Taxonomy and diversity of the sponge fauna from the Agulhas Bank hard reef complex off the South African south coast

By

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Examination Copy

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ABSTRACT

Hard benthic reefs are increasingly known as highly diverse biodiversity ecosystems where sponges constitute a significant proportion of inhabitants. Nearly 60% of the seabed over the inner and outer shelf on the Agulhas Bank comprises hard substrata, and in combination with the convergence of the cold Benguela and the warm Agulhas Currents serves to create a nutrient rich zone supporting multiple fish nurseries. Here we provide the first information on hard benthic reef habitats on the subphotic zone of the Agulhas Bank complex marine protected area, which include Alphard Banks, 72 Mile Reef and 45 Mile Reef, as well as the shallow reefs which include 12 Mile Reef, 6 Mile Reef, 7 Mile Reef, Marthas Reef and inshore reefs within the Tsitsikamma marine protected area. The aim is to provide baseline data on the sponge fauna by identifying species and describing the biodiversity of these hard reefs between the depths 10–200 m on the south coast of South Africa. This is because little is known about the benthic invertebrate diversity on these reefs due to their limited limited commercial value. A total of 362 sponges were collected from multiple surveys onboard the RV *Ellen Kuzwayo* between 2005–2009. We identified 111 taxa from the collection including geographical range extensions for approximately 70 species. The collection also included 17 demospongiae species new to science and 3 species belong to the Calcarea and Homoscleromorpha respectively. The families Ancorinidae (7 spp.), Geodiidae (7 spp.), Halichondriidae (7 spp.), Axinellidae (10 spp.), Microcionidae (13 spp.) and the genera *Stelletta (5 spp.), Tedania, Haliclona* (6 spp.) and *Clathria* (9 spp.) were dominant, in terms of number of species and frequency of occurrence in the samples. The region is more species rich than previously suspected, particularly Alphard Banks where the majority of sponges are located. I describe here principally 11 demosponges collected during these surveys. The results of this project will support a broader programme aimed at systematic conservation planning for the marine environment along the south coast of southern Africa. This is important as marine conservation efforts shift offshore. I conclude with an overview of what is known and what still needs to be discovered to further enhance our understanding of the sponge biodiversity on the south coast hard reefs and South Africa on a whole.

KEYWORDS

Agulhas Bank,

Hard reef complex,

Taxonomy,

Biodiversity,

Marine Protected Areas,

Porifera

DECLARATION

I declare that Taxonomy and diversity of the sponge fauna from the Agulhas Bank hard reef complex off the South African South Coast, is my own work. The content has never been submitted to any university before and all the sources used are acknowledged.

Imogen Weideman

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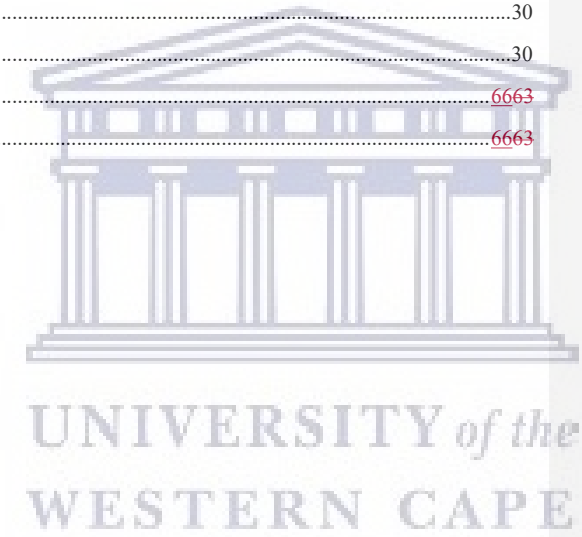

