and the bleaching event are examined and discussed in terms of perturbations in the proxy records.

1.4.4 Chapter 5

Chapter 5 is a general discussion of the research described in previous chapters. In this chapter I also discuss the fidelity of tissue thickness variation to changes in the health and growth of corals. I examine the ramifications of a variable tissue layer in the context of density band formation and I speculate on the fidelity of proxy environmental records derived from coral skeletons that do not take this into account. I also the use the results of the previous chapters to synthesise a model of energy allocation for corals and to describe a hypothetical stress management protocol for corals. The usefulness of tissue thickness variation as the basis for a reactive monitoring technique is discussed. I speculate about possible future directions for research to further elucidate the nature of the tissue layer of massive *Porites* and the potential for tying such knowledge into proxy environmental reconstructions.