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# The Taxonomy of Demospongiae (Porifera) from the Bay of Plenty, New Zealand – Connecting Linnaean and Phylogenetic Classification

A thesis submitted in partial fulfilment

of the requirements for the degree

of

Master of Science

in Biological Sciences

at

The University of Waikato

by

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The University of Waikato

2015



THE UNIVERSITY OF WAIKATO Te Whare Wananga o Waikato "Food is brought to them, waste is taken away. For them in their eternal abyss, with its time-like stream, there is no hurry, there is no return. Such an organism becomes a mere living screen between the used half of the universe and the unused half – a moment of active metabolism between the unknown future and the exhausted past" (Bidder 1923).

### ABSTRACT

The ability to accurately identify species is prerequisite for assessing levels of biological diversity and a fundamental requirement for ecological research. In recent times, there has been a shortfall in biologists who practice traditional alpha taxonomy, leading to difficulties in assessment of biodiversity in some taxonomic groups. The use of a molecular DNA barcoding approach has been suggested as a tool that can be used to complement and accelerate traditional alpha taxonomy, without supplanting or invalidating existing taxonomic practices.

Two different techniques were used to identify organisms, molecular and alpha taxonomy. This thesis addressees several questions relating to sponge systematics. Research was focused on three areas; (1) record sponge biodiversity from the Bay of Plenty region, (2) undertake a systematic revision of the fauna correlating 'historical' taxonomy with a modern phylogenetic assessment (3) determine whether identifications based on genetic barcoding are congruent with those produced via traditional morphological methods (alpha taxonomy), and to assess the use of molecular techniques for Demospongiae species identifications.

This was the first focused research on sponge diversity in the Bay of Plenty region. Fifty five species are described in this research. Of these, there are up to three new families, three new genera and thirty four species which are undescribed and deemed new to science. However, a more conservative estimate with grouped specimens suggests that there is a minimum of at least one new family, one new genus, and eighteen new species that are undescribed and deemed new to science. In summary, we conclude that for New Zealand Demospongiae, sequence variation present in the barcoding region of the COI gene is sufficient to allow for the identification individuals to their nominate species. The use of mtDNA barcoding can without doubt complement classical morphological taxonomy and accelerate the identification process and may in fact revive an interest classical morphological taxonomy.

### ACKNOWLEDGEMENTS

I would wholeheartedly like to thank my supervisors Professor Chris Battershill, Dr Phil Ross and Dr Ian Hogg for their unstinting devotion and enthusiasm which helped me to produce this thesis. All of whom have shown infinite patience, assistance and inspiration in the face of ignorance, failure and constant distraction. Chris is one of the most inspirational gentlemen I have ever met. His passion and selfless dedication was expressed through his patience in teaching me the science and art of sponge taxonomy (despite the appallingly difficult task of classifying sponges). Secondly, I would like to thank Phil Ross for his constant motivational speeches. I am extremely grateful to have had Phil as my supervisor as he provided me with advice, countless hours of discussion, knowledge and above all, his moral support. I am in debt to Ian Hogg, who provided me with invaluable advice, criticism and guidance. Without his help this period would have been far more difficult than it was.

The technical support I received during my research at the University of Waikato's Coastal Marine Field Station was exceptional. I would especially like to thank David Culliford and Rex Fairweather for whom no problem was too big and who put up with my complaining when things were not going according to plan. Thanks to David for being my dive buddy, photographer, skipper and friend among other things. Thanks to Rex for sharing your taxonomic knowledge with me, for taking care of all the last minute things I'd forget to organise, and for your willingness to always help me, no matter what the problem was.

Of the staff and students at the University of Waikato, special thanks to: Clare Beet who was exceptionally helpful, providing countless hours of guidance in the Biosystematics Laboratory, I really could not have done this without her. A special thanks to Kim Pritchard for your proof reading and referencing tips. Kim was always available to help me no matter what the time of day or night. To Te Puea Dempsey for proof reading everything! To Dr Barry O'Brien for assisting me with electron and light microscopy, thank you. To Gemma Collins for assisting me with data submission to BOLD. I am also grateful to my friend Peter Randrup for assisting me with spicule measurements in a room which often exceeded temperatures of over 10,000°C during summer months. Thanks to Matthew Bennion, Kayne Tiehuis, Lance Litchfield and Rob Besaans.

I would sincerely like to thank the University of Waikato for providing me with a master's research scholarship; it made my time at the University a whole lot easier than it would have been.

It is difficult to express the depth of my gratitude to such people as my parents, Patricia and Patrick Mc Cormack for their love, support and encouragement when I needed it the most. Thanks Dad for being an all-round top man. Thanks Mam for always looking after me. Without my parents, I literally would not be here today. Go raibh mile maith agat gach duine ~ Thank you everyone.

# TABLE OF CONTENTS

| ABSTRACT          | i    |
|-------------------|------|
| ACKNOWLEDGEMENTS  | ii   |
| TABLE OF CONTENTS | iv   |
| LIST OF FIGURES   | vi   |
| LIST OF TABLES    | viii |

| Chapter 11 |                        |   |
|------------|------------------------|---|
| GENER      | AL INTRODUCTION        | 1 |
| 1.1        | Background             | 1 |
| 1.2        | Modern taxonomic tools | 3 |
| 1.3        | Organisation of thesis | 3 |

| Chapter 2                                | 5  |
|--|----|
| SPECIES DESCRIPTIONS OF THE DEMOSPONGIAE | 5  |
| 2.1 Introduction                         | 5  |
| 2.2 Materials and Methodology            |    |
| 2.2.1 Field Collections                  | 8  |
| 2.2.2 Specimen preparation               | 10 |
| 2.2.3 Taxonomic procedure                | 11 |
| 2.3 Species descriptions                 | 19 |
| ORDER CHORISTIDA                         | 19 |
| ORDER HADROMERIDA                        | 30 |
| ORDER AXINELLIDA                         | 39 |
| ORDER POECILOSCLERIDA                    | 42 |

| ORDER HALICHONDRIDA    | 92  |
|------------------------|-----|
| ORDER HAPLOSCLERIDA    | 104 |
| ORDER NEPHELIOSPONGIDA | 131 |
| ORDER DICTYOCERATIDA   | 136 |
| ORDER EPIPOLASIDA      | 141 |
| 2.4 Discussion         | 143 |

| Chapter | <sup>-</sup> 3 |                                    | 148 |
|---------|----------------|------------------------------------|-----|
| MOLEC   | ULAR           | SYSTEMATICS                        | 148 |
| 3.1     | Intr           | oduction                           | 148 |
| 3.2     | Me             | thods                              | 149 |
| 3.2     | .1             | Study sites and sample collection: | 149 |
| 3.2     | 2              | Genetic analysis                   | 150 |
| 3.2     | 3              | Phylogenetic analysis              | 151 |
| 3.3     | Res            | ults                               | 152 |
| 3.3     | .1             | COI sequence divergence            | 153 |
| 3.3     | .2             | Phylogenetic analysis              | 153 |
| 3.3     | .3             | Non sequence trends                | 155 |
| 3.4     | Disc           | cussion                            | 162 |

| Chapter 4          |  |  |
|--------------------|--|--|
| •                  |  |  |
| GENERAL DISCUSSION |  |  |

| REFERENCES |     |
|------------|-----|
| PLATES     | P.i |

## LIST OF FIGURES

- Figure 3.2 : Neighbour-joining tree for 32 specimens of Bay of Plenty sponges constructed using the Barcode of Life Database for identification (families are indicated on the right and orders are underlined). Numbers next to branches indicate percentage non parametric bootstrap support (>50%) from 1000 pseudoreplicates. Individuals

which are genetically dissimilar to other specimens within the same taxonomic group are denoted by an Asterisk (\*) ...... 158

# LIST OF TABLES

| Table 2.1: Spicule dimensions of Stelletta crater             | 21 |
|---|----|
| Table 2.2: Spicule dimensions of Unknown n.sp.1               | 24 |
| Table 2.3: Spicule dimensions of Stelletta maori              | 27 |
| Table 2.4: Spicule dimensions of Stelletta sandalinun.        | 29 |
| Table 2.5: Spicule dimensions of Cliona celata.               | 32 |
| Table 2.6: Spicule dimensions of Cliona n.sp.1                | 34 |
| Table 2.7: Spicule dimensions of Cliona sp.                   | 36 |
| Table 2.8: Spicule dimensions of Polymastia fusca.            | 38 |
| Table 2.9: Spicule dimensions of Acanthoclada sp              | 41 |
| Table 2.10: Spicule dimensions of Crella encrustans.          | 44 |
| Table 2.11: Spicule dimensions of Tetrapocillon sp.           | 46 |
| Table 2.12: Spicule dimensions of <i>Tedania</i> n. sp. 1     | 50 |
| Table 2.13: Spicule dimensions of Tedania sp.                 | 52 |
| Table 2.14: Spicule dimensions of <i>Tedania battershilli</i> | 54 |
| Table 2.15: Spicule dimensions of <i>Tedania</i> sp           | 56 |
| Table 2.16: Spicule dimensions of Coelosphaeridae sp.         | 59 |
| Table 2.17: Spicule dimensions of Carmia n sp.1               | 62 |
| Table 2.18: Spicule dimensions of Carmia n sp.2               | 65 |
| Table 2.19: Spicule dimensions of Carmia n sp.3               | 67 |
| Table 2.20: Spicule dimensions of <i>Carmia</i> n sp. 1 (1)   | 70 |
| Table 2.21: Spicule dimensions of <i>Carmia</i> n sp. 1 (2)   | 73 |
| Table 2.22: Spicule dimensions of <i>Carmia</i> n. sp. 2 (1)  | 76 |
| Table 2.23: Spicule dimensions of <i>Carmia</i> n. sp. 3 (1)  | 78 |
| Table 2.24: Spicule dimensions of <i>Carmia</i> n. sp. 3 (2)  | 81 |
| Table 2.25: Spicule dimensions of <i>Carmia</i> n. sp. 3 (3)  | 83 |

| Table 2.26: Spicule dimensions of Carmia n. sp. 3 (4).85                                |
|---|
| Table 2.27: Spicule dimensions of Carmia n. sp. 3 (5).    87                            |
| Table 2.28: Spicule dimensions of Carmia n. sp. 3 (6).    89                            |
| Table 2.29: Spicule dimensions of <i>Ciocalypta polymastia</i>                          |
| Table 2.30: Spicule dimensions of Halichondria n. sp. 1                                 |
| Table 2.31: Spicule dimensions of Halichondria sp                                       |
| Table 2.32: Spicule dimensions of Halichondria sp                                       |
| Table 2.33: Spicule dimensions of Halichondria moorei                                   |
| Table 2.34: Spicule dimensions of Hymeniacidon hauraki.       103                       |
| Table 2.35: Spicule dimensions of Sigmadocia fragilis.    105                           |
| Table 2.36: Spicule dimensions of Haliclona sp. 1    108                                |
| Table 2.37: Spicule dimensions of Haliclona n.sp. 2    110                              |
| Table 2.38: Spicule dimensions of Haliclona n.sp. 3 cf. tenacior       112              |
| Table 2.39: Spicule dimensions of Haliclona n. sp. 4 cf. brøndstedi       114           |
| Table 2.40: Spicule dimensions of <i>Haliclona</i> n.sp. 5 <i>cf. heterofibrosa</i> 116 |
| Table 2.41: Spicule dimensions of Haliclona n.sp. 6 cf. heterofibrosa                   |
| Table 2.42: Spicule dimensions of Haliclona heterofibrosa.       120                    |
| Table 2.43: Spicule dimensions of Haliclona sp.    122                                  |
| Table 2.44: Spicule dimensions of Adocia n. sp. 1    124                                |
| Table 2.45: Spicules dimensions of Adocia n. sp. 2                                      |
| Table 2.46: Spicule dimensions of Callyspongia ramosa.    128                           |
| Table 2.47: Spicule dimensions of <i>Callyspongia</i> n. sp.1 <i>cf. irregularis</i>    |
| Table 2.48: Spicule dimensions Xestospongia n. sp. 1    132                             |
| Table 2.49: Spicule dimensions of Nepheliospongiidae sp.       135                      |
| Table 2.50: Spicule dimensions of <i>Dictyoceratida</i> sp                              |
| Table 2.51: Spicule dimensions of Dysideidae sp.    140                                 |

| Table 2.52: Spicule dimensions of Teth | ya aurantium 142 |
|--|------------------|
|--|------------------|

- Table 3.2: Mean pairwise distances among Demospongiae taxa collected in the Bay of Plenty, New Zealand. For individuals within species, n = number of species which had two or more individuals; species within genus, n = number of genera with two or more species; species among genera, n = number of genera analysed within a family; species among families, n = total number of species analysed....... 157

### **Chapter 1**

### **GENERAL INTRODUCTION**

### 1.1 Background

Over the past century taxonomy, the science of identification, classification and naming of organisms has undergone fundamental conceptual changes (Vences *et al.* 2013). For some modern biologists, traditional taxonomy has been increasingly disregarded and considered an out-dated method of routine species identification, in favour of molecular based approaches (Wheeler *et al.* 2004). Unfortunately, for a growing number of groups, taxonomic studies are chronically underfunded and traditional alpha taxonomists have been themselves described as endangered species (Daugherty *et al.* 1990; Wheeler *et al.* 2004).

Traditional alpha-taxonomy has different interpretations. Therefore, within the framework of combined taxonomic methods it is incorrect to split taxonomy into different components, each being assigned a different weight of importance (Decraemer & Backeljau 2015). Preferably, taxonomy should be holistic, delimiting and describing taxa, based on divergent characters and all available methodology and information. Therefore taxonomy cannot and should not be approached without considering evolutionary relationships (Decraemer & Backeljau 2015).

Sponge systematics is generally acknowledged as being appallingly difficult. Sponges are morphologically plastic and conspecifics potentially phenotypically different. This plasticity makes identifications based solely on growth form and colour alone prone to error. Traditionally, taxonomists have depended upon morphological features such as colour, shape and skeletal elements (spicules) to identify sponges (Hooper 2002; Battershill 2010). The limitations of such classifications have become apparent to sponge researchers in particular, who have attempted to use these systematic arrangements in their own research context. Although the relatively unstructured sponge body form has been highly successful in an evolutionary context, the evolution of sponge taxonomy has been less successful, on account of a lack of conserved gross morphological characters for species delineation (Bergquist & Wells, 1983).

#### Chapter 1

Sponges are among the most primitive multicellular organisms currently in existence on the planet. They are sessile animals and lack true tissues and organs (Battershill 2010). Sponge morphology can be influenced by the available space, water current velocity, habitat, and the formation and slope of the substrate. Sponges can develop as thin or thick encrusting forms, as massive or globular arrangements, or as erect or lobate forms (Battershill 2010). Sponges inhabiting high energy environments with greater currents and wave formations often retreat to more compressed forms. Moreover, sponges which occupy areas with limited flow are often larger and more open to increase their surface area to volume ratio for suspension feeding (Battershill 2010).

Species identifications using exclusively classical taxonomic methods have several limitations. For example, the misidentification of taxa due to phenotypic plasticity among species is often unavoidable and the likelihood of incorrectly describing cryptic taxa as a result of homeoplasy is high. Moreover, morphological keys are rarely available for all life history stages of a given taxon. Accordingly, a high level of taxonomic expertise is required to morphologically identify many sponges (Valentini *et al.* 2009).

Aristotle, in 384 B.C. was one of the first naturalists to describe a sponge (Johnson 1884). Two millennia later, Linnaeus provided binomial nomenclature, the Linnaean system of classification. This system, developed during the late 18<sup>th</sup> and early 19<sup>th</sup> century, is a system by which all organisms are now named and a tool with which to describe relationships between species. Today, two hundred years later, this is the system that forms the basis of modern alpha taxonomy (Hooper & Van Soest 2002). Few would disagree that sponge systematics based solely on alpha taxonomy is in poor shape. In fact, Phylum Porifera, is one of the few phyla to still have unresolved ordinal level classifications (McCormack *et al.* 2002). However, all is not lost. Modern taxonomic tools, such as DNA barcoding techniques, promise to bring some order and certainty to the dark art of sponge classification (Hebert *et al.* 2003a; Hebert *et al.* 2003b; Meier *et al.* 2006; Miller 2007).

### **1.2 Modern taxonomic tools**

Genetics and the DNA barcoding approach has been used to complement and accelerate traditional alpha taxonomy, without supplanting or invalidating existing taxonomic practices (Bucklin *et al.* 2011). The mitochondrial DNA cytochrome c oxidase subunit (COI) gene locus has become the DNA marker of choice for many taxonomic groups both for identification and resolving phylogenetic relationships (Hebert *et al.* 2003a).

Chemotaxonomy, differentiating species based on their (usually bioactive) secondary metabolite chemical profile, has been suggested as a mechanism by which sponges may be assigned to certain taxonomic groupings for many years (Bergman & Feeney, 1950; Bergquist, 1979; Bergquist & Hartman, 1969). However the biosynthesis of such metabolites can now be traced to microbial symbionts or shared metabolic pathways, hence the taxonomic level by which chemotaxonomic techniques can be applied is generally recognised to be broad and hence there are limitations in the technique (see review by Erpenbeck and van Soest, 2006).

### **1.3 Organisation of thesis**

The main body of this thesis comprises two chapters (2 and 3) examining the biodiversity of Bay of Plenty sponges and their classification. In **Chapter two I** deal with the systematics and diversity of sponges in the Bay of Plenty, presenting a survey of sponge biological diversity in two key habitats: the first, an estuary (Tauranga Harbour); the second, an open coast rocky reef (Karewa Island) off shore from the harbour. Based on the sponge collections made during these surveys, I undertake a systematic revision of the fauna correlating 'historical' taxonomy with the modern phylogenetic assignments. Furthermore, I give consideration to workers in the biochemical, ecological and general marine biology fields, by incorporating notes on macroscopic features, reproduction and biochemistry where possible.

In **Chapter three** I present a genetic analysis sponge biodiversity based on subsamples from the collections made across the locations described above. Specifically, I sequence the Bay of Plenty sponge collection for the mitochondrial COI gene and use the Barcode of Life Database to assign taxonomic information to gene sequences. Phylogenetic analysis tools are then used to model evolutionary relationships among sequenced sponge taxa. I test the effectiveness of COI as a gene for species level sponge identification and compare the utility of classical taxonomy based on the Linnaean system of classification and modern molecular systematics approach using DNA barcoding in providing identifications and classifications for the Bay of Plenty sponge collection. Finally, I compare higher level sponge classification as described by both molecular and alpha methods, using molecular data to identify elements of contemporary sponge alpha taxonomy that may be in need of revision.

My overall aim in this thesis was to shed light on how close or disparate the two main systems for taxonomic species assignments are while at the same time carrying out the first focused research on sponge diversity in the Bay of Plenty region.

The knowledge generated by this research will substantiate the relative usefulness of these techniques and she light on a possible best practice for marine invertebrate biosystematics for the lowest metazoa. The diversity recorded and the affiliations (warm or cold temperate) of the species identified will add to the understanding of the biogeography of the Bay of Plenty Region.

### Chapter 2

### **SPECIES DESCRIPTIONS OF THE DEMOSPONGIAE**

### 2.1 Introduction

Over the past century, the science of taxonomy involving the identification, classification and naming of organisms has undergone fundamental conceptual changes (Vences *et al.* 2013). For some modern biologists, taxonomy has been increasingly disregarded and considered an out-dated method of classification, without epistemological underpinning (Wheeler *et al.* 2004). Unfortunately, for a growing number of groups taxonomic studies are chronically underfunded, in favour of more modern molecular approaches and traditional taxonomists have been described as an endangered species (Daugherty *et al.* 1990; Wheeler *et al.* 2004). It is essential that the art and science of taxonomy be kept alive, as there is no method of back tracking modern molecular identifications.

There is a need to update current traditional Porifera systematics in New Zealand's literature. We have attributed to Bergquist (1961, 1968, 1970, 1978, 1980; Bergquist & Warne, 1980; Bergquist & Fromont, 1988) the role of most of the pioneering work in the field of sponge systematics in New Zealand. More recently, notable authors who have followed Bergquist's work closely have contributed to sponge systematics in this country (Pritchard. et al. 1984; Bergquist & Fromont 1988; Kelly-Borges & Bergquist 1997; Cook & Bergquist 1999; Battershill *et al.* 2010). However, there is a need to update old classical taxonomy from Bergquist's era to the modern Systema Porifera, which is the current international authority for sponge systematics (Hooper & Van Soest 2002). This current study aims to provide a comprehensive review of Porifera from New Zealand described by Bergquist's early studies, and to update them to currently accepted binomial nomenclature using the World Register of Marine Species Database (WoRMS 2015). The study will do this with a focus on a new collection of sponges from a hitherto unexamined region from the sponge taxonomic perspective: the Tauranga Harbour and coast.

Sponges belonging to the phylum Porifera are comprised of aquatic sedentary filter feeding animals (Fromont 1985). They fall within the subkingdom metazoan

and have no true tissues or organs, but show significant independence from one another (Battershill et al., 2010). Sponges are mainly found on rocky or coral reefs, but can also be found in soft substrate, often commensal on shells or boring into calcareous material. Many sponges have interesting interspecific interactions with other organisms, e.g. encrusting on the backs of crabs and the shells of bivalves (Fromont 1985). They are one of the most important animal groups living in benthic ecosystems because they filter vast quantities of water and form the basis of stable productive ecosystems (Battershill et al., 2010). Porifera is currently recognized as having four classes, with Demospongiae contributing to >90% of recent species (Fromont 1985). All orders of the Demospongiae in New Zealand have been revised in the past, with the largest contribution coming from Bergquist working on orders of Axinellida, Halichondrida, Haplosclerida, Nepheliospongidae, Poecilosclerida and orders belonging to the subclass Tetractinomorpha (Bergquist 1968; Bergquist 1970; Bergquist & Warne 1980; Bergquist & Fromont 1988). Now, with extensive subtidal collections of sponges from around the Bay of Plenty, a taxonomic review of most Demospongiae orders is possible.

Levi (1957) stated that the phylum Porifera is one of the last major groups of Metazoa in which the ordinal level of classification has still not been clearly defined. Difficulties in Dempospongiae systematics has been attributed to the relatively simple bauplan of this taxon, resulting in a shortage of characters required for a robust phylogenetic reconstruction (Erpenbeck *et al.* 2006). Sponge systematics has traditionally been based almost completely on skeletal traits, such as skeletal mineral elements (spicules) (Erpenbeck & Wörheide 2007). Fromont (1990) found that the most useful morphological character for separating families belonging to the Haplosclerida order was the organisation of the internal skeleton, its spicule composition and their quantities. Morphological delineation of sponge species is hindered by the lack of fixed diagnostic morphological characters and our limited knowledge of phenotypic plasticity among Porifera species (Bergquist & Warne 1980; Andreakis *et al.* 2012).

The inherent limitations of classical (alpha) taxonomy based identifications systems and the declining number of taxonomists signals the need for a novel approach to sponge taxonomy (Hebert *et al.* 2003a). Over the past decade, classical (alpha) taxonomy has been completely overturned; consequently a

review of the state of the art has been suggested among taxonomists (Cárdenas *et al.* 2012). One study by advocates of DNA barcoding, Hebert *et al.* (2003a), suggested the use of DNA based technologies to bridge the gap between classical taxonomic practices and the need for routine species identifications. Phylogenetic assignment advancements have been triggered by the advent of powerful novel molecular techniques (Vences *et al.* 2013). DNA barcoding has become increasingly attractive as a method of species identification in terms of cost, speed and objectivity (von Crautlein *et al.* 2011). DNA barcodes provide clear and comparable analyses that can be repeated by anybody, even untrained taxonomists, which are particularly important for sponges, as few taxonomists currently practice in New Zealand. Additionally, unlike classical taxonomy, DNA barcoding can analyse fragmented samples within all the life stages of organisms (von Crautlein *et al.* 2011). Therefore, DNA barcoding techniques could be used to complement classical taxonomy by speeding up identifications without superseding or replacing the diminishing art of sponge taxonomy.

A chemosystematics approach has been suggested as an alternate method of sponge identification and was initially proposed by (Bergman 1949, 1962) and later by Bergquist (1978). As an example, chemicals from the *Carmia* genus have been particularly well studied in New Zealand and are characteristic of this genus. Specifically, secondary metabolites such as Peteamine and Peluroside A have been isolated from New Zealand *Carmia* species, which has assisted in the identification of these species (Northcote *et al.* 1991; Page *et al.* 2005a; Page *et al.* 2005b; Page *et al.* 2011). The use of chemical signatures, which can also complement traditional taxonomy, has historically shown great potential (Bergquist 1978). However, the role of sponge microbial symbionts in shared metabolic pathways that underpin the biosynthesis of many of the 'signature chemicals' raises concerns for the robustness of these chemically based procedures.

The intention of this study is to facilitate current taxonomic descriptions and to provide a platform for future taxonomic work to be undertaken on the Demospongiae in New Zealand, while generating discussion among fellow researchers on these revisions. This chapter presents a series of concise rediscriptions of existing species from New Zealand, with full taxonomic descriptions of species which are new to science. Descriptions are also supported by generic placements, affinities and relationships of each ordinal, familial, genera and species level classifications which are summarised after the spicule descriptions. Moreover, the descriptions of auxiliary material to further support other 'poriferologists' or general researchers is provided for all specimens. This chapter makes use of alpha taxonomic techniques, which is the science of defining taxa on the basis of shared phenotypic characteristics and provides taxonomic names for these groups.

The primary aim of this chapter is to record the biodiversity of sponge fauna from two key habitats of the Bay of Plenty (a harbour and a coastal reef system), and based on this, to undertake a systematic revision of the fauna correlating 'historical' taxonomy with the modern phylogenetic assignments. Furthermore, consideration is given to workers in the biochemical, ecological and general marine biology fields, by incorporating notes on macroscopical features, reproduction and biochemistry where possible.

### 2.2 Materials and Methodology

#### 2.2.1 Field Collections

Sponges were collected from four sites around the Bay of Plenty: Karewa Island on the north eastern coast of the North Island of New Zealand, and three sites within Tauranga Harbour: Bridge Marina, Salisbury Wharf and Pilot Bay (Fig. 2.1). Sponges were all collected sub-tidally using SCUBA between January and October 2014. Sections were cut from whole individuals using a sharp scalpel, which allowed regrowth and survival. Ecological data were recorded including minimum and maximum depths, habitat, substrate, abundance, mucus emission, internal and external colour, size ranges and symbiotic coverage *in situ*. Additionally, details of gross and surface morphology, pigmentation, texture and dimensions of sponges in life were recorded. Site GPS coordinates were recorded using a WGS84 format in decimal degrees. Where possible, *in situ* photographs were taken of specimens. Photographs were also taken immediately after exposure to air, to determine whether oxidation initiated changes in pigmentation, Photographs were also taken after preservation in spirit (initially 90% ethanol). The locality and depth ranges are described with the systematic description for each individual specimen. Colour in life and after preservation in spirit was recorded using the Munsell system (Munsell 1942).



Figure 2.1: Aerial map of all dive sites sampled for sponges within the Bay of Plenty region (1:65,000).

#### 2.2.2 Specimen preparation

Upon collection, specimens were assigned unique field identification codes, placed within individual zip lock bags and then immediately placed on ice in a 105 litre chilly bin for transport. Specimen subsamples for taxonomic assignment were preserved in 500 ml glass jars initially fixed directly in 90% ethanol solution. At a later date, specimens were resorted and the fixative was drained and replaced with 100% ethanol. Sponge identification required two forms of histological preparation: first, a spicule preparation, to determine the diversity of spicules in the skeleton; and a second, a perpendicular thick section through the sponge tissue, to determine the structure of the skeleton, spicular geometry, the structure of the water-canal system, and other aspects of histology (Hooper 2000).

#### **Spicule preparation:**

For spicule preparations a nitric acid digestion was used. Small fragments ensuring a cross section of surface, matrix and (where possible) basal material (5 mm) of each specimen were placed in individual 15 ml, graduate, conical bottomed CELLSTAR<sup>®</sup> tubes. Several drops of nitric acid were placed on the fragments within a fume hood. The solution was then centrifuged in a Heraeus Sepatech centrifuge until all organic matter was dissolved. This was followed by three washes of distilled water and a final wash of 100% ethanol. Spicules were allowed to settle for approximately two minutes between each wash to avoid losing microscleres. The spicule extract was air dried on a slide and mounted using Canada balsam.

Spicule preparations for scanning electron microscopy (SEM) were dissolved in nitric acid and centrifuged as described above. Spicules were mounted on carbon coated glass cover slips; coated in sputter-deposited gold particles gold coated in sputter and viewed on a Hitachi S-4700 cold field emission SEM.

#### Thick section preparations:

Mineral skeletal arrangements were examined using thick hand-cut sections. A perpendicular section (50-100  $\mu$ m thickness) representing both the choanosomal and ectosomal skeletons were cut from a larger, preserved fragment, using a new, clean scalpel. Thick sections of 1.0 - 1.5 mm thick section of sponge ensuring both the ectosome and choanosomal skeletons were represented. Spicule dimensions were visualised using a Canon EOS 60D camera mounted on an Olympus BH-2 microscope. A 1 mm stage micrometre was photographed under

each objective. Images were then inserted into ImageJ software (Abramoff 2004) where a line tool was used to draw a selected line at a known length of 100  $\mu$ m. ImageJ was used to calculate the dimensions of spicules within each specimen. Where necessary, spicules were photographed using scanning electron microscopy. Photography of thick sections was undertaken using a Nikon SMZ745 compound microscope and a Canon EOS 60D camera.

#### 2.2.3 Taxonomic procedure

External morphological characteristics were examined firstly using in situ photographs, before carefully removing the voucher specimen from ethanol. Spicules and thick sections were examined, and contrasted against referenced material for the South Pacific region. Where a sponge was determined to be a potential new species and may require a new name, it is denoted by 'n.sp'. Where there is more than one 'n.sp.', these are numbered consecutively. Where there is 'n.sp. cf.', this denotes that this specimen may potentially become a new species; however, it is similar, and should be compared with the species mentioned thereafter by examining the type specimens and similar species for the genus (this was beyond the scope of this MSc but will be followed up in subsequent studies). All specimens were identified to species where possible and usually to at least genus using mainly the following key references: Bergquist, 1968; Bergquist, 1970; Bergquist 1980; Bergquist & Warne, 1980; Bergquist & Fromont, 1988; Kelly-Borges & Bergquist, 1997. Assignments were then further updated using the World Register of Marine Species Database (WoRMS 2015), to currently realign higher order taxonomy to accepted binomial nomenclature. However, when species names are used in the text, they are usually referring to species names described in Bergquist's publications, as in many instances, these monographs provide the only description of sponge species described from New Zealand. Where there was no taxonomic descriptions for orders, families or genera within the aforementioned literature from Bergquist, the reader is directed to Hooper, et al., (2002) for definitions. Figures for all specimens are kept in order of collection, but are cited based on the sequence of descriptions to keep the ordinal classifications of sponges together in the sequence of the Systema. Hence, orders of sponges were listed with a taxonomic format following the order of appearance in Hooper et al., (2002). The following criteria are used throughout the species descriptions.

#### **Description:**

External morphological features are recorded from observations of live specimens *in situ*, as contraction of tissue frequently causes a loss of features after exposure to air, or preservation in spirit. Sponge taxonomists are usually aware of phenotypic plasticity among specimens, however earlier descriptions of sponges by Bergquist and others (pre 1980's) relied largely on dredged and damaged specimens that had been landed. Unique micro-environmental conditions can also result in flexible, even uncommon external features in sponges. For example, *Cliona celata* specimens observed within a sheltered low wave environment in Pilot Bay are usually thickly encrusting; however, specimens observed at Karewa Island, in an area of high wave exposure, are usually thinly encrusting (this study). Hence, given this morphological flexibility, the shapes of a sponge can only usually be described in general terms, such as amorphous, massive, encrusting or ramose (Fromont 1985).

#### **Dimensions:**

Sponge sizes can be determined by skeletal arrangement and composition which assist in diagnosis. For example, the plumose non-anastomosing skeleton found in species of *Hymedesmia* limits the development of a sponge to a thick encrustation (Fromont 1985). The dimensions of all specimens collected were measured and representative samples of different sized individuals were indicative of the population. With this being said, we were mindful that there were many sponges at the alpha stages of their lifecycle. Bergquist and Warne (1980) suggested that skeletal structure and size can also be representative of the ecophenotypic response to environmental pressures such as wave exposure.

#### **Colour:**

Pigmentation on individuals may appear different depending on the light, and camera exposure conditions *in situ*. Nevertheless, colour differences may be used as additional systematic characteristics. The chemistry of some sponges cause pigment transformations after death or exposure to air. As an example, pigments in *Cliona celata* and *Iophon species* oxidise in air, changing from orange or yellow to purple, and then further changing to a brown colour morph after preservation in spirit. The majority of sponges, however, remain the same colour. For instance, *Tedania battershilli* occurs in different shades of orange in life and may become only slightly faded after preservation (Fromont 1985). Colour

examples and codes always refer to the system from the Munsell Colour Book (Munsell 1942).

#### **Texture:**

The consistency of sponges was recorded, which can assist in determining whether sponges are composed largely of mineral skeleton or spongin fibres. Sponges composed primarily of mineral skeletons are likely to be firm and crisp, e.g. *Stelletta maori*. On the other hand, sponges composed of largely spongin fibres are easily compressed and can be hard to tear e.g. species within the Dendroceratida order. Sponges with sand grains incorporated into their structure are likely to be brittle and incompressible e.g. *Chondropsis* species. Certain species are characterised by mucus production, namely *Carmia* and *Tedania* species (Fromont 1985).

#### Surface:

Three dimensional surface structures are developed in relation to oscula and ostia inhalant and exhalent canals. Species such as *Haliclona fragilis* develop characteristic fistulose structures. Similarly, the leathery vertucose macroscopical features on *Cliona celata* are characteristic of that species.

#### Skeleton:

The architecture of the mineral skeleton can be used in conjunction with spicule type, numbers of size classes and to some extent, abundance to characterise sponges to ordinal, familial, genera or species taxonomic levels. For diagrammatic representations of skeletal nomenclature used throughout this study refer to Boury-Esnault and Rutzler (1997). The "choanosomal" skeleton refers to the internal region of the sponge; the "ectosomal skeleton" refers to the superficial region of the sponge, with an immediate sub-surface skeleton.

#### **Spicules:**

Spicule size and morphological characteristics determine whether they are classed as megascleres or microscleres. However, there are no set sizes to these two types of spicules and megascleres have been defined as larger spicules which are macroscopically visible and contribute to the skeletal framework within the sponge, whereas smaller ones, microscleres, are packed between tracks of megascleres, supporting the soft parts (Hooper 2000). Megascleres act as the main structural component within the skeleton, whereas microsleres are generally packers between tracts of spicules, or are scattered among interstitial compartments or within ectosomal layers. A megasclere pointed at one end is monactinal; if pointed at one end and rounded at the other, it is referred to as diactinal. Spicule types and morphological features are shown in Figures 2.2, to 2.5. Terms relating to the arrangement of spicules and position within the skeletal structure are defined in Boury Esnault (1997).

Spicule morphology and geometry are relatively consistent within a species and can be used to determine the identity of taxa. Ten measurements of each spicule type, average and ranges gave an acceptable accuracy with minimum effort in a study by (Fromont 1985). Specifically, this study found that the degree of error decreased only slightly with an increased number of length measurements (Fromont 1985). As a result, ten measurements of each spicule type were taken within this study.



Figure 2.2.2: This page and opposite: a relatively small section of some of the possible spicule types (megascleres and microscleres); note sizes are not to scale. Diagram from *New Zealand Coastal Marine Invertebrates* (Battershill *et al.*, 2010).



Figure 2.3: This page and opposite: a relatively small section of some of the possible spicule types (megascleres and microscleres); note sizes are not to scale. Diagram from New Zealand Coastal Marine Invertebrates (Battershill *et al.*, 2010).



Figure 2.4: Diagrammatic representation of megasclere types found within the order *Poecilosclerida*. a-f, diactinal spicules: a, oxea; b, hastate oxea; c, strongyloxea; d, strongyle; e, tylote, f, tylote with spinned heads. g-l, monactinal spicules: g, style; h, anisostrongyloxea; i, tylostyle; j, subtylostyle; k, acanthostyle; l, acanthostyle with verticillate spining. Diagram from (Bergquist & Fromont 1988).



Figure 2.5: Diagrammatic representation of microsclere types found within the order *Poecilosclerida*. a, microstyle; b, microxea; c, raphide; d, trichodragmata; e, onychaete; f, toxa; g, tetrapocilli; h, sigma, C-shape; i, sigma, S-shape; j, sigma, hook-shape; k, comma; l, sigma with spines; m, inequieneded bipocilli, front view; n, inequieneded bipocilli, side view; o, arcuate isochelae, side view; p, arcuate isochelae, front view; q, anchorate isochelae, side view; r, anchorate isochelae, front view; u, birotulate chelae; v, palmate isochelae, front view; w, palmate isochelae, side view; x, palmate anisochelae, side view; y, palmate anisochelae, front view; z, placochelae, side view; bb. rosette of isochelae. Diagram from (Bergquist & Fromont 1988)

### 2.3 Species descriptions

General Note: In order not to be repetitive, the reader is referred to the primary literature for the Ordinal and Family level descriptions. These have not been changed or reviewed in this work. Rather, the assignments at genus and species level are linked between the original descriptions and the new ordinal level configuration as per the SYSTEMA/WoRMS 2015.