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CHAPTER 1

INTRODUCTION

Biodiversity can be described as 'the variability among living organisms from all sources including, inter alia, terrestrial, marine and other aquatic ecosystems and the ecological complexes of which they are part; this includes genetic diversity, diversity within species, between species, communities and ecosystems" (UNEP, 2002: Article 2). Life on earth is incredibly rich and diverse, with approximately 8.7 million terrestrial and marine species described to date, with millions more to be described or discovered (Mora et al, 2011) Despite over 250 years of taxonomic classification ~~(Linnaeus, 1958)~~, an estimated 86% of terrestrial, and 91% of marine, species still remain to be discovered (Mora et al. 2011). This is especially true in the case of the deep-sea, which is likely to be the greatest contributor to our lack of knowledge in the marine environment (Levin et al. 2019). In South Africa, approximately 12,914 marine species have been recorded, although many taxa, particularly those of small body size, and from deeper areas (>100 m) remain poorly documented (Gibbons et al. 1999; Griffiths et al. 2010). A lack of knowledge about biodiversity hampers our understanding of the true impacts of anthropogenic activities, many of which are driving the loss of marine biodiversity in our oceans (Moberger, 2017).

Threats to biodiversity

Industrialization started properly during the 19th century, and this has had a number of consequences on species, populations, communities and ecosystems, whether terrestrial or marine. For example, in the past 40 years alone an estimated 487 marine species have been trans-located worldwide in an attempt to protect approximately 242 species from extinction (Swan et al. 2015). Nearly half of these species have been coastal invertebrates, and a further 30% are marine plants or algae (Swan et al. 2016). When a species is lost from an ecosystem, changes in the way that materials and energy flow through that system change and this in turn may lead to other organisms flourishing or disappearing, and the ecosystem runs the risk of collapsing (Green et al, 2014).

Primary industrial activities that pose a risk to benthic invertebrate communities in South African waters include the demersal trawl, crustacean trawl, demersal longline and rock lobster trap fisheries and extractive mining operations, specifically for marine diamonds, petroleum (oil and gas) and minerals (Atkinson & Sink, 2008). Most of these industrial activities are known to negatively impact communities on the seabed across the world with a considerable amount of ongoing research being done to quantify impacts and develop mitigation measures or best practice guidelines to minimise damage (Grieve et al. 2015; Kaiser et al. 2016, Miller et al. 2018).

Globally, demersal trawl fisheries are known to impact the seabed and benthic communities (Hughes et al. 2014, Hiddink et al. 2017). Although such fisheries generally take place in unconsolidated,

homogenous habitats, there are areas where sensitive, biogenic ecosystems continue to be impacted by trawl fisheries e.g. seamounts and deep reefs (Clark et al. 2016). An analysis conducted by Sink et al (2012) reported that 27 of 136 mapped marine habitat types are exposed to trawling in South Africa's EEZ with 12 of these being likely to host dense aggregations of fragile, sessile animals forming biogenic features. These include canyons, steep slopes and rocky and gravel areas that are known to provide habitat for vulnerable, slow-growing, sessile species and are areas essential for many spawning and juvenile fish. Other types of fishing activities in South Africa that may result in damaging interactions with sessile, fragile benthic species include the crustacean trawl fishery, demersal longline fishery and fisheries that deploy traps, such as the rock lobster fisheries (Atkinson & Sink, 2008).

Around South Africa, petroleum exploration and production activities are mostly concentrated on the Agulhas Bank, however several wells have been drilled on the west coast and large areas are under lease for production (Ibhubesi Gas Field) and further exploration (www.petroleumagencysa.com, 2019). Seabed impacts of these activities include localised habitat destruction, disturbance, smothering and risk of catastrophic pollution, should an oil spill occur as a result of an uncontrolled release of hydrocarbons (Sink et al. 2012a). The only known *in situ* research conducted in South Africa investigating benthic impacts of petroleum activities showed a limited area (< 250 m radius) of impact around a wellhead (Sink et al, 2010). Nonetheless, should an area of petroleum interest intersect with that of a sessile, fragile community, it is likely that the seabed fauna will be negatively impacted or destroyed during exploration and/or extraction.

Biological invasions pose a significant threat to ecosystem integrity and biodiversity and the rate at which marine organisms are introduced to new environments is high around South Africa (Mead et al, 2011). Currently, 84 marine alien species have been identified in South African waters (Sink et al. 2012; Mead et al,2011); eight are classified as invasive and 34 as cryptogenic species (Sink et al. 2012; Mead et al. 2011). Although most of these introduced species are confined to sheltered sites, such as harbours, bays, lagoons and estuaries and two species, the Mediterranean mussel (*Mytilus galloprovincialis*) and the Pacific barnacle (*Balanus glandula*) have become widespread. Taxa with the largest numbers of locally introduced species are crustaceans, molluscs, ascidiaceans, and cnidarians (see Mead et al. 2011). At least five introduced species are documented from deep waters. The anemone *Metridium senile* has established significant populations in deeper waters up to a depth of 120 m, especially associated with oilrigs on the Agulhas Bank, but the impacts of these populations on the associated reef habitats are unknown (Sink et al. 2010). The Sea Anemone, *Sagartia elegans*, probably a new introduction, was common at both the FA oil platform and on infrastructure in the Oribi/Oryx field on the Agulhas Bank during the initial survey period in 2009.

Why biodiversity is important

Marine biodiversity is important because every ecosystem is incredibly complex and has a multitude of roles and activities (Gamfeldt, Hildebrand & Jonsson, 2008): some may be nurseries for species, others may be niche habitats, or simply be areas of great diversity, all of which are crucial for the survival of species and the wellbeing of the marine realm (Heyman, et al. 2008). Areas with great diversity become aesthetically pleasing and people flock there for viewings, this in turn often raises funds for conservation and awareness (Tribot et al. 2016). New marine resources are also constantly being discovered, often in the form of chemical compounds that are studied in the attempt to produce new drugs for diseases and ailments (Malve, 2016). Sponges have been considered as a reservoir for pharmacologically and biotechnologically important microbes (Villegas-Plazas, et al 2019). A healthy ecosystem supplies us with food security but is also natural defense against coastal erosion (Belley & Snelgrove, 2016).

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Managing biodiversity

Many different government and non-government agencies in South Africa have dedicated programs to aid conservation efforts. The United Nations Environmental Program (UNEP) along with Ramsar and others, have annual sittings where current marine ecosystems and their statuses are assessed (Germani & Salpin, 2007). Amongst the best tools to help conserve marine biodiversity are Marine Protected Areas (MPAs), which have the advantage of benefiting adjacent fisheries and other commercial activities as marine organisms are seldom contained within the MPA. One crucial MPA function is to prevent the over exploitation of marine species, (Kenchingtin, Ward & Hegerl, 2003) and Rising and Heal (2014) noted that if an MPA increased its size by 1%, the fish population could grow by 1%. Currently South Africa has 22 MPAs in place, with more being proposed. That said, the area protected when all 22 are combined still results in less than 0.5% of the ocean falling under conservation efforts compared to the 8% of terrestrial. By 2028 the target is set for 15% of our ocean and coastline to fall under MPAs (Sink, 2018).

In South Africa, the first MPA was declared in 1964 (the Tsitsikamma National Park MPA) and by 2009, 23 MPAs had been declared around South Africa, covering 23% of the length of the coastline of which approximately 10% were "no-take" areas. Following the declaration of the large Prince Edward Islands MPA in the Southern Ocean (2013) the overall level of protection in South African waters exceeded the 10% target of the 2020 Global Target in the Decadal Plan of the Convention of Biodiversity, however only 0.4% of the area of the mainland exclusive economic zone (EEZ) was protected. The full diversity of South Africa's marine systems was thus greatly underrepresented in the MPA network, with more than 40% of recognized marine ecosystem types not represented at all, and with representation of offshore areas of the mainland's EEZ especially lacking. To address this, a proposal for a network of new MPAs that would advance marine habitat representation and protection of threatened marine ecosystems and species, was developed in 2014,

and gazetted for public comment in 2016. In 2019, 20 new MPAs were finally declared, which brought overall protection of the mainland's EEZ to 5%.

This recent MPA network advanced habitat representation from 60% to 94%, with 46 of the 54 ecosystem types being included and significantly advances representation of benthic as well as pelagic ecosystem types. Apart from increasing the representation of habitats under protection, the expanded network contributes to protection of threatened and vulnerable ecosystems and species, supports fisheries management and ecotourism, and provides a platform for research and monitoring. Most of the new MPAs also included some degree of overlap with existing Ecologically or Biologically Significant Marine Areas (EBSAs).