### **ORDER CHORISTIDA**

Definition: Original description found in Sollas (1885).

### FAMILY ANCORINIDAE

Definition: Original description found in Gray (1867).

### SUBFAMILY STELLETTINAE

Definition: Found in Schmidt (1870).

#### <u>Genus Stelletta</u>

Definition: Found in Bergquist, 1968, p. 44.

Stelletta crater (Dendy 1924) Spon00011 (Dendy 1924) (Plate 12, A-F).

| As Per Bergquist:       | As PER WORMS 2015:  |
|-------------------------|---------------------|
| Restricted synonymy:    | Updated synonymy.   |
| (Bergquist, 1968).      |                     |
| Class: Demospongiae     | Class: Demospongiae |
| Order: Choristida       | Order: Astrophorida |
| Family: Ancorinidae     | Family: Ancorinidae |
| Subfamily: Stellettinae |                     |
| Genus: Stelletta        | Genus: Stelletta    |
| Species: crater         | Species: crater     |
|                         |                     |

**Material examined**: Spon00011, Karewa Island, Bay of Plenty, 10.8 m. It should be noted that *Stelletta crater* and *Desmacella dendyi* were grouped together in this description as they had an intimate commensal relationship (see below).

Desmacella dendyi (Laubenfels 1936)

(Plate 12, A-F).

Spon00011

**Note:** This species does not belong to the Choristida order; however, it was commensal on the top of *Stelletta crater*. Consequently, descriptions were presented together for these specimens.

| AS PER BERGQUIST:            | AS PER WORMS 2015:     |
|------------------------------|------------------------|
| Restricted synonymy:         | Updated synonymy.      |
| (Bergquist & Fromont, 1988). |                        |
| Class: Demospongiae          | Class: Demospongiae    |
| Order: Poecilosclerida       | Order: Poecilosclerida |
|                              | Suborder: Mycalina     |
| Family: Biemnidae            | Family: Desmacellidae  |
| Genus: Desmacella            | Genus: Desmacella      |
| Species: dendyi              | Species: dendyi        |

**Material examined**: Spon00011, Karewa Island, Bay of Plenty, 10.8 m. It should be noted that *Stelletta crater* and *Desmacella dendyi* were grouped together in this description as they had an intimate commensal relationship.

**Description**: *Stelletta crater* is often cup shaped, however; this specimen was turbinate, resembling an inverted cone. The orange surface was due to the encrusting sponge *D. dendyi*. The second description is of *D. dendyi* which was completely covering *S. crater* with a thinly encrusting layer.

**Dimensions**: *D. dendyi* was 4 cm in length; width 2.5 cm; thickness 1.5 cm. *S.* crater was 2.5 cm in length; thickness 0.5 cm.

**Colour**: In life *S. crater's* colour was unknown as it was covered, in spirit reddish yellow 2.5Y5/4. In life *D. dendyi* was yellow-red 5.0YR7/10, in spirit yellow-red-yellow 10.0YR7/4.

**Texture**: *Stelletta crater* was incompressible and sandy to touch. *D. dendyi* was firm, and incompressible.

**Surface**: The surface of *S. crater* was intimately connected to *D. dendyi*, with triaenes protruding into the ectosomal skeleton of *D. dendyi*. The surface of *D. dendyi* was granular and microscopically hispid (Plate 12, A).

**Skeleton**: *S. crater* had spicules which radiated into its dermis, and had plagiotriaenes protruding into the choanosomal skeleton of *D. dendyi* (Plate 12, C). *Desmacella dendyi* had a fan shaped ectosomal skeleton composed of tylostyles. The skeleton also protruded into *S. crater's* choanosomal skeleton.

#### Spicules:

Megascleres: Tylostyles- slightly curved tylostyles, with characteristically welldeveloped tylote ends.

Microscleres: Sigmas- thin sigma's with a C-shape, or hooked ends.

For spicule dimensions see Table 2.1.

**Remarks**: It was recognised that *S. crater* is commonly found with *D. dendyi* commensal on its pinacoderm. Spicules from *S. crater* were growing intimately through *S. crater*, which can be seen as a light-dark band of plagiotriaenes (Plate 12, C-D).

| Locality   |       | Tylostyles<br>(µm)  | Sigmas<br>(µm) | Oxypheraster<br>(µm) |
|--|-------|---------------------|----------------|----------------------|
| Spon00011,<br>Karewa Island,<br>Bay of Plenty,<br>10.8 m | x     | 199 X 7.4           | 25             | 18                   |
|  | Range | 117-364 X<br>4.7-11 | 22-29          | 16-21                |

 Table 2.1: Spicule dimensions of Stelletta crater.

#### Chapter 2

### **Unknown family n.sp. 1** Spon00024

(Plates 28, F, 29, A-C).

| As Per Bergquist:                    | As PER WORMS 2015:  |  |  |
|--------------------------------------|---------------------|--|--|
|                                      | Updated synonymy.   |  |  |
| Class: Demospongiae.                 | Class: Demospongiae |  |  |
| Order: cf. Choristida or Hadromerida | Order: Unknown      |  |  |
| Family: Unknown                      | Family: Unknown     |  |  |
| Genus: Unknown                       | Genus: Unknown      |  |  |
| Species: n.sp.                       | Species: Unknown    |  |  |

Material examined: Spon00024, Karewa Island, Bay of Plenty, 10.8 m.

**Description**: This was a thickly encrusting sponge. There was an abundance of zoanthids commensal on the surface of the specimen *in situ*. After preservation in spirit, encrusting zoathids appear like they are a part of the sponge, forming small clusters on the surface. This specimen was competing for space with other sponges in a species rich habitat.

**Dimensions**: Length 4 cm; width 2 cm; thickness 1.5 cm. *In situ* specimens over 10 cm in diameter and 3 cm thick have been observed.

**Colour**: In life purple-blue purple 10.0PB3/2, in spirit purple-blue purple 10.0PB2/2.

Texture: Soft with a rubbery texture and difficult to compress.

**Surface**: Smooth with zoanthids (*Epizoanthus* sp.) scattered haphazardly over the surface (Plates 28, F, 29, A) in what appears to be a commensal relationship.

**Skeleton**: Tightly packed, thick, choanosomal skeleton, with megascleres arranged in uniform rows, all pointing in the same direction, perpendicular to the surface (Plate 29, B, C). Spicules are held together by spongin fibres.

#### Spicules:

Megascleres: Styles: single size class, straight styles. Strongyles were long and thick.

Microscleres: Microxyspherasters in low abundance. Tylasters in low abundance. For spicule dimensions see Table 2.2.

**Remarks**: There were a large number of broken spicules within this sample; with megascleres as large as 1.5 mm. The epibiont zoanthids on this specimen were

also growing on neighbouring thickly encrusting species of sponges at this site. The ecological relationship between the zoanthids and sponges requires further work to determine how they are interacting. The spicular configuration of this specimen did not match any known genera found within New Zealand shallow water environments (reported to date). There were elements of the sponge's spiculation and skeletal arrangement that would put this family either into the Choristida or Hadromerida. More work is required on this specimen where a wider range of type specimens need to be made available.
| Locality  |       | Styles<br>(µm)        | Strongyles<br>(µm)   | Tylasters<br>(µm) | Microxyspherasters<br>(µm) |
|---|-------|-----------------------|----------------------|-------------------|----------------------------|
| Spon00024,<br>Karewa Island, Bay of Plenty,<br>10.8 m | x     | 911 X 21              | 524 X 25             | 13                | 62                         |
|   | Range | 84-1582 X<br>2.2-35.7 | 375-635 X<br>14.9-46 | 11.3-13.8         | 43-71                      |

Table 2.2: Spicule dimensions of Unknown n.sp.1

*Stelletta maori* (Dendy 1924) Spon00010 (Plate 11, A-F) Spon00012 (Plates 13, A-F, 14, A-F, 15, A-D).

| As Per Bergquist:      | AS PER WORMS 2015:  |
|------------------------|---------------------|
| Restricted synonymy:   | Updated synonymy.   |
| (Bergquist, 1968).     |                     |
| Class: Demospongiae    | Class: Demospongiae |
| Order: Choristida      | Order: Astrophorida |
| Family: Ancorinidae    | Family: Ancorinidae |
| Subfamily: Stelletinae |                     |
| Genus: Stelletta       | Genus: Stelletta    |
| Species: maori         | Species: maori      |
|                        |                     |

**Material examined**: Spon00010, Karewa Island, Bay of Plenty, 10.8 m. Note: Spon00010 is the same species as Spon00012, therefore it was grouped into this description.

**Description**: Both specimens were caliculate shaped *in situ*. Sizes and shapes of this species can vary. For instance, morphological forms include massive, vasiform, or pillow-like formations (Bergquist 1968).

**Dimensions**: Length 7 cm; width 4.5 cm; thickness 3 cm. *In situ* specimens over 10cm have been observed. Spon00012 was 3.5 cm in length; width 4.5 cm; thickness 1.7 cm.

**Colour**: In life Spon00010 was purple 5.04/8, in spirit yellow-red-yellow 10.0YR7/4. In life and in spirit Spon00012 was the same colour as Spon00010 in its respective states.

**Texture**: Both specimens have grainy textures that are firm but slightly compressible.

**Surface**: The surface of both specimens is slightly hispid and coarsely granular (Plate 11, B). There were no oscules or pores visible on the dermis.

**Skeleton**: The architecture of the ecotosomal skeleton is congruent with *S. maori* described in Bergquist (1968), with a dermal layer of euasters, in addition to an abundance of euasters that are scattered throughout the choanosomal skeleton

(Plate 11, C, D). The dermal layer of euasters was "ill" defined in both specimens, as per the description of *Stelletta maori* in Dendy (1924).

## **Spicules:**

Megascleres: Dichotriaenes- these are plagiotriaenes in which clads are bifurcate. Dichotriaenes form a dense layer of dermal armour, with clads facing the dermis. The lengths and widths of dichotriaenes varied greatly. Plagiotriane- There was much fewer plagiotrienes present in comparison to the abundance of dichotriaenes. These also varied in size, but were usually of similar sizes to plagiotriaenes. Oxeas varied greatly in size.

Microscleres: Oxypherasters- the rays were smooth without sharply pointed ends. There were no particular size classes of oxypherasters. Euasters- had 11-12 spines, and sharply pointed rays.

For spicule dimensions see Table 2.3.

**Remarks**: Asters were found throughout the specimen, including euasters, oxypherasters and a single oxyaster. It was difficult to distinguish between the variations in spines of each aster type, based on the presence and absence of spines.

| Locality  |                         | Oxeas         | Tylostyles | tyles Plagiotriaenes | Dichotriaenes | Euasters    | Oxypherasters |
|---|-------------------------|---------------|------------|----------------------|---------------|-------------|---------------|
| Locanty   |                         | ( <b>µm</b> ) | (µm)       | (μm)                 | (µm)          | (µm)        | (µm)          |
| Spirits Bay, 11-20 fm<br>Dendy Type   | x                       |               |            |                      | 1800 X 68     |             | 20-66         |
| Average <i>Stelletta maori</i> spicule<br>dimensions from specimens<br>collected in New Zealand<br>(Bergquist, 1968). | x                       | 231 X 8.9     |            |                      | 2159 X 37     |             | 12            |
|   | Dongo                   | 206-255 X     |            |                      | 1792-2619 X   |             | 0.2.16        |
|   | Kange                   | 8-9.7         |            |                      | 28-43         |             | 9.2-10        |
| Spon00010,  |                         |               |            |                      |               |             |               |
| Karewa Island,  |                         |               |            |                      |               |             |               |
| Bay of Plenty,  | $\overline{\mathbf{X}}$ | 246 X 8       | 263 X 8.3  | 847 X 118            | Broken        | 20.5 X 18.6 | 18.3 X 16.2   |
| 10.2 m  |                         |               |            |                      |               |             |               |
|   |                         |               |            |                      |               |             |               |
|   | Panga                   | 133-360 X     | 187-295 X  | 399-1380 X           | Broken        | 19-25 X     | 17-20 X       |
|   | Range                   | 6.8-9.2       | 4.1-15     | 48-230               | 16.7-23       |             | 15-17.6       |

 Table 2.3: Spicule dimensions of Stelletta maori.

| Chapter 2                            | Species description of the Demospongiae |
|--------------------------------------|---|
| Stelletta sandalinum (Brønsted 1924) | (Plates 21, D-F, 22, A-C).              |
| Spon00017                            |   |
| As Per Bergquist:                    | As Per WoRMS 2015:                      |
| Restricted synonymy:                 | Updated synonymy.                       |
| (Bergquist 1968).                    |   |
| Class: Demospongiae                  | Class: Demospongiae                     |
| Order: Choristida                    | Order: Astrophorida                     |
| Family: Ancorinidae                  | Family: Ancorinidae                     |
| Subfamily: Stelletinae               |   |
| Genus: Stelletta                     | Genus: Stelletta                        |
| Species: sandalinum                  | Species: sandalinum                     |

Material examined: Spon00017, Karewa Island, Bay of Plenty, 10.8 m.

**Description**: The sponge was amorphous to massive, with a flattened hispid surface. The specimen resembled the original type specimen described in Bergquist (1968) which was cushion-shaped flattened and hispid.

**Dimensions**: Length 3 cm; width 1.5 cm; thickness 0.5 cm. Specimens over 7 cm in diameter by 3 cm thick have been observed at this site.

**Colour**: In life pale white, in spirit pale white.

Texture: The sponge was tough and relatively hard to compress.

**Surface**: Rough to touch, with a stony, hispid dermal layer and a cushion-shaped body.

**Skeleton**: The skeletal structure was fascicular fibrous with plagiotriaenes fanning outwards towards the dermis (Plate 22, A).

#### **Spicules:**

Megascleres: Plagiotriaenes- were broken in the spicule slides; however, ends were clearly visible. Oxeas- of a single size class.

Microscleres: Oxyasters- these spicules were generally scarce throughout the spicule slide. Strongylospherasters- were abundant in the spicule slide.

For spicule dimensions see Table 2.4.

| Locality   |                         | Plagiotriaenes<br>(µm) | Oxeas<br>(µm) | Small<br>oxyaster<br>(µm) | Strongylospheraster<br>(µm) |
|--|-------------------------|------------------------|---------------|---------------------------|-----------------------------|
| Slipper I., low water  |                         | 2250 X 80              | 2500 X 52     |                           | 8                           |
| Brønsted Type (Bergquist, 1968).<br>Average Stelletta sandalinum spicule |                         |                        |               |                           |                             |
| dimensions from specimens collected                                      | $\overline{\mathbf{X}}$ | 2120 X 64              | 2170 X 51     | 15                        | 6                           |
| in New Zealand (Bergquist, 1968).  |                         |                        |               |                           |                             |
|  | Range                   | 1644-2654 X            | 1753-2590 X   | 11 5-20                   | 4 5-7 4                     |
|  | Runge                   | 53-73                  | 47-57         | 11.5 20                   | 1.5 7.1                     |
| Spon00017, Karewa Island,  | $\overline{\mathbf{x}}$ | Broken                 | 125 X 2.9     | 12.4                      | 4.4                         |
| Day of Flenty, 10.6 III.   | Range                   | Broken                 | 111-131 X     | 10-16.2                   | 4.2-4.5                     |
|  | Tunge                   | 210101                 | 2-3.7         | 10 10.2                   |                             |

 Table 2.4: Spicule dimensions of Stelletta sandalinun.

# **ORDER HADROMERIDA**

Definition: Found in Hooper & Van Soest 2002, p. 169.

# FAMILY CLIONAIDAE

Definition: Found in Hooper & Van Soest, 2002, p.173.

#### Genus Cliona

Definition: Found in Bergquist, 1968. p. 29.

| Cliona celata (Grant 1826)             | (Plate 3, A-E),     |
|--|---------------------|
| Spon00002                              |                     |
| <u>A Per Bergquist</u> :               | As PER WORMS 2015:  |
| Restricted synonymy: (Bergquist, 1968) | Updated synonymy.   |
| Class: Demospongiae                    | Class: Demospongiae |
| Order: Hadromerida                     | Order: Hadromerida  |
| Family: Clionaidae                     | Family: Clionidae   |
| Genus: Cliona                          | Genus: Cliona       |
| Species: celata                        | Species: celata     |

**Material examined**: Spon00002, Pilot Bay, Tauranga Harbour, 10.6 m. References made to the material described in Bergquist & Fromont (1988); particularly their collections. *Cliona celata* is found throughout New Zealand from sheltered shallow embayment's to deep shore environments that are heavily exposed form the Three Kings through to the Chatham Islands and internationally. A restricted synonymy is provided in Bergquist & Fromont (1988), and recent reviews of the genus are provided (Schönberg 2000).

**Description**: When compared to specimens described in (Bergquist 1968), *C. cellata* found in Pilot Bay is typical of Clionids found in New Zealand and internationally. Although the specimen examined here was thick, there were also thick and thin encrusting specimens, in addition to bio-eroding individuals, at the site where this specimen was collected.

**Dimensions**: Length 12 cm; width 9 cm; thickness 5 cm; taken as a section of a larger sponge, specimens over a meter in diameter of thin to thickly encrusting sponges have been observed in this region.

**Colour**: In life 5.0Y8/8, in spirit 5.0YR4/2–3/4. Internal and external colours are analogous. Note: this specimen immediately changed colour on exposure to air, and further changed colour in spirit. *Cliona celata* usually undergo oxidation in air to become purple, interestingly, this specimen did not turn purple but became orange when exposed to air.

**Texture**: Leathery, difficult to tear, slightly compressible.

**Surface**: A lumpy low relief surface. There are vurrucose macroscopical structures with ostia on the surface of these structures, with larger oscula interspersed between grooves (Plate 3, B-D).

**Skeleton**: Disordered mass of spicules, with a confused skeletal structure lacking clear tracts or fibres.

Spicules: (Plate 3, A).

Megascleres: Tylostyles- One size class of marginally curved tylostyles with oval to globular surfaces at the base.

For spicule dimensions see Table 2.5.

**Remarks**: Spon00002 has all the components of a *Cliona* sp., including a single spicule type of tylostyle megascleres. Spiculation was assessed closely as the pigmentation change observed on exposure to air was different from that normally seen in *C. celata*, hence some comment on closely related species is appropriate. There were some small acanthostyles, in low abundance, thus these were considered foreign.

| Locality                                   |                         | Large<br>tylostyles<br>(µm) |
|--|-------------------------|-----------------------------|
| France (Topsent 1900a)                     | Range                   | 180-360 X 3-9               |
| East Haven Conn. YPM 767<br>(Hartman 1958) | $\overline{\mathbf{x}}$ | 323 X 9.1                   |
|  | Dongo                   | 213-377 X                   |
|  | Kallge                  | 7-11.9                      |
| Goat Island Bay (Bergquist 1968)           | $\overline{\mathbf{X}}$ | 298 X 8.1                   |
| <b>u</b> stage                             |                         | 260-339 X                   |
|  | Range                   | 7.2-9.4                     |
| Narrow neck (Bergquist 1968)<br>α stage    | x                       | 294 X 8.6                   |
|  |                         | 217-350 X                   |
|  | Range                   | 5.7-11.5                    |
| Gt. Barrier (Bergquist 1968)<br>γ stage    | x                       | 268 X 7.6                   |
|  |                         | 200-290 X                   |
|  | Range                   | 5.7-8.9                     |
| Spon00002                                  |                         |                             |
| Pilot Bay, Tauranga,                       | $\overline{\mathbf{X}}$ | 284 X 9.7                   |
| 10.6 m, γ stage                            |                         |                             |
|  | Deres                   | 246-307 X                   |
|  | Kange                   | 6.3-12.3                    |
|  |                         |                             |

Table 2.5: Spicule dimensions of *Cliona celata*.

#### Species description of the Demospongiae

Chapter 2

*Cliona* n. sp.1 *cf. celata* Spon00019 (Plates 23, D-F, 24, A-C).

| As Per Bergquist:          | As Per WoRMS 2015:  |
|----------------------------|---------------------|
| Restricted synonymy:       | Updated synonymy.   |
| (Bergquist, 1968).         |                     |
| Class: Demospongiae        | Class: Demospongiae |
| Order: Hadromerida         | Order: Hadromerida  |
| Family: Clionaidae         | Family: Clionidae   |
| Genus: Cliona              | Genus: Cliona       |
| Species: n.sp.1 cf. celata | Species: celata     |

Material examined: Spon00019, Karewa Island, Bay of Plenty, 10.8 m.

**Description**: This specimen was thinly encrusting on a rock wall at a wave exposed site near Karewa Island. The specimen appeared to have been slightly damaged due to predation.

**Dimensions**: Length 5 cm; width 3.5 cm; thickness 0.7 cm. *In situ* specimens over 20 cm in diameter and 4 cm thick have been observed.

**Colour**: In life yellow-red-yellow 10.0YR8/8, in spirit yellow-red-yellow 10.0YR 4/4.

Texture: Leathery and firm.

**Surface**: A lumpy low relief surface. There is a structure with ostia on the surface of the projections and larger oscula interspersed between grooves (Plate 24, D).

**Skeleton**: Disordered mass of spicules, with a plumoreticulate to confused skeletal structure lacking clear tracts or fibres (Plate 23, D, E).

### Spicules:

Megascleres: Subtylostyles- tylostyles with one end pointed and the other with an obvious knob (Plate 24, C). Tylostyles- of normal form.

For spicule dimensions see Table 2.6.

**Remarks**: This specimen is believed to be a different species of *Cliona* as it has subtylostyles and tylostyles as opposed to only having tylostyles, which is congruent of other *Cliona celata* specimens collected by Bergquist. It should be noted that further molecular studies may support this differentiation. There did not appear to be any difference in colour *in situ* or in spirit between the two

specimens of *Cliona* which were collected at Pilot Bay (Spon00002) versus Karewa Island (Spon00019). From other studies of Cliona sponges in this region, (Webb MSc thesis in prep), there appear to be at least two variations of the *Cliona celata* morphotype based on chemistry. This taxonomic assignment will be expanded to support future chemical ecological and chemotaxonomic research.

| Locality               |                         | Subtylostyes | Tylostyle |  |
|------------------------|-------------------------|--------------|-----------|--|
| Locality               |                         | (µm)         | (µm)      |  |
| Spon00019,             |                         |              |           |  |
| Karewa Island,         | $\overline{\mathbf{X}}$ | 273 X 8.9    | 175 X 4.9 |  |
| Bay of Plenty, 10.8 m. |                         |              |           |  |
|                        | Dongo                   | 265-281 X    | 126-279 X |  |
|                        | Kange                   | 8-9.7        | 4-4.8     |  |
|                        |                         |              |           |  |

Table 2.6: Spicule dimensions of *Cliona* n.sp.1

Chapter 2

Spon00041

Cliona n. sp. 2 cf. celata

(Plates 39, A-F, 40, A).

| As Per Bergquist:    | As Per WoRMS 2015:  |
|----------------------|---------------------|
| Restricted synonymy: | Updated synonymy.   |
| (Bergquist, 1968)    |                     |
| Class: Demospongiae  | Class: Demospongiae |
| Order: Hadromerida   | Order: Hadromerida  |
| Family: Clionaidae   | Family: Clionidae   |
| Genus: Cliona        | Genus: Cliona       |
| Species: cf. celata  | Species: Unknown    |

**Material examined**: Spon00041, Pilot Bay, Tauranga Harbour, 15 m. References made to the material described in Bergquist & Fromont (1988); particularly their collections. *Cliona celata* is found throughout New Zealand from sheltered shallow embayment's to deep shore environments that are heavily exposed form the Three Kings through to the Chatham Islands and internationally. A restricted synonymy is provided in Bergquist & Fromont (1988), and recent reviews of the genus are provided (Schönberg 2000).

**Description**: The specimen collected here was a thinly encrusting species that was found on rock in Pilot Bay. The specimen was difficult to remove from the substrate without tearing the body apart.

**Dimensions**: Length 2 cm; width 1 cm; thickness 0.2 cm.

**Colour**: In life yellow green-yellow 10.0Y8/8, in spirit white.

Texture: Leathery, soft, difficult to tear and slightly compressible.

**Surface**: The sponge has a smooth surface with small chimney like projections on sections of the dermis (Plate 39, A-C).

**Skeleton**: The choanosomal skeleton has a plumoreticulate structure with a bouquet of megascleres at the ends of the spicule tracts that pierce the dermis making the sponge microscopically hispid (Plate 39, E). There is an abundance of sand grains which have been incorporated into the choanosomal skeletal structure. The ectosomal skeleton has both a palisade of megascleres aligned outwards in addition to megascleres which are aligned parallel to the surfaces.

# **Spicules:**

Megascleres: This specimen has two size classes of tylostyles. The smaller size classes are straight and the larger class are slightly curved (Plate 39, F). For spicule dimensions see Table 2.7.

**Remarks**: This specimen appeared slightly different to *Cliona celata* and *Cliona n.sp1* (described above) in that it lacked a verrucose surface, had a plumoreticulate choanosomal skeleton and had two size classes of tylostyles. The colour was also unusual for Cliona celata and no oxidation of pigments was observed.

| Locality              |       | Large<br>tylostyles | Small<br>tylostyles      |  |
|-----------------------|-------|---------------------|--------------------------|--|
| Spon00041, Pilot Bay, | x     | (μm)<br>325 X 7.1   | <b>(μm)</b><br>154 X 5.1 |  |
| Bay of Plenty, 5 m    | _     | 255-408 X           | 132-178 X                |  |
|                       | Range | 5.5-8.5             | 4.2-6                    |  |

Table 2.7: Spicule dimensions of *Cliona* sp.

# FAMILY SUBERITIIDAE

Definition: Found in Hooper & Van Soest 2002, p. 227.

## Genus Polymastia

Definition: Found in Bergquist, 1968, p. 21.

*Polymastia fusca* (Bergquist 1968) Spon00020 (Plates 24, D-F, 25, A-B).

| As Per Bergquist:    | As Per WoRMS 2015:    |
|----------------------|-----------------------|
| Restricted synonymy: | Updated synonymy.     |
| (Bergquist, 1968)    |                       |
| Class: Demospongiae  | Class: Demospongiae   |
| Order: Hadromerida   | Order: Hadromerida    |
| Family: Subertiidae  | Family: Polymastiidae |
| Genus: Polymastia    | Genus: Polymastia     |
| Species: fusca       | Species: fusca        |
|                      |                       |

Material examined: Spon00020, Karewa Island, Bay of Plenty, 10.8 m.

**Description**: A spherical specimen with a consistent characteristic vurrucose layer over the entire dermis.

**Dimensions**: Length 5 cm; width 4 cm; thickness 1cm. Specimens over 10 cm in diameter have been observed at this site.

**Colour**: In life green-yellow green 10.0GY6/4, in spirit green-yellow green 10.0GY2/2.

**Texture**: Firm and fleshy.

**Surface**: Leathery with a smooth vurrucose consistency.

**Skeleton**: The choanosomal skeleton is composed of stout radiating tracts of large subtylostyles, with subtylostyles condensed thickly in the endosome (Plate 24, F). The choanosomal skeleton is dendritic in that it has single ramifying fibres that branch but rarely anastomose. The ectosomal skeleton is a thick palisade of subtylostyles arranged tangentially to the surface.

# Chapter 2

# Spicules:

Megascleres: Subtylostyles- there were two size classes of subtylostyles present: large and small classes. Styles- were predominately straight.

For spicule dimensions see Table 2.8.

| Locality   |                         | Medium<br>subtylostyles<br>(µm) | Small<br>subtylostyles<br>(µm) | Styles<br>(m) |
|--|-------------------------|---------------------------------|--------------------------------|---------------|
| Burges Bay, Kawau (littoral)<br>Type   | $\overline{\mathbf{X}}$ | 435 X 8                         | 140 X 3.2                      |               |
|  | Range                   | 380-480 X                       | 101-159 X                      |               |
|  | Range                   | 8                               | 1.6-4.6                        |               |
| Average <i>Polymastia fusca</i><br>spicule dimensions from<br>specimens collected in New<br>Zealand (Bergquist, 1968). | x                       | 431 X 6.6                       | 162 X 4.2                      |               |
|  | Range                   | 299-516 X                       | 142-182                        |               |
|  | itunge                  | 5.6-8                           |                                |               |
| Spon00020, Karewa Island,<br>Bay of Plenty, 10.8 m.  | $\overline{\mathbf{X}}$ | 363 X 5.5                       | 162 X 3.4                      | 211 X 3.4     |
|  | Panga                   | 267-495 X                       | 97-201 X                       | 167-294 X     |
|  | Kallge                  | 2.3-9.3                         | 2.4-4                          | 2.4-4         |

# Table 2.8: Spicule dimensions of Polymastia fusca.

# **ORDER AXINELLIDA**

Note: This order is not accepted in Hooper & Van Soest, 2002. Consequently an updated definition for this order could not be located.

The order Axinellida was created for sponges with axially strengthened skeletons and oviparous reproduction. These features are no longer considered indicators for a monophyletic grouping as per (Hooper & Van Soest 2002). Families assigned to this order have been dispersed among three currently recognized orders, Halichondrida, Hadromerida and Poecilosclerida. The genus *Axinella* and the Axinellidae family are presently assigned to Halichondrida. However, recent molecular studies indicate the presence of a monophyletic group largely similar in content to Levi (1953). Orders may need to be revised to update current nomenclature.

# FAMILY DESMOXYIDAE

Definition: Found in Bergquist, 1970, p. 22.

Genus Acanthoclada

Definition: Found in Bergquist, 1970, p. 22.

# Acanthoclada prostrata

Spon00047 (Plates 45, D-F, 46, A-C). Spon00052 (Plate 50, A-E)

| As Per Bergquist:    | As Per WoRMS 2015:   |
|----------------------|----------------------|
| Restricted synonymy: | Updated synonymy.    |
| (Bergquist 1970)     |                      |
| Class: Demospongiae  | Class: Demospongiae  |
| Order: Axinellida    | Order: Halichondrida |
| Family: Desmoxyidae  | Family: Heteroxyidae |
| Genus: Acanthoclada  | Genus: Acanthoclada  |
| Species: prostrata   | Species: Unknown     |

**Material examined**: Spon00047, Pilot Bay, Tauranga Harbour, 15 m. Spon00052, Pilot Bay, Tauranga Harbour, 15 m. **Description**: The Spon00047 specimen was massive to amorphous with a bright orange colour in life. The surface has conulose projections with a microscopically hispid uneven pinacoderm. This sponge was found encrusting on rock in Pilot Bay in approximately 15 meters of water. The Spon00052 specimen was globular *in situ* with similar small conulose projections on the dermal membrane to Spon00047. Note: Spon00047 and Spon00052 are the same species; therefore they were grouped together in this description.

**Dimensions**: Spon00047 was 3 cm in length; width 1.5 cm. Spon00052 was 3.4 cm in length; width 2.5 cm; thickness 1 cm.

**Colour**: In life Spon00047 is yellow-red 5.0YR7/10, in spirit yellow 5.0Y8/6. The colour of Spon00052 in life is unknown, in spirit yellow 5.0Y8/4.

**Texture**: The Spon00047 specimen has a course texture and is relatively ridged when compressed; however, it is easy to pull apart. The texture of Spon00052 is somewhat softer on the surface compared to the Spon00047 specimen, although they appear morphologically similar

**Surface**: The Spon00047 specimen has many small conulose structures which gives an uneven macroscopically hispid appearance (Plate 45, E). The Spon00052 specimen has the same conulose structures present on the dermis; however, they appear slightly thicker than Spon00047 (Plate 50, A).

**Skeleton**: The skeletons in both the specimens were plumoreticulate to lax. There were distinguishable plumose tracts of megascleres; however, this arrangement becomes vague as the abundance of megascleres increases throughout the choanosome. Spongin B is also present around the fibres. The ends of tracts radiate to form dense bundles which appear to protrude from the dermis.

#### Spicules:

Both specimens had the same spicule types and classes.

Megascleres: Styles- large which made them difficult to measure under light microscopy. Rhabdostyles- large with broad shafts.

Microscleres: Birotules- these were originally described in this study as dentate stigmata's due their curvature and size (Plate 46, B, C). However, it was later confirmed that they are in fact birotules, due to their nail-like heads as per Bergquist, 1970 p 22.

Cladoxas- these had a smooth curved shaft, and occurred throughout the sponge. For spicule dimensions see Table 2.9. **Remarks:** This specimen had all of the characteristics of *Acanthoclada prostrata* as per Bergquist, 1970; however, it lacked a spicule complement of oxeas, consequently it requires further confirmation with the type specimen of *A. prostrata*.

| Locality                                      |                         | Styles                | Rhabdostyles        | Birotules | Cladotoxas |
|---|-------------------------|-----------------------|---------------------|-----------|------------|
|   |                         | (µm)                  | (µm)                | (µm)      | (µm)       |
| North Channel,                                |                         |                       |                     |           |            |
| 10 fm, Bergquist, 1970                        | $\overline{\mathbf{X}}$ | 1206 X<br>7.2         | 420 X 7             | 66        | 90 X 4.9   |
| (Holotype).                                   |                         |                       |                     |           |            |
|   | Range                   | 960-<br>1320 X        | 213-600 X           | 52-72 X   | 80-96 X    |
|   | Tunge                   | 1.2-9.2               | 5.7-8.1             | 3.4-3.8   | 4.6-5.6    |
| Spon00047, Pilot Bay,<br>Bay of Plenty, 15 m. | x                       | 275 X<br>8.1          | 262 X 9             | 46        | 140        |
|   | Range                   | 224-<br>338 X<br>5-10 | 188-384 X<br>7-14.9 | 35-61     | 92-188     |

 Table 2.9: Spicule dimensions of Acanthoclada sp.

# **ORDER POECILOSCLERIDA**

Definition: Found in Bergquist & Fromont, 1988, p.17.

# FAMILY CRELLIDAE

Definition: Found in Bergquist & Fromont, 1988, p. 75.

#### Genus Crella

Definition: Found in Bergquist & Fromont, 1988, p. 76.

*Crella incrustans* (Carter 1885)

(Plates 1, A-F, 2, A-F)

Spon00001

Spon00003

| As Per Bergquist:           | As Per WoRMS 2015:     |
|-----------------------------|------------------------|
| Restricted synonymy:        | Updated synonymy.      |
| (Bergquist & Fromont 1988). |                        |
| Class: Demospongiae         | Class: Demospongiae    |
| Order: Poecilosclerida      | Order: Poecilosclerida |
|                             | Suborder: Myxillina    |
| Family: Crellidae           | Family: Crellidae      |
| Genus: Crella               | Genus: Crella          |
| Species: incrustans         | Species: incrustans    |
|                             |                        |

**Material examined**: Spon00001, Pilot Bay, Tauranga Harbour, 10.6 m References made to the material described in Bergquist & Fromont (1988) particularly collections from Harington Point, Dunedin, Leigh Reef and the Canyons at Leigh. Note Spon00003 is the same species as Spon00001, therefore it was grouped into this description.

**Description**: The sponge is a thickly encrusting to amorphous, massive sponge with a soft and fleshy texture found encrusting on rock. The sponge was surrounded by other sponges and encrusting organisms. Hydroids were present as epibionts and the sponge was found living next to red algae (species of algae unknown). Crella incrustans was common within Pilot Bay. Unlike Spon00001 there were no hydroids found on Spon00003.

**Dimensions**: Length 12 cm; width 10 cm; and 4 cm high turrets ridges. *In situ* specimens over 50 cm in diameter by 5 cm thick have been observed. Spon00003 was 9 cm in length; width 8 cm; thickness 4 cm.

**Colour**: Spon00001 was orange-brown to yellow-red-yellow 10YRY7/10 in life, to yellow-red-yellow 10YR5/4 inside and out in spirit. Both Spon00003 and Spon00001 were the same colours in life and after preservation in spirit.

**Texture**: Firm but compressible sponge, relatively easily torn, little mucus on damage. The surface feels fibrous.

**Surface**: Lumpy, relatively smooth, oscula relatively obvious haphazardly scattered over the sponge's surface, ridges and tending towards conulose projections of various dimensions (Plate. 2E, F). The overall shape and texture is known to be variable according to environmental exposure.

**Skeleton**: The choanosomal skeleton is a plumose to plumose reticulate structure of collagen fibres around which oxeas are aligned and acanthostyles are tangentially echinating (Plate 1, C, F). The choanosome is characterised by frequent large openings. Towards the surface the plumose columns align perpendicularly forming a dense sub-dermal layer. Note: the skeletal structure was similar in both Spon00001 and Spon00003 specimens.

**Spicules**: (Plates 1, A, B, D, E, 2, A-D)

Megascleres: Hastate oxeas - Smooth oxeas with hastate ends invariably straight, ends can be slightly different (Plate 2, A-C). Acanthostyles- of two size classes, spinned all over, with no verticillate spinning. Some acanthostyles are slightly bent (Plate 1, D). As observed by Bergquist & Fromont (1988) curved acanthostyle are observed in the pinacoderm, straighter versions are found throughout the matrix. It should be noted that the spicule descriptions are for the spicules found in the Spon00001 specimen.

Microscleres: Small isochelae - arcuate to palmate isochelae.

For spicule dimensions see Table 2.10.

**Remarks**: I had a close look at other *Crella* as reported by Bergquist & Fromont (1988), and through close association with Chris Battershill (who gathered most of this material for Bergquist & Fromont, (1988)); I have concluded that this species is evidently similar to *C. incrustans*. Differences from *C. incrustans* observed elsewhere, were that the specimens collected in this study were more yellow in life at 10YRY7/10 compared to 7.5R5/10 respectively (Bergquist & Fromont, 1988). The specimen collected came away from the substrate without the basal layer, so confirmation of a collagen basal mat with echinating acanthostyles, as per Hallmann (1914) cannot be confirmed.

| Locality  |                         | Oxea<br>(µm)      | Large<br>ancanthostyles<br>(µm) | Small<br>acanthostyles<br>(µm) | Isochelae<br>(µm) |
|---|-------------------------|-------------------|---------------------------------|--------------------------------|-------------------|
| ТҮРЕ  |                         |                   | 150 X 4                         | •                              |                   |
| North Cape,   |                         | 176 X 5           | (not divided into distinct      |                                | 16                |
| 26-55 m (Dendy 1924).   |                         |                   | categories)                     |                                |                   |
| ТҮРЕ  | $\overline{\mathbf{x}}$ | 174 X 5 3         | 141 X 8 7                       | 83 X 7                         | 15.3              |
| Remeasured (Bergquist & Fromont, 1988).   | л                       | 17 <b>-</b> X 3.5 | 141 A 0.7                       | 03 A /                         | 15.5              |
|   | Denes                   | 160-197           | 130-155                         | 58-100                         | 14 17             |
|   | Kange                   | X 5-6             | X 7.5-9.5                       | X 5.5-8                        | 14-1/             |
| Average <i>Crella incrustans</i> spicule dimensions<br>from specimens collected in New Zealand<br>(Bergquist & Fromont, 1988) | $\overline{\mathbf{x}}$ | 174 X 5.4         | 145 X 8.1                       | 86 X 6.7                       | 17                |
|   | D                       | 156 -188          | 134-156                         | 64-104                         | 15.10             |
|   | Range                   | X 4.2-6.1         | X 6.3-9                         | X 4.6-7.9                      | 15-19             |
| Spon00001,  |                         |                   |                                 |                                |                   |
| Pilot Bay, Tauranga,  | $\overline{\mathbf{X}}$ | 190 X 4.5         | 157 X 9                         | 95 X 7.3                       | 15.4              |
| 10.6 m  |                         |                   |                                 |                                |                   |
|   | _                       | 180-195           | 157-158                         | 87-100                         | 14 7-16 1         |
|   | Kange                   | X 4-5             | X 8.7-9.5                       | X 6.6-7.8                      | 1, 10.1           |

 Table 2.10: Spicule dimensions of Crella encrustans.

# FAMILY DESMACIDONIDAE

Definition: Found in Bergquist & Fromont, 1988, p. 36.

## Genus Tetrapocillon

Définition: Found Bergquist & Fromont, 1988, p. 46.

Tetrapocillon n.sp. 1 cf. novaezelandiae

Spon00050

(Plate 49, A-F).

| As Per Bergquist:                 | As PER WORMS 2015:     |
|-----------------------------------|------------------------|
| Restricted synonymy:              | Updated synonymy.      |
| (Bergquist & Fromont 1988).       |                        |
| Class: Demospongiae               | Class: Demospongiae    |
| Order: Poecilosclerida            | Order: Poecilosclerida |
|                                   | Suborder: Mycalina     |
| Family: Desmacidonidae            | Family: Guitarridae    |
| Genus: Tetrapocillon              | Genus: Tetrapocillon   |
| Species: n.sp. cf. novaezelandiae | Species: Unknown       |

Material examined: Spon00050, Pilot Bay, Tauranga Harbour, 15 m.

**Description**: The sponge is amorphous to massive with a smooth soft dermal layer, and a leathery texture. In life the surface has visible oscules which are at the apex of conulose structures but these become flattened and retracted in spirit. This sponge was competing for space with other encrusting sponge species.

**Dimensions**: Length 5.5 cm; width 5 cm; thickness 1.2 cm. *In situ* specimens above 10 cm have been observed.

Colour: In life yellow 5.0Y8/2 to yellow 5.0Y8/8, in spirit same colour as in life.

**Texture**: Soft and velvety to touch, the dermal layer is firm but pliable and easier to tear beneath.

**Surface**: Oscules were visible, protruding from the dermis in life (Plate 49, A), however, these retracted within the preserved sample, lying flush with the surface (Plate 49, C).

**Skeleton**: The sponge has a reticulated fibrous choanosomal skeleton (Plate 49, D). The fibres are cored by megascleres with an abundance of spongin type B.

The skeletal structure appears confused in areas with large quantities of spicules. The ectosomal skeleton has a thick layer of tangential spicules.

### Spicules:

Megascleres: Styles- some subtylote, and some like oxeas. Strongyles- normal form. Tetrapocilli- normal form.

Microscleres: Small sigmas- very small spinned, C-shape sigmas. Birotules- spiny and curved.

For spicule dimensions see Table 2.11.

**Remarks**: This sponge is believed to belong to the *Tetrapocillon* genus due to the irregular reticulating skeleton and the presence of tetrapocilli microscleres which are characteristic of this genus. This specimen however has distinct papillae, unlike *T. novaezelandiae*. Moreover, this sponge had strongyles which are not typically found in *novaezelandiae* spp. With the aforementioned description these differences may be sufficient to warrant a description of a new species. Additionally, this specimen was a more khaki colour as opposed to jet black typically found on *T. novaezelandiae*.

| Locality  |       | Styles<br>(µm)       | Strongyles<br>(µm) | Tetrapocilli<br>(µm) | Small<br>sigmas<br>(µm) | Birotule<br>(µm) |
|---|-------|----------------------|--------------------|----------------------|-------------------------|------------------|
| Spon00050,<br>Pilot Bay,<br>Bay of Plenty,<br>15 m. | x     | 170 X 4.6            | 200-9              | 44                   | 11.6                    | 62               |
|   | Range | 117-258 X<br>1.3-9.4 | 183-215 X<br>8.3-9 | 41-48                | 9.1-14.5                | 58-67            |

 Table 2.11: Spicule dimensions of Tetrapocillon sp.

### Chapter 2

# Genus Chondropsis

Definition: Found in Bergquist & Fromont, 1988, p. 42.

Chondropsis n. sp. 1 cf. topsenti

(Plate 59, A-F).

Spon00061

| As Per Bergquist:           | As Per WoRMS 2015:     |
|-----------------------------|------------------------|
| Restricted synonymy:        | Updated synonymy.      |
| (Bergquist & Fromont 1988). |                        |
| Class: Demospongiae         | Class: Demospongiae    |
| Order: Poecilosclerida      | Order: Poecilosclerida |
|                             | Suborder: Myxillina    |
| Family: Desmacidonidae      | Family: Chondropsidae  |
| Genus: Chondropsis          | Genus: Chondropsis     |
| Species: n.sp. cf. topsenti | Species: Unknown       |
|                             |                        |

Material examined: Spon00061, Bridge Marina, Tauranga Harbour, 6 m.

**Description**: The sponge is amorphous to massive, with a transparent layer of pinacoderm around the periphery of the oscules after preservation in spirit. The whole specimen is soft and compressible. Oscules appear at the apex of conulose projections in spirit.

Dimensions: Length 9 cm; width 4 cm; thickness 2 cm.

Colour: In life yellow 5.0Y5/4, in spirit yellow 5.0Y6/4.

**Texture**: Firm and compressible. No mucus present when removed from the water. Specimen is coarse, brittle and easy to tear. Oscules appear at the apex of conulose projections in spirit.

**Surface**: The surface has grooves and slight elevations, however, these forms are inconsistent due to the generally amorphous structure of the specimen.

**Skeleton**: The choanosomal skeleton is heavily infested with remnants of tube worms. There are few consistent structures within the endosome with sand grains scattered throughout the entire specimen (Plate 59, C-E). However, in some segments of the endosome there appears to be a slight reticulation of sand grains towards the surface.

## Spicules:

Fragments of many different spicule types were found. Nevertheless, these are likely foreign.

**Remarks**: This specimen had strongyles; however, they were a larger size class to spicules found in *Chondropsis topsenti* (Bergquist, 1988). It may also be a *Dysidia* sp. based on its composition of strongyles and oxeas. This specimen appeared to be borrowing fragments of spicules from other species, as there was no set type of spicules, or any fixed arrangement within the choanosomal skeleton. Further work is needed on examining the fibre construction and a better specimen is needed for this.

# FAMILY TEDANIIDAE

Definition: Found in Bergquist & Fromont, 1988, p. 57.

## <u>Genus Tedania</u>

Definition: Found in Bergquist & Fromont, 1988, p. 57.

*Tedania n.* **sp. 1** *cf. diversirhaphidiophora* (Plates 19, C-F, 20, A-F, 21, A-C). Spon00016

| As Per Bergquist:                        | As Per WoRMS 2015: |                        |  |
|--|--------------------|------------------------|--|
| Restricted synonymy: (Bergquist          | &                  | Updated synonymy.      |  |
| Fromont 1988).                           |                    |                        |  |
| Class: Demospongiae                      |                    | Class: Demospongiae    |  |
| Order: Poecilosclerida                   |                    | Order: Poecilosclerida |  |
|  |                    | Suborder: Myxillina    |  |
| Family: Tedaniidae                       |                    | Family: Tedaniidae     |  |
| Genus: Tedania                           |                    | Genus: Tedania         |  |
| Species: n. sp. cf. diversirhaphidiophot | ra                 | Species: unknown       |  |
|  |                    |                        |  |

Material examined: Spon00016, Karewa Island, Bay of Plenty, in 10.8m of water.

**Description**: The growth form of this specimen is digitate, with a heavily infested community of commensal hydroids. Finger like projections are found growing through the thick matt of hydroids.

**Dimensions**: Length 5 cm; width 3 cm; with 4 cm high fistules. *In situ* specimens over 10 cm in diameter by 3 cm thick have been observed.

Colour: In life yellow-red 5.0YR7/10, in spirit pale white.

Texture: Surface is soft and compressible.

**Surface**: The surface is covered by a transparent dermal layer after preservation in spirit, and is smooth to touch. The finger like projections are both rounded and flattened throughout the specimen after preservation in spirit (Plate 19, D).

**Skeleton**: The choanosomal skeleton appears to have an alveolate structure (Plate 20, A). Megascleres are generally confused, and arranged around choanosomal cavities.

# Spicules:

Megascleres: Styles- slightly curved (Plate 20, B). Strongyles- with spinned heads (Plate 20, C, E). Onychaetes- of two size classes. Finely spinned, asymmetric onychaetes (Plate 20, D).

For spicule dimensions see Table 2.12.

**Remarks**: This specimen appears to be dissimilar to other members of the *Tedania* genus due to its digitate growth form. This may be a function of the infestation of commensal hydroids, but other specular dissimilarities suggest a closer examination of this specimen with the type *T. diversirhaphidiophora* is warranted.

|                        |                         |                | ······             |                    |
|------------------------|-------------------------|----------------|--------------------|--------------------|
| Locality               |                         | Styles<br>(µm) | Strongyles<br>(µm) | Onychaetes<br>(µm) |
| Spon00016              |                         |                |                    |                    |
| Karewa Island,         | $\overline{\mathbf{X}}$ | 329 X 7.3      | 241 X 5.9          | 142 X 3.8          |
| Bay of Plenty, 10.8 m. |                         |                |                    |                    |
|                        | Danga                   | 320-377 X      | 176-312 X          | 100-172 X          |
|                        | капge                   | 6.5-8.5        | 4-8                | 2-6                |

Table 2.12: Spicule dimensions of *Tedania* n. sp. 1

Chapter 2

*Tedania* n. sp.2 Spon00029 (Plates 32, F, 33, A-F, 34, A).

| As Per Bergquist:                        | As Per WoRMS 2015:       |
|--|--------------------------|
| Restricted synonymy:                     | Updated synonymy.        |
| (Bergquist & Fromont 1988).              |                          |
| Class: Demospongiae                      | Class: Demospongiae      |
| Order: Poecilosclerida                   | Order: Poecilosclerida   |
|  | Suborder: Myaxillina     |
| Family: Tedaniidae                       | Family: Tedaniidae       |
| Genus: Tedania                           | Genus: Tedania (Tedania) |
| Species: n.sp. cf. diversirhaphidiophora | Species: Unknown         |
|  |                          |

Material examined: Spon00029, Salisbury Wharf, Tauranga Harbour, 5 m.

**Description**: The sponge is a brightly orange thickly encrusting specimen, which was growing in a rounded mould. There was a large amount of mucus exuded from the sponge after removal.

**Dimensions**: Length 10 cm; width 7 cm; thickness 3 cm as a section of a larger sponge.

**Colour**: In life red-yellow-red 10.0R6/10, in spirit red-yellow-red 10.0R6/10 to yellow-red-yellow 10.0YR8/4.

**Texture**: Soft and compressible, with a gelatinous like choanosome. A large amount of mucus was exuded from the specimen after collection and the specimen remained slimy after preservation in spirit.

**Surface**: The surface is smooth and rounded with small macroscopically visible oscules present on the dermis (Plate 33, B).

**Skeleton**: The choanosomal skeleton consists of plumose tracts of spicules which meander towards the surface, only occasionally intersecting (Plate 33, C-E).

### Spicules:

Megascleres: Tylostyles- slightly bent with very fine spines (Plate 29, A). Strongyles- of normal form. Oxeas- thick and slender forms. There were some acanthostyles present within the choanosomal skeleton but these were found in low abundances and were consequently considered foreign (Plate 33, F).

Microscleres: Onychaetes- spinned and thin.

For spicule dimensions see Table 2.13.

**Remarks**: What appeared to be raphides were in fact onychaetes with spinned styles. This specimen was dissimilar to *Tedania* n.sp.1 in that it did not have a digitate growth form and had no oxeas present within its skeletal architecture.

| Locality  |       | Tylostyles<br>(µm)   | Strongyles<br>(µm)   | Oxeas<br>(µm)      | Onychaetes<br>(µm) |
|---|-------|----------------------|----------------------|--------------------|--------------------|
| Spon00029,<br>Salisbury Wharf,<br>Bay of Plenty,<br>5 m | x     | 251 X 7.9            | 202 X 4.9            | 135 X 7.2          | 128 X 2.8          |
|   | Range | 157-288 X<br>6.6-9.3 | 175-270 X<br>4.5-5.4 | 116-175 X<br>5-8.9 | 121-334 X<br>2-3.4 |

 Table 2.13: Spicule dimensions of *Tedania* sp.

Chapter 2

*Tedania battershilli* (Bergquist & Fromont 1988) (Plates 22, D-F, 23, A-C). Spon00018

| AS PER BERGQUIST:           | As Per WoRMS 2015:                      |
|-----------------------------|---|
| Restricted synonymy:        | Updated synonymy.                       |
| (Bergquist & Fromont 1988). |   |
| Class: Demospongiae         | Class: Demospongiae                     |
| Order: Poecilosclerida      | Order: Poecilosclerida                  |
|                             | Suborder: Myxillina                     |
| Family: Tedaniidae          | Family: Tedaniidae                      |
| Genus: Tedania              | Genus: Tedania                          |
|                             | Subgenus: Tedania (Tedania)             |
| Species: battershilli       | Species: Tedania (Tedania) battershilli |
|                             |   |

Material examined: Spon00018, Karewa Island, Bay of Plenty, 10.8 m.

**Description**: The sponge is thickly encrusting specimen with filamentous red algae encrusting on its surface. The specimen was found encrusting subtidally on rock at Karewa Island. There was a moderate level of mucus discharged from the specimen upon removal from water.

**Dimensions**: Length 6 cm; width 2 cm; thickness 1 cm. Specimens above 15 cm in diameter were observed *in situ*.

Colour: In life red yellow-red 10.R6/10, in spirit pale white to yellow-red yellow 10.0YR8/4.

Texture: Firm, and slimy when compressed.

**Surface**: Smooth and slimy to touch. Oscules 1< mm are visible on the flattened dermal layer. The surface is microscopically hispid.

**Skeleton**: The skeleton is plumoreticulate with tracts of styles reticulating to the dermis (Plate 23, A). The ectosomal skeleton was congruent with *Tedania battershilli* with tangential aligned megascleres placed at right angles to the surface.

# **Spicules:**

Megascleres: Styles- slightly curved to straightened styles. Tylotes - smooth, with rounded ends.

Microscleres: two size classes of onychaets.

For spicule dimensions see Table 2.13.

**Remarks**: *Tedania battershilli* has a distinctive bright orange colour, compact texture and typically produces mucus after collection which was congruent with this specimen.

| Locality                     | Style<br>(µm            |           | Tylotes<br>(µm) | Large<br>onychaetes<br>(µm) | Small<br>onychaetes<br>(µm) |  |
|------------------------------|-------------------------|-----------|-----------------|-----------------------------|-----------------------------|--|
| Average Tedania battershilli |                         |           |                 |                             |                             |  |
| spicule dimensions from      |                         |           |                 |                             |                             |  |
| specimens collected in New   | $\overline{\mathbf{X}}$ | 261 X 5   | 265 X 4.5       | 134                         | 51                          |  |
| Zealand (Bergquist &         |                         |           |                 |                             |                             |  |
| Fromont, 1988).              |                         |           |                 |                             |                             |  |
|                              | Range                   | 231-296 X | 225-288 X       | 119-150                     | 42-63                       |  |
|                              | Kange                   |           | 3.4-4.9         | 117 150                     | 42 05                       |  |
| Spon00018,                   |                         |           |                 |                             |                             |  |
| Karewa Island,               | $\overline{\mathbf{X}}$ | 148 X 3.4 | 263 X 6.5       | 118 X 4.2                   | 42 X 1.5                    |  |
| Bay of Plenty, 10.8 m.       |                         |           |                 |                             |                             |  |
|                              | Dongo                   | 120-229 X | 246-290 X       | 110-132 X                   | 36-48 X                     |  |
|                              | Kallge                  | 2.1-6.2   | 4.7-7.8         | 3.4-5.7                     | 1.2-1.9                     |  |

#### Table 2.14: Spicule dimensions of *Tedania battershilli*.

Chapter 2

*Tedania n.* sp. 3 cf. battershilli Spon00048

(Plates 46, D-F, 47, A-D).

| As Per Bergquist:           | As Per WoRMS 2015:     |
|-----------------------------|------------------------|
| Restricted synonymy:        | Updated synonymy.      |
| (Bergquist & Fromont 1988). |                        |
| Class: Demospongiae         | Class: Demospongiae    |
| Order: Poecilosclerida      | Order: Poecilosclerida |
|                             | Suborder: Myxillina    |
| Family: Tedaniidae          | Family: Tedaniidae     |
| Genus: Tedania              | Genus: Tedania         |
| Species: cf. battershilli   | Species: Unknown       |
|                             |                        |

Material examined: Spon00048, Pilot Bay, Tauranga Harbour, 15 m.

**Description**: This specimen is amorphous to massive, with conulose projections. It was found covered in fine sediment with oscules projecting through the layer of mud.

**Dimensions**: Length 5.5 cm; width 4 cm; thickness 2.4 cm.

Colour: In life yellow-red 5.0YR7/10, in spirit yellow 5.0Y8/2.

**Texture**: A firm sponge, which exuded a large amount of mucus upon collection. **Surface**: The surface of the sponge has large conulose structures over the dermis *in situ* (Plate 46, D). Oscules became retracted after removal from water and preservation in spirit to create a smooth surface (Plate 46, E, F). The surface was slimy and slippery in spirit.

**Skeleton**: The skeleton is difficult to distinguish due to the degraded tissue sample in the thick section. However, it appeared to have a plumose structure, with an abundance of larva which were scattered throughout the choanosomal skeleton (Plate 47, A, B).

# Spicules:

Megascleres: Tyles- some slightly spiny. Strongyles- normal form.

Microscleres: Onychaetes which are slightly spiny.

For spicule dimensions see Table 2.15.

**Remarks**: The conulose structure of this specimen was unlike any other *Tedania battershilli* specimens previously described.

| Locality             |                         | Styles<br>(µm) | Strongyles<br>(µm) | Onychaetes<br>(µm) | Raphides<br>(µm) |
|----------------------|-------------------------|----------------|--------------------|--------------------|------------------|
| Spon00048,           |                         |                |                    |                    |                  |
| Pilot Bay,           | $\overline{\mathbf{x}}$ | 267 X 4.9      | 235 X 5.6          | 132 X 1.8          | 126 X 1.8        |
| Bay of Plenty, 15 m. |                         |                |                    |                    |                  |
|                      | Range                   | 244-290 X      | 179-283 X          | 118-141 X          | 121-130 X        |
|                      |                         | 3.8-6.5        | 2.9-4.1            | 1.3-2.3            | 1.6-2            |

 Table 2.15: Spicule dimensions of Tedania sp.

# FAMILY COELOSPHAERIDA

Definition: Found in Bergquist & Fromont, 1988, p. 47.

## Genus Forcepia

Definition: Found in Hooper & Van Soest 2002, p. 523.

| Coelosphaeridae sp. cf. Forcepia sp. | (Plates 30, D-F, 31, A-D). |
|--------------------------------------|----------------------------|
| Spon00027                            |                            |
| As Per Hentschel                     | As Per WoRMS: 2015         |
| Restricted synonymy:                 | Updated synonymy.          |
| (Hentschel, 1911).                   |                            |
| Class: Demospongiae                  | Class: Demospongiae        |
| Order: Poecilosclerida               | Order: Poecilosclerida     |
|                                      | Suborder: Myxillina        |
| Family: Coelosphaeridae              | Family: Coelosphaeridae    |
| Genus: cf. Forcepia                  | Genus: Unknown             |
| Species: n.sp.                       | Species: Unknown           |

Material examined: Spon00027, Salisbury Wharf, Tauranga Harbour, 5 m.

**Description**: The sponge was found thickly encrusting commensal on an abandoned speckled whelk (*Cominella adspersa*) on a sand flat.

**Dimensions**: Length 5.5 cm; width 5 cm; thickness 4.5 cm. This specimen's growth may have been limited by the available surface area on the spotted whelk (*C. adspersa*) it was commensal on.

Colour: In life yellow-red 5.0YR7/10, in spirit yellow 5.0Y8/4.

**Texture**: The sponge has a grainy texture and is difficult to tear.

**Surface**: The sponge is rugose, having a rough and ridged granular surface. The specimen's surface area appears to have been limited to the surface area of the shell in which it was commensal on (Plate 30, E, F).

**Skeleton**: The choanosomal skeleton was plumoreticulate, with two size classes of styles within the spicular tracts (Plate 31, A, B). The ectosomal skeleton was composed of spicule brushes composed of acanthostyles.

# **Spicules:**

Megascleres: Acanthostyles- were straight and spiny with no verticcillate spining and fusiform points (Plate 31, C). Styles- had smooth styles, rounded spiny bases, fusiform points and were straight. Oxeas- two size classes, the smaller size class had bent oxeas, and the larger class had straight oxeas. Subtylostyles- were large, slightly curved with spiny heads. Tylostyles- one size class, straight with spherical heads.

For spicule dimensions see Table 2.16.

**Remarks**: This sponge had a plumoreticulate choanosomal skeleton with tylostyles which is similar to that found in the *Forcepia* genus described in Hentschel (1911).

| Locality         |          | Acanthostyles<br>(µm) | Styles<br>(µm) | Large oxeas<br>(µm) | Small oxeas<br>(µm) | Subtylostyles<br>(µm) | Tylostyles<br>(µm) |
|------------------|----------|-----------------------|----------------|---------------------|---------------------|-----------------------|--------------------|
| Spon00027,       |          |                       |                |                     |                     |                       |                    |
| Salisbury Wharf, | v        | 84 2 X 10 2           | 215 X 5 0      | 383 X 10 2          | 152 X 8 5           | 232 X 12 1            | 138 X 5 8          |
| Bay of Plenty,   | Λ        | 04.2 A 10.2           | 215 A J.9      | 363 A 10.2          | 152 A 0.5           | 232 A 12.1            | 136 A 3.6          |
| 5 m              |          |                       |                |                     |                     |                       |                    |
|                  | Danas    | 76-93 X               | 181-271 X      | 371-391 X           | 141-170 X           | 209-265 X             | 126-149 X          |
| Kange            | 7.1-15.3 | 3.4-10.7              | 7.9-12.7       | 7.3-9.6             | 10.1-13.2           | 4.7-6.6               |                    |

Table 2.16: Spicule dimensions of *Coelosphaeridae* sp.
# FAMILY MYCALIDAE

Definition: Found in Bergquist & Fromont, 1988, p. 17.

## <u>Genus Carmia</u>

Definition: Found in Bergquist & Fromont, 1988, p. 21.

**Note:** There were 12 *Carmia* spp. collected within this study; however, these species were morphologically similar and had few skeletal characteristics which could be used to differentiate species (Bergquist & Fromont, 1988). The choanosomal skeletons within *Carmia macilenta*, *C. tasmani* and *C. hentscheli* all have plumose branching tracts (Bergquist & Fromont, 1988). As a result, spicule types and sizes were used as the main distinguishing features between *Carmia* spp. within this study. Further work is planned to examine the chemotaxonomy of this genus given that it is commonly associated with strongly bioactive compounds (see Discussion Chapter).

# Carmia n.sp.1 cf. tasmani (Plates 55, A-F, 56, A-B). Spon00057

**Note:** All of the specimens described within 'Group one' are likely the same species, and appear dissimilar to other *Carmia* spp. described in Bergquist & Fromont (1988). Accordingly, they were grouped together to allow the reader to compare between specimens. Further work is required with comparison to type specimens (unavailable for this study). Chemotaxonomic work is also planned as a future research component.

| As Per Bergquist:         |            |   | As PER WORMS 2015:        |
|---------------------------|------------|---|---------------------------|
| Restricted synonymy:      | (Bergquist | & | Updated synonymy.         |
| Fromont 1988).            |            |   |                           |
| Class: Demospongiae       |            |   | Class: Demospongiae       |
| Order: Poecilosclerida    |            |   | Order: Poecilosclerida    |
|                           |            |   | Suborder: Mycalina        |
| Family: Mycalidae         |            |   | Family: Mycalidae         |
| Genus: Carmia             |            |   | Genus: Mycale             |
|                           |            |   | Subgenus: Mycale (Carmia) |
| Species: n.sp. cf. tasman | i          |   | Species: Unknown          |
|                           |            |   |                           |

Material examined: Spon00057, Bridge Marina, Tauranga Harbour, 6 m.

**Description**: This sponge is an amorphous to massive specimen and is heavily encrusted with a commensal spaghetti bryozoan (*Zoobotryon verticillatum*). The pinacoderm was covered by dark areas larvae which appear to have migrated there to be released as free swimming larvae.

**Dimensions**: Length 9.5 cm; width 6 cm; thickness 4 cm of a section of a larger sponge.

Colour: In life red yellow-red 10.0R2/2, in spirit yellow 5.0Y8/4.

**Texture**: Soft, compressible and pulls apart easily.

**Surface**: Specimen had a smooth but uneven surface and a transparent dermal membrane, through which pores are visible.

**Skeleton**: The sponge had poorly developed spicular tracts, additionally obscured by polychaete tubes creating meandering bundles throughout the entire choanosomal skeleton (Plate 56, C-E). Sigmas were visible interstitially throughout the choanosome.

#### Spicules:

Megascleres: Subtylostyles- swelling is slightly visible. Palmate anisochelae with three size classes.

Microscleres: Sigmas- large C and S shaped sigmas. Toxas- large range in size classes.

For spicule dimensions see Table 2.17.

**Remarks**: The surface has visible oscula and a transparent dermal membrane. It should be noted that this specimen has an abundance of larvae throughout its entire body. This specimen was collected on the 9<sup>th</sup> of September 2014, which may be when it begins to reproduce or release free swimming larvae. There is one large size class of sigmas that are of similar sizes to *C. tasmani* (Bergquist & Fromont, 1988). Furthermore, this specimen had large, medium and small size classes of palmate anisochelae of similar sizes to *C. tasmani*. Interestingly, this specimen had small dark patches on its surface and upon close inspection these patches were confirmed to be larvae, which may have been ready for release. This specimen was similar to the descriptions of *C. tasmani*, which warranted a separation from other species which are potentially new species described in this study. However, more work is needed with the type specimen of *C. tasmani* to confirm this.

| Locality            |                         | Subtylostyles<br>(µm) | Large palmate<br>anisochelae<br>(µm) | Medium palmate<br>anisochelae<br>(µm) | Small palmate<br>Anisochelae<br>(µm) | Sigmas<br>(µm) | Toxas<br>(µm) |
|---------------------|-------------------------|-----------------------|--------------------------------------|---------------------------------------|--------------------------------------|----------------|---------------|
| Spon00057,          |                         |                       |                                      |                                       |                                      |                |               |
| Bridge Marina,      | $\overline{\mathbf{X}}$ | 225 X 5.6             | 42                                   | 25                                    | 13.8                                 | 99             | 61            |
| Bay of Plenty, 6 m. |                         |                       |                                      |                                       |                                      |                |               |
|                     | Range                   | 220-229 X<br>5-6.2    | 38-45                                | 20-30                                 | 12.1-16.7                            | 91-102         | 37-78         |

Table 2.17: Spicule dimensions of *Carmia* n sp.1

Carmia n.sp.2 cf. tasmani Spon00062 (Plate 60, A-F).

| As Per Bergquist:           | As Per WoRMS 2015:        |
|-----------------------------|---------------------------|
| Restricted synonymy:        | Updated synonymy.         |
| (Bergquist & Fromont 1988). |                           |
| Class: Demospongiae         | Class: Demospongiae       |
| Order: Poecilosclerida      | Order: Poecilosclerida    |
|                             | Suborder: Mycalina        |
| Family: Mycalidae           | Family: Mycalidae         |
| Genus: Carmia               | Genus: Mycale             |
| Species: n.sp. cf. tasmani  | Subgenus: Mycale (Carmia) |
|                             | Species: Unknown          |
|                             |                           |

Material examined: Spon00062, Bridge Marina, Tauranga Harbour, 6 m.

**Description**: The sponge has a smooth surface and was found thickly encrusting beneath a marina competing for space with other species of sponges. The sponge was somewhat triangular to amorphous shaped *in situ*. There is a visible surface infestation of polychaete worms.

Dimensions: Length 7.5 cm; width 5.5 cm; thickness 1.5 cm.

Colour: In life yellow-red-yellow 10.YR4/4 to yellow-red-yellow 10.0YR7/8, in spirit yellow 5.0Y8/6.

Texture: This specimen had a grainy texture.

**Surface**: The surface was smooth with no surface projections and a transparent dermal membrane. There are dark sand filled pores scattered over the dermis as a result of a polychaete worm infestation (Plate 60, A, B).

**Skeleton**: The sponge has a plumoreticulate skeleton, with a confused mass of interstitial megascleres (Plate 60, C, D). The skeletal arrangement appeared to have been disrupted by polychaete tubes, which made it difficult to distinguish spicule tracts.

# **Spicules:**

Megascleres: Sutylostyles- characteristic size ranges in comparison to other *Carmia* spp. C and S shaped sigmas of one size class.

Microscleres: Palmate anisochelae- three size classes.

For spicule dimensions see Table 2.18.

**Remarks**: This species was similar to *C. tasmani* in that it had one large size class of sigmas similar to that found in *C. tasmani*. Furthermore, this specimen had large, medium and small size classes of palmate anisochelae which is congruent with *C. tasmani*. There were sand grains incorporated into the dermal membrane, which was similar to specimens of *C. tasmani* described in Bergquist & Fromont (1988). The surface has visible oscula and a transparent dermal membrane. It should be noted that the surface of this specimen is smooth without any irregular projections. This specimen was of such divergence from the other spices which were similar to *C. tasmani* described in this study that that it warranted a separate description. Further work is needed to determine whether it is in fact *Carmia tasmani*.

| Locality            |                         | Subtylostyles<br>(µm) | Large palmate<br>anisochelae<br>(µm) | Medium palmate<br>anisochelae<br>(µm) | Small palmate<br>Anisochelae<br>(µm) | Large<br>sigmas<br>(µm) | Toxas<br>(µm) |
|---------------------|-------------------------|-----------------------|--------------------------------------|---------------------------------------|--------------------------------------|-------------------------|---------------|
| Spon00062,          |                         |                       |                                      |                                       |                                      |                         |               |
| Bridge Marina,      | $\overline{\mathbf{x}}$ | 221 X 4.7             | 40                                   | 25                                    | 13.4                                 | 91                      | 185           |
| Bay of Plenty, 6 m. |                         |                       |                                      |                                       |                                      |                         |               |
|                     | Range                   | 228-214 X<br>4.7-6.2  | 39-41                                | 22-29                                 | 12.4-15.1                            | 83-95                   | 78-310        |

 Table 2.18: Spicule dimensions of Carmia n sp.2

*Carmia* **n.sp.3** *cf. tasmani* (Plates 51, F, 52, A-E). Spon00054

| As Per Bergquist:           | As PER WORMS 2015:        |
|-----------------------------|---------------------------|
| Restricted synonymy:        | Updated synonymy.         |
| (Bergquist & Fromont 1988). |                           |
| Class: Demospongiae         | Class: Demospongiae       |
| Order: Poecilosclerida      | Order: Poecilosclerida    |
|                             | Suborder: Mycalina        |
| Family: Mycalidae           | Family: Mycalidae         |
| Genus: Carmia               | Genus: Mycale             |
|                             | Subgenus: Mycale (Carmia) |
| Species: n.sp. cf. tasmani  | Species: Unknown          |

Material examined: Spon00054, Bridge Marina, Tauranga Harbour, 6 m.

**Description**: The specimen is soft and amorphous to massive found encrusting underneath a marina in a species rich sponge community competing for space.

**Dimensions**: Length 9 cm; width 4 cm; thickness 3.5 cm as a section of a larger sponge. The specimen was approximately 15 cm in length *in situ*.

Colour: In life yellow-red-yellow 10.0YR5/4, in spirit yellow 5.0Y8/4.

Texture: Soft, easily compressed and easy to tear.

**Surface**: The surface is smooth but uneven with oscules 1-3 mm in diameter (Plate 52, A).

**Skeleton**: The choanosomal skeleton is plumoreticulate. The spicules form dermal brushes at the surface (Plate 52, A).

#### **Spicules:**

Megascleres: Subtylostyles- slender, with rounded subtylote heads and a small compression beneath the head.

Microscleres: Palmate anisochelae- of two size classes and normal form (no small size class present). Large S and C shaped sigmas. Toxas- fine and long with a wide central flexure.

For spicule dimensions see Table 2.19.

**Remarks**: Spicule dimensions of this specimen are dissimilar to other *Carmia* spp. described in Bergquist & Warne (1980), as this specimen has a single small

size class of anisochelae. This specimen is more distinctively different from *C*. *tasmani* than the others above.

| Locality         |       | Subtylostyles<br>(µm) | Large<br>palmate<br>anisochelae<br>(µm) | Medium<br>palmate<br>anisochelae<br>(µm) | Sigmas<br>(µm) | Toxas<br>(µm) |
|------------------|-------|-----------------------|---|--|----------------|---------------|
| Spon00054        |       |                       |   |  |                |               |
| Bridge Marina,   | v     | 220 X 6 8             | 42                                      | 27                                       | 100            | 257           |
| Bay of Plenty, 6 | Λ     | 239 A 0.8             | 42                                      | 21                                       | 100            | 231           |
| m.               |       |                       |   |  |                |               |
|                  | Range | 233-247 X<br>5.8-7.6  | 39-46                                   | 24-30                                    | 95-106         | 116-327       |

Table 2.19: Spicule dimensions of *Carmia* n sp.3

*Carmia* **n.sp.1** (1) (Group one) Spon00055

(Plates 52, F, 53, A-E).

| As Per Bergquist:           | As Per WoRMS 2015:        |
|-----------------------------|---------------------------|
| Restricted synonymy:        | Updated synonymy.         |
| (Bergquist & Fromont 1988). |                           |
| Class: Demospongiae         | Class: Demospongiae       |
| Order: Poecilosclerida      | Order: Poecilosclerida    |
|                             | Suborder: Mycalina        |
| Family: Mycalidae           | Family: Mycalidae         |
| Genus: Carmia               | Genus: Mycale             |
|                             | Subgenus: Mycale (Carmia) |
| Species: n.sp.1             | Species: Unknown          |
|                             |                           |

Material examined: Spon00055, Bridge Marina, Tauranga Harbour, 6 m.

**Description**: The specimen is soft and amorphous to massive found encrusting underneath a marina in a species rich sponge community. Hydroids were commensal on the surface of this specimen, which appears to be a common occurrence within this genus.

Dimensions: Length 7 cm; width 5 cm; thickness 2 cm.

Colour: In life yellow 5.0Y6/4, in spirit yellow 5.0Y8/4.

Texture: Soft and compressible and pulls apart easily.