EBSAs are defined areas of the oceans that have special ecological properties. They are identified and described under the Convention on Biological Diversity (CBD) through a technical and scientific exercise on the basis of a set of seven scientific criteria (Clark, 2014). These include "Uniqueness or rarity"; "Special importance for life history of species"; "Importance for threatened, endangered or declining species and/or habitats"; "Vulnerability, fragility, sensitivity or slow recovery"; "Biological productivity"; "Biological diversity"; and "Naturalness". While not a general strategy for protecting all marine habitats and communities, EBSAs are a tool for calling attention to areas that have particularly high ecological or biological importance and that should be considered by decision makers working towards ecosystem objectives, e.g. they could be treated with a higher than usual degree of risk-averseness. Potential management interventions that are encouraged by the CBD to manage EBSAs and their underlying features include MPAs and Other Effective Area-based Conservation Measures (OECMs) (Convention on Biological Diversity, 2009). That focused portions of most of South Africa's EBSAs have been incorporated in the country's expanded MPA network supports the assertion that EBSAs can contribute towards achieving Aichi Target 11.

Descriptions of 17 areas that meet the criteria for EBSAs in South Africa were formally endorsed by the CBD Conference of the Parties in October 2014. These included 11 EBSAs that were contained within the Exclusive Economic Zone (EEZ) and six that extended into other country's EEZ's or into Areas Beyond National Jurisdiction (ABNJ). Since 2016 the country's EBSAs have been under review, and a Systematic Conservation Planning (SCP) approach has been used to assist with identifying new potential areas, which were then assessed in terms of EBSA criteria, and also to delineate their boundaries or to revise the boundary delineations of existing EBSAs (Sink et al 2012, 2019). As part of this process, descriptions of three newly described EBSAs (namely Protea Seamount Cluster, Cape Point to Cape Agulhas, Tsitsikamma-Robberg) have been proposed. Eight of the existing or proposed EBSAs in South Africa have been motivated for largely, or partly, on the basis of scoring highly in terms of the "Vulnerability, fragility, sensitivity or slow recovery" EBSA criterion. For most, this was based on the presence of benthic invertebrate communities that are highly applicable to this category. Examples include structurally complex and habitat forming cold water corals, habitat-forming sponges, hydrocorals, gorgonions, bryozoans and others. Of all of

these, sponges are the most under studied; their significance and representation will be further explored in this study.

What are Sponges?

Sponges are sedentary, filter-feeding metazoans that live in both marine and freshwater environments, where they can be an essential and highly diverse component of marine benthic communities (Berquest 1978; Hooper and van Soest, 2002; van Soest et al. 2012). They are often the most important filter-feeding organisms in benthic marine habitats, and they play an important role in ocean food webs (De Goeij et al. 2017). Some species may possess algal and bacterial symbionts and so are capable of fixing nitrogen or carbon (Schubauer et al. 1985). Further, since sponges may contribute a significant amount of nitrogen through excretion to the surrounding waters, they can be an important source of nutrients to primary producers in nutrient impoverished waters (Schubauer et al, 1985).

The oldest preserved sponge specimen dates back to the Cambrian era (Steiner et al. 1993), with little to no evolution taking place in the past 540–480 million years. Sponges are found in all the world's oceans at depths ranging from euryhaline estuaries/intertidal environments to the deep-sea; from the tropics to the highest latitudes and from rocky reef communities to muddy bottoms (Hooper & Van Soest 2002; Van Soest et al. 2012).

Owing to the fact that sponges are sessile, they are susceptible to predators. They have adapted their defences to such a measure that some of them have developed chemical mechanisms to ward of predators. The chemicals produced by sponges render them as un-desirable food source for fish and other organisms. Having said this, some nudibranchs can consume sponges and these have the ability to metabolize toxins (e.g. Latrunculin A found in sponges of the Genus *Negombata*), and use them to ward off their own enemies (Cheney et al., 2012). The secondary metabolites found in sponges have the potential to be used by the pharmaceutical industry (Wörheide & Erpenbeck, 2007; Pöppe et al. 2010; Wörheide et al. 2012) and ongoing studies are being conducted into their use as antibacterial, antiviral, antifungal, antimalarial, and antitumor agents, and they are targets for immune suppressive, cancer, TB and cardiovascular research. Many of these chemicals may help with sponge –community interactions (Uriz~~,~~ 1991).