**Surface**: The sponge had a smooth but irregular surface with some small parallel ridges and grooves (Plate 53, A).

**Skeleton**: The choanosomal skeleton is plumoreticulate (Plate 53, B, C). The spicules form dermal brushes at the surface. There are plumose spicule tracts with single spicules position haphazardly between the tracts. The ectosomal skeleton is not visible on the thick section sampled.

#### Spicules:

Megascleres: Subtylostyles- short and slender, with elongated tapering points.

Microscleres: Palmate anisochelae- identical in form, with two size classes (no small size class present). C and S shaped sigmas. Toxas- rare with central flexures.

For spicule dimensions see Table 2.20.

**Remarks**: The spicule dimensions of this specimen are dissimilar to other *Carmia* spp. described in Bergquist & Warne (1980), as there is a smaller size class of anisochelae. There was an abundance of larvae present within the endosome of this specimen.

| Locality            | ocality (µm)            |                      | Large palmate<br>anisochelae<br>(µm) | Medium palmate<br>anisochelae<br>(µm) | Sigmas<br>(µm) | Toxas<br>(µm) |
|---------------------|-------------------------|----------------------|--------------------------------------|---------------------------------------|----------------|---------------|
| Spon00055,          |                         |                      |                                      |                                       |                |               |
| Bridge Marina,      | $\overline{\mathbf{X}}$ | 235 X 6.8            | 43                                   | 25                                    | 100            | 112           |
| Bay of Plenty, 6 m. |                         |                      |                                      |                                       |                |               |
|                     | Range                   | 206-290 X<br>5.1-8.2 | 40-46                                | 15-28                                 | 93-109         | 56-155        |

 Table 2.20: Spicule dimensions of Carmia n sp. 1 (1)

*Carmia* **n.sp.1** (2) (Group one) Spon00077 (Plates 74, F, 75, A-F).

| As Per WoRMS 2015:        |
|---------------------------|
| Updated synonymy.         |
|                           |
| Class: Demospongiae       |
| Order: Poecilosclerida    |
| Suborder: Mycalina        |
| Family: Mycalidae         |
| Genus: Mycale             |
| Subgenus: Mycale (Carmia) |
| Species: Unknown          |
|                           |

Material examined: Spon00077, Bridge Marina, Tauranga Harbour, 6 m.

**Description**: The sponge was massive to amorphous and found encrusting beneath a marina. There was a colony of hydroids encrusting on the surface of the specimen, in addition to dark lines caused by an infestation of commensal polychaete worms. The specimen appeared to be regressed which may be an indication that it was at the end of its life cycle.

**Dimensions**: Length 6 cm; width 3 cm; thickness 2.5 cm as a section of a larger sponge.

**Colour:** In life yellow 5.0Y6/6, in spirit yellow 5.0Y8/4.

Texture: Soft, compressible and easy to tear.

**Surface**: The surface was uneven with regressed pores and grooves which appeared as a loss of pinacoderm tissue. *In situ* the sponge had wide open oscules which were both flush with the surface and slightly elevated. There were epibiont hydroids on this specimen (Plate 75, F).

**Skeleton**: The choanosomal skeleton was damaged due to a heavy infestation of polychaete worms; therefore it was unable to be described adequately (Plate 75, C, D). Moreover, there was an abundance of larvae buds scattered throughout the skeletal structure.

#### Spicules:

Megascleres: Subtylostyles- of normal form.

Microscleres: Palmate anisochelae of two size classes (smaller size class absent). C and S shaped sigmas. Toxas- slender and curved. There were rhabdostyles found in low abundances and were consequently considered foreign (Plate 75, E). For spicule dimensions see Table 2.21.

**Remarks**: Spicule dimensions are dissimilar to other *Carmia* spp. described in Bergquist & Warne (1980), as this specimen has a small size class of anisochelae. There was an abundance of polychaete worms within the choanosomal cavities which may provide information regarding the life cycle of this species.

| Locality   |                         | Subtylostyles<br>(µm) | Large palmate<br>anisochelae<br>(µm) | Medium palmate<br>anisochelae<br>(µm) | Sigmas<br>(µm) | Toxas<br>(µm)    |
|--|-------------------------|-----------------------|--------------------------------------|---------------------------------------|----------------|------------------|
| Spon00077, Bridge Marina,<br>Bay of Plenty, 6 m. | $\overline{\mathbf{x}}$ | 219 X 6.8             | 44                                   | 25                                    | 106            | 82 X 2.5         |
|  | Range                   | 194-237 X<br>5.7-8.6  | 47-44                                | 21-29                                 | 102-111        | 73-92 X<br>2-3.6 |

 Table 2.21: Spicule dimensions of Carmia n sp. 1 (2)

# Carmia n.sp.2 (1) (Group two)

(Plates 53, F, 54, A-F).

#### Spon00056

**Note:** The specimen described in 'Group two' appears dissimilar to other *Carmia* spp. described in Bergquist & Fromont (1988) and throughout this study, consequently it was classified as a separate group.

| As Per Bergquist:           | As PER WORMS 2015:     |
|-----------------------------|------------------------|
| Restricted synonymy:        | Updated synonymy.      |
| (Bergquist & Fromont 1988). |                        |
| Class: Demospongiae         | Class: Demospongiae    |
| Order: Poecilosclerida      | Order: Poecilosclerida |
|                             | Suborder: Mycalidae    |
| Family: Mycalidae           | Family: Mycalidae      |
| Genus: Carmia               | Genus: Mycale (Carmia) |
| Species: n.sp.2             | Species: Unknown       |

Material examined: Spon00056, Bridge Marina, Tauranga Harbour, 6 m.

**Description**: The sponge is an amorphous to massive specimen that is soft to touch. This specimen was found encrusting underneath a marina in a species rich sponge community. The growth form of this specimen was larger than all other *Carmia* species described in this study at 140 cm *in situ*.

Dimensions: Length 13 cm; width 11 cm; with 6 cm high ridges.

Colour: In life red yellow-red 10.0R2/2, in spirit yellow 5.0Y8/4.

Texture: Soft and compressible, and easy to tear.

**Surface**: The surface is smooth, with furrowed meandering channels, with hydroids encrusting mainly on the apex of the ridges (Plate 54, A). There are distinct ridges and grooves that are prominent over the entire surface.

**Skeleton**: The choanosomal skeleton is plumoreticulate, however, this was difficult to recognize due to the commensal tube worms obstructing the architecture of the skeleton (Plate 54, B-C).

# **Spicules:**

Megascleres: Subtylostyles- typical sizes ranges as per other *Carmia* spp. (Plate 54, E).

Microscleres: Anisochelae- two size classes only. Sigmas- two sizes classes, S and C shaped. Toxas- thin with a central flexure.

For spicule dimensions see Table 2.22.

**Remarks**: This specimen had larger sigmas compared to *C. macilenta, C. tasmani* and *C. hentscheli*. There was a larger range of toxas within this specimen. This specimen had clearly defined ridges on its surface that appear dissimilar to any other *Carmia* spp. (Bergquist & Fromont, 1988). The note of interest is the larger size class of sigmas, which are uncharacteristic of any other species of *Carmia* described in New Zealand (Bergquist & Fromont 1988). The larger growth form of this specimen may be an ecophenotype in response to low wave exposure. There was an abundance of larvae present within the endosome of this sponge.

| Locality            |                         | Subtylostyles<br>(µm) | Large palmate<br>anisochelae<br>(µm) | Medium palmate<br>anisochelae<br>(µm) | Large sigmas<br>(µm) | Small<br>sigmas<br>(µm) | Toxas<br>(µm) |
|---------------------|-------------------------|-----------------------|--------------------------------------|---------------------------------------|----------------------|-------------------------|---------------|
| Spon00056,          |                         |                       |                                      |                                       |                      |                         |               |
| Bridge Marina,      | $\overline{\mathbf{X}}$ | 205 X 5.7             | 43                                   | 27                                    | 98                   | 29                      | 159           |
| Bay of Plenty, 6 m. |                         |                       |                                      |                                       |                      |                         |               |
|                     |                         | 189-222 X             | 40-44                                | 21.22                                 | 01.101               | 22.42                   |               |
|                     | Range                   | 4.7-6.5               |                                      | 21-33                                 | 81-101               | 22-42                   | 43-261        |

Table 2.22: Spicule dimensions of *Carmia* n. sp. 2 (1)

*Carmia* **n.sp.3** (1) (Group three).

(Plate 80, A-F).

Spon00073

**Note:** All of the specimens described within 'Group three' are likely the same species, and appear dissimilar to other *Carmia* spp. described in Bergquist & Fromont (1988). Accordingly, they were grouped together to allow the reader to compare between specimens.

| As Per Bergquist:           | As Per WoRMS 2015:        |
|-----------------------------|---------------------------|
| Restricted synonymy:        | Updated synonymy.         |
| (Bergquist & Fromont 1988). |                           |
| Class: Demospongiae         | Class: Demospongiae       |
| Order: Poecilosclerida      | Order: Poecilosclerida    |
|                             | Suborder: Mycalina        |
| Family: Mycalidae           | Family: Mycalidae         |
| Genus: Carmia               | Genus: Mycale             |
|                             | Subgenus: Mycale (Carmia) |
| Species: n.sp.              | Species: Unknown          |

Material examined: Spon00073, Bridge Marina, Tauranga Harbour, 6 m.

**Description**: The specimen was soft and amorphous and was found encrusting on a broken shell. There was a heavy abundance of larvae present in the endosome, with a visible network of tubes constructed by polychaete worms. The sponge had large broad open oscules which were flush with the dermis *in situ*.

**Dimensions**: Length 4.5 cm; width 5 cm; height 4 cm as a section of a larger sponge. Massive sponges above 6 cm have been observed at this site.

Colour: In life yellow 5.0Y7/4, in spirit yellow 5.0Y8/4.

Texture: The living specimen was soft, spongy and easy to tear.

**Surface**: The surface is smooth but irregular, oscular pores 1-2 mm in diameter are visible in material that has been preserved. The oscules were widely opened *in situ* (Plate 80, A). However, they became retracted after preservation in spirit (Plate 80, B).

**Skeleton**: The skeleton is plumose with columns of aligned megascleres (Plate 80, C, D). The ectosomal skeleton is composed of subtylostyles which are situated parallel to the pinacoderm.

#### Spicules:

Megascleres: Subtylostyles- sizes are characteristic of other *Carmia* spp. (Plate 80, E). Microscleres: One size class of large anisochelae only. Sigmas- C-shape and S-shape of a single size class; C shaped sigmas were abundant and S shaped sigmas were scarce within the spicule slide. There was allow abundance of birutules within the sample, however these were considered foreign (Plate 80, F). For spicule dimensions see Table 2.23.

**Remarks**: This specimen only has one size class of large palmate anisochelae, which is different to other *Carmia* species described in Bergquist & Fromont (1988). The skeleton is plumose with columns of aligned megascleres. There is an abundance of larvae present in the endosome of this specimen.

| Locality   |       | Subtylostyles<br>(µm) | Palmate<br>anisochelae<br>(µm) | Sigmas<br>(µm) |
|--|-------|-----------------------|--------------------------------|----------------|
| Spon00073, Bridge Marina,<br>Bay of Plenty, 6 m. | x     | 220 X 7.2             | 39 X 13.8                      | 57             |
|  | Range | 212-228 X<br>5.9-8.8  | 36-41 X<br>11.2-15.8           | 47-67          |

Table 2.23: Spicule dimensions of Carmia n. sp. 3 (1)

*Carmia* **n.sp.3** (2) (Group three) Spon00074

(Plates 72, A-F, 73, A).

| <u>As Per Bergquist</u> :   | As Per WoRMS 2015:        |
|-----------------------------|---------------------------|
| Restricted synonymy:        | Updated synonymy.         |
| (Bergquist & Fromont 1988). |                           |
| Class: Demospongiae         | Class: Demospongiae       |
| Order: Poecilosclerida      | Order: Poecilosclerida    |
|                             | Suborder: Mycalina        |
| Family: Mycalidae           | Family: Mycalidae         |
| Genus: Carmia               | Genus: Mycale             |
|                             | Subgenus: Mycale (Carmia) |
| Species: n.sp.              | Species: Unknown          |
|                             |                           |

Material examined: Spon00074, Bridge Marina, Tauranga Harbour, 6 m.

**Description**: The sponge is amorphous to massive, found encrusting beneath a marine in a highly populated sponge community in approximately one meter of water. There were hydroids commensal on the surface of this specimen. This specimen was degraded *in situ*.

**Dimensions**: Length 8 cm; width 7 cm; thickness 4 cm as a section of a larger sponge.

Colour: In life yellow 5.0Y6/4, in spirit yellow 5.0Y8/4.

**Texture**: Soft and spongy in spirit. The sponge was compressible and easy to tear. **Surface**: The surface appeared to be retracted in life, with holes throughout the dermis and pieces of pincoderm tissue missing. Overall, the surface was granular with irregular projections (Plate 72, A, B).

**Skeleton**: The skeleton is a plumose structure, with columns of aligned spicules (Plate 72 C, D). Sigmas are visibly scattered throughout the interstitial spaces in the choanosomal skeleton. The ectosomal skeleton is composed of subtylostyles arranged parallel to the dermis.

# **Spicules:**

Megascleres: Subtylostyles- slender, some are slightly bent (Plate 72, E).

Microscleres: Anisochelae- one large size class. Toxas of a small size class in comparison to other *Carmia* spp.

For spicule dimensions see Table 2.24.

**Remarks**: This specimen has a single size class of large palmate anisochelae, which is different to other *Carmia* species described in Bergquist & Fromont, 1988. The skeleton is plumose with columns of aligned megascleres. Interestingly, although the specimen appeared to be regressed and dying, there was an abundance of larvae scattered sub-dermally and throughout the choanosomal skeleton. This may be an indicator that this particular species has a seasonal life cycle.

| Looglitz   |       | Subtylostyles        | Anisochelae | Sigmas            | Toxas              | Raphide              |
|--|-------|----------------------|-------------|-------------------|--------------------|----------------------|
| Locanty  |       | (µm)                 | (µm)        | (µm)              | (µm)               | (µm)                 |
| Spon00074, Bridge Marina,<br>Bay of Plenty, 6 m. | x     | 226 X 6.5            | 37          | 37 X 109          | 82 X 2.1           | 256 X 2.6            |
|  | Range | 204-245 X<br>5.7-7.9 | 27-52       | 27-52 X<br>97-121 | 69-88 X<br>1.9-2.3 | 245-273 X<br>2.3-3.3 |

 Table 2.24: Spicule dimensions of Carmia n. sp. 3 (2)

*Carmia* **n.sp.3** (3) (Group three). Spon00076

(Plates 74, A-E).

| As Per Bergquist:           | As PER WORMS 2015:        |
|-----------------------------|---------------------------|
| Restricted synonymy:        | Updated synonymy.         |
| (Bergquist & Fromont 1988). |                           |
| Class: Demospongiae         | Class: Demospongiae       |
| Order: Poecilosclerida      | Order: Poecilosclerida    |
|                             | Suborder: Mycalina        |
| Family: Mycalidae           | Family: Mycalidae         |
| Genus: Carmia               | Genus: Mycale             |
|                             | Subgenus: Mycale (Carmia) |
| Species: n.sp.              | Species: Unknown          |
|                             |                           |

Material examined: Spon00076, Bridge Marina, Tauranga Harbour, 6 m.

**Description**: This specimen was soft and amorphous with a relatively even surface. There were no organisms encrusting this specimen, and there was an abundance of larva present within the endosome. It should be noted that there was a large infestation of commensal polycahete worms within the choanosomal skeleton.

**Dimensions**: Length 7 cm; width 4.5 cm; thickness 4.5 cm, as a section of a larger sponge. Specimens above 10 cm were observed at this site.

Colour: In life yellow 5.0Y5/4, in spirit yellow 5.0Y8/4.

**Texture**: The sponge is soft, spongy and easy to tear.

**Surface**: The surface was smooth but irregular with projections without definitive shape (Plate 74, A, B). Pores were not visible within the preserved specimen.

**Skeleton**: The skeleton is plumose, with tracts of spicules occurring in spiral formations, although this may be the result of the large infestation of polychaete worms disrupting the endosomal tissue within this specimen (Plate 74, C, D). Subtylostyles are confused between spiral tracts and sigmas are found interstitially. The infestation of polychaete worms within the choanosomal structure made it difficult to distinguish the skeletal arrangement.

#### Spicules:

Megascleres: Subtylostyles- size ranges characteristic of other Carmia spp.

Microscleres: Anisochelae- one large size class. Sigmas- C shaped only.

For spicule dimensions see Table 2.25.

**Remarks**: This specimen has a single size class of large palmate anisochelae, which is dissimilar to other *Carmia* species described in Bergquist & Fromont (1988). The skeleton is plumose with columns of aligned megascleres. There was an abundance of larva scattered sub-dermally and throughout the choanosomal skeleton.

| Locality   |       | Subtylostyles<br>(µm) | Palmate<br>anisochelae<br>(µm) | Sigmas<br>(µm) |
|--|-------|-----------------------|--------------------------------|----------------|
| Spon00076, Bridge Marina,<br>Bay of Plenty, 6 m. | x     | 222 X 6.1             | 40                             | 99             |
|  | Range | 202-241 X<br>4.8-7.9  | 34-44                          | 88-109         |

Table 2.25: Spicule dimensions of *Carmia* n. sp. 3 (3).

*Carmia* **n.sp.3** (4) (Group three). Spon00075

(Plate 73, B-F).

| As Per Bergquist:           | As Per WoRMS 2015:        |
|-----------------------------|---------------------------|
| Restricted synonymy:        | Updated synonymy.         |
| (Bergquist & Fromont 1988). |                           |
| Class: Demospongiae         | Class: Demospongiae       |
| Order: Poecilosclerida      | Order: Poecilosclerida    |
|                             | Suborder: Mycalina        |
| Family: Mycalidae           | Family: Mycalidae         |
| Genus: Carmia               | Genus: Mycale             |
|                             | Subgenus: Mycale (Carmia) |
| Species: n.sp.              | Species: Unknown          |
|                             |                           |

Material examined: Spon00075, Bridge Marina, Tauranga Harbour, 6 m.

**Description**: The sponge is amorphous to massive with a soft but uneven surface and was found encrusting beneath a marina in approximately one meter of water. There are distinct tracts of dark sand created by polychaete worms visible subdermally through the transparent dermal membrane.

**Dimensions**: Length 3.5 cm; width 2 cm; thickness 1 cm, as a section of a larger sponge.

Colour: In life yellow 5.0Y8/8, in spirit yellow 5.0Y8/4.

Texture: Soft and compressible.

**Surface**: The surface is irregular with elevated projects without definite shape (Plate 73, B, C). There are dark vein-like processes just below the dermis, as a result of a polychaete worm infestation.

**Skeleton**: The choanosomal skeleton was plumose, with subtylostyles scattered interstitially between tracts, in addition to sigmas haphazardly placed within interstitial areas (Plate 73, D, E).

# Spicules:

Megascleres: Subtylostyles- size class's typical of other Carmia spp.

Microscleres: Palmate anisochelae- one large size class only. C shaped sigmas. For spicule dimensions see Table 2.26. **Remarks**: This specimen has a single size class of large palmate anisochelae, which is unlike other *Carmia* species described in Bergquist & Fromont (1988). The skeleton is plumose with columns of aligned megascleres. There was an abundance of larvae scattered sub-dermally and throughout the choanosomal skeleton.

| Locality   |       | Subtylostyles<br>(µm) | Palmate<br>anisochelae<br>(µm) | Sigmas<br>(µm) |
|--|-------|-----------------------|--------------------------------|----------------|
| Spon00075, Bridge Marina,<br>Bay of Plenty, 6 m. | x     | 231 X 6.6             | 33                             | 86             |
|  | Range | 207-249 X<br>5.8-8.1  | 21-45                          | 50-98          |

Table 2.26: Spicule dimensions of *Carmia* n. sp. 3 (4).

*Carmia* **n.sp.3** (5) (Group three). Spon00070

(Plate 68, A-F).

| As Per Bergquist:           | As Per WoRMS 2015:        |
|-----------------------------|---------------------------|
| Restricted synonymy:        | Updated synonymy.         |
| (Bergquist & Fromont 1988). |                           |
| Class: Demospongiae         | Class: Demospongiae       |
| Order: Poecilosclerida      | Order: Poecilosclerida    |
|                             | Suborder: Mycalina        |
| Family: Mycalidae           | Family: Mycalidae         |
| Genus: Carmia               | Genus: Mycale             |
|                             | Subgenus: Mycale (Carmia) |
| Species: n.sp.              | Species: Unknown          |
|                             |                           |

Material examined: Spon00070, Bridge Marina, Tauranga Harbour, 6 m.

**Description**: This sponge was thinly encrusting commensal on top of a living oyster shell (*Crassostrea gigas*). The specimen was living in approximately one meter of water beneath a wooden marina at a wave exposed site. The sample for this specimen was small as most of the organic material collected was infested with other organisms such as ascidians and bryozoans, thus was consequently removed.

**Dimensions**: Length 5 cm; width 4 cm; thickness 1.5 cm which was the surface area of the shell this specimen was commensal on.

Colour: In life yellow 5.0Y5/5, in spirit yellow 5.0Y8/4.

Texture: Soft and compressible with a spongy texture.

**Surface**: Smooth surface, with no projections present. No oscules were visible (Plate 68, A, B).

**Skeleton**: The skeleton is plumoreticulate, with subtylostyles aligned unidirectionally in thick multispicular tracts (Plate 68, C, D). Some sigmas are dispersed within the choanosome (Plate 68, C, D).

# **Spicules:**

Megascleres: Subtylostyles- size classes are typical of other Carmia spp.

Microscleres: Palmate anisochelae- one large size class only. S shaped and C shaped sigmas.

For spicule dimensions see Table 2.27.

**Remarks**: This specimen only has one size class of large palmate anisochelae, which is different to other *Carmia* species described in Bergquist & Fromont (1988). The skeleton is plumose with columns of aligned megascleres. There was an abundance of larvae scattered sub-dermally and throughout the choanosomal skeleton.

| Locality   |       | Subtylostyles<br>(µm) | Palmate<br>anisochelae<br>(µm) | Sigmas<br>(µm) |
|--|-------|-----------------------|--------------------------------|----------------|
| Spon00070, Bridge Marina,<br>Bay of Plenty, 6 m. | x     | 225 X 6.3             | 32                             | 110            |
|  | Range | 216-230 X<br>5.3-7.3  | 24-43                          | 95-118         |

Table 2.27: Spicule dimensions of Carmia n. sp. 3 (5).

Species description of the Demospongiae

*Carmia* **n.sp.3** (6) (Group three). Spon00066 (Plates 63, E-F, 64, A-F).

| As Per Bergquist:           | As Per WoRMS 2015:        |
|-----------------------------|---------------------------|
| Restricted synonymy:        | Updated synonymy.         |
| (Bergquist & Fromont 1988). |                           |
| Class: Demospongiae         | Class: Demospongiae       |
| Order: Poecilosclerida      | Order: Poecilosclerida    |
|                             | Suborder: Mycalina        |
| Family: Mycalidae           | Family: Mycalidae         |
| Genus: Carmia               | Genus: Mycale             |
|                             | Subgenus: Mycale (Carmia) |
| Species: n.sp.              | Species: Unknown          |

Material examined: Spon00066, Bridge Marina, Tauranga Harbour, 6 m.

**Description**: This specimen is soft and amorphous growing underneath a marina in a heavily populated sponge community. The specimen appears similar in gross morphology to *Carmia hentscheli* (Bergquist & Fromont, 1988, p. 145).

**Dimensions**: Length 8 cm; width 5 cm; thickness 2.5 cm.

Colour: In life yellow 5.0Y5/4, in sprit yellow 5.0Y8/4.

**Texture**: Soft, compressible and easy to tear.

**Surface**: The surface appears uneven with a granular appearance. The surface appears to be lacking some of its pinacoderm, and appears regressed, which may have been due to damage or predation, or signalling the end of this specimens life cycle. Oscules are found lying flush with the surface and are on average 1 mm in diameter.

**Skeleton**: The skeleton is plumose with tracts of aligned subtylostyles (Plate 64, A). There is a confused mass of spicules between the choanosomal tracts. The construction of the skeleton is difficult to distinguish due to the infestation of polychaete worms (Plate 64, B).

# **Spicules:**

Megascleres: Subtylostyles- size classes are typical of other *Carmia* spp. (Plate 64, C, E, F).

Microscleres: Palmate anisochelae- one large size class only. C and S shaped sigmas (Plate 64, D).

For spicule dimensions see Table 2.28.

**Remarks**: This specimen only has one size class of large palmate anisochelae, which is different to other *Carmia* species described in Bergquist & Fromont (1988). The skeleton is plumose with columns of aligned megascleres. There was an abundance of larvae scattered sub-dermally and throughout the choanosomal skeleton. Additionally, there was a heavy infestation of worms

| Locality   |                         | Subtylostyles<br>(µm) | Palmate<br>anisochelae<br>(µm) | Sigmas<br>(µm) |
|--|-------------------------|-----------------------|--------------------------------|----------------|
| Spon00066, Bridge Marina,<br>Bay of Plenty, 6 m. | $\overline{\mathbf{x}}$ | 218 X 5.5             | 43                             | 95             |
|  | Range                   | 211-223 X<br>5-6      | 42-44                          | 92-98          |

Table 2.28: Spicule dimensions of *Carmia* n. sp. 3 (6).

| Chapter 2                               | Species description of the Demospongiae |  |  |  |
|---|---|--|--|--|
| Carmia n.sp.4 cf. hentscheli (Group fou | r). (Plate 58, A-F).                    |  |  |  |
| Spon00060                               |   |  |  |  |
| As Per Bergquist:                       | As Per WoRMS 2015:                      |  |  |  |
| Restricted synonymy:                    | Updated synonymy.                       |  |  |  |
| (Bergquist & Fromont 1988).             |   |  |  |  |
| Class: Demospongiae                     | Class: Demospongiae                     |  |  |  |
| Order: Poecilosclerida                  | Order: Poecilosclerida                  |  |  |  |
| Family: Mycalidae                       | Family: Mycalidae                       |  |  |  |
| Genus: Carmia                           | Genus: Mycale                           |  |  |  |
|   | Subgenus: Mycale (Carmia)               |  |  |  |
| Species: Unknown                        | Species: Unknown                        |  |  |  |

Material examined: Spon00060, Bridge Marina, Tauranga Harbour, 6 m.

**Description**: This sponge was a massive with small conulose projections occurring over its surface. The specimen was found in one meter of water encrusting under a marina. The preserved material remains a dark purple and is therefore characteristic of this specimen.

Dimensions: Spon00060 was 6 cm in length; width 6 cm; thickness 3 cm.

Colour: In life purple 5.0P2/2, in spirit purple 5.0P2/2.

**Texture**: The sponge is soft but uneven and easy to pull apart.

**Surface**: The surface is lamellate with plate like erect ridges and small spinned conulose projections spread over the dermis.

**Skeleton**: The endosome has an abundance of thick spongin fibres; therefore the skeletal structure is difficult to distinguish. Oscules are 2-5 mm in diameter and are the only features which can be described when observing the water vascular system and skeleton without further removal of sponging tissue (Plate 58, C).

#### **Spicules:**

There are many different types of megascleres, which are believed to be borrowed from other sponges. Consequently they were not measured.

**Remarks**: This specimen was difficult to identify in that it had a mixture of many types and classes of spicules, which are believed to be borrowed from a neighbouring species. Moreover, the skeletal structure appeared to be mainly composed of thick spongin fibres, which masked spicules within the skeleton. Originally this specimen was placed within the Dictyodendrillidae family as it had an abundance of what appeared to be 'borrowed' spicules of many types, oval

choanocyte chambers and was a dark purple colour in life (Bergquist 1980). Nevertheless, this specimen did not possess a contrasting pale or uniformly pigmented soft tissue congruent with species within the Dictyodendrillidae family (Bergquist 1980). Moreover, this sponge was similar in gross morphology to *Carmia hentscheli* for the following reasons: 1) it was dark purple in life; 2) it was soft and compressible and easy to pull apart; 3) it had sand grains and other foreign material incorporated into its choanosomal structure; 4) it was characterised by a reduced spicule density 5) and it was similar in gross morphology and colour to *C. hentscheli*. The note of omission was that that the choanosomal skeleton was not visible due to an abundance of spongin fibres, therefore it could not be identified as a reticulate skeleton, nor did it contain particular spicule types. Consequently, it was grouped into the *Carmia* genus, and referred to *C. hentscheli* for comparison.

# **ORDER HALICHONDRIDA**

Definition: Found in Hooper & Van Soest 2002, p. 721.

# FAMILY HALICHONDRIIDAE

Definition: Found in Hooper & Van Soest 2002, p. 787.

## Genus Ciocalypta

Definition: Found in Bergquist, 1970, p. 34.

*Ciocalypta polymastia* (Lendenfeld 1888) Spon00005 (Plates 4, F, 5, A-F, 6, A).

| As Per Bergquist:       | As Per WoRMS 2015:      |  |
|-------------------------|-------------------------|--|
| Restricted synonymy:    | Updated synonymy.       |  |
| (Bergquist 1970).       |                         |  |
| Class: Demospongiae     | Class: Demospongiae     |  |
| Order: Halichondrida    | Order: Halichondrida    |  |
| Family: Halichondriidae | Family: Halichondriidae |  |
| Genus: Ciocalypta       | Genus: Ciocalypta       |  |
| Species: polymastia     | Species: polymastia     |  |

**Material examined:** Spon00005, Pilot Bay, Tauranga Harbour, 10.6 m References made to material described in Bergquist (1970).

**Description:** The *C. polymastia* specimen found in Pilot Bay is typical in comparison to other *C. polymastia* examined in New Zealand (Bergquist, 1970). This specimen has fistules or finger-like projections ubiquitous of the polymastias. Although the specimen collected here was massive, there were thickly encrusting specimens at this site.

**Dimensions:** Length 2 cm; width 0.5 cm, as a section of a larger sponge; the entire specimen was over 20 cm in situ.

Colour: In life the colour is pale yellow 5.0Y8/8; and is almost white in spirit.

Texture: Stiff, but compressible.

**Surface:** Smooth surface with small oscules 1 mm in diameter lying flush with the surface. The sponge had a papillae shape (Plate 4, F).

**Skeleton:** Plumose skeleton with spicule tracts radiating obliquely (Plate 5, F). The ecotosomal skeleton had a tangential structure with spicules arranged parallel to the surface.

Spicules: (Plate 5, A-D).

Megascleres: Subtylostyles- two size classes of subtylostyles various spicules were slightly curved, while others were straight.

For spicule dimensions see Table 2.29.

**Remarks:** Based on the descriptions provided by Bergquist (1970), and (Battershill *et al.*, 2010), I have concluded that this specimen was similar to *C. polymastia*. Specimens collected in this study were different from *C. polymastia* described elsewhere in that they contained oxeas. Furthermore, the specimen was more yellow in life at (5.0Y8/8) compared to *C. polymastia* (Y-R 7/10) described in Bergquist (1970).

|   |                         | Large         | Medium        |
|---|-------------------------|---------------|---------------|
| Locality                                    |                         | subtylostyles | subtylostyles |
|   |                         | (µm)          | (µm)          |
| North Channel,<br>10 fm<br>(Bargauist 1070) | x                       | 635 X 16.2    | 328 X 6.5     |
| (Bergquist 1970).                           | 5                       | 605-677 X     | 271-385 X     |
|   | Range                   | 12.7-22       | 5.7-7         |
| Spon00005<br>Pilot Bay, Tauranga,<br>10.6 m | $\overline{\mathbf{x}}$ | 579 X 19      | 205 X 6.7     |
| 10.0 m                                      | Danga                   | 551-608 X     | 161-329 X     |
| Känge                                       | 14.9-22.8               | 2.9-11.6      |               |

 Table 2.29: Spicule dimensions of Ciocalypta polymastia.

#### <u>Genus Halichondria</u>

Definition: Found in Bergquist, 1970. p. 32.

#### Halichondria n.sp.1

(Plate 65, A-F).

Spon00067

| As Per Bergquist:       | As Per WoRMS 2015:      |  |
|-------------------------|-------------------------|--|
| Restricted synonymy:    | Updated synonymy.       |  |
| (Bergquist 1970)        |                         |  |
| Class: Demospongiae     | Class: Demospongiae     |  |
| Order: Halichondrida    | Order: Halichondrida    |  |
| Family: Halichondriidae | Family: Halichondriidae |  |
| Genus: Halichondria     | Genus: Halichondria     |  |
| Species: n.sp.          | Species: Unknown        |  |
|                         |                         |  |

Material examined: Spon00067, Bridge Marina, Tauranga Harbour, 6 m.

**Description**: This specimen was a thickly encrusting specimen, with an uneven wrinkled and folded surface. It was found encrusting underneath a marine in approximately one meter of water. This specimen had a distinctive yellow colour in life and had no mucus present after collection.

**Dimensions**: Length 7 cm; width 4.5 cm; thickness 2 cm.

Colour: In life yellow 5.0Y7/6 to yellow 5.0Y6/6, in spirit yellow 5.0Y8/4.

Texture: Grainy, easy to tear and difficult to compress.

**Surface**: Irregularly wrinkled and folded surface (Plate 65, A, B). The sponge has a transparent dermal membrane after preservation in spirit (Plate 65, B). The surface is microscopically hispid.

**Skeleton**: The skeleton is confused to alveolate throughout the choanosome and becomes plumose near the dermis (Plate 65, C, D). The subtylostyles form a dense layer at right angles to the surface and protrude from the dermis appearing microscopically hispid.

#### Spicules:

Megascleres: Oxeas- of a single size class. The subtylostyles are of normal form. For spicule dimensions see Table 2.30.

#### **Remarks:**

Although the speculation was confused as per Halichondrid features, the spicule complement of oxeas and subtylostyles was dissimilar enough to listed New Zealand Halichondrids to warrant further investigation. The spiculation was confused as per usual Halichondrid features, the spicule complement was dissimilar enough to listed New Zealand Halichondrids to warrant further investigation.

| Locality   |       | Large<br>oxeas<br>(µm) | Subtylostyles<br>(µm) |
|--|-------|------------------------|-----------------------|
| Spon00067, Bridge Marina,<br>Bay of Plenty, 6 m. | x     | 55 X 1.1               | 200 X 7.3             |
|  | Dongo | 47-75 X                | 159-196 X             |
| Kange  | Kange | 0.7-1.2                | 4-8.3                 |

 Table 2.30: Spicule dimensions of Halichondria n. sp. 1
*Halichondria* **n.sp.2** *cf. panacea* Spon00007

(Plates 8, D-F, 9, A-F).

| As Per Bergquist:          | As Per WoRMS 2015:      |
|----------------------------|-------------------------|
| Restricted synonymy:       | Updated synonymy.       |
| (Bergquist, 1970).         |                         |
| Class: Demospongiae        | Class: Demospongiae     |
| Order: Halichondrida       | Order: Halichondrida    |
| Family: Halichondriidae    | Family: Halichondriidae |
| Genus: Halichondria        | Genus: Halichondria     |
| Species: n.sp. cf. panacea | Species: Unknown        |

**Material examined**: Spon00007, Pilot Bay Tauranga Harbour, 10.6 m References made to the material described in Bergquist (1970).

**Description**: Bergquist (1970) specified that *H. panicea* varies considerably in form throughout New Zealand. Massive or encrusting specimens with delicate detachable dermal membranes and flush oscula are found on the east coast of the North Island of New Zealand (Bergquist, 1970). In comparison, sponges found on the west coast have been described as possessing tough skin-like dermal membranes, with prominent, thick lipped oscula raised and aligned along ridges (Bergquist 1970). This fits the description for this species, as it was collected on the east coast, was thinly encrusting, and had flush oscules.

Dimensions: Length 3 cm; width 2 cm; thickness 3 mm.

**Colour**: In life, the colour is greenish yellow 7.5Y8/10; and almost white in spirit. **Texture**: Soft, easily torn.

**Surface**: The surface was entirely smooth with scarcely visible oscula chimneys, which may have been related to the exposed oceanic currents which it may have experienced encrusting at this site (Plate 8, F).

**Skeleton**: The choanosomal skeleton has isodictyal reticulation with triangular meshes composed of oxeas and styles. The ecotosomal skeleton displayed an alveolate structured skeleton (Plate 9, B, C).

#### Spicules:

Megascleres: Oxeas- two size classes, straight or slightly curved diactinal spicules (Plate 8, E). These were a fusiform shape. Styles- Slightly curved with one end

pointed, and the other end blunt, in addition to others styles possessing distinct tylote ends.

For spicule dimensions see Table 2.31.

**Remarks**: Occasionally style-like modifications occur within species of the *Halichondria* genus (Hooper & Van Soest 2002). This was evident in the specimen studied here, with style like modification occurring at low frequencies. Moreover, (Hooper & Van Soest 2002) diagnoses species belonging to the *Halichondria* genus as having oxeas with gradually tapering sharp points; which is consistent with this specimen (9, D-F).

| Looplity  |                         | Oxeas                | Styles               |
|---|-------------------------|----------------------|----------------------|
| Locality  |                         | (µm)                 | (µm)                 |
| Average <i>Halichondria</i><br><i>panacea</i> spicule dimensions<br>from specimens collected in<br>New Zealand (Bergquist<br>1970). | x                       | 348 X 6.7            | 213 X 4.8            |
|   | Range                   | 317-415 X<br>5.4-8.6 | 182-302 X<br>4.3-5.7 |
| Spon00007,<br>Pilot Bay, Tauranga,  | $\overline{\mathbf{X}}$ | 325 X 6              | 264 X 4.6            |
| 10.0 m  | Range                   | 247-294 X<br>3.3-6.8 | 247-294 X<br>3.3-6.8 |

Table 2.31: Spicule dimensions of *Halichondria* sp.

Chapter 2 Species description of the Demospongiae Halichondria n.sp.3 (Plate 40, B-F). Spon00042 As Per Bergquist: As PER WORMS 2015: Restricted synonymy: Updated synonymy. (Bergquist 1970) Class: Demospongiae Class: Demospongiae Order: Halichondrida Order: Halichondrida Family: Halichondriidae Family: Halichondriidae Genus: cf. Halichondria Genus: Unknown Species: n.sp. Species: Unknown

Material examined: Spon00042, Pilot Bay, Tauranga Harbour, 15 m.

**Description**: The sponge was a thickly encrusting specimen with visible elevated conulose oscules *in situ*. The oscules became retracted after preservation in spirit, but were clearly visible. The specimen was found thickly encrusting on a single stone in Pilot Bay.

Dimensions: Length 10.5 cm; width 7 cm; thickness 1cm.

Colour: In life yellow 5.0Y8/8, in spirit white.

Texture: The texture was soft, spongy and easily torn.

**Surface**: The surface was smooth but irregular with a thin transparent dermal layer, through which punctate pores are visible after preservation in spirit (Plate 40, D). The oscules were slightly elevated from the dermis *in situ* but become regressed after preservation in spirit (Plate 40, B-D).

**Skeleton**: Interestingly, the ectosomal skeleton within this specimen was dissimilar to any other skeleton described in Boury Esnault (1997). For instance, this specimen has net-like multispicular tracts, which were interconnected to form a dermal layer over the surface of the sponge. As a result the multispicular tracts often diverge from one another to become tangentially arranged. The net-like skeleton may be an ecophenotypic response to an increase in predation, wave exposure or scouring from movement of the rock on which it was encrusting. The ectosomal skeleton is an alveolate to confused structure.

#### Spicules:

Megascleres: Styles- slightly curved. Tylostyles- curved. Oxeas- long and curved.

For spicule dimensions see Table 2.32.

**Remarks:** Although the general speculation is Halichondriid, there are some unique structural components not seen in this family such that a new genus is suggested. For instance, this specimen has net-like multispicular tracts, which were interconnected to form a dermal layer over the surface of the sponge. Indeed this specimen requires more detailed examination with suitable type specimens for the Order.

| Tuble 2.52. Spicare differisions of Huncholmin a sp. |                         |                     |                      |                    |  |
|--|-------------------------|---------------------|----------------------|--------------------|--|
| Locolity   |                         | Styles              | Tylostyles           | Oxeas              |  |
| Locality   |                         | (µm)                | (µm)                 | (µm)               |  |
| Spon00042, Pilot Bay,<br>Bay of Plenty, 15 m         | $\overline{\mathbf{x}}$ | 239 X 7.7           | 161 X 5.8            | 315 X 7.9          |  |
|  | Range                   | 177-275 X<br>5.6-11 | 129-189 X<br>5.3-6.8 | 271-384 X<br>6-9.7 |  |

Table 2.32: Spicule dimensions of Halichondria sp.

*Halichondria moorei* (Bergquist 1961) Spon00044 (Plates 42, C-F, 43, A-B) Spon00046 (Plates 44, D-F, 45, A-C) Spon00049 (Plates 47, E, F, 48, A-F)

| As Per Bergquist:       | As Per WoRMS 2015:      |
|-------------------------|-------------------------|
| Restricted synonymy:    | Updated synonymy.       |
| (Bergquist 1970).       |                         |
| Class: Demospongiae     | Class: Demospongiae     |
| Order: Halichondrida    | Order: Halichondrida    |
| Family: Halichondriidae | Family: Halichondriidae |
| Genus: Halichondria     | Genus: Halichondria     |
| Species: moorei         | Species: moorei         |

Material examined: Spon00044, Pilot Bay, Tauranga Harbour, 15 m.

**Description**: Spon00044 was a globular to massive shaped specimen found encrusting on a rock in Pilot Bay. This specimen was heavily infested with polychaete worms. The Spon00049 specimen was massive to amorphous with prominent oscules lying flush with the surface. This specimen did not have an infestation of polychaete worms. Spon00046 was massive to amorphous with a visible infestation of polychaete worms on its pinacoderm. All three specimens had slightly transparent dermal membranes, with visible surface reticulation. Note: Spon00046 and Spon00044 are clearly the same species, thus they were grouped into this description.

**Dimensions**: Spon00044 was 11 in length; width 7 cm; thickness 3.5 cm. Spon00049 was 9 cm in length; width 7 cm; thickness 4 cm. *In situ* specimens above 15 cm have been observed. Spon00046 was 11 in length; width 7.5 cm; thickness 3.5 cm.

**Colour:** All specimens were yellow 5.0Y8/4 to white in life, in spirit yellow 5.0Y8/4.

Texture: All specimens are soft and fleshy, rather fragile.

**Surface**: The surface of all specimens is irregularly wrinkled with mammillate projections (Plate 42, C, D). Oscules range from 1-3 mm in diameter. Specimens

infested with polychaete worms have oscules filled with dark sand that appear as dots throughout the pinacoderm.

**Skeleton**: The endosomal skeleton is a confused mass of oxeas which protrude from the endopinacocytes (Plate 42, F). The ectosomal skeleton is a tangential formation of spicules which can be distinguished with the naked eye.

#### Spicules:

Megascleres: Oxeas- three size classes. The small class were straight and thick, the medium class were straight and thin, and the larger class were slightly curved. For spicule dimensions see Table 2.33.

**Remarks:** These specimens were well representative of the genus and species of *Halichondria moorei*. The irregularly wrinkled surface with oscules flush with the surface is characteristic of this specimen. As a result it was instantly identified based on its superficial features.

These specimens are well representative of the genus and species *Halichondia moorei*.

| Locality  |                         | Large oxeas | Small oxeas |
|---|-------------------------|-------------|-------------|
| Locality  |                         | (µm)        | (µm)        |
| Pt. Chevalier (type)  | $\overline{\mathbf{X}}$ | 628 X 13.2  |             |
|   | D                       | 300-800 X   |             |
|   | Range                   | 5-17        |             |
| Average <i>Halichondria moorei</i><br>spicule dimensions from<br>specimens collected in New<br>Zealand (Bergquist, 1970). | $\overline{\mathbf{x}}$ | 612 X 14    |             |
|   | Range                   | 339-750 X   |             |
|   | Kange                   | 6.2-17.3    |             |
| Spon00044, Pilot Bay,<br>Bay of Plenty, 15 m  | $\overline{\mathbf{x}}$ | 697 X 16.5  | 339 X 7.5   |
|   | Danga                   | 640-775 X   | 307-382 X   |
|   | Kange                   | 12-23       | 6.8-9.1     |

Table 2.33: Spicule dimensions of Halichondria moorei.

# FAMILY HYMENIACIDONIDAE

Definition: Found in Hooper & Van Soest 2002, p. 787. Note: Has been updated to *Halichondriidae* in Hooper & Van Soest (2002). **Genus** *Hymeniacidon* 

Definition: Found in Hooper & Van Soest, 2002, p. 807.

*Hymeniacidon hauraki* (Brønsted 1924) (Plates 43, C-F, 44, A-C). Spon00045

| As Per Bergquist:        | As PER WORMS 2015:     |
|--------------------------|------------------------|
| Restricted synonymy:     | Updated synonymy.      |
| (Bergquist 1970)         |                        |
| Class: Demospongiae      | Class: Demospongiae    |
| Order: Halichondrida     | Order: Halichondrida   |
| Family: Hymeniacidonidae | Family: Dictyonellidae |
| Genus: Hymeniacidon      | Genus: Stylissa        |
| Species: hauraki         | Species: haurakii      |
|                          |                        |

Material examined: Spon00045, Pilot Bay, Tauranga Harbour, 15 m.

**Description**: The body has finely serrated conulose formations giving a jagged appearance. A large amount of mucus was exuded from the specimen after removal. The dermal membrane is raised by small conulose projections that appear over the surface of the larger serrated finger-like conulose processes.

**Dimensions**: Length 7 cm; width 7 cm; bearing fistules that were 3.5 cm high.

**Colour**: In life yellow 5.0Y8/6, in spirit yellow 5.0Y8/4.

**Texture**: Soft and fleshy.

**Surface**: The surface is jagged with irregular, hispid rows of conules; each conule is lifted by a dermal brush of styles that penetrate the tips (Plate 43, C-E). The dermal membrane becomes transparent after preservation in spirit (Plate 43, E).

**Skeleton**: The choanosomal skeleton is a confused mass of styles, which form ascending tracts (Plate 43, F).

## **Spicules:**

Megascleres: Oxeas- of a single size class slightly curved. Styles- slightly curved styles (Plate 44, B, C).

For spicule dimensions see Table 2.34.

**Remarks**: This species is most common on deep reef habitats where sand levels do not exceed 15 cm in areas of coarse sand or shell Battershill & Bergquist (1984).

| Locality                                     |                         | Oxeas         | Styles     |
|--|-------------------------|---------------|------------|
| Locanty                                      |                         | ( <b>µm</b> ) | (µm)       |
| Prendstad North (type)                       | Danga                   |               | 400-800 X  |
| Brøndsted, North (type)                      | Kange                   |               | up to 14   |
| Takatu Channel, 6 fm                         | $\overline{\mathbf{X}}$ |               | 720 X 12.8 |
|  | Dense                   |               | 605-847 X  |
|  | Kange                   |               | 5.7-15     |
| Spon00045, Pilot Bay,<br>Bay of Plenty, 15 m | $\overline{\mathbf{x}}$ | 340 X 8.6     | 758 X 16   |
| с с, -                                       |                         | 337-344 X     | 744-777 X  |
|  | Kange                   | 7.1-10.4      | 13-21      |

Table 2.34: Spicule dimensions of Hymeniacidon hauraki.

# **ORDER HAPLOSCLERIDA**

Definition: Found in Bergquist & Warne, 1980, p. 12.

# FAMILY ADOCIIDAE

Definition: Found in Bergquist & Warne, 1980, p. 19.

#### <u>Genus Sigmadocia</u>

Definition: Found in Bergquist & Warne, 1980, p.22.

*Sigmadocia fragilis* (Plates 15, E-F, 16, A). Spon00013

| As Per Bergquist:        | As Per WoRMS 2015                     |
|--------------------------|---------------------------------------|
| Restricted synonymy:     | Updated synonymy.                     |
| (Bergquist & Warne 1980) |                                       |
| Class: Demospongiae      | Class: Demospongiae                   |
| Order: Haplosclerida     | Order: Haplosclerida                  |
|                          | Suborder: Haplosclerina               |
| Family: Adociidae        | Family: Chalinidae                    |
| Genus: Sigmadocia        | Genus: Haliclona                      |
| Species: fragilis        | Subgenus: Haliclona (Gellius)         |
|                          | Species: Haliclona (Gellius) fragilis |

Material examined: Spon00013, Karewa Island, Bay of Plenty, 10.8 m.

**Description**: A fragile sponge with fragile oscular fistules ascending from the upper surface. Its shape was massive to slightly spherical.

**Dimensions**: Length 4.5 cm; width 2.5 cm; 1.5 cm high fistules. *In situ* specimens over 10cm were observed.

**Colour**: In life the colour was not discernible with a poor quality picture; in spirit the sponge was white to yellow-red-yellow 10.0YR7/4.

Texture: Crisp and fragile.

**Surface**: Smooth surface with small oscular chimneys which were elevated from the dermis. The surface was covered in commensal hydroids (Plate 15, E).

**Skeleton**: The choanosomal skeleton was isodictyal with both multispicular and unispicular connections (Plate 15, F). The dermis is composed of spicule tracts

that run at right angles to the surface. The dermal skeleton has unispicular reticulation with a non-uniform structure.

#### Spicules:

Megascleres: Oxeas- of one size class

Microscleres: Sigmas- Some sigmas were much smaller than the larger oxeas (e.g. 10.3 microns compared to 28.3 microns). There was an abundance of C-shape sigmas.

For spicule dimensions see Table 2.35.

**Remarks**: There were some styles found within the sample but these are believed to be from a foreign source. Additionally, there were two asters found within the slide, which are believed to be foreign.

| Locality   |                         | Oxeas                 | Sigmas |
|--|-------------------------|-----------------------|--------|
| Locality   |                         | (µm)                  | (µm)   |
| Average Sigmadocia fragilis                        |                         |                       |        |
| Spicule dimensions from specimens                  |                         | 242 V 12              |        |
| collected in New Zealand                           | Х                       | 343 A 12              |        |
| (Bergquist &Warne 1980).                           |                         |                       |        |
|  | Range                   | 305-341               | 11-14  |
| Spon00013,<br>Karewa Island, Bay of Plenty,10.8 m. | $\overline{\mathbf{x}}$ | 214 X 10              | 12     |
|  | Range                   | 192-236 X<br>8.8-11.9 | 11-13  |

Table 2.35: Spicule dimensions of Sigmadocia fragilis.

# FAMILY HALICHLONIDAE

Definition: Found in Bergquist & Warne, 1980, p. 12.

#### Genus Haliclona

Definition: Found in Bergquist & Warne, 1980, p.12.

# Haliclona n. sp. 1 cf. fragilis

(Plates 3, F, 4, A-E).

Spon00004

| As Per Bergquist:           | As PER WORMS 2015:      |
|-----------------------------|-------------------------|
| Restricted synonymy:        | Updated synonymy.       |
| (Bergquist & Warne 1980)    |                         |
| Class: Demospongiae         | Class: Demospongiae     |
| Order: Haplosclerida        | Order: Haplosclerida    |
|                             | Suborder: Haplosclerina |
| Family: Haliclonidae        | Family: Chalinidae      |
| Genus: Haliclona            | Genus: Haliclona        |
| Species: n.sp. cf. fragilis | Species: Unknown        |

**Material examined**: Spon00004, Pilot Bay, Tauranga Harbour, 10.6 m References made to the material described in Bergquist & Warne (1980).

**Description**: A massive sponge with oscula that is flush with the pinacoderm. Spongin can be found at the ends of the nodes on some spicule meshes. The specimen was heavily colonised by red algae epibionts.

**Dimensions**: Length 6 cm; width 5 cm; thickness 4 cm, as a section of a larger sponge. Specimens over 100 cm were observed within the Tauranga Harbour.

**Colour**: In life the colour is 5.0Y8/4 and appears to be the same colour in spirit 5.0Y8/4.

**Texture**: Soft and friable; easily torn; easily compressed and brittle when picked up. No slime present.

**Surface**: Transparent dermal membrane, through which ostia and oscula are clearly visible. Ostia were haphazardly located over the dermis, with congregations of oscula visible.

**Skeleton**: The skeletal network appears to have isodictyal to anisotropic reticulation (Plate 4, B). There did not appear to be any unispicular reticulation in the skeletal arrangement similar to that found in *H. fragilis* (Bergquist & Warne, 1980 (Plate 4, B). Rectangular and polygonal skeletal arrangements were also present in loosely constructed meshes. The specimen contained reticulated multispicular primary lines, which were often connected by unispicular secondary lines and did not appear to have an ectosomal skeleton.

Spicules: (Plate 4, A).

Megascleres: Hastate oxeas- Large thick oxeas, straight or slightly curved with no mucronate ends visible. There did not appear to be a large range between thicknesses of oxeas.

For spicule dimensions see Table 2.36.

Remarks: Bergquist & Warne (1980) stated that sponges found within the Haliclonidae family are the most difficult to classify of all of the Haplosclerida. There are few visible characters to differentiate among species and there is great morphological variability. This species had some of the components of Haliclona fragilis in that it was soft and limp; easily torn and relatively compressible (Bergquist & Warne, 1980). The surface characteristics of this specimen were not congruent with that of *H. fragilis* as they lacked cylindrical aggregates of oscula (Bergquist & Warne, 1980; Plate 3 F; Plate 4 A). Unlike *H. fragilis* this specimen did not have any mucronate oxeas and did not have a large range in the widths of oxeas (Bergquist & Warne, 1980). Additionally, unlike H. fragilis this specimen was not visibly slimy or hispid, and the skeletal structure was more irregular than that shown in the photographs of H. fragilis described by Bergquist & Warne (1980) (Plate 4 D). There was unispicular reticulation; however, it was extremely dense. The skeletal structure was a combination of an isodictyal to anisotropic arrangement (Bergquist & Warne 1980; Hooper & Van Soest 2002). In some parts of the sponge the skeletal structure was clearly isodictyal (Plate 4 D); however, anisotropic skeletal arrangements were also visible in other sections.

| Ĩ   |                         | 1         |
|---|-------------------------|-----------|
| Locality  |                         | Oxeas     |
| Locality  |                         | (µm)      |
| (Holotype, H. fragilis)   |                         |           |
| Wairepo Lagoon  | _                       | 175 X O   |
| (Bergquist & Warne 1980)  | X                       | 175 X 9   |
|   | _                       | 131-197 X |
|   | Range                   | 2-13      |
| Average <i>Haliclona fragilis</i><br>spicule dimensions from<br>specimens collected in New<br>Zealand (Bergquist & Warne<br>1980) | x                       | 139 X 6.5 |
|   |                         | 111-166 X |
|   | Range                   | 1-10      |
| Spon00004<br>Pilot Bay, Tauranga,   | $\overline{\mathbf{x}}$ | 156 X 6.5 |
| 10.6 m  |                         |           |
|   | Danas                   | 148-168 X |
|   | Kange                   | 5.7-7.6   |

| <b>Table 2.36:</b> | Spicule | dimensions | of Haliclona | sp. 1 |
|--------------------|---------|------------|--------------|-------|
|--------------------|---------|------------|--------------|-------|

| Chapter 2                | Species description of the Demospongiae |
|--------------------------|---|
| Haliclona n.sp. 2        | (Plates 36, D-F, 37, A-F).              |
| Spon00033                |   |
| As Per Bergquist:        | As Per WoRMS2015:                       |
| Restricted synonymy:     | Updated synonymy.                       |
| (Bergquist & Warne 1980) |   |
| Class: Demospongiae      | Class: Demospongiae                     |
| Order: Haplosclerida     | Order: Haplosclerida                    |
|                          | Suborder: Haplosclerina                 |
| Family: Haliclonidae     | Family: Chalinidae                      |
| Genus: Haliclona         | Genus: Haliclona                        |
| Species: n.sp.           | Species: Unknown                        |

Material examined: Spon00033, Salisbury Wharf, Tauranga Harbour, 5 m.

**Description**: This specimen is amorphous with an uneven surface. The sponge was found encrusting on Salisbury Wharf in approximately five meters of water. Segments of the sponge appeared degraded *in situ*. Moreover, oscula were protuberant *in situ* and after removal; however, oscules became regressed after preservation in spirit.

**Dimensions**: Length 9 cm; width 7 cm; thickness 1.5 cm as a section of a larger sponge

**Colour**: In life yellow green-yellow 10.0Y8/8 to white, in spirit yellow 5.0Y6/4. **Texture**: Soft and compressible.

**Surface**: The sponge has a slightly regressed uneven dermal layer which appears degraded (Plates 36, D-F, 37, E, F).

**Skeleton**: This specimen is heavily infested with polychaete worms; as a result the skeletal architecture has been modified to the point where it is indistinguishable (Plate 37, A, B). Small sections of the skeleton do however; appear to have a confused organisation.

#### Spicules:

Megascleres: Oxeas- of normal form (Plate 37, C, D).

For spicule dimensions see Table 2.37.

**Remarks:** The skeleton was degraded in this specimen, but despite this there were differences to the arrangement of fibres that suggested the specimen could not be assigned to known species without further work probably including a fresh collection from the site.

| Looplity  |       | Oxeas                |  |
|---|-------|----------------------|--|
| Locanty   |       | (µm)                 |  |
| Spon00033, Salisbury Wharf,<br>Bay of Plenty, 5 m | x     | 131 X 5.4            |  |
|   | Range | 117-145 X<br>4.2-7.3 |  |

 Table 2.37: Spicule dimensions of Haliclona n.sp. 2

Haliclona n.sp. 3 *cf. tenacior* Spon00030 (Plates 34, B-F, 35, A-C). Spon00063 (Plate 61, A-F) Spon00035 (Plates 37, E, F, 38, A-F).

| As Per Bergquist:           | As PER WORMS 2015:      |
|-----------------------------|-------------------------|
| Restricted synonymy:        | Updated synonymy.       |
| (Bergquist & Warne 1980)    |                         |
| Class: Demospongiae         | Class: Demospongiae     |
| Order: Haplosclerida        | Order: Haplosclerida    |
|                             | Suborder: Haplosclerina |
| Family: Haliclonidae        | Family: Chalinidae      |
| Genus: Haliclona            | Genus: Haliclona        |
| Species: n.sp. cf. tenacior | Species: Unknown        |

**Material examined**: Spon00030, Salisbury Wharf, Tauranga Harbour, 5 m. Spon00063, Bridge Marina, Tauranga Harbour, 6 m. Spon00035, Salisbury Wharf, Tauranga Harbour, 5 m. Note specimens of Spon00030, Spon00063 and Spon00035 are the same species, thus they were grouped into this description.

**Description**: All specimens are thickly encrusting to massive, usually infested with commensal tube worms. They have slightly raised oscules that range from 1-3 mm in diameter. Spon00063 had broad barrel shaped oscules and a transparent dermal membrane after preservation in spirit. Spon00035 had broader coulose shaped oscules that were slightly elevated from the dermis (Plate 38, A).

**Dimensions**: Spon00030 was 7 cm in length; width 5cm; thickness 2 cm. Spon00063 was 7.5 cm in length; width 5.5 cm; thickness 1.5 cm. Spon00035 was 6 cm in length and 4 cm width.

**Colour**: Spon00030 in life yellow 5.0Y7/6, in spirit yellow5.0Y7/4. Spon00063 in life yellow-red-yellow 10.YR4/4 to yellow-red-yellow 10.0YR7/8, in spirit yellow 5.0Y8/6. Spon00030 in life yellow 5.0Y7/6, in spirit yellow5.0Y7/4.

**Texture**: The texture of all specimens was soft, spongy, compressible and easy to tear.

Surface: All specimens are encrusting to massive sponges with short, rounded oscular papillae that are slightly raised form the surface (Plate 34, B-D).

Spon00030 is noticeable covered by red algae, which appear as thick hair like projections after preservation in spirit. Spon00063 has elevated barrel shaped oscules (Plate 61, A, B). The Spon00035 specimen had broader conulose oscules (Plate 38, A).

**Skeleton**: The skeletal organisation of all specimens is a unispicular isodictyal network at the basic level; however, in most instances there is a multispicular network (Plates 34, E, F, 35, A). The choanosomal skeleton within Spon00035 was heavily infested with tube worms (Plate 38, B-D).

#### **Spicules:**

Megascleres: Oxeas- withy slightly flexed shafts in all specimens (Plate 38, E, F). For spicule dimensions see Table 2.38. (Note spicule dimensions for all three specimens were similar in size, consequently a single representation of Spon00063 was used.

**Remarks:** The skeletal structure and spicule morhphology is consistent with *H*. *tenacior* but there is enough difference to warrant a check with the type specimen.

| Locality   |                         | Oxeas     |  |
|--|-------------------------|-----------|--|
| Locality   |                         | (µm)      |  |
| Spon00030, Salisbury Wharf,<br>Bay of Plenty 5 m | x                       | 124 X 5   |  |
|  | Range                   | 120-133 X |  |
|  |                         | 3.36-6.6  |  |
| Spon00063,                                       |                         |           |  |
| Bridge Marina, Tauranga,                         | $\overline{\mathbf{X}}$ | 240 X 11  |  |
| 6 m  |                         |           |  |
|  | Range                   | 145-99 X  |  |
|  |                         | 8.4-2.2   |  |

Table 2.38: Spicule dimensions of Haliclona n.sp. 3 cf. tenacior

Haliclona n.sp. 4 cf. brøndstedi Spon00071

(Plate 69, A-F).

| <u>As Per Bergquist</u> :     | As Per WoRMS 2015:            |
|-------------------------------|-------------------------------|
| Restricted synonymy:          | Updated synonymy.             |
| (Bergquist & Warne 1980)      |                               |
| Class: Demospongiae           | Class: Demospongiae           |
| Order: Haplosclerida          | Order: Haplosclerida          |
|                               | Suborder: Haplosclerina       |
| Family: Haliclonidae          | Family: Chalinidae            |
| Genus: Haliclona              | Genus: Haliclona              |
|                               | Subgenus: Haliclona (Gellius) |
| Species: n.sp. cf. brøndstedi | Species: Unknown              |

Material examined: Spon00071, Bridge Marina, Tauranga Harbour, 6 m.

**Description**: The sponge is an amorphous specimen with a large number of conical oscular turrets on its surface.

**Dimensions**: Length 6 cm; width 4 cm; thickness 2 cm as a section of a larger sponge.

Colour: In life yellow 5.0Y8/5, in spirit yellow 5.0Y8/4.

Texture: The specimen is soft and friable.

**Surface**: Evenly and minutely hispid. The majority of oscules are flush with the surface or slightly elevated. The dermal membrane is transparent, through which oscular pores are visible after preservation in spirit (Plate 69, B).

**Skeleton**: The skeleton consists of a unispicular to multispicular network of megascleres (Plate 69, C, D). The networks do not always have a regular quadratic network, and are often strewn in a disorientated way throughout the choanosome (Plate 69, C, D).

#### Spicules:

Megascleres: Oxeas- slightly curved (Plate 69, E, F).

For spicule dimensions see Table 2.39.

**Remarks:** This specimen had most of the characteristics of *H. brøndstedi*, however, there were no toxa microscleres, which is the defining feature which

separates this species from the rest of the *Haliclona* genus (Bergquist & Warne, 1980).

| Locality   |       | Oxeas<br>(µm) |
|--|-------|---------------|
| Spon00071, Bridge Marina,<br>Bay of Plenty, 6 m. | x     | 131 X 7       |
|  | Range | 123-148 X     |
|  |       | 5.7-10.4      |

Table 2.39: Spicule dimensions of Haliclona n. sp. 4 cf. brøndstedi

| Haliclona n.sp. 5 cf. heterofibrosa |  |
|-------------------------------------|--|
| Spon00068                           |  |

(Plate 66, A-F).

| As Per Bergquist:                | As Per WoRMs 2015:      |
|----------------------------------|-------------------------|
| Restricted synonymy:             | Updated synonymy.       |
| (Bergquist & Warne 1980)         |                         |
| Class: Demospongiae              | Class: Demospongiae     |
| Order: Haplosclerida             | Order: Haplosclerida    |
|                                  | Suborder: Haplosclerina |
| Family: Haliclonidae             | Family: Chalinidae      |
| Genus: Haliclona                 | Genus: Haliclona        |
| Species: n.sp. cf. heterofibrosa | Species: Unknown        |

Material examined: Spon00068, Bridge Marina, Tauranga Harbour, 6 m.

**Description**: The sponge was a thickly encrusting individual that was found in the subtidal zone. The sponge has barrel shaped oscules that are slightly raised or flush with the dermis.

**Dimensions**: Length 6 cm; width 3 cm; thickness 3 cm. *In situ* specimens above 5 cm have been observed.

Colour: In life yellow 5.0Y8/4, in spirit is the same colour as in life.

Texture: Soft, limp and easy to pull apart.

**Surface**: The surface is smooth and granular, with oscules that are either flush with the dermis or slightly raised. Some oscules are broad and barrel shaped extending out from the rest of the pinacoderm (Plate 66, A, B).

**Skeleton**: This specimen has simple quadratic reticulation of oxeas with predominately multispicular tracts (Plate 66 C, D). The multispicular tracts are placed one to two spicules apart. Bergquist & Warne, 1980 found a correlation between wave exposure and skeletal arrangement. It is probable that spicule reinforcement of the skeleton in *H. heterofibrosa* is an ecophenotypic response to increased wave exposure (Bergquist & Warne, 1980), which may explain the multispicular reinforcement within this specimen.

## **Spicules:**

Megascleres: Oxeas- slightly curved (Plate 66, E). Euasters were also found in a low abundance (Plate 66, F).

For spicule dimensions see Table 2.40.

**Remarks:** The skeletal structure and spicule morphology is consistent with *H*. *heterofibrosa* but there is enough difference to warrant a check with the type specimen.

LocalityLarge oxeasSpon00068, Bridge Marina,<br/>Bay of Plenty, 6 m. $\overline{x}$ 131 X 6.6117-131 X<br/>Range117-131 X<br/>3.7-8

Table 2.40: Spicule dimensions of Haliclona n.sp. 5 cf. heterofibrosa

Haliclona n.sp. 6 cf. heterofibrosa Spon00064

(Plate 62, A-F).

| As Per WoRMS 2015:      |
|-------------------------|
| Updated synonymy.       |
|                         |
| Class: Demospongiae     |
| Order: Haplosclerida    |
| Suborder: Haplosclerina |
| Family: Chalinidae      |
| Genus: Haliclona        |
| Species: Unknown        |
|                         |

Material examined: Spon00064, Bridge Marina, Tauranga Harbour, 6 m.

**Description**: The sponge was an amorphous specimen with oscules that are flush with the surface.

**Dimensions**: Length 6.5 cm; width 6 cm.

Colour: In life yellow 5.0Y8/4, in spirit was the same colour as in life.

**Texture**: The sponge has a grainy texture, is relatively firm and easy to pull apart.

**Surface**: Oscules are flush with the surface and are up to 4 cm in this specimen in life (Plate 62, A, B). There is a transparent layer of pinacoderm tissue around the oscules after preservation in spirit (Plate 62, B). The pinacoderm is relatively transparent, through which small pores are visible. The surface is uneven and granular.

**Skeleton**: A simple quadratic reticulation of oxeas is the typical skeletal arrangement, which is reinforced with multispicular tracts. The quadratic structures extend to one spicule in length (Plate 62, C, D).

#### Spicules:

Megascleres: Oxeas- slightly flexed.

For spicule dimensions see Table 2.41.

**Remarks**: This specimen had a skeletal arrangement similar to that of *Haliclona heterofibrosa*. The structure was more ridged compared to the other specimen (Spon00068) that was compared to *H. heterofibrosa* in this study. The note of omission, however, for this specimen is that Bergquist & Warne (1980) found that

*H. heterofibrosa* may change its phenotype and skeletal arrangement depending on the severity of wave exposure resulting in a ridged specimen.

| Locality   |       | Large oxeas          |  |
|--|-------|----------------------|--|
| Locality   |       | (µm)                 |  |
| Spon00064, Bridge Marina,<br>Bay of Plenty, 6 m. | x     | 162 X 9.1            |  |
|  | Range | 152-170 X<br>7.8-9.8 |  |

 Table 2.41: Spicule dimensions of Haliclona n.sp. 6 cf. heterofibrosa

Haliclona heterofibrosa (Lundbeck 1902) Spon00059 (Plate 57, A-F) Spon00065 (Plate 63, A-D)

| As Per Bergquist:        | As Per WoRMS 2015:                    |
|--------------------------|---------------------------------------|
| Restricted synonymy:     | Updated synonymy.                     |
| (Bergquist & Warne 1980) |                                       |
| Class: Demospongiae      | Class: Demospongiae                   |
| Order: Haplosclerida     | Order: Haplosclerida                  |
|                          | Suborder: Haplosclerina               |
| Family: Haliclonidae     | Family: Chalinidae                    |
| Genus: Haliclona         | Genus: Haliclona                      |
|                          | Subgenus: Haliclona (Rhizoniera)      |
| Species: heterofibrosa   | Species: Haliclona (Rhizoniera) rosea |

**Material examined**: Spon00059, Bridge Marina, Tauranga Harbour, 6 m. Spon00065, Bridge Marina, Tauranga Harbour, 6 m. Note specimens Spon00059 and Spon00065 are the same species, thus they were grouped into this description. **Description**: The Spon00059 sponge is a thickly encrusting specimen and invested on two oyster shells (*Crassostrea gigas*). This specimen had hydroids which were commensal on its surface. This specimen holds a large amount of water when removed from the water. The Spon00065 specimen was a thickly encrusting specimen with raised barrel shaped oscules.

**Dimensions**: Spon00059 was 8 cm in length; width 5.5 cm; thickness 3 cm. Spon00065 was 5cm in length; width 4 cm; thickness 3 cm. Specimens above 10 cm have been observed in length have been observed *in situ*.

**Colour:** In life yellow-red-yellow 10.0YR6/4, in spirit yellow 5.0Y8/4. In life yellow 5.08/6, in spirit yellow 5.0Y8/6.

Texture: Soft and spongy. The specimen is delicate as it is easy to tear.

**Surface**: The majority of oscules are slightly raised from the surface and are barrel shaped in the Spon00059 specimen (Plate 57, A, B). Some oscules were conulose and elevated further, with small pores occurring near the apex. The surface of this specimen is shaggy and slightly granular. The Spon00065 specimen also has elevated barrel shaped oscules on its surface (Plate 63, A, B).

**Skeleton**: The skeleton is a simple quadratic reticulation of oxeas, but the skeleton is also reinforced with multi- and uni-spicular reticulation that can extend to four spicules in length in both specimens (Plates 57, C-E, 63, C, D). The shaggy surface may be a response to the multispicular reticulation (Bergquist & Warne, 1980).

#### Spicules:

Megascleres: Oxeas- two size classes, slightly curved in both specimens.

For spicule dimensions see Table 2.42.

#### **Remarks:**

This specimen is a good example of *Haliclona heterofibrosa with* The skeleton is a simple quadratic reticulation of oxeas, but the skeleton is also reinforced with multi and uni-spicular reticulation that can extend to four spicules in length in both specimens (Plates 57, C-E, 63, C, D). This skeletal architecture is characteristic of *H. heterofibrosa*.

| Logality                        |                         | Large oxeas | Medium oxea |  |
|---------------------------------|-------------------------|-------------|-------------|--|
| Locanty                         |                         | (µm)        | (µm)        |  |
| Spon00059, Bridge Marina,       | v                       | 139 X 7 9   | 112 X 3 6   |  |
| Bay of Plenty, 6 m.             | л                       | 137 A 1.7   | 112 X 5.0   |  |
|                                 | D                       | 131-146 X   | 117-4 X     |  |
|                                 | Kange                   | 7.5-8       | 2.9-4       |  |
| Average Haliclona heterofibrosa |                         |             |             |  |
| spicule dimensions from         |                         |             |             |  |
| specimens collected in          | $\overline{\mathbf{X}}$ | 136 X 7.1   |             |  |
| New Zealand                     |                         |             |             |  |
| (Bergquist & Fromont. 1988).    |                         |             |             |  |
|                                 | D                       | 127-152 X   |             |  |
|                                 | Kange                   | 7.1         |             |  |

 Table 2.42: Spicule dimensions of Haliclona heterofibrosa.

| Chapter 2                | Species description of the Demospongiae |
|--------------------------|---|
| Haliclona n.sp. 7        | (Plate 56, C-F).                        |
| Spon00058                |   |
| As Per Bergquist:        | As Per WoRMS 2015:                      |
| Restricted synonymy:     | Updated synonymy.                       |
| (Bergquist & Warne 1980) |   |
| Class: Demospongiae      | Class: Demospongiae                     |
| Order: Haplosclerida     | Order: Haplosclerida                    |
|                          | Suborder: Haplosclerina                 |
| Family: Haliclonidae     | Family: Chalinidae                      |
| Genus: Haliclona         | Genus: Haliclona                        |
| Species: n.sp            | Species: Unknown                        |

Material examined: Spon00058, Bridge Marina, Tauranga Harbour, 6 m.

**Description**: The sponge is soft and amorphous with raised uneven surface projections and was heavily infested with commensal polychaete worms.

Dimensions: Length 11 cm; width 7 cm; thickness 5.5 cm.

Colour: In life yellow 5.0Y5/4, in spirit yellow 5.0y8/4 to white.

Texture: Soft, friable and easy to pull apart.

**Surface**: Smoot but uneven surface with elevated projections of irregular shapes (Plate 56, C, D). The sponge has a transparent dermal membrane, through which pores are visible after preservation in spirit (Plate 56, D).

**Skeleton**: The sponge had isodictyal reticulation within its choanosomal skeleton. **Spicules:** 

Megascleres: Oxeas- large size class only.

For spicule dimensions see Table 2.43.

**Remarks:** This specimen is different enough from other *Haliclona* species examined above to list separately. It also requires comparison with type specimens of common New Zealand Haliclonids especially *H. heterofibrosa*. It had some of the characteristics of *H. heterofibrosa* as it appeared similar morphologically with a smooth but uneven surface and had isodictyal reticulation within its choanosomal skeleton.

| 1  |                         | 1           |  |
|--|-------------------------|-------------|--|
| Locality   |                         | Large oxeas |  |
| Locanty  |                         | (µm)        |  |
| Spon00058, Bridge Marina,<br>Bay of Plenty, 6 m. | $\overline{\mathbf{x}}$ | 152 X 7.5   |  |
|  | Dongo                   | 140-167 X   |  |
|  | Kallge                  | 6.7-8.5     |  |

Table 2.43: Spicule dimensions of *Haliclona* sp.

#### Genus Adocia

Definition: Found in Bergquist & Warne, 1980, p.20.

| Adocia n. sp. 1              | (Plate 67, A-F).                |
|------------------------------|---------------------------------|
| Spon00069                    |                                 |
| As Per Bergquist:            | As PER WORMS 2015:              |
| Restricted synonymy:         | Updated synonymy.               |
| (Bergquist & Warne 1980)     |                                 |
| Class: Demospongiae          | Class: Demospongiae             |
| Order: Haplosclerida         | Order: Haplosclerida            |
|                              | Suborder: Haplosclerina         |
| Family: Adociidae            | Family: Chalinidae              |
| Genus: Adocia                | Genus: Haliclona                |
| Species: n.sp. cf. venustina | Subgenus: Haliclona (Haliclona) |

Material examined: Spon00069, Bridge Marina, Tauranga Harbour, 6 m.

**Description**: The sponge was thickly encrusting to massive with flush oscules and or tall chimneys with pores occurring at the apex of these oscules. The sponge was found sub-tidally encrusting on a marina in a wave exposed environment.

**Dimensions**: Length 5.5 cm; width 2 cm; thickness 4 cm as a section of a larger sponge.

Colour: In life yellow 5.0Y7/4, in spirit yellow 5.0Y8/4.

**Texture**: The texture is firm and friable.

**Surface**: The surface uneven, and appears punctate due to the appearance of microscopic pores, which can be seen through the slightly transparent dermal membrane. Oscules are both flushed with the surface and situated at the apex of tall chimneys (Plate 67, A, B).

**Skeleton**: The choanosomal skeleton has a unispicular to multispicular simple quadratic reticulation. The reticulation is separated by 1-2 oxeas in length (Plate. 67, C, D).

#### Spicules:

Megascleres: Oxeas- two size classes, slightly curved (Plate 67, E). There were some birotules found within the thick section, however, these were in low abundance and were therefore considered foreign (Plate 67, F) For spicule dimensions see Table 2.44.

**Remarks:** The presence of birotules within this specimen may have been foreign. Further work is required to determine whether these do in fact, belong to this species.

| Locality   |       | Large oxeas          | Small oxeas        |
|--|-------|----------------------|--------------------|
|  |       | (µm)                 | (µm)               |
| Spon00069, Bridge Marina,<br>Bay of Plenty, 6 m. | x     | 152 X 8.7            | 91 X 2.6           |
|  | Range | 151-152 X<br>7.6-9.6 | 82-96 X<br>1.7-3.8 |

Table 2.44: Spicule dimensions of Adocia n. sp. 1

| Chapter 2                     | Species description of the Demospongiae |
|-------------------------------|---|
| Adocia n.sp. 2                | (Plates 41, A-F, 42, A-B).              |
| Spon00043                     |   |
| AS PER BERGQUIST:             | As Per WoRMS 2015:                      |
| Restricted synonymy           | Updated synonymy.                       |
| (Bergquist & Warne 1980)      |   |
| Class: Demospongiae           | Class: Demospongiae                     |
| Order: Haplosclerida          | Order: Haplosclerida                    |
|                               | Suborder: Haplosclerina                 |
| Family: Adocia                | Family: Chalinidae                      |
| Genus: Adocia                 | Genus: Haliclona                        |
| Species: n.sp. cf. venustina. | Species: n.sp. cf. venustina            |

Material examined: Spon00043, Pilot Bay, Tauranga Harbour, 15 m.

**Description**: The sponge is a spherical shape with a punctate surface and raised oscules both *in situ* and after preservation. The specimen was found encrusting in the subtidal zone in a sheltered environment in Pilot Bay.

Dimensions: Length 10 cm; width 7 cm; thickness 6 cm.

Colour: In life yellow-red-yellow to white, in spirit yellow 5.0Y7/4.

Texture: Firm and crumbly; crisp.

**Surface**: The sponge is a spherical shape with broad elevated oscules in life (Plate 41, A). The oscules become regressed after preservation in spirit, but still appear slightly raised (Plate 41, B). There is a thin transparent dermal layer which is punctate.

**Skeleton**: The skeleton has multispicular aligned tracts of oxeas which occur in thick bundles and are aligned parallel to each other. The tracts often join and appear plumoreticulate (Plate 41, D-F). Oxeas appear scattered in interstitial areas, and in some instances the oxeas appear to be arranged in an alveolate formation around oscules.

#### Spicules:

Megascleres: Oxeas- of a single size class only (Plate 42, A, B).

For spicule dimensions see Table 2.45.

**Remarks**: This specimen was more globular shaped than other *Adocia* species described within this study.

|  | Oxeas |                     |
|--|-------|---------------------|
| Locality                                     |       | (µm)                |
| Spon00043, Pilot Bay,<br>Bay of Plenty, 15 m | x     | 155 X 8.7           |
|  | Range | 123-180 X<br>3.4-12 |

 Table 2.45: Spicules dimensions of Adocia n. sp. 2

# FAMILY CALLYSPONGIIDAE

Definition: Found in Bergquist & Warne, 1980, p. 24.

#### Genus Callyspongia

Definition: Found in Bergquist & Warne, 1980, p.24.

| Callyspongia ramosa (Gray 1843)   | (Plates 35, D-F, 36, A-C). |  |
|-----------------------------------|----------------------------|--|
| Spon00032                         |                            |  |
| As Per Bergquist:                 | As Per WoRMS 2015:         |  |
| Restricted synonymy: (Bergquist & | Updated synonymy.          |  |
| Warne 1980)                       |                            |  |
| Class: Demospongiae               | Class: Demospongiae        |  |
| Order: Haplosclerida              | Order: Haplosclerida       |  |
|                                   | Suborder: Haplosclerina    |  |
| Family: Callyspongiidae           | Family: Callyspongiidae    |  |
| Genus: Callyspongia               | Genus: Callyspongia        |  |
| Species: ramosa                   | Species: ramosa            |  |
|                                   |                            |  |

Material examined: Spon00032, Salisbury Wharf, Tauranga Harbour, 5 m.

**Description**: Arborescent, erect branching cylindrical branches which have both tube and flattened shaped branches (Plate 35, D-F). The oscules are aligned in a linear pattern along the branches of this specimen which is characteristic of *Callyspongia ramosa*.

**Dimensions**: The branches are 6 mm in diameter and extending 60 cm high.

Colour: In life yellow 5.0Y8/4, in spirit yellow 5.0Y8/4.

Texture: Firm and elastic.

**Surface**: Smooth and even, with oscules flush with the dermis arranged linearly along the branches. The tips of branches appear to have microscopic hair like projections of tissue after preservation in spirit (Plate 35, D-F).

**Skeleton**: The skeleton has a radial vertical reticulation of multispicular primary fibres with unispicular secondary's (Plate 36, A, B). There appears to be oxeas scattered interstitially between radial tracts.

## **Spicules:**

Megascleres: Oxeas- two size classes (Plate 36, C). The larger class were thin and bent, the smaller class were thick and straight.

For spicule dimensions see Table 2.46

Oxeas were straight, and bonded together with collagen. A classic C. ramose specimen.

| Locality   |                         | Large oxeas | Small oxeas |
|--|-------------------------|-------------|-------------|
| U U  |                         | (µm)        | (µm)        |
| Average <i>Callyspongia ramosa</i><br>spicule dimensions from<br>specimens collected in New<br>Zealand (Bergquist & Warne,<br>1980). | x                       |             | 58.3 X 4.2  |
|  | Danaa                   |             | 52-63.8 X   |
|  | Kange                   |             | 2.5-6       |
| Spon00032, Salisbury Wharf,<br>Bay of Plenty, 5 m  | $\overline{\mathbf{X}}$ | 127 X 9.6   | 58 X 8.4    |
|  | Domas                   | 122-131 X   | 53-61 X     |
|  | Kange                   | 8.5-10.7    | 6.5-10.3    |
|  |                         |             |             |

 Table 2.46: Spicule dimensions of Callyspongia ramosa.

Callyspongia n. sp.1 cf. irregularis Spon00015

(Plates 18, C-F, 19, A-B).

| As Per Bergquist:              | As Per WoRMS 2015:      |
|--------------------------------|-------------------------|
| Restricted synonymy:           | Updated synonymy.       |
| (Bergquist & Warne 1980)       |                         |
| Class: Demospongiae            | Class: Demospongiae     |
| Order: Haplosclerida           | Order: Haplosclerida    |
|                                | Suborder: Haplosclerina |
| Family: Callyspongiidae        | Family: Callyspongiidae |
| Genus: Callyspongia            | Genus: Callyspongia     |
| Species: n.sp. cf. irregularis | Species: Unknown        |

Material examined: Spon00015, Karewa Island, Bay of Plenty, 10.8 m.

**Description**: This is a sausage shaped specimen with a linear row of oscules along its single branch. There is no *in situ* photograph available for this specimen.

**Dimensions**: Length 4 cm; width 1.5 cm; with fistules 1.5 cm in height.

Colour: In life purple-blue purple 10.0PB6/8, in spirit yellow 5.0Y8/4.

**Texture**: Soft, spongy and difficult to tear.

**Surface**: The surface is similar to the of *Callyspongia ramosa* with a smooth, but slightly uneven surface with oscules flush with the surface and aligned in a linear series along the branch (Plate 18, C).

**Skeleton**: The main skeleton has rectangular multispicular reticulation (Plate 18, E). There are no unispicular secondary tracts within this specimen and spongin is scarce. There is no clear distinction between primary and secondary tracts. The rectangular reticulation often becomes disorientated with no clear shape (Plate 18, F). Many isolated spicules often occur throughout the endosome.

## Spicules:

Megascleres: Oxeas- short and stout.

There were some styles present in the spicule mount, however, these were found in a low abundance and were consequently classified as foreign.

For spicule dimensions see Table 2.47.

**Remarks**: The colour is not quite correct and it has a smooth surface rather than a roughened surface. There were two strongyles present, however, these were believed to be foreign.

| Localit             | y                |     |    |                         | Oxeas<br>(µm) |
|---------------------|------------------|-----|----|-------------------------|---------------|
| pon000              | 15,              |     |    |                         |               |
| Karewa<br>Plenty,10 | Island,<br>.8 m. | Bay | of | $\overline{\mathbf{x}}$ | 97 X 5.1      |
|                     |                  |     |    | D                       | 89-106 X      |
|                     |                  |     |    | Range                   | 3.8-6.4       |

Table 2.47: Spicule dimensions of Callyspongia n. sp.1 cf. irregularis

# **ORDER NEPHELIOSPONGIDA**

Definition: Found in Bergquist & Warne, 1980, p. 34. Note: Has been updated to the suborder *Petrosina* in Hooper & Van Soest (2002).

## FAMILY NEPHELIOSPONGIIDAE

Definition: Found in Bergquist & Warne, 1980, p. 35.

#### <u>Genus Xestspongia</u>

Definition: Found in Bergquist & Warne, 1980, p.36.

| Xestospongia n.sp. 1       | (Plates 6, B-F, 7, A-F, 8, A-C). |
|----------------------------|----------------------------------|
| Spon00006                  |                                  |
| As Per Bergquist:          | As PER WORMS 2015:               |
| Restricted synonymy:       | Updated synonymy.                |
| (Bergquist & Warne 1980)   |                                  |
| Class: Demospongiae        | Class: Demospongiae              |
| Order: Nepheliospongida    | Order: Haplosclerida             |
|                            | Suborder: Petrosina              |
| Family: Nepheliospongiidae | Family: Petrosiidae              |
| Genus: Xestospongia        | Genus: Xestospongia              |
| Species: n.sp.             | Species: Unknown                 |

Material examined: Spon00006, Pilot Bay, Tauranga Harbour, 10.6 m.

**Description**: Thick encrusting sponge on a calcified shell (formerly encrusted with *Corallina* sp.) which it has incorporated into its structure on a sand flat in Pilot Bay. The sponge is friable and there is a slight sheen on the surface when dry through light bouncing off a flat layer of dermal spicules. It has an almost silvery shine to it. It has a thin dermis that is like developed skin and is tissue like when wet.

**Dimensions**: Length 3.5 cm; width 2 cm; thickness1 cm.

Colour: In life, colour is pale yellow2.5Y8/8; and is 2.5Y8/4 in spirit.

Texture: The specimen was stable, friable and easily compressed.

**Surface**: The surface was lumpy and slightly raised. Oscula are visible through the transparent dermal surface and lay flush with the surface (Plate 7, A).
**Skeleton**: The choanosomal skeleton has an alveolate/isotropic structure. Oxeas are tangentially echinating from fibres surrounding the choanosomal cavities (Plate 7, A, B). The ectosomal skeleton is characterised by frequent small openings, and spicules are arranged parallel to the surface in a tangential manner (Plate 7, C).

Spicules: (Plates 7, 8, D, E).

Megasceleres: Oxeas- three size classes of hastate oxeas were present, with thick shafts towards the proximal section. The oxeas were also evenly tapered towards each end as per the description of oxeas found in *Adocia parietaliodes* (Bergquist 1980).

For spicule dimensions see Table 2.48.

**Remarks**: Originally I classified this species within the *Adociidae* family based on the appearance of isodictyal reticulation within the skeletal structure. However, it was later realised that the specimen had no special dermal skeleton and only oxeas as spicules. Furthermore, there did not appear to be isodictyal reticulation, rather there was a reticulation that was often obscured by isolated spicules and the pattern was hard to discern. The oscula are scattered over the surface which is similar to that found within *Xestospongia novaezealandiae* (Bergquist & Warne, 1980).

| Locality             |                         | Large oxeas<br>(µm) | Medium<br>oxeas<br>(µm) | Small oxeas<br>(µm) |
|----------------------|-------------------------|---------------------|-------------------------|---------------------|
| Spon00006,           |                         |                     |                         |                     |
| Pilot Bay, Tauranga, | $\overline{\mathbf{X}}$ | 606 X 11.6          | 356 X 7.5               | 254 X 5.4           |
| 10.6 m               |                         |                     |                         |                     |
|                      | - D                     | 690-14.8 X          | 320-393 X               | 166-292 X           |
|                      | Range                   | 777-18.8            | 4.6-9.1                 | 2.3-8.4             |

 Table 2.48: Spicule dimensions Xestospongia n. sp. 1

Nepheliospongiidae n. sp. 1 Spon00014 (Plate 16, B-F, 17, A-F, 18, A-B).

| As Per Bergquist:          | As PER WORMS 2015:   |
|----------------------------|----------------------|
| Restricted synonymy:       | Updated synonymy.    |
| (Bergquist & Warne 1980)   |                      |
| Class: Demospongiae        | Class: Demospongiae  |
| Order: Nepheliospongida    | Order: Haplosclerida |
| Family: Nepheliospongiidae | Suborder: Petrosina  |
| Genus: cf. Petrosia        | Family: Petrosiidae  |
| Species: n.sp.             | Genus: Unknown       |
|                            | Species: Unknown     |

Material examined: Spon00014, Karewa Island, Bay of Plenty, 10.8 m.

**Description**: Spherical to massive sponge with oscula lying flush with the dermis. Oscula are scattered over the surface of the sponge and are approximately 0.5 cm in height.

**Dimensions**: Length 3 cm; width 1.5 cm. *In situ* specimens over 10 cm in diameter by 5 cm thick have been observed.

**Colour**: In life white to yellow-red-yellow 10.0YR8/4. In spirit, white to yellow 5.0Y8/4.

**Texture**: Brittle and stony.

**Surface**: The oscules are flush with the surface. There is a transparent dermal layer with oscules 1-4 mm scattered haphazardly over the dermis (Plate 16, C).

**Skeleton**: The skeletal arrangement is isotropic and becomes isodictyal with an increase in the number of spicules (Plates 16, F, 17, A, B).

Spicules:

Megascleres: Oxeas- two size classes of thick oxeas (Plate 17, F). Styles- slightly curved.

For spicule dimensions see Table 2.49.

**Remarks**: It is believed to be a new genus as it has all of the characteristics of the family Nepheliospongiidae with a dominance of mineral skeleton and isotropic reticulation. However, this specimen did not fit the description of any of the

genera within this family for New Zealand as it had oxeas with stylote ends, which in this instance were classified as styles.

| T 194                  |                         | Oxeas     | Styles    |
|------------------------|-------------------------|-----------|-----------|
| Locanty                |                         | (µm)      | (µm)      |
| Spon00014,             |                         |           |           |
| Karewa Island,         | $\overline{\mathbf{X}}$ | 189 X 6.2 | 211 X 13  |
| Bay of Plenty, 10.8 m. |                         |           |           |
|                        | D                       | 116-213 X | 119-230 X |
|                        | Kange                   | 4.2-7.1   | 13-15     |

 Table 2.49: Spicule dimensions of Nepheliospongiidae sp.

# **ORDER DICTYOCERATIDA**

Definition: Found in Bergquist 1980, p. 451.

| Dictyoceratida n.sp.  | (Plates 25, C-F, 26, A-D). |
|-----------------------|----------------------------|
| Spon00021             |                            |
| As Per Bergquist:     | As Per WoRMS 2015:         |
| Restricted synonymy:  | Updated synonymy.          |
| (Bergquist 1980)      |                            |
| Class: Demospongiae   | Class: Demospongiae        |
| Order: Dictyoceratida | Order: Dictyoceratida      |
| Family: Unknown       | Family: Unknown            |
| Genus: Unknown        | Genus: Unknown             |
| Species: Unknown      | Species: Unknown           |

Material examined: Spon00021, Karewa Island, Bay of Plenty, 10.8 m.

**Description**: The sponge is a spherical to thickly incrusting shape with elevated oscules and a surface covered in small regularly occurring mammilate projections. The sponge was found in a small cave at a sheltered site near Karewa Island. This specimen was the only individual of this species at this site.

Dimensions: Length 4 cm, width 2 cm, thickness 0.5 cm.

Colour: In life pale white, in spirit pale white to yellow-red 5.0YR6/10.

Texture: Soft, compressible and elastic.

**Surface**: The surface is mammilated with relatively small protrusions on the exterior. This specimen has elevated broad conical shaped oscules interspersed with flattened areas of pinacoderm (Plate 25, C, D).

**Skeleton**: The skeleton is composed of a complex anastomosing network of spongin fibres that are interconnected (Plate 25, F). Megascleres are interspersed within the interstitial regions of the networks of fibres.

#### Chapter 2

#### **Spicules:**

Megascleres: Foreign. Tylostyles- normal form. Acanthostyles- spinned styles, with no verticillate spines (Plate 26, B). Oxeas- two size classes, one was small curved, and the other was larger and straight. Styles- curved styles of the same size class.

For spicule dimensions see Table 2.50.

**Remarks**: This specimen is a dictyoceratid with a variety of different spicules (all foreign). It did not fit into the familial level of classification as it was dissimilar to all of the other families described within the Dictyoceratids (Bergquist, 1980). After preservation in spirit this specimen had a distinct pale external colour and was light brown internally.

| Locality       | Tylostyles |           | A conthectulor (um) | Large oxea | Small oxeas | Styles    |
|----------------|------------|-----------|---------------------|------------|-------------|-----------|
| Locality       |            | (µm)      | Acanthostyles (µm)  | (µm)       | (µm)        | (µm)      |
| Spon00021,     |            |           |                     |            |             |           |
| Karewa Island, |            | 101 V 2 7 | 60 V 5              | 157 V 2 2  | 62 V 4 7    | 102 V / 9 |
| Bay of Plenty, | Х          | 121 A 3.7 | 09 A 3              | 157 X 3.2  | 03 A 4./    | 102 A 4.8 |
| 10.8 m         |            |           |                     |            |             |           |
|                | D          | 114-129 X | 61-87 X             | 81-253 X   | 54-88 X     | 89-114 X  |
|                | Kange      | 2.8-4.5   | 3.1-5.8             | 1.1-4.6    | 2.4-6.9     | 2.8-7     |

 Table 2.50: Spicule dimensions of Dictyoceratida sp.

## FAMILY DYSIDEIDAE

Definition: Found in Bergquist 1980, p. 481.

#### Genus Dysidea

Definition: Found in Bergquist 1980, p. 481.

| Dysideidae n.sp.      | (Plates 29, D-F, 30, A-C). |
|-----------------------|----------------------------|
| Spon00026             |                            |
| As Per Bergquist:     | As PER WORMS 2015:         |
| Restricted synonymy:  | Updated synonymy.          |
| (Bergquist 1980)      |                            |
| Class: Demospongiae   | Class: Demospongiae        |
| Order: Dictyoceratida | Order: Dictyoceratida      |
| Family: Dysideidae    | Family: Dysideidae         |
| Genus: cf. Dysidea    | Genus: Unknown             |
| Species: n.sp.        | Species: Unknown           |

Material examined: Spon00026, Salisbury Wharf, Tauranga Harbour, 5 m.

**Description**: The sponge is a massive to amorphous specimen with dermal ridges covered in larger conulose projections with oscules situated at the apex of the projections. The sponge was found encrusting subtidally at the bottom of a wooden wharf pile (Plate 29, D).

**Dimensions**: Length 12 cm; width 9 cm; thickness 4 cm.

**Colour**: In life yellow 5.0Y8/4 to white, in spirit yellow 5.0Y4/4 to yellow 5.0Y2/2.

Texture: Soft and easily torn.

**Surface**: The surface is composed of both large broad slightly conulose ridges with oscules at the top of these projections (Plate 29, D-F). There are many smaller macroscopical projections covering the entire dermis. *In situ* these raised projections appear paler in colour than the rest of the dermis.

**Skeleton**: The skeletal structure of this specimen was damaged by a heavy infestation of polychaete worms (Plate 30, A, B); consequently the skeletal structure could not accurately be described. However, there was a large amount of sand grains present, interspersed among megascleres which appeared to be foreign.

#### **Spicules:**

Megascleres: Foreign. Oxeas- one size class slightly curved.

For spicule dimensions see Table 2.51.

**Remarks**: The sponge contained a variety of spicule types which is congruent with the Dictyoceratid order. It was also relatively easy to tear, and had small conulose projections raised up by the underlying skeleton.

| Locality          |                         | Oxeas<br>(µm) |
|-------------------|-------------------------|---------------|
| Spon00026,        |                         |               |
| Salisbury Wharf,  | $\overline{\mathbf{X}}$ | 261 X 7.8     |
| Bay of Plenty, 5m |                         |               |
|                   | D                       | 141-44 X      |
|                   | Range                   | 5.9-9         |

 Table 2.51: Spicule dimensions of Dysideidae sp.

## **ORDER EPIPOLASIDA**

Definition: The Epipolasida order is currently not accepted on the WoRMS database. This order was changed into a subclass of Demospongiae (Tetractinomorpha); however, this subclass is also currently unaccepted; therefore there are no definitions available for these two taxa.

## FAMILY THYIDAE

Definition: Found in Hooper & Van Soest 2002, p. 245).

#### Genus Tethya

Definition: Found in Bergquist, 1968, p. 35.

*Tethya aurantium* (Topsent 1900b) Spon00023 (Plates 27, D-F, 28, A-E).

| As Per Bergquist:    | As Per WoRMS 2015:  |  |
|----------------------|---------------------|--|
| Restricted synonymy: | Updated synonymy.   |  |
| (Bergquist 1968).    |                     |  |
| Class: Demospongiae  | Class: Demospongiae |  |
| Order: Epipolasida   | Order: Hadromerida  |  |
| Family: Tethyidae    | Family: Tethyidae   |  |
| Genus: Tethya        | Genus: Tethya       |  |
| Species: aurantium   | Species: aurantium  |  |

Material examined: Spon00023, Karewa Island, Bay of Plenty, 10.8 m.

**Description**: The sponge was globular with basal rooting processes *in situ*. This sponge is one of the most common sponge species found in New Zealand. This specimen was found sub-tidally encrusting on a rock face on a relatively exposed site.

Dimensions: Length 3.5 cm; width 2.5 cm; thickness 1.7 cm.

Colour: In life yellow-red 5.0YR7/10, in spirit yellow-red 5.0YR8/4.

Texture: The pinacoderm is firm, but the endosome is slightly compressible.

**Surface**: The surface has small mammilate polygonal projections, and is slightly smooth in some parts of the dermis (Plate 28, B).

**Skeleton**: The choanosomal skeleton is composed of thick compact bundles of strongyloxeas, which are aligned perpendicular to the surface and radiate out from the centre to the dermis (Plate 28, A). There is also a separate cortex in between each row of aligned megascleres composed of microspherasters and tylasters. Both of the microsclere types are abundant in the endosome.

#### **Spicules:**

Megascleres: Strongyloxeas- of normal form. Tylasters- of normal form. Microscleres: Microxyspheraster (spherasters) - of normal form. For spicule dimensions see Table 2.52.

| Locality  |       | Strongyloxea<br>(µm) | Microspheraster<br>(spherasters)<br>(µm) | Tylasters<br>(µm) |
|---|-------|----------------------|--|-------------------|
| AverageTethyaaurantiumspiculedimensionsfromspecimenscollectedinNewZealand(Bergquist & Fromont, 1988). | x     |                      |  |                   |
|   | Range |                      |  |                   |
| Spon00023,<br>Karewa Island,<br>Bay of Plenty,<br>10.8 m  | x     | 518 X 11             | 61                                       | 13                |
| 10.0 m  | Range | 330-943 X<br>9-13    | 46-69                                    | 11.3-14           |

 Table 2.52: Spicule dimensions of Tethya aurantium.

## 2.4 Discussion

Internationally the Porifera remain a highly under-represented taxa in new marine research programs, and hence the potential for novel species discoveries is high (Goodwin et al. 2012). Specifically, only 8608 sponge species have currently been described, although global diversity of sponges is expected to increase to 12,000 valid species by the end of the century (Van Soest et al. 2012). Approximately 730 sponge species have either been described from New Zealand waters or are in the process of being described (Battershill et al., 2010). At the beginning of this study, Demospongiae biodiversity characteristics within the Bay of Plenty were relatively unknown, with the majority of previous sponge research occurring throughout other regions of New Zealand (Bergquist 1970; Bergquist 1980; Bergquist & Warne 1980; Bergquist & Fromont 1988; Kelly-Borges & Bergquist 1997; Alvarez et al. 2002). No focused biosystematics work on the Porifera has ever been done in the Bay of Plenty. This is the first survey of sponge biodiversity within this region and has identified an exceptionally large diversity of sponge fauna, even though collections were limited to a relatively small geographic focal area within the Tauranga Harbour and Karewa Island on the immediate outer coast.

Fifty five species are described in this research. Of these, there are up to three new families, three new genera and thirty four species which are undescribed and deemed new to science. However, a more conservative estimate with grouped specimens suggests that there is a minimum of at least one new family, one new genera, and eighteen new species that are undescribed and deemed new to science. As such, 36% of all specimens collected are undescribed. The sponges identified had affinities mostly with northern warm temperate even subtropical fauna (Battershill et al 2010), although a few (*Forcepia* like species) indicated the influence of southern oceanic inflow events into the Bay of Plenty.

This suggests a high level of sponge diversity within northern New Zealand waters. Reasons for this high level of biodiversity are due to the diverse area studied, with many different habitats together with a complex oceanography including cold temperate and warm temperate intrusions into the Bay of Plenty (Ridgway & Greig 1986). Furthermore, many sponges collected in this study were thinly encrusting on bedrock, often overlooked or not sampled by other means.

Studies have reported that encrusting sponge biodiversity remains largely undescribed elsewhere also, due to remote sampling methods such as dredging and trawling, even in well frequented areas such as the United Kingdom. (Picton & Goodwin 2007; Goodwin *et al.* 2011; Goodwin *et al.* 2012).

Sponge taxonomic description began in the early 19<sup>th</sup> century, but little emphasis was placed on relationships or kinships within this phylum, apart from creating primary divisions based on the chemical composition of the skeleton (Bowerbank 1864; Bowerbank 1886). Bowerbank's classifications were elaborated by Gray (1867) and established a large number of new families, particularly the Poecilosclerida. Bowerbank was among the first researchers to use spicule characters to form diagnostic tools for nominating sponges into orders, genera and species (Fromont 1985).

Sponge systematics has traditionally been based almost completely on skeletal traits, such as skeletal mineral elements (spicules) (Erpenbeck & Wörheide 2007). Fromont (1990) found that the most useful morphological character for separating families belonging to the Haplosclerida order was the organisation of the internal skeleton, its spicule composition and their quantities. These characters allowed separation of sponges into familial and species level classifications (Bergquist & Warne 1980; Fromont 1990). Similarly, within this current study the morphology and composition of microscleres (e.g. palmate aniochelae) were used as the main morphological character to separate new species belonging to the *Carmia* genus.

Morphological delineation of sponge species is hindered by the lack of fixed diagnostic morphological characters and our limited knowledge of phenotypic plasticity among Porifera species (Bergquist & Warne 1980; Andreakis *et al.* 2012). For example, Bergquist and Warne (1980) found some Haplosclerida species were influenced by an ecophenotypic response to environmental influences such as wave exposure. Sponge body form is extremely variable and can be influenced by accessible space, water current velocity, habitat, and the nature and slope of substratum (Erpenbeck & Wörheide 2007; Battershill *et al.* 2010) Similarly, sponges within this study had varying phenotypic characteristics within and among species. As an example, unknown *Haliclona* species had analogous oxeas, but the arrangement of the skeletal structure changed, depending on the environment that they were collected from (e.g. wave exposed versus

sheltered habitat). The task is to determine where what is an intrinsic skeletal feature and what has been influenced by the micro-environment. The use of cytological features for sponges systematics have been examined in the past (Boury-Esnault *et al.* 1994), although these characters were suggested as inadequate methods to address broader phylogenetic questions within the Porifera (Erpenbeck & Wörheide 2007).

The major difficulties with traditional sponge systematics is the interpretation of homology, whereby some prominent characters that appear similar are in fact analogous features that do not necessarily reflect phylogenetic relationships, or translate into morphological identification (Hooper & Van Soest 2002). There is still a long way to go given the plastic morphology of sponges and variable spicule elements. Although there is some information regarding the reproductive seasons of marine sponge, little is known about their reproductive efforts and patterns of structural and histological change. Consequently, little is known about how fast sponges speciate (Fromont 1990; Ereskovsky 2000). A mixture of both sexual and asexual reproduction methods blurs the edges of sponge species.

Hebert et al. (2003a) suggested that species identification based solely on traditional (alpha) taxonomy has four major limitations. First, both phenotypic plasticity and genetic variability in the characters nominated for species recognition can lead to misidentifications. Second, this approach often overlooks morphologically cryptic species, which are common among sponges (Xavier et al. 2010; Andreakis et al. 2012; de Paula et al. 2012). Third, since morphological data is often only available for adult taxa, many individuals cannot be identified. Finally, the use of updated morphological keys often requires such a high level of expertise that misdiagnoses are common (Hebert et al. 2003a). Nevertheless, past and present sponge taxonomic work has contributed substantially to advancing knowledge on sponge morphologies, their structural, physiological and biochemical mechanisms (Hooper & Van Soest 2002). Classical taxonomic work has also contributed to answering fundamental questions such as understanding biosynthesis of chemicals, evolution of eukaryotic immunology, cellular theory and totipotency, gene function of the metazoan and the extent of marine biodiversity (Hooper & Van Soest 2002).

Classical (alpha) taxonomic descriptions are extremely valuable as they provide basic information regarding phenotypic, ecological, behavioural and evolutionary explanations of data which is gathered from molecular analyses (Decraemer & Backeljau 2015). It is important to track the changes of species names over the process of modernisation of taxonomy. For example, biogeographic studies will require taxonomic descriptions based on morphology to back-track species comparisons. At the present time, sponge systematics is facing novel challenges with the introduction of new molecular based technologies (Cardenas et al. 2012). Over the past decade classical (alpha) taxonomy has been completely overturned; consequently a review of the state of the art has been suggested among taxonomists (Cárdenas et al. 2012). In comparison to other phyla, phylogenetic relationships within Porifera are still largely unresolved (McCormack et al. 2002; Cárdenas et al. 2012). DNA barcoding has become increasingly attractive as a method of species identification in terms of cost, speed and objectivity (von Crautlein et al. 2011). DNA barcodes provide clear and comparable analyses that can be repeated by anybody, even untrained taxonomists. Additionally, unlike classical taxonomy DNA barcoding can analyse fragmented samples within all the life stages of organisms (von Crautlein et al. 2011). Nevertheless, the primary goal of DNA barcoding is to identify an unknown specimen in terms of known classification (Miller 2007; Stoeckle 2008) and to complement and not supersede or replace existing taxonomic practices based on morphological characters.

The primary limitation of this research is the lack of chemical work, which could extend the study from classical and molecular comparisons to include chemotaxonomic assessment. This was beyond the scope of the project time-wise. I suggest that further work be undertaken using biochemical, ecological and reproductive techniques to further understand the phylogeny and affinities of sponges in New Zealand.

Biological compounds were suggested as alternative methods to morphological identification in sponge systematics with the pioneering chemosystematics work undertaken by work by Bergman (Bergman 1949; Bergman 1962) followed by Bergquist (1978) who elaborated on this work. Although publications over the past decade have decreased, due to apparent difficulties with the identification of the actual producers of the secondary metabolites (sponges or symbionts), homologization of pathways and experimental difficulties. Interestingly,

chemosystematic work has led to the discovery and development of marine natural compound from sponges. Specifically, novel secondary metabolites such as Peteamine and Peluroside A have been isolated from the New Zealand *Carmia* species (Northcote *et al.* 1991; Page *et al.* 2005a; Page *et al.* 2005b; Page *et al.* 2011). In fact, the phylum Porifera have the highest discovery rates of marine natural products among all marine and terrestrial phyla (Munro *et al.* 1999). Future chemical ecology and chemosystematic work on sponges may lead to the revival of a chemosystematics approach both in New Zealand and internationally.

## **Chapter 3**

## **MOLECULAR SYSTEMATICS**

## 3.1 Introduction

The ability to accurately identify species is prerequisite for assessing levels of biological diversity and a fundamental requirement for ecological research (Hogg & Hebert 2004; Hogg et al. 2009). In recent times, there has been a shortfall in biologists who practice traditional alpha taxonomy, leading to difficulties in assessment of biodiversity in some taxonomic groups. For example, in sponges, systematics has traditionally been based mostly on skeletal structure and spicule geometry (McCormack et al. 2002). In particular, the dimensions of structural spicules (megascleres) and smaller packing spicules (microscleres) have been used as important taxonomic characters of sponge classification (Sollas 1888; Lendenfeld 1889). Sponge identification is generally perceived as appallingly difficult as taxonomists are faced with great morphological variability among species which are prone to hoopla's, and often have few macroscopical characters to work with (Bergquist & Warne 1980; Hooper & Van Soest 2002; Worheide et al. 2004). Accordingly, there is some concern that our planets sponge biodiversity is vanishing faster than our accumulated mass of taxonomic expertise can catalogue it (Bucklin et al. 2011).

The use of a molecular DNA barcoding approach has been suggested as a tool that can be used to complement and accelerate traditional alpha taxonomy, without supplanting or invalidating existing taxonomic practices (Bucklin *et al.* 2011). The mitochondrial DNA cytochrome *c* oxidase subunit (COI) gene locus has become the marker of choice as for many taxonomic groups as an effective identification tool (Hebert *et al.* 2003a). Extensive research has been undertaken using the mitochondrial gene in phylogenetic research (Desalle *et al.* 1987; Adachi *et al.* 1993; Oliveira *et al.* 2008; Schaffer *et al.* 2010; Zhang *et al.* 2011). However, further work is required to validate the use of COI for species level identifications (Meier *et al.* 2006; Elias *et al.* 2007; Hogg *et al.* 2009)

The use of the COI gene for species identification within the phylum Porifera has been used in the past (Schroder *et al.* 2003; Huang *et al.* 2008b; Lavrov *et al.* 

2008; Cardenas *et al.* 2010). However, there remains important gaps in the knowledge of the Demospongiae with species level identifications difficult to achieve (Hooper & Van Soest 2002). A molecular identification tool would assist in providing adequate species identifications which could be used in combination with traditional taxonomy to construct a comprehensive reference library.

Here, we focus on the class Demospongiae which is both ecologically and taxonomically diverse and includes 85% of recent Porifera species (Hooper & Van Soest 2002). Obtaining DNA reference sequences from taxa within the Bay of Plenty will provide a platform from which more widespread sampling can be undertaken. Additionally, an accelerated identification processes would assist targeted research on Demospongiae species with secondary metabolites which are of commercial interest to the pharmaceutical and agrichemical industries (Bergman 1951; Ireland *et al.* 1993; Munro *et al.* 1999; Duckworth & Battershill 2003).

The aim of this chapter is to determine whether identifications based on genetic barcoding are congruent with those produced via traditional morphological methods (alpha taxonomy), and to assess the use of molecular techniques for Demospongiae species identifications. Phylogenetic analyses using different reconstruction methods are also used to determine whether COI can infer phylogeny at deeper taxonomic levels.

### **3.2 Methods**

#### **3.2.1** Study sites and sample collection:

Sponges were collected from four sites in the Bay of Plenty, three within the Tauranga Harbour (Bridge Marina, Salisbury Wharf, Pilot Bay) and one on the open coast (Karewa Island; Fig. 3.1). Scuba diving was used at each site for collection of sponges. The selected sponges were photographed and sections cut from whole individuals using a sharp scalpel to allow for their continued regrowth and survival. Following collection, specimens were frozen and preserved in 90% ethanol and stored at the University of Waikato's Coastal Marine Field Station Laboratory (Hooper 2000). A total of 95 specimens of sponges were analysed. All specimens were taxonomically identified based on morphological references

(Bergquist 1961; Bergquist 1968; Bergquist 1970; Bergquist 1980; Bergquist & Warne 1980; Bergquist & Fromont 1988; Kelly-Borges & Bergquist 1997). For each specimen a 3 mm section of pinacoderm tissue was removed for DNA extraction (Harrison 1972). Each sample was placed in an individual well of a 96 well microplate. Samples were then sent to the Canadian Centre for DNA Barcoding (CCDB) at the University of Guelph for genetic analyses.

### **3.2.2 Genetic analysis**

Total genomic DNA was extracted using a Glass Fibre Plate DNA Extraction (AcroPrep) method (Ivanova et al. 2006) at CCDB. Polymerase Chain Reactions (PCRs) carried out at CCDB were comprised of a 10.5 µl reaction containing 2 µl ddH20, 6.25 µl 10% trehalose, 1.25 µl 10X buffer, 0.625 µl 50 mM MgCl2,0.0625 μl 10 mM dNTPs, 0.06 μl Polymerase (5 U/μl), 0.125 μl 10 μM primer A, 0.125 μl 10 μM primer B and 2 μl of template DNA per well (Ivanova & Grainger 2015). A 658 bp fragment of the mitochondrial CO1-5P gene was amplified using 1.0mM concentrations of the primers dgLCO-1490 (sequence 5'-GGTCAACAAATCATAAAGAYATYGG-3') and dgHCO-2198 (sequence 5'-TAAACTTCAGGGTGACCAAARAAYCA-3') (Meyer 2003). PCR thermocyler conditions included preliminary denaturing at 94°C for 30 sec, followed by annealing at 45-50°C for 40 sec, and extension at 72°C for 1 min, followed by 30-35 cycles of 94°C for 30 sec, 51-54°C for 40 sec, and 72°C for 1 min, with a finishing extension at 72°C for 10 min, followed by indefinite hold at 4°C.

PCR products were then sequenced in both directions using the same primers as the PCR. All sequences were aligned and visually inspected using MUSCLE, in Geneious v.6.1.2 (Drummond et al. 2011). Specimens were verified as being Porifera using the Barcode of Life Data Systems (BOLD) identification engine, and a minimum of 500 bp to determine the most likely match. All primer sequences were removed from sequence fragments before further analysis. All sequences were uploaded and available on the BOLD database (www.boldsystems.org) under project Sponges of New Zealand (NZSPG).

### 3.2.3 Phylogenetic analysis

A chi-square test was used, as executed in PAUP\* 4.0b10 (Swofford 2002), to determine whether the hypothesis of equal base frequencies among sequences was violated on all sites; parsimony-informative sites only; and with the third codon position only. A Maximum Likelihood phylogram was constructed with the most appropriate substitution model determined using JModel test 2.1.1 (Darriba et al. 2012). The model employed was GTR + I + G (-1nL = 5, 144.667) with A = 0.3083, C = 0.0894, G0.2065, T = 0.3957. The settings were: 11 substitution schemes (88 models), base frequencies +F, rate variation +I, +G, set to BioNJ. A heuristic search function was used with an as-is addition sequence. Searches were conducted in MEGA ver 5.2.2 (Tamura et al. 2011). Neighbour Joining trees were also constructed to visualise sequences for all specimens. Duplicates were retained to show which haplotypes were repeatedly being successfully sequenced. An available COI sequence from a member of the Clathrinida order was utilised as an outgroup taxon (BOLD accession number NC\_02113) in both analyses. Non-parametric bootstrap analyses with 1000 pseudoreplicates (Felsenstein 1981) were also used on ML and NJ trees to assess support for nodes on the trees. Other settings were set to default in MEGA. NJ and ML trees were overlaid with species names based on morphological identification. Binomial names obtained from morphological identifications were updated to currently accepted binomial nomenclature using the WoRMS Database (World Register of Marine Species 2015).

Pair-wise genetic distances between all specimens were also calculated in MEGA ver 5.2.2. This analysis was conducted using the Jukes-Cantor model (Jukes & Cantor 1969). *Cliona celata* (NZSPG031) had a relatively short sequence at 462 bp; consequently, this sequence was removed from the analysis. Two additional *C. celata* specimens were 575 bp long; consequently, all sequences were trimmed at both ends, resulting in 575 bp (191) codons, with reads in both directions.



Figure 3.1: Map of the Bay of Plenty, New Zealand, showing SCUBA diving sampling locations.

### **3.3 Results**

There was a 34% sequence success rate with 32 of the 95 samples successfully sequenced. For those successfully sequenced, a 575 bp fragment of COI gene was recovered from 32 Demospongiae specimens. No insertion, deletion or stop codon was detected and sequences were unambiguous. Of the 575 bp fragment used, 280 characters were constant, 23 variable but uninformative and 272 parsimony informative. The average nucleotide composition for all taxa (excluding outgroup) showed an A-T bias 63.5% (A=24.9%, C=14.9%, G=21%, T=38.6%), and base frequencies were homogenous across all sites ( $X^{2}_{96}$  =86.5, p=0.74). However, as base frequencies were not homogenous for parsimony informative sites (A-T =68.3%,  $X^{2}_{96}$  =181.1, p=<0.001) nor third codon positions (A-T=74.68%,  $X^{2}_{96}$ =219.8, p<0.001), homology of base pairs was rejected. Thirty two species were represented by two or more specimens, thus allowing within species variation to be determined.

## **3.3.1 COI sequence divergence**

Thirty two species were represented by two or more individuals allowing an analysis of within species variation. Within each of the species examined there were no divergences between conspecific individuals (Table 1). In contrast, divergences among species exceeded 0.05% in all cases. The most divergent taxa were *Xestospongia* sp. (NZSPG006), which had a mean sequence divergence of 25% and *Polymastia fusca* (NZSPG019), which had a mean sequence divergence of 22% from other taxa (Table. 2). Conspecific individuals in most instances had identical sequences and individuals of *Cliona celata* had identical sequences for different locations (e.g. Pilot Bay versus Karewa Island).

## 3.3.2 Phylogenetic analysis

Neighbour Joining (NJ) and Maximum Likelihood (ML) trees using morphological identifications are shown in Figures 3 and 5. NJ and ML trees using BOLD identifications are shown in Figures 2 and 4. Tree constructions for NJ and ML trees showed broadly similar topology and node support (Figures. 3.3 to 3.5). The trees estimated from both methods of analysis showed that families and genera, with the exception of species within the Petrosiidae and Chalinidae families (NJ, ML), were monophylectic (Figures. 3.3 to 3.5). Both analyses resulted in the same six morphologically identified taxonomic orders and a fifth unknown group, some of which did not agree with currently accepted families. The Haplosclerida order was polyphyletic containing a distinct clade of three families (Petrosiidae, Chalinidae, Callyspongiidae), and another monophyletic order (Dictyoceratida) including members of an unknown family. A third order (Poecilosclerida) consisted of four families (Desmacellidae, Tedaniidae, Crellidae, Mycalidae) and was polyphyletic and monophyletic in NJ and ML trees respectively (Figures 3.3 and 3.5). A fourth monophyletic order (Astrophorida) was composed of a single family (Ancorinidae). The fifth polyphyletic order (Hadromerida) consisted of species from four families (Clionidae, Tethyidae, unknown family, Polymastiidae). Interestingly bootstrap support between the fifth order (Hadromerida) and taxa belonging to a sixth unknown order had 99 and 100% bootstrap support in NJ and ML trees respectively (Figs. 3.3, 3.5).

These NJ and ML analyses showed that species in families represented by more than one taxon usually formed cohesive assemblages. Linking nodes between families within the Haplosclerida order were 62% and 77% in NJ and ML trees, respectively. However, some Haplosclerida species were associated with other genera (*Haliclona* sp.) and one species (*Xestospongia* sp.) was located externally to other members of this order (Fig. 3.3, 3.5). Linking nodes between families in the Hadromerida order were 77 and 94% in NJ and ML trees, respectively; however, one member of this order (*Polymastia fusca*) was located externally from the rest of these families within this order (Figs. 3.3, 3.5).

Both analyses (NJ and ML) yielded fairly congruent trees (Figures 3.3, 3.5). The ML tree (Fig. 3.5) was slightly better resolved than the NJ tree, especially with respect to resolving deeper ordinal level classifications. Species identifications using BOLD allowed identification of some unknown species. Specifically, species NZSPG023, NZSPG021, NZSPG089, NZSPG074 and NZSPG008 were unknown using morphological identification but were identified down to genera levels (Hymeniacidon, Halichondria) using BOLD identification. Similarly, unknown species (NZSPG086, NZSPG020) were both identified as Spongia species using BOLD. However, there was some incongruence between morphological and genetic identification methods. For example, one specimen morphologically identified as Xestospongia sp. (NZSPG006) was identified on BOLD as Scopalina ruetzleri (Figures. 3.2 to 3.5). Similarly, morphologically identified specimens of Haliclona (Gellius) fragilis (NZSPG078, NZSPG012) were identified as Petrosia species using BOLD identification. Three species (NZSPG105, NZSPG079 and NZSPG013) were morphologically separated into two families (Chalinidae, Petrosiidae) but were grouped into a single family (Chalinidae) based on their COI sequences (Figures. 3.2, 3.3, 3.4 and 3.5). Furthermore, morphologically identified Mycale species (NZSPG050, NZSPG102) were genetically identified as Chelon aplysilla erecta (Figs. 3.2 to 3.5). Finally, one specimen which was morphologically identified as *Desmacella* dendyi was identified as Dictyonella incisa from its COI sequence.

### **3.3.3** Non sequence trends

Of the 95 specimens examined, 32 were successfully sequenced. Four specimens matched sequences from foreign phyla (Phaeophyta, Herokotophyta, Haptophyta and Anellida) and a further 71 samples were unable to be sequenced. This was a reoccurring trend with only two of the 21 specimens belonging to the *Mycale* genus successfully sequenced. Moreover, only one of the 13 *Haliclona* spp. was able to be successfully sequenced. Three *Halichondria moorei* and two members of the *Ciocalypta* genus were not able to be successfully sequenced.

Table 3.1: Locality of collection, specimen numbers from the Barcode of Life Data Systems (BOLD), and field identification numbers for the sponge species used in this study.

| Species                      | COI       | Field ID  | Collection site |
|------------------------------|-----------|-----------|-----------------|
| Demospongiae sp.             | NZSPG023  | Spon00024 | Karewa Island   |
| Demospongiae sp.             | NZSPG021  | Spon00022 | Karewa Island   |
| Demospongiae sp.             | NZSPG089  | Spon00024 | Karewa Island   |
| Hadromerida sp.              | NZSPG074  | Spon00008 | Pilot Bay       |
| Hadromerida sp.              | NZSPG008  | Spon00008 | Pilot Bay       |
| Tethya aurantium             | NZSPG088  | Spon00023 | Karewa Island   |
| Cliona celata                | NZSPG002  | Spon00002 | Pilot Bay       |
| Cliona celata                | NZSPG084  | Spon00019 | Karewa Island   |
| Cliona celata                | NZSPG068  | Spon00002 | Pilot Bay       |
| Xestospongia sp.             | NZSPG006  | Spon00002 | Pilot Bay       |
| Polymastia fusca             | NZSPG019  | Spon00020 | Karewa Island   |
| Stelletta crater             | NZSPG076  | Spon00011 | Karewa Island   |
| Stelletta sandalinum         | NZSPG082  | Spon00017 | Karewa Island   |
| Stelletta maori              | NZSPG011  | Spon00012 | Karewa Island   |
| Stelletta maori              | NZSPG077  | Spon00012 | Karewa Island   |
| Stelletta maori              | NZSPG075  | Spon00010 | Karewa Island   |
| Stelletta maori              | NZSPG009  | Spon00010 | Karewa Island   |
| Haliclona (Gellius) fragilis | NZSPG078  | Spon00013 | Karewa Island   |
| Haliclona (Gellius) fragilis | NZSPG012  | Spon00013 | Karewa Island   |
| Callyspongia ramosa          | NZSPG029  | Spon00032 | Salisbury Wharf |
| Callyspongia sp.             | NZSPG080  | Spon00015 | Karewa Island   |
| Haliclona sp.                | NZSPG105  | Spon00063 | Bridge Marina   |
| Petrosiidae sp.              | NZSPG079  | Spon00014 | Karewa Island   |
| Petrosiidae sp.              | NZSPG013  | Spon00014 | Karewa Island   |
| Dictyoceratida sp.           | NZSPG086  | Spon00021 | Karewa Island   |
| Dictyoceratida sp.           | NZSPG020  | Spon00022 | Karewa Island   |
| <i>Mycale</i> sp.            | NZSPG050  | Spon00060 | Bridge Marina   |
| <i>Mycale</i> sp.            | NZSPG102  | Spon00060 | Bridge Marina   |
| Desmacella dendyi            | NZSPG010  | Spon00011 | Karewa Island   |
| Tedania battershilli         | NZSPG017  | Spon00018 | Karewa Island   |
| <i>Tedania</i> sp.           | NZSPG081  | Spon00016 | Karewa Island   |
| Tedania sp.                  | NZSPG015  | Spon00016 | Karewa Island   |
| Crella incrustans            | NZSPG003  | Spon00003 | Pilot Bay       |
| Outgroup                     |           |           |                 |
| Clathrinida sp.              | NC_021113 |           |                 |
|                              |           |           |                 |

#### Chapter 3

Table 3.2: Mean pairwise distances among Demospongiae taxa collected in the Bay of Plenty, New Zealand. For individuals within species, n = number of species which had two or more individuals; species within genus, n = number of genera with two or more species; species among genera, n = number of genera analysed within a family; species among families, n = total number of species analysed.

|                 | Inc | lividuals within | <u>S</u> | pecies within | <u>S</u> 1 | pecies among | Species among |            |  |  |
|-----------------|-----|------------------|----------|---------------|------------|--------------|---------------|------------|--|--|
|                 |     | species          |          | genus         |            | genera       | families      |            |  |  |
| Family          | n   | Divergence       | n        | Divergence    | n          | Divergence   | n             | Divergence |  |  |
| Petrosiidae     | 1   | 0.00             |          |               | 2          | 0.25         | 2             | 0.18       |  |  |
| Chalinidae      | 1   | 0.00             | 1        | 0.15          |            | 0.09         | 3             | 0.14       |  |  |
| Callyspongiidae |     |                  | 1        | 0.09          |            |              | 2             | 0.12       |  |  |
| Unknown         | 1   | 0.00             |          |               |            |              | 2             | 0.14       |  |  |
| Mycalidae       | 1   | 0.00             |          |               |            |              |               |            |  |  |
| Polymastiidae   |     |                  |          |               |            |              |               |            |  |  |
| Ancorinidae     | 1   | 0.00             | 1        | 0.10          |            |              | 3             | 0.14       |  |  |
| Clionidae       | 1   | 0.00             |          |               |            |              |               |            |  |  |
| Tethyidae       |     |                  |          |               |            |              |               |            |  |  |
| Hadromerida     | 1   | 0.00             |          |               |            |              |               |            |  |  |
| Unknown         | 1   | 0.00             |          |               |            |              |               |            |  |  |
| Desmacellidae   |     |                  |          |               |            |              |               |            |  |  |
| Tedaniidae      | 1   | 0.00             | 1        | 0.05          |            |              | 2             | 0.10       |  |  |
| Crellidae       |     |                  |          |               |            |              |               |            |  |  |

If desired, comparison can be made with the genetic distances within and among species using Table 3.3.



Figure 3.2 : Neighbour-joining tree for 32 specimens of Bay of Plenty sponges constructed using the Barcode of Life Database for identification (families are indicated on the right and orders are underlined). Numbers next to branches indicate percentage non parametric bootstrap support (>50%) from 1000 pseudoreplicates. Individuals which are genetically dissimilar to other specimens within the same taxonomic group are denoted by an Asterisk (\*)



Figure 3.3: Neighbour-joining tree for 32 specimens of Bay of Plenty sponges overlaid with morphological identifications (families are indicated on the right and orders are underlined). Numbers next to branches indicate percentage non parametric bootstrap support (>50%) from 1000 pseudoreplicates. Individuals which are genetically dissimilar to other specimens within the same taxonomic group are denoted by an Asterisk (\*).



Figure 3.3: Maximum likelihood tree for 32 specimens of Bay of Plenty sponges constructed using the Barcode of Life Database for identification (families are indicated on the right and orders are underlined). Numbers next to branches indicate percentage non parametric bootstrap support (>50%) from 1000 pseudoreplicates. Individuals which are genetically dissimilar to other specimens within the same taxonomic group are denoted by an Asterisk (\*).



Figure 3.4: Maximum likelihood tree for 32 specimens of Bay of Plenty sponges overlaid with morphological identifications (families are indicated on the right and orders are underlined). Numbers next to branches indicate percentage non parametric bootstrap support (>50%) from 1000 pseudoreplicates. Individuals which are genetically dissimilar to other specimens within the same taxonomic group are denoted by an Asterisk (\*).

## 3.4 Discussion

In all cases where sequences were able to be generated, it was possible to successfully distinguish known Demospongiae species using the COI gene. In most instances, COI sequences uncovered patterns of divergence among species that agreed with current alpha taxonomy which is based exclusively on morphology (Chapter 2). For the entire sequence data set, the A-T nucleotide bias was 63%, which is similar to that previously reported in Demospongiae (Lavrov et al. 2008; Wang & Lavrov 2008). In all cases, there was no sequence divergence within species, although within species replication was limited. Among species, divergences in all cases exceeded 0.05%. Similar levels of divergence have been reported elsewhere, with Schroder et al. (2003) recording identical sequences within species and only 1-2% different between species in different families of Demosponges. This suggests that low species divergence (<3%) is the rule within recognised Demospongiae species. High interspecific divergence was detected from a single Xestospongia sp. (NZSPG006), which had a mean sequence divergence of 25% from other taxa in the Petrosiidae family. This value is much higher than the mean interspecific divergence reported for Porifera (4.93%). Moreover, Polymastia fusca (NZSPG019) also had an exceptionally high divergence of 22% from other taxa within the Hadromerida order. It is likely that further study will reveal that both of these species are currently novel undescribed sibling species. Interestingly, two morphologically different specimens of *Cliona* spp. that were identified as possibly being new species based on their spicule composition (Chapter 2), were genetically identified as *Cliona celata* based on their COI sequences. Cryptic speciation has been previously reported within the allegedly cosmopolitan species of Cliona celata (Xavier et al. 2010; de Paula et al. 2012). Morphologically cryptic species are also well known among other Demospongiae species (Blanquer & Uriz 2008; Xavier et al. 2010; Andreakis et al. 2012; de Paula et al. 2012).

Congeneric species divergences were generally greater (range 0.05-0.15%; Table 2) than within species divergences. However, congeneric species divergences within this study (mean 0.09%) appear lower than those reported for all other animal taxa (Hebert *et al.* 2003b). Mitochondrial DNA data reinforces previous work that the COI molecular marker yielded a coherent and robust phylogeny for

separating closely related Demospongiae species, even if it could only provide resolution down to the genus level (Duran *et al.* 2004; Cardenas *et al.* 2010).

Neighbour Joining (NJ) and Maximum likelihood (ML) trees have different inherent assumptions, advantages and limitations. For example, NJ is not as robust as ML as it does not integrate a model of evolution and does not allow the input of allele frequencies. However, NJ does allow for the rapid assessment of a large dataset. If the two methods resolve similar relationships there seems little reason to not include NJ analyses with the inclusion of bootstrap values (Munch *et al.* 2008). The NJ and ML analyses constructed for Porifera within this study showed that species in families represented by more than one taxon usually formed congruent cohesive assemblages for both analyses.

ML and NJ analyses both grouped the 32 species and resulted in the same six morphologically identified taxonomic orders and a fifth unknown order, some of which did not agree with currently accepted families based on alpha taxonomy. The Haplosclerida order was polyphyletic in the phylogenetic trees but contained a distinct clade of three families (Petrosiidae, Chalinidae, Callyspongiidae). Interestingly, this tree structure supports previous work (McCormack *et al.* 2002; Nichols 2005) showing that taxonomy within the Haplosclerida is contentious with discrepancies to former phylogenies. McCormack (et al, 2002) also found that there was no support for the monophyly of the Haplosclerida and almost all taxa (families, genera) within this order were polyphyletic, although a different gene (28S rRNA) was used. In fact, McCormack (*et al.*, 2002) found that *Haliclona, Callyspongia*, and *Xestospongia* were quite divergent. Their results support the findings of this study with *Xestospongia* sp. (NZSPG006) having a mean sequence divergence of 25% from other taxa within the Haplosclerida order.

While the Haplosclerida is well defined morphologically, it has been previously split into two orders Nepheliospongidae and Haplosclerida (Bergquist & Warne 1980). This suggests that the current placement of species within the Haplosclerida may need reconsideration. I caution, however, that this assumption is based on a limited number of Haplosclerida specimens (n = 8), and a single gene (COI). Furthermore, a larger number of taxa should be studied with both COI and 28S rRNA genes to gain a better understanding of these divergences.

Another monophyletic order (Dictyoceratida) contained one previously undescribed species (Figures 3.3, 3.5). Previous work (Nichols 2005) on the Dictyoceratida found that this order was largely reconstructed as monophyletic. A third order (Poecilosclerida) was polyphyletic and monophyletic in NJ and ML trees respectively. A previous study of the Poecilosclerida using the COI gene (Nichols 2005) found that the Poecilosclerida order was reconstructed as monophyletic, which is in agreement with the ML analyses within this study. A fourth order (Astrophorida) was monophyletic in both analyses and was composed of a single family (Ancorinidae). The fifth order (Hadromerida) was polyphyletic in both analyses. Our data on Hadromerida also supports previous work (Nichols 2005) showing that this order is not supported as monophyletic under any data partition (Nichols 2005). Interestingly, bootstrap support between the fifth order (Hadromerida) and taxa belonging to a sixth unknown order had 99 and 100% bootstrap support in NJ and ML trees respectively. I suspect further taxonomic work based on morphology will group this unknown order within the Hadromerida.

The primary goal of DNA barcoding is to identify an unknown specimen in terms of known classification (Miller 2007; Stoeckle 2008) and to complement and not supersede or replace existing taxonomic practices based on morphological characters. This utility of DNA barcoding was evident within this study as both analyses (NJ, ML) yielded fairly congruent trees that in most instances matched morphospecies. The COI gene worked for resolving most specimens down to a genus level classification in both NJ and ML trees. As an example, species NZSPG023, NZSPG021, NZSPG089, NZSPG074 and NZSPG008 were unknown using morphological identification but were identified down to genera (Hymeniacidon, Halichondria) using BOLD identification. This suggests that COI sequence divergence can be used in combination with classical alpha taxonomy in order to accelerate the identification process of Demosponges. However, a paucity of sponge sequences on BOLD currently limits the effectiveness of this tool. Possibly because this was the first record of sponge barcoding in the Bay of Plenty region there were few species level sequence matches in the BOLD database. Another concern with the use of BOLD is that the sequences submitted to this repository may have been misidentified which has been documented in other databases (Harris 2003). However, this can be avoided by only using

identifications from researchers who are proficient at identifying Demospongiae taxa (Huang *et al.* 2008a). Further barcoding of sponges in the Bay of Plenty region will provide more species sequences for comparisons on BOLD, and are thus likely to accelerate the identification process of sponges.

The use of the COI gene to identify Demospongiae species has some limitations. In this study, only 32 of the 95 specimens examined were successfully sequenced. This included four specimens with sequences matching foreign phyla (indicating non-target amplification or sample contamination) and a further 59 samples for which the COI gene could not be either amplified or sequenced. Only two of the 21 specimens belonging to the Mycale genus were successfully sequenced. There are several possible explanations for our consistent inability to sequence this genus. First, secondary metabolites may be inhibiting or interfering with the barcoding pipeline, possibly during the DNA extraction or PCR amplification stages. Marine sponges often produce secondary metabolites (bio-actives) (Soest & Braekman 1999; Miller et al. 2001), with highly potent metabolites, for example, Peloruside A and Pateamine, previously isolated from species of the Mycale genus in New Zealand (Northcote et al. 1991; West et al. 2000; Page et al. 2005a). An alternative explanation for our inability to sequence so many taxa is the suitability of the non-specific HCO/LCO primers used in the COI amplification process. It is possible that these primers simply do not work for many sponges and that some effort should to be put into developing sponge specific primers if further sponge biodiversity work is to be conducted. Regardless, further studies with larger sample sizes specifically targeting hard to sequence groups such as Mycale, will further test these hypotheses around sequencing failures.

The utility of the mtDNA COI gene for phylogeographic studies of sponges is also hindered by its slow evolutionary rate compared to other animals where mtDNA evolve faster than nuclear DNA (Worheide *et al.* 2004; Voigt *et al.* 2012). However, studies on two independent genes (COI, 28SrDNA) used together have been reported to show relatively congruent phylogenies within Demospongiae suggesting that COI may in fact be a suitable marker (McCormack *et al.* 2002; Cardenas *et al.* 2010). For this study, the COI gene was chosen because it is well studied and understood and because it is the preferred gene of the Canadian Centre for DNA Barcoding (CCDB) where sequencing was

conducted. Consequently, the CCDB has well developed protocols for this gene (Huang *et al.* 2008b; Cardenas *et al.* 2010; Bucklin *et al.* 2011; Sperling *et al.* 2012; Voigt *et al.* 2012), although in this case it is apparent that CCDB protocols do not work well for all taxonomic groups.

In summary, we conclude that for New Zealand Demospongiae, sequence variation present in the barcoding region of the COI gene is sufficient to allow for the identification individuals to their nominate species. The use of mtDNA barcoding can without doubt complement classical morphological taxonomy and accelerate the identification process and may in fact revive an interest classical morphological taxonomy. The low within-species divergences recorded here indicate the need to analyse additional samples and taxa across a wider geographic region to further ascertain whether Demospongiae can be separated to the species level using the COI gene (Hogg et al. 2009). We suggest that sponge specific primers should be developed in an effort to target sponge taxa that were not amplified using LCO/HCO. This may increase the sequencing success rate and therefore the utility of the technique, resulting in additional identifications to augment the results of this study. In conclusion, the use of COI will assist in the identification of previously unidentifiable morphospecies and the identification of cryptic species, for example, *Xestospongia* sp. As a result of these findings we suggest barcoding be incorporated into classical taxonomy as a tool to reduce the time and cost required by poriferologists to accurately identify Demospongiae species.

| fragments. In   | nclud | ing t          | he C            | lath            | arin | ida (        | outg     | roup             | (PO      | RBC           | 076             | ). Sp           | onge                  | e ord         | lers        | are         | indic    | ated    | at th    | e top   | o and   | l fam       | ilies a       | re inc        | licate        | d to the left |
|-----------------|-------|----------------|-----------------|-----------------|------|--------------|----------|------------------|----------|---------------|-----------------|-----------------|-----------------------|---------------|-------------|-------------|----------|---------|----------|---------|---------|-------------|---------------|---------------|---------------|---------------|
|                 |       | Dictyoceratida | Poecilosclerida | Poecilosclerida |      | Astrophorida |          |                  |          | Haplosclerida | Poecilosclerida | Poecilosclerida |                       | Haplosclerida | Hadromerida | Hadromerida |          | Unknown |          | Unknown |         | Hadromerida | Haplosclerida | Haplosclerida | Haplosclerida |               |
| Haplotypes      |       |                | _               | -               |      |              |          |                  |          | 100           | 88888           |                 | •.•.•.•               |               |             |             |          |         |          | xxxx    |         |             |               | 8             | - <b>00</b>   |               |
|                 | NZSPG | 86             | 20 10           | 50              | 102  | 76           | 82 7     | 7 75             | 9        | 11 6          | 3               | 17              | 81 1                  | 5 19          | 88          | 2           | 84 68    | 89      | 23 21    | 74      | 8       | 78 12       | 29 8          | 0 79          | 13 105        | 1             |
| Unknown         | 86    |                |                 |                 |      |              |          |                  |          |               |                 |                 |                       |               |             |             |          |         |          |         |         |             |               |               |               |               |
| Unknown         | 20    | 0.00           |                 |                 |      |              |          |                  |          |               |                 |                 |                       |               |             |             |          |         |          |         |         |             |               |               |               |               |
| Demacellidae    | 10    | 0.52 0         | .52             |                 |      |              |          |                  |          |               |                 |                 |                       |               |             |             |          |         |          |         |         |             |               |               |               |               |
| Mycalidae       | 50    | 0.44 0         | .44 0.45        | ;               |      |              |          |                  |          |               |                 |                 |                       |               |             |             |          |         |          |         |         |             |               |               |               |               |
| Mycalidae       | 102   | 0.44 0         | .44 0.45        | 0.00            |      |              |          |                  |          |               |                 |                 |                       |               |             |             |          |         |          |         |         |             |               |               |               |               |
| Ancorinidae     | 76    | 0.48 0         | .48 0.43        | 0.39            | 0.39 |              |          |                  |          |               |                 |                 |                       |               |             |             |          |         |          |         |         |             |               |               |               |               |
| Ancorinidae     | 82    | 0.48 0         | .48 0.38        | 0.41            | 0.41 | 0.12         |          |                  |          |               |                 |                 |                       |               |             |             |          |         |          |         |         |             |               |               |               |               |
| Ancorinidae     | 77    | 0.49 0         | .49 0.36        | 0.42            | 0.42 | 0.11 0.      | .07      | _                |          |               |                 |                 |                       |               |             |             |          |         |          |         |         |             |               |               |               |               |
| Ancorinidae     | 75    | 0.49 0         | .49 0.36        | 6 0.42          | 0.42 | 0.11 0.      | .07 0.00 | 0                |          |               |                 |                 |                       |               |             |             |          |         |          |         |         |             |               |               |               |               |
| Ancorinidae     | 9     | 0.49 0         | .49 0.36        | 6 0.42          | 0.42 | 0.11 0.      | .07 0.00 | 0 0.00           |          |               |                 |                 |                       |               |             |             |          |         |          |         |         |             |               |               |               |               |
| Ancorinidae     | 11    | 0.48 0         | .48 0.36        | 6 0.41          | 0.41 | 0.11 0.      | .07 0.00 | 0 0.00           | 0.00     | ~ .           |                 |                 |                       |               |             |             |          |         |          |         |         |             |               |               |               |               |
| Petrosiidae     | 6     | 0.44 0         | .44 0.38        | 0.34            | 0.34 | 0.33 0.      | .32 0.32 | 2 0.32           | 0.32 0.3 | 31            |                 |                 |                       |               |             |             |          |         |          |         |         |             |               |               |               |               |
|                 | 3     | 0.40 0         | .40 0.34        | 0.30            | 0.30 | 0.33 0.      | .32 0.32 | 2 0.32           | 0.32 0.3 | 31 0.29       |                 |                 |                       |               |             |             |          |         |          |         |         |             |               |               |               |               |
| Tedaniidae      | 17    | 0.43 0         | .43 0.30        | 0.31            | 0.31 | 0.35 0.      | .35 0.34 | 4 0.34           | 0.34 0.3 | 35 0.27       | 0.16            | 0.05            |                       |               |             |             |          |         |          |         |         |             |               |               |               |               |
|                 | 81    | 0.42 0         | .42 0.28        | 0.31            | 0.31 | 0.35 0.      | .33 0.34 | 4 0.34           | 0.34 0.  | 33 0.26       | 0.15            | 0.05            |                       |               |             |             |          |         |          |         |         |             |               |               |               |               |
|                 | 15    | 0.42 0         | 42 0.20         | 0.31            | 0.31 | 0.35 0.      | .33 0.34 | 4 0.34           | 0.34 0.  | 33 0.20       | 0.15            |                 |                       | <b>`</b>      |             |             |          |         |          |         |         |             |               |               |               |               |
| Thothyidaa      | 19    | 0.46 0         | 40 0.30         | 0.30            | 0.30 | 0.31 0.      | 29 0.30  | 7 0 27           | 0.30 0.  | 29 0.20       | 0.20            | 0.29 0          | 0.29 0.2              | 9             |             |             |          |         |          |         |         |             |               |               |               |               |
| Clionidae       | 2     | 0.47 0         | 47 0.30         | 0.29            | 0.29 | 0.30 0.      | 31 0.2   | 0.21             | 0.27 0.  | 20 0 26       | 0.23            | 0.22 0          | 0.21 0.2              | 3 0 21        | 0 15        |             |          |         |          |         |         |             |               |               |               |               |
| Clionidae       | 84    | 0.46 0         | 40 0.30         | 0.31            | 0.31 | 0.31 0.      | 31 0.2   | 9 0.29<br>0 0 20 | 0.29 0.  | 29 0.20       | 0.23            | 0.22 0          | 123 0.2               | 3 0.21        | 0.15        | 0 00        |          |         |          |         |         |             |               |               |               |               |
| Clionidae       | 68    | 0.46 0         | 46 0 35         | 0.31            | 0.31 | 0.31 0.      | 31 0.2   | a n 2a           | 0.20 0.  | 20 0.20       | 0.23            | 0.22 0          | 1 23 0.2              | 3 0 21        | 0.15        | 0.00        | 00       |         |          |         |         |             |               |               |               |               |
| Unknown         | 89    | 0.40 0         | 40 0.31         | 0.31            | 0.31 | 0.31 0       | 29 0 29  | 9 0 29           | 0.20 0.  | 29 0.26       | 0.23            | 0.22 0          | 24 0 2                | 4 0 22        | 0.10        | 0.18 0      | 18 0 18  |         |          |         |         |             |               |               |               |               |
| Unknown         | 23    | 0.40 0         | 40 0.31         | 0.31            | 0.31 | 0.31 0       | 29 0 29  | 9 0 29           | 0.29 0   | 29 0 26       | 0.23            | 0.24 0          | ) 24 0.2              | 4 0 22        | 0.19        | 0.18 0      | 18 0 18  | 0.00    |          |         |         |             |               |               |               |               |
| Unknown         | 21    | 0.40 0         | .40 0.31        | 0.31            | 0.31 | 0.31 0.      | 29 0.29  | 9 0.29           | 0.29 0.3 | 29 0.26       | 0.23            | 0.24 0          | 0.24 0.24             | 4 0.22        | 0.19        | 0.18 0      | .18 0.18 | 0.00 0  | .00      |         |         |             |               |               |               |               |
| Unknown         | 74    | 0.50 0         | .50 0.35        | 0.33            | 0.33 | 0.32 0.      | .33 0.32 | 2 0.32           | 0.32 0.3 | 32 0.28       | 0.27            | 0.25 C          | 0.25 0.2              | 5 0.27        | 0.23        | 0.21 0      | .21 0.21 | 0.13 0  | .13 0.13 | 3       |         |             |               |               |               |               |
| Unknown         | 8     | 0.50 0         | .50 0.35        | 0.33            | 0.33 | 0.32 0.      | .33 0.32 | 2 0.32           | 0.32 0.3 | 32 0.28       | 0.27            | 0.25 0          | .25 0.2               | 5 0.27        | 0.23        | 0.21 0      | .21 0.21 | 0.13 0  | .13 0.13 | 3 0.00  |         |             |               |               |               |               |
| Chalinidae      | 78    | 0.42 0         | .42 0.35        | 0.30            | 0.30 | 0.32 0.      | 29 0.2   | 7 0.27           | 0.27 0.3 | 27 0.25       | 0.22            | 0.21 0          | ).21 0.2 <sup>-</sup> | 1 0.24        | 0.23        | 0.20 0      | .20 0.20 | 0.18 0  | .18 0.18 | 3 0.23  | 0.23    |             |               |               |               |               |
| Chalinidae      | 12    | 0.42 0         | .42 0.35        | 0.30            | 0.30 | 0.32 0.      | 29 0.2   | 7 0.27           | 0.27 0.3 | 27 0.25       | 0.22            | 0.21 0          | ).21 0.2 <sup>-</sup> | 1 0.24        | 0.23        | 0.20 0      | .20 0.20 | 0.18 0  | .18 0.18 | 3 0.23  | 0.23 0. | 00          |               |               |               |               |
| Callyspongiidae | 29    | 0.39 0         | .39 0.34        | 0.28            | 0.28 | 0.30 0.      | .28 0.26 | 6 0.26           | 0.26 0.3 | 26 0.24       | 0.25            | 0.22 0          | 0.22 0.22             | 2 0.23        | 0.23        | 0.21 0      | .21 0.21 | 0.21 0  | .21 0.21 | 0.24    | 0.24 0. | 18 0.18     |               |               |               |               |
| Callyspongiidae | 80    | 0.42 0         | .42 0.32        | 0.28            | 0.28 | 0.29 0.      | .30 0.29 | 9 0.29           | 0.29 0.3 | 28 0.26       | 0.24            | 0.24 0          | .24 0.24              | 4 0.22        | 0.23        | 0.22 0      | .22 0.22 | 0.21 0  | .21 0.21 | 0.22    | 0.22 0. | 16 0.16     | 0.09          |               |               |               |
| Petrosiidae     | 79    | 0.40 0         | .40 0.33        | 0.27            | 0.27 | 0.29 0.      | .29 0.28 | 8 0.28           | 0.28 0.3 | 28 0.25       | 0.23            | 0.22 0          | .22 0.22              | 2 0.22        | 0.22        | 0.21 0      | .21 0.21 | 0.21 0  | .21 0.21 | 0.23    | 0.23 0. | 17 0.17     | 0.06 0.0      | 4             |               |               |
| 🖶 Petrosiidae   | 13    | 0.40 0         | .40 0.33        | 0.27            | 0.27 | 0.29 0.      | .29 0.28 | 8 0.28           | 0.28 0.3 | 28 0.25       | 0.23            | 0.22 0          | .22 0.22              | 2 0.22        | 0.22        | 0.21 0      | .21 0.21 | 0.21 0  | .21 0.21 | 0.23    | 0.23 0. | 17 0.17     | 0.06 0.0      | 4 0.00        |               |               |
| 🕒 Chalinidae    | 105   | 0.41 0         | .41 0.34        | 0.26            | 0.26 | 0.27 0.      | .28 0.2  | 7 0.27           | 0.27 0.2 | 27 0.24       | 0.23            | 0.22 0          | 0.22 0.22             | 2 0.21        | 0.21        | 0.21 0      | .21 0.21 | 0.21 0  | .21 0.21 | 0.21    | 0.21 0. | 15 0.15     | 0.07 0.0      | 5 0.03 0      | .03           | -             |

Table 3.3 : Genetic matrix generated in MEGA ver 5.2.2 for all 33 haplotypes (Sp86 - Sp0105), from 34 individual specimens for 575 bp mtDNA (COI)fragments. Including the Clatharinida outgroup (PORBC076). Sponge orders are indicated at the top and families are indicated to the left
# Chapter 4

# **GENERAL DISCUSSION**

Issues with estimating the severity of the 'global biodiversity crisis' involving the rapid loss of species are being exacerbated by the inability of researchers to accurately identify species. The art and science of classical sponge taxonomy is diminishing with the retirement, and lack of replacement, of many of the world's leaders in this discipline. The risk is that for many taxonomic groups, species identifications can only be made by a few experts, and our current loss of biodiversity is occurring faster than taxonomic experts can catalogue it (Hogg *et al.* 2009; Bucklin *et al.* 2011). Recently, Bell *et al.* (2015) suggested that the assessment of information available on endangered and threatened sponges, for example, is hindered by the difficulty of sponge taxonomy and the unresolved status of many sponge taxa.

Molecular systematics using DNA based identification founded on the mitochondrial gene, cytochrome c oxidase subunit 1(COI), has been suggested as a tool which can be used to assist in the resolution of species level classifications within the animal kingdom (Desalle *et al.* 1987; Adachi *et al.* 1993; Hebert *et al.* 2003b). In this study, molecular sequenced data provided an indication of the phylogenetic relationships within five orders of the class Demospongiae (Poecilosclerida, Dictyoceratida, Haplosclerida, Astrophorida, and Hadromerida).

In all cases where sequences were able to be generated, it was possible to successfully distinguish known Demospongiae species using the COI gene. The hypothetical phylogeny produced is highly consistent, confirming that the families, Petrosiidae, Chalinidae and Callyspongiidae, form a cluster within the Haplosclerida order and another monophyletic order, Dictyoceratida, which includes members of an unknown family. Previous work (Nichols 2005) on the Dictyoceratida found that this order should be largely reconstructed as monophyletic. However, a single *Xestospongia* sp. located within the Petrosiidae family had a high sequence divergence from other Haplosclerida taxa and was thus grouped externally from the other taxa. Interestingly, the notion of polyphyly

within Haplosclerida is supported by previous work (McCormack et al. 2002; Nichols 2005) showing there was no evidence for monophyly of the Haplosclerida, and almost all taxa (families, genera) within this order were polyphyletic. This conclusion was based on use of a different gene (28S rRNA). McCormack et al., (2002) also found a similar relationship in Xestospongia sp., being quite divergent from other taxa within this order. While the Haplosclerida is well defined morphologically, it has been previously split into two orders Nepheliospongidae and Haplosclerida (Bergquist & Warne 1980). It would be interesting to further test the affinities of families within the Haplosclerida order using both COI and 28S rRNA genes to gain a better understanding of these complex and seemingly conflicting affinities. A third order (Poecilosclerida) consisted of four families (Desmacellidae, Tedaniidae, Crellidae, Mycalidae) and was monophyletic in ML analyses. A previous study of the Poecilosclerida using the COI gene (Nichols 2005) found that the Poecilosclerida order was reconstructed as monophyletic, which is in agreement with the ML analyses within this study.

The fifth order (Hadromerida) was polyphyletic in both analyses using morphological techniques. My data and analyses on Hadromerida are in alignment with the previous work by Nichols (2005) showing that this order is not supported as monophyletic under any data partition. (Nichols, 2005) The present study indicates strong support for the species of an unknown order (NZSPG023, NZSPG021, and NZSPG069) which could not be morphologically identified, and may potentially be assigned to the Hadromerida order following confirmation with further taxonomic work. It would be interesting to undertake further taxonomic work based on morphology to determine if this unknown species does in fact belong within the Hadromerida.

The dominant attitude among the supporters of molecular systematics is that molecular characters (sequence data) are fundamentally superior to morphological identifications to estimate phylogenies (Hori & Osawa 1987; Patterson 1987; Kelly-Borges 1991). Whilst it has been demonstrated that sequence data have great potential to explore relationships at higher taxonomic levels such as that of phylum, class, order, familial, and genera (McCormack *et al.* 2002; Lavrov *et al.* 2008; Cardenas *et al.* 2010; Andreakis *et al.* 2012), gene based phylogenies are

169

beginning to create major discrepancies between phylogenetic hypotheses based on molecular and morphological characters within Demospongiae species (McCormack *et al.* 2002; Xavier *et al.* 2010; Andreakis *et al.* 2012). However, phylogenies which have been generated using molecular systematics usually confirm morphological identifications (Chapter three) (Hogg & Hebert 2004; Hogg *et al.* 2009; Cardenas *et al.* 2010).

In the evaluation of Demospongiae classification presented here, morphological characteristics at the genus and species levels have been principally useful in assigning specimens to species and in differentiating species within genera. As an example, three groups of *Carmia (Mycale)* species were sorted based on the presence and dimensions of palmate anisochelae. While the shape and size characteristics of the anisochelae were consistently different among the 3 groupings, there remained additional (albeit subtle) variability in spicular configuration of other spicules within each group, possibly denoting further species or alternatively indicating micro-environmental variability is specular size and shape. More work including experimental transplant projects is needed to explore just how plastic speculation is in this Genus (as well as others).

Morphological based identifications of sponge species is therefore invariably hindered by the lack of fixed diagnostic morphological characters and the limited knowledge of phenotypic plasticity among sponges. As a further example, members of the *Haliclona* genus have homologous spicules of one major megasclere type -oxeas, (hence 'Haplo'-sclerida), and have very few macroscopic features which could differentiate among species. Consequently, many of the *Haliclona* species described here require further validation to confirm if they are in fact new species. The Haplosclerids are notoriously difficult to assign because of this (Bergquist & Fromont, 1988).

The presence of subtyostyles found within a single species of *C. celata* was used as a diagnostic morphological character to warrant the description of potentially new species (NZSPG084, NZSPG006). However, molecular systematics revealed that this specimen was in fact a *C. celata* species as there were no divergences between these individuals. This is an excellent example of the ability to use molecules to uncover morphologically cryptic species and to speed up the identification process of sponges. Given the difficulties in identifying morphologically plastic species, new DNA barcoding techniques would also reduce the amount of time and thus money required for accurately identifying sponges.

Fifty-five species are described in this current study. Based on classical morphological characters up to three new families, three new genera and 34 species are indicated as undescribed and potentially new to science, certainly new recordings for the region. However, a conservative estimate, where specimens are grouped based on basic spicule type (bearing in mind possible micro-environmental influence on spicule size), suggests that from the collections made in this study, there is a minimum of at least one new family, one new genera, and 18 new species that are to be described fully with reference to type specimens (outside the scope of this study) and therefore possibly deemed new to science.

The COI identification systems have applications for assigning morphologically plastic, cryptic species and to reinforce classical taxonomic descriptions. Where species boundaries are blurred through hybridization causing introgression, potentially complementary genes may be required. Mitochondrial gene identifications will provide phylogenetic resolution in areas where much taxonomic work struggles to separate taxa. Furthermore, a database system such as BOLD will provide a partial solution to the dwindling numbers of expert taxonomists and create a platform which will allow even untrained biologists to identify species to reasonably well refined 'operational taxonomic units' (OTUs) at the genus level. However, I strongly believe that DNA barcoding based approaches are equally as important as classical morphological identifications which are fundamental for referencing a species.

The use of the COI gene to identify Demospongiae species has some limitations however. In this study, only 32 of the 95 specimens examined were successfully sequenced. This included four specimens with sequences matching foreign phyla (indicating non-target amplification or sample contamination) and a further 59 samples for which the COI gene could not be either amplified or sequenced. Only two of the 21 specimens belonging to the *Carmia (Mycale)* genus were successfully sequenced. There are several possible explanations for our consistent

inability to sequence this genus. First, secondary metabolites, known to be prevalent in this genus Northcote *et al.* (1991) may be inhibiting or interfering with the barcoding pipeline, possibly during the DNA extraction or PCR amplification stages.

In comparison, the inherent limitations of classical (alpha) taxonomy based identification systems and the declining number of experienced taxonomists signals the need for a novel approach to sponge taxonomy (Hebert *et al.* 2003a). Over the past decade, classical (alpha) taxonomy has been completely overturned; consequently a review of the state of the art has been suggested among taxonomists (Cárdenas *et al.* 2012)

The taxonomic species descriptions in this study provide a comprehensive review of the classical pioneering work undertaken in New Zealand by Bergquist (1961, 1968, 1970, 1978,1980), Bergquist & Warne (1980), and Bergquist & Fromont (1988) - with updated species names to currently accepted binomial nomenclature. This study has highlighted a further need to update all taxa within the aforementioned literature to the global authority on sponge systematics compiled by Hooper and Van Soest (2002) in an attempt to stabilize New Zealand sponge nomenclature to the revised international Porifera classification. Species identification based solely on traditional (alpha) taxonomy has four major limitations. Firstly, both phenotypic plasticity and genetic variability in the characters nominated for species recognition can lead to misidentifications. Second, this approach often overlooks morphologically cryptic species, which are common among sponges (Xavier et al. 2010; Andreakis et al. 2012; de Paula et al. 2012). Third, since morphological data is often only available for taxa that are in the adult stages of their life cycle, many individuals cannot be identified (Hebert et al. 2003a). Finally, the use of updated morphological keys often requires such a high level of expertise that misdiagnoses are again common (Hebert et al. 2003a). Nevertheless, sponge taxonomic work has contributed substantially to advancing knowledge on sponge morphologies, their structural, physiological and biochemical mechanisms and attributes (Hooper & Van Soest 2002).

In conclusion, molecular characters should be used to complement and not supersede or replace existing taxonomic practices (Miller 2007; Stoeckle 2008). The use of molecular techniques provides a potentially robust tool for testing higher taxonomic classifications, which can be used in combination with biochemical, reproductive, and morphological character sets (Kelly-Borges 1991). Studies which enhance our understanding of phylogenetic relationships should aim to incorporate characters from both morphological and molecular DNA based technologies and empirically test these methods together. This will ultimately provide a more robust understanding of the diversity and phylogeny of the Phylum Porifera, and thus connect the old Linnaean classification with modern molecular based techniques.

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185

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Spon00008, Spon00022, Spon00028, & Spon00053 are included in the following plates, but are not presented in text as samples were too degraded to be described.



# Plate 1.

A-F, *Crella incrustans* (Spon00001, Spon00003): A, acanthostyle, X 400; B, spicules, X 200; C, choanosomal skeleton with tangentially echinating acanthostyles, X 50; D, acanthostyle, X 200; E, spicules, X 100; F, choanosomal skeleton with plumose reticulation, X 50.





A-F, *Crella incrustans* (Spon00001, Spon00003): A, oxea, X 300; B, apex of oxea, X 1800; C, oxea, X 300; D, slightly curved oxea, X 350; E, *in situ* thickly encrusting to massive specimen; F, specimen after preservation in spirit, displaying a slightly translucent pinacoderm.



#### Plate 3.

A-E, *Cliona celata* (Spon00002): A, tylostyles, X 100; B, specimen before preservation in spirit, displaying orange colouration; C, *in situ* specimen displaying adjacent pores in a massive form; D, specimen after preservation in spirit, displaying a polygonally grooved vurrucose surface; E, choanosomal skeleton displaying a confused skeletal structure with tylostyles, X 50.

F, Haliclona sp. (Spon00004): F, hastate oxeas, X 200.



#### Plate 4.

A-E, *Haliclona* sp. (Spon00004): A, spicules, X 40; B, choanosomal skeleton with isodictyal to anisotropic reticulation, X 50; C, *in situ* specimen displaying heavy epibiont coverage of red algae; D, specimen before preservation in spirit, displaying a dermal membrane, through which ostia and oscula are clearly visible; E, specimen after preservation in spirit.

F, Ciocalypta polymastia (Spon00005): F, surface view of specimen showing papillae.





A-F, *Ciocalypta polymastia* (Spon00005): A, spicules X 300; B, spicules, X 300; C, subtylostyles, X 40; D, subtyolostyle, X 100; E, *in situ* specimen displaying finger like papilla; F, plumose skeleton with primary tracts, X 50.



Plate 6.

A, Ciocalypta polymastia (Spon00005): A, tangential skeleton.

B-F, *Xestospongia* sp. (Spon00006): B, acanthostyles, X 200; C, oxea, X 350; D, oxea, X 300; E, oxea apex, X 3500; F, hastate oxea, X 200.



#### Plate 7.

A-F, *Xestospongia* sp. (Spon00006): A, whole specimen after being preserved in spirit; B, *in situ* specimen; C, ectosomal skeleton displaying perpendicular spicules, X 50; D, oxea, X 350; E, oxea, X 200; F, apex of an oxea, X 3500.



## Plate 8.

A-C, *Xestospongia* sp. (Spon00006): A, choanosomal skeleton displaying echinating oxeas and ostia pores, X 50; B, ectosomal skeleton displaying alveolate skeleton, X 50; C, ectosomal skeleton displaying ostia.

D-F, *Halichondria* n.sp.2 *cf. panacea* (Spon00007): D, spicules, X 40; E, oxea, X 100; F, section of specimen after preservation in spirit.



Plate 9.

A-F, *Halichondria* n.sp.2 *cf. panacea* (Spon00007): A, whole specimen *in situ* (note it is the encrusting specimen growing on the left of the other yellow encrusting specimen above); B, isodictyal choanosomal skeleton, X 50; C, ectosomal surface displaying frequent pores, X 50; D, oxea, X 250; E, oxea, X 300; F, spicules, X 80.



Plate 10.

A-F, Spon00008: A, thinly encrusting specimen; B, specimen after preservation in spirit; C, spicules, X 100; D, tylostyle, X 200; E, choanosomal alveolate plumoreticulate skeletal structure, X 50; F, choanosomal plumoreticulate skeletal structure.



# Plate 11.

A-F, *Stelletta maori* (Spon00010): A, *in situ* specimen; B, specimen after preservation in spirit; C, choanosomal skeleton with a dermal layer of euasters, X 50; D, choanosomal spicule structure, X 50; E, spicules, X 100; F, plagiotriaene, X 40.





A-F, *Stelletta crater* and *Desmacella dendyi* (Spon00011): A, whole specimen *in situ*; B, specimen after preservation in spirit; C, choanosomal skeleton with tracts of aligned spicules, X 50; D, choanosomal skeleton, X 50; E, spicules, X 400; F, sigma X 400.



## Plate 13.

A-F, *Stelletta maori* (Spon00012): A, whole specimen *in situ*; B, specimen after preservation in spirit; C, ectosomal skeleton structure, X 50; D, choanosomal skeleton with protruding spicules, X 50; E, euaster, X 400; F, plagiotriaene, X 100.



Plate 14.

A-F, *Stelletta maori* (Spon00012): A, spicules, X 200; B, oxea, X 100; C, apex of plagiotriaene with euaster, X 350; D, euaster, X 3500; E, entire plagiotriaene, X 90; F, euasters attached to plagiotriaenes with sponging fibres, 800.



Plate 15.

A-D, *Stelletta maori* (Spon00012): A, broken euasters, X 4500; B, euasters, X 1800; C, apex of an unusual plagiotriaene, X 500; D, euaster with spines, X 1000.

E-F, *Sigmadocia fragiis* (Spon00013): E, specimen after preservation in spirit; F, alveolate skeletal structure, X 50.



## Plate 16.

A, Sigmadocia fragilis (Spon00013): A, spicules, X 100.

B-F, *Nepheliospongiidae* sp. (Spon00014): B, whole specimen *in situ*; C, specimen after preservation in spirit; D, spicules, X 100; E, euaster, X 1000; F, alveolate choanosomal skeletal structure, X 50.



## Plate 17.

A-F, *Nepheliospongiidae* sp. (Spon00014): A, alveolate choanosomal skeletal structure, X 50; B, alveolate choanosomal skekelta structure, X 50; C, spicules, X 250; D, cluster of spicules, X 100; E, spicule shaft close up with spongin fibres, X 10000; F, oxea, X 500.




A-B, *Nepheliospongiidae* sp. (Spon00014): A, slightly curved oxea, X 500; B, style, X 600.

C-F, *Callyspongia* sp. (Spon00015): C, specimen after preservation in spirit; D, rectangular multispicular reticulation skeletal structure, X 50; E, disorientated skeletal structure, X 50; F, anisotropic skeletal structure, X 50.

P-xviii





A, B, Callyspongia sp. (Spon00015): A, spicules, X 200; B, styles, X 400.

C-F, *Tedania n.* sp. 1 *cf. diversirhaphidiophora* (Spon00016): C, whole specimen *in situ*; D, specimen after preservation in spirit; E, alveolate choanosomal skeletal structure, X 50; F, alveolate choanosomal skeletal structure, X 50.



Plate 20.

A-F, *Tedania n.* sp. 1 *cf. diversirhaphidiophora* (Spon00016): A, alveolate choanosomal skeletal structure, X 50; B, spicules, X 100; C, strongyle with a spined head, X 8000; D, spicules, X 2500; E, strongyles with spined heads, X 600; F, style, X 350.



Plate 21.

A-C, *Tedania n.* sp. 1 *cf. diversirhaphidiophora* (Spon00016): A, smooth style apex, X 11000; B, spicules, X 2000; C, internal structure of a style, X 9000.

D-F, *Stelletta sandalinum* (Spon00017): D, whole specimen *in situ*; E, specimen after preservation in spirit; F, choanosomal spicule tracts with plagiotriaenes, X 50.





A-C, *Stelletta sandalinum* (Spon00017): A, choanosomal spicule tracts with plagiotriaenes, X 50; B, euaraster, X 1000; C, plagiotriaene, X 40.

D-F, *Tedania battershilli* (Spon00018): D, whole specimen *in situ*; E, specimen after preservation in spirit; F, choanosomal plumoreticulate skeleton, X 50.



#### Plate 23.

A-C, *Tedania battershilli* (Spon00018): A, plumoreticulate choanosomal skeleton, X 50; B, choanosomal plumoreticulate skeleton, X 50; C, spicules, X 200.

D-F, *Cliona* n. sp.1 *cf. celata* (Spon00019): D, whole specimen *in situ*; E, specimen after preservation in spirit; F, alveolate choanosomal skeleton, X 50.





A-C, *Cliona* n. sp.1 *cf. celata* (Spon00019): A, ectosomal skeleton, X 50; B, choanosomal skeleton with echinating spicules, X 50; C, subtylostyles, X 100.

D-F, *Polymastia fusca* (Spon00020): D, whole specimen *in situ*; E, specimen after preservation in spirit; F, choanosomal skeleton with stout radiating tracts, X 50.





A, B, Polymastia fusca (Spon00020): A, spicules, X 200; B, tylostyle, X 200.

C-F, *Dictyoceratida* sp. (Spon00021): C, whole specimen *in situ*; D, specimen after preservation in spirit; E, anastomosing choanosomal skeleton, X 50; F, anastomosing choanosomal skeleton, X 50.





A-D, *Dictyoceratida* sp. (Spon00021: A, anastomosing choanosomal skeleton, X 50; B, arcuate isochelae, X 1000; C, spicules, X 200; D, subtylote, X 100.

E-F, Spon00022 (This specimen was removed from the descriptions as it was too degraded): E, whole specimen *in situ*; F, specimen after preservation in spirit.



# Plate 27.

A-C, Spon00022 (This specimen was removed from the descriptions as it was too degraded): A, fascicular fibrous skeleton, X 50; B, spicules, X 100; C, microstyle, X 100.

D-F, *Tethya aurantium* (Spon00023): D, whole specimen *in situ*; E, cut cross section of choanosomal spicule bouquet, X 50; F, middle section of choanosomal spicule bouquet, X 50.

P-xxvii





A-E, Spon00023: A, choanosomal spicule tracts with echinating spicules, X 50; B, specimen after preservation in spirit; C, euasters, X 100; D, small euaster, X 1000; E, tylostyle, X 100. F, Unknown family n.sp. 1 (Spon00024):

F, Unknown family n.sp. 1 (Spon00024): whole specimen in situ.



Plate 29.

A-C, Unknown family n.sp. 1 (Spon00024): A, whole specimen after preservation in spirit; B, alveolate choanosomal skeleton, X 50; C, alveolate choanosomal skeleton, X 50.

**D-F**, *Dysideidae* sp. (Spon00026): **D**, whole specimen *in situ*; **E**, specimen before preservation in spirit; **F**, specimen after preservation in spirit.

P-xxix



Plate 30.

A-C, *Dysideidae* sp. (Spon00026): A, choanosomal skeleton with primary fibres, X 50; B, choanosomal skeleton with primary fibres, X 50; C, oxea, X 100.

**D-F**, *Coelosphaeridae* sp. *cf. Forcepia* sp. (Spon00027): **D**, whole specimen *in situ*; **E**, specimen before preservation in spirit; **F**, specimen after preservation in spirit.





A-D, *Coelosphaeridae* sp. *cf. Forcepia* sp. (Spon00027): A, plumoreticulate choanosomal skeleton, X 50: B, plumoreticulate choanosomal skeleton, X 50; C, euaster and acanthostyle, X 400; D, spicules, X 100.

E-F, Spon00028: E, whole specimen in situ; F, specimen before preservation in spirit.



#### Plate 32.

A-E, Spon00028: A, basal section of specimen after preservation in spirit; B, plumoreticulate choanosomal skeleton, X 50; C, plumoreticulate choanosomal skeleton, X 50; D, spicules, X 100; E, acanthostyle, X 200.

F, Tedania n. sp. 2 (Spon00029): F, whole specimen in situ.



Plate 33.

A-F, *Tedania* n. sp. 2 (Spon00029): A, specimen before preservation in spirit; B, specimen after preservation in spirit; C, plumoreticulate choanosomal skeleton, X 50; D, choanosomal spicule tracts, X 50; E, choanosomal skeleton displaying pores, X 50; F, acanthostyle, X 200.





A, Tedania n. sp. 2 (Spon00029): A, spicules, X 200.

B-F, *Haliclona* sp. (Spon00030): B, whole specimen *in situ*; C, specimen before preservation in spirit; D, specimen after preservation in spirit; E, choanosomal skeleton with a unispicular isodictyal network, X 50; F, choanosomal skeleton with a three dimensional tube as a result of a tube worm infestation, X 50.

P-xxxiv



Plate 35.

A-C, *Haliclona* sp. (Spon00030): A, choanosomal skeleton with a three dimensional tube as a result of a polychate infestation, X 50; B, spicules, X 100; C, tylostyle, X 100.

**D-F**, *Callyspongia ramosa* (Spon00032): **D**, whole specimen *in situ*; **E**, specimen before preservation in spirit; **F**, specimen after preservation in spirit.



# Plate 36.

A-C, *Callyspongia ramosa* (Spon00032): A, choanosomal skeleton with radial vertical reticulation, X 50; B, choanosomal skeleton with radial vertical reticulation, X 50; C, oxea, X 400.

**D-F**, *Haliclona* n.sp.2 (Spon00033): **D**, whole specimen *in situ*; **E**, specimen before preservation in spirit; **F**, specimen after preservation in spirit.





A-F, *Haliclona* sp. (Spon00033): A, confused spicule skeleton, X 50; B, confused choanosomal skeleton, X 50; C, spicules, X 200; D, raphide, X 400.

E, F, *Haliclona* sp. (Spon00035): E, whole specimen *in situ*; F, specimen before preservation in spirit.

P-xxxvii



Plate 38.

A-F, *Haliclona* sp. (Spon00035): A, specimen after preservation in spirit; B, isodictyal reticulation, X 50; C, isodictyla reticulation, X 50; D, isodictyal reticulation, X 50; E, spicules, X 200; F, oxea, X 200.





A-F, *Cliona n.* sp. 2 *cf. celata* (Spon00041): A, whole specimen *in situ*; B, specimen before preservation in spirit; C, specimen after preservation in spirit; D, choanosomal skeleton, X 50; E, choanosomal spicules tracts, X 50; F, spicules, X 100.



Plate 40.

A, Cliona n. sp. 2 cf. celata (Spon00041): A, tylostyle, X 200.

B-F, *Halichondria* n.sp.3 (Spon00042): B, whole specimen *in situ*; C, specimen before preservation in spirit; D, specimen after preservation in spirit; E, net like choanosomal skeleton, X 50; F, net like choanosomal skeleton, X 50.



Plate 41.

A-F, *Adocia* sp. (Spon00043): A, whole specimen *in situ*: B, specimen before preservation in spirit; C, specimen after preservation in spirit; D, plumoreticulate choanosomal skeleton, X 50; E, plumoreticulate choanosomal skeleton, X 50; F, plumoreticulate choanosomal skeleton which echinating megascleres, X 50.





A-B, Adocia sp. (Spon00043): A, oxeas, X 200; B, spicules, X 100.

C-F, *Halichondria moorei* (Spon00044): C, whole specimen *in situ*; D, specimen before preservation inspirit; E, specimen after preservation in spirit; F, choanosomal skeleton with echinating spicules, X 50.





A, B, Halichondria moorei (Spon00044): A, spicules, X 100; B, tylostyle, X 200.

C-F, *Hymeniacidon hauraki* (Spon00045); C, whole specimen *in situ*; D, specimen before preservation in spirit; E, specimen after preservation in spirit; F, choanosomal skeleton with confused structure, X 50.





A-C, *Hymeniacidon hauraki* (Spon00045): A, confused choanosomal skeleton, X 50; B, style, X 100; C, spicules, X 40.

D-F, *Halichondria moorei* (Spon00046): D, whole specimen *in situ*; E, specimen before preservation in spirit; F, specimen after preservation in spirit.



Plate 45.

A-C, *Halichondria moorei* (Spon00046): A, choanosomal skeleton with echinating spicules, X 50; B, choanosomal skeleton, X 50; C, spicules, X 40.

D-F, *Acanthoclada* n. sp. 1 (Spon00047): D, specimen before preservation in spirit; E, specimen after preservation in spirit; F, lax choanosomal skeleton, X 50.



Plate 46.

A-C, *Acanthoclada* n. sp. 1 (Spon00047): A, lax choanosomal skeleton, X 50; B, spicules, X 200; C, spicules, X 200.

D-F, *Tedania* n.sp.3 *cf. battershilli* (Spon00048): D, whole specimen *in situ*; E, specimen before preservation in spirit; F, specimen after preservation in spirit.



Plate 47.

A-D, *Tedania* n.sp.3 *cf. battershilli* (Spon00048): A, alveolate choanosomal skeleton, X 50; B, alveolate choanosomal skeleton, X 50; C, spinispirae, X 200; D, subtylotes, X 200.

E-F, *Halichondria moorei* (Spon00049): E, specimen *in situ*; F, specimen before preservation in spirit.



Plate 48.

A-F, *Halichondria moorei* (Spon00049): A, specimen after preservation in spirit; B, confused choanosomal skeleton, X 50; C, confused choanosomal skeleton, X 50; D, spicules, X 200; E, raphide, X 100; F, spicules, X 200.



Plate 49.

A-F, *Tetrapocillon* n.sp. 1 *cf. novaezealandia* (Spon00050): A, whole specimen *in situ*; B, specimen before preservation in spirit; C, specimen after preservation in spirit; D, reticulated fibrous choanosomal skeleton, X 50; E, tetrapocilli, X 400; F, spicules, X 200.



#### Plate 50:

A-E, *Acanthoclada prostrata* (Spon00052): A, specimen after preservation in spirit; B, lax choanosomal skeleton, X 50; C, spicules, X 100; D, spicules, X 100; E, two spinispirae, X 200.

F, Spon00053: F, whole specimen in situ.



Plate 51.

A-E, Spon00053: A, specimen after preservation in spirit; B, plumose choanosomal skeleton, X 50; C, confused choanosomal skeleton, X 50; D, acanthostyle, X 100; E, tylostyle, X 40;

F, Carmia n.sp.3 cf. tasmani (Spon00054): F, specimen before preservation in spirit.



# Plate 52.

A-E, *Carmia* n.sp.3 *cf. tasmani* (Spon00054): A, specimen after preservation in spirit; B, plumoreticulate choanosomal skeleton, X 50; C, plumoreticulate choanosomal skeleton, X 50; D, spicules, X 200; E, spicules, X 200.

F, Carmia n.sp.1 (1) (Spon00055): F, specimen before preservation in spirit.



Plate 53.

A-E, *Carmia* n.sp.1 (1) (Spon00055): A, specimen after preservation in spirit; B, plumoreticulate choanosomal skeleton, X 50; C, plumoreticulate choanosomal skeleton D, spicules, X 100; E, tylostyle, X 200.

F, Carmia n.sp.2 (1) (Spon00056): F, specimen before preservation in spirit.


Plate 54.

A-F, *Carmia* n.sp.2 (1) (Spon00056): A, specimen after preservation in spirit; B, paucispicular choanosomal skeleton, X 50; C, paucispicular choanosomal skeleton, X 50; D, paucispicular choanosomal skeleton, X 50; E, spicules, X 200; F, tylostyle, X 400.



Plate 55.

A-F, *Carmia* n.sp.1 *cf. tasmani* (Spon00057): A, specimen before preservation in spirit; B, specimen after preservation in spirit; C, choanosomal skeleton with poorly developed spicule tracts, X 50; D, choanosomal skeleton with poorly developed spicule tracts, X 50; E, choanosomal skeleton with poorly developed spicule tracts, X 50; F, spicules, X 200.



Plate 56.

A-B, Carmia n.sp.1 cf. tasmani (Spon00057): A, spicules, X 200; B, spicules, X 100.

C-F, *Haliclona* sp. (Spon00058): C, specimen before preservation in spirit; D, specimen after preservation in spirit; E, isodictyal choanosomal skeleton, X 50; F, isodictyal choanosomal skeleton, X 50.





A-F, *Haliclona heterofibrosa* (Spon00059): A, specimen before preservation in spirit; B, specimen after preservation in spirit; C, simple quadratic choanosomal skeleton, X 50; D, simple quadratic choanosomal skeleton, X 50; E, simple quadratic choanosomal skeleton, X 50; F, spicules, X 100.



Plate 58.

A-F, *Carmia* n.sp.4 *c.f. hentscheli* (Spon00060): A, specimen before preservation in spirit; B, specimen after preservation in spirit; C, choanosomal skeleton, X 50; D, acanthostyle, X 200; E, strongyle, X 200; F, tylostyle, X 100.



Plate 59.

A-F, *Chondropsis* n. sp. 1 *cf. topsenti* (Spon00061): A, specimen before preservation in spirit; B, specimen after preservation in spirit; C, choanosomal skeleton, X 50; D, choanosomal skeleton, X 50; E, choanosomal skeleton, X 50; F, strongyloxea, X40.



Plate 60.

A-F, *Carmia* n.sp.2 *cf. tasmani* (Spon00062): A, specimen before preservation in spirit; B, specimen after preservation in spirit; C, plumoreticulate choanosomal skeleton, X 50; D, plumoreticulate choanosomal skeleton, X 50; E, spciueles, X 200; F, spicules, X 400.



Plate 61.

A-F, *Haliclona* sp. (Spon00063): A, specimen before preservation in spirit; B, specimen after preservation in spirit; C, unispicular isodictyal network, X 50; D, unispicular isodictyal network, X 50; E, unispicular choanosomal tracts, X 50; F, spicules, X 200.



Plate 62.

A-F, *Haliclona* n.sp.6 *cf heterofibrosa* (Spon00064); A, whole specimen before preservation in spirit; B, specimen after preservation in spirit; C, choanosomal skeleton with isodictyal reticulation, X 50; D, choanosomal isodictyal reticulation, X 50; E, spicules, X 100; F, spicules, X 200.



Plate 63.

A-D, *Haliclona heterofibrosa* (Spon00065): A, specimen before preservation in spirit; B, specimen after preservation in spirit; C, simple quadratic choanosomal skeleton, X 50; D, simple quadratic choanosomal skeleton, X 50.

E, F, *Carmia* n.sp.3 (6) (Spon00066): E, specimen before preservation in spirit; F, specimen after preservation in spirit.



Plate 64.

A-F, *Carmia* n.sp.3 (6) (Spon00066): A, plumose choanosomal skeleton with tracts of aligned megascleres, X 50; B, plumose choanosomal skeleton with an infestation of ctube worms, X 50; C, spicules, X 100; D, s-shaped sigma, X 300; E, spicules, X 200; F, subtylostyle, X 200.



Plate 65.

A-F, *Halichondria* n.sp.1 (Spon00067): A, specimen before preservation in spirit; B, specimen after preservation in spirit; C, confused choanosomal skeleton, X 50; D, confused choanosomal skeleton, X 50; E, spicules, X 40; F, tylostyle, X 200.



Plate 66.

A-F, *Haliclona* n.sp.5 *cf heterofibrosa* (Spon00068): A, specimen before preservation in spirit; B, specimen after preservation in spirit; C, choanosomal skeleton with quadratic multispicular reticulation, X 50; D, choanosomal skeleton with quadratic multispicular reticulation, X 50; E, spicules, X 100; F, microxyaster, X 400.



## Plate 67.

A-F, *Adocia* sp. (Spon00069): A, specimen before preservation in spirit; B, specimen after preservation in spirit; C, unispicular choanosomal skeleton, X 50; D, unispicular choanosomal skeleton, X 50; E, spicules, X 100; F, spinispirae, X 200.



Plate 68.

A-F, *Carmia* n.sp.3 (5) (Spon00070): A, specimen before preservation in spirit; B, section of specimen after preservation in spirit; C, plumoreticulate choanosomal skeleton, X 50; D, plumoreticulate choanosomal skeleton, X 50; E, spicules, X 200; F, rabdostyle, X 200.



Plate 69.

A-F, *Haliclona* n.sp.4 *cf brøndstedi* (Spon00071): A, specimen *in situ*; B, specimen after preservation in spirit; C, unispicular choanosomal skeleton, X 50; D, unispicular choanosomal skeleton, X 50: E, spicules, X 100; F, oxea, X 200.



## Plate 72.

A-F, *Carmia* n.sp.3 (2) (Spon00074): A, whole specimen *in situ*; B, specimen after preservation in spirit; C, choanosomal spicule tracts, X 50; D, choanosomal spicule tracts, X 50; E, spicules, X 100; F, spicule structure, X 200.





A, Carmia n.sp.3 (2) (Spon00074): A, spinispirae, X 400.

B-F *Carmia* n.sp.3 (4) (Spon00075): B, whole specimen *in situ*; C, specimen after preservation in spirit; D, plumose choanosomal spicule tracts, X 50; E, plumose choanosomal spicule tracts, X 50; F, oxea, X 40.



## Plate 74.

A-E, *Carmia* n.sp.3 (3) (Spon00076): A, an amorphous specimen *in situ*; B, specimen after preservation in spirit; C, plumose choanosomal skeleton heavily infested with polychaete worms, X50; D, plumose choanosomal skeleton heavily infested with polychaete worms, X50; E, spicules, X 100.

F, Carmia n.sp.1 (2) (Spon00077): F, whole specimen in situ.

P-lxxii



Plate 75.

A-F, *Carmia* n.sp.1 (2) (Spon00077): A, specimen after preservation in spirit; B, choanosomal spicule tracts, X 50; C, choanosomal spicule tracts, X 50; D, choanosomal spicule tracts, X 50; E, rabdostyle, X 200; F, whole specimen *in situ*.



Plate 30.

A-F, *Carmia* n.sp.3 (1) (Spon00073): A, whole specimen *in situ*; B, specimen after preservation in spirit; C, plumose choanosomal skeleton with spicule tracts, X 50; D, plumose choanosomal skeleton with spicule tracts, X 50; E, spicules, X 100; F, birotule, X 400.