Sponges are sensitive to the quality of the environment, and are among those taxa that can be used effectively to assess the well-being of marine communities and ecosystems (Carballo et al. 1996; New, 1994). However, ignorance regarding the identity of sponges negates the value of these organisms as useful indicators of environmental health. This is of particular concern for South Africa, owing to the high levels of endemism that can be expected there (Emmanuel et al. 1992; Luter et al ., 2014).

Less is known about South African sponges than their sessile counterparts like corals and hydroids (Millard, 1975). To date, about 9 200 valid species of sponge are ~~recognised~~recognized from around the world (and incorporated into the World Porifera Database among almost 20,000 sponge entries). The vast

majority of these belong to the Class Demospongiae and more than half are yet to be identified. There are around 15000 species (Amina & Musayeib, 2018; Van Soest et al. 2019).

Although sponges are currently divided into four distinct classes (Figure 1), 25 orders, 128 families and 680 genera (Hooper & Van Soest 2000; Van Soest et al. 2019), many of the higher taxa are disputed following new insights obtained from molecular systematic methods, which ~~is causing a rethink about~~ can be used as a extra characteristic along with morphological characteristics (Morrow & Cardenas, 2015). Currently, four major classes of marine sponges are recognized (van Soest et al. 2012, Redmond et al. 2013; Metobole et al. 2017 & van Soest et al. 2018): Calcarea, Demospongiae, Hexactinellida and Homoscleromorpha (van Soest et al. 2019).

Calcarean sponges are simple tubular sponges with a skeleton comprised of calcium carbonate (calcite) spicules. These spicules are beautifully symmetrical and resemble miniature javelins (monaxons), pickaxes (tetraxons) or tridents (triaxons). Spicules are not divided or categorised by ~~size~~ microscleres and megascleres as in the other two classes (Wörheide et al. 2012). Calcareous sponges occur mostly in shallow water, with just a few species known from the deep sea (Wörheide et al. 2012; Willenz et al. 2014). Approximately, 675 valid species have been described, representing just 7.5% of all living sponges (Hooper & Van Soest 2002; Wörheide et al. 2012; Redmond et al. 2013; Willenz et al. 2014 & Van Soest et al. 2019). Biodiversity is unlikely to be fully described due to lack of taxonomic expertise

Hexactinellid sponges are also called the "glass sponges". They are exclusively marine and are generally more common in deeper waters (200 – 6 000 m). They can also occur in cold fjords (Mackie and Singla, 1983), and submarine caves at shallower depths (Boury-Esnault et al. 1994). These sponges have siliceous hexactine spicules (six-pointed) in both megasclere and microsclere categories, which may be free in the tissue or fused into an intricate lattice to form a rigid elegant skeleton. Hexactinellids are primarily distinguished by the lack of a superficial cellular pinacoderm and a mesohyl matrix (Bergquist, 1976). Currently, 700 extant species are considered valid, representing 7% of all sponges described to date (Reiswig, 2002; Dohrmann et al. 2008; Van Soest et al. 2012; Redmond et al. 2013, Van Soest et al. 2019). This number is questionable and is believed to be an underestimate for a number of reasons: 1) they are found to occupy remote habitats; 2) experts working on this group of sponges are few 3) the deep-sea is still largely unexplored, and 4) vast museum collections await revision (Dohrmann et al. 2008, Wörheide et al. 2012). Glass sponges are remarkably different from the other three main classes of sponges in many aspects of their biology. This includes their syncytial tissue organization and triaxonic spicule symmetry which clearly distinguish them from the other three major sponge groups and make them one of the best-supported higher level metazoan monophyla (Dohrmann et al. 2008, Wörheide et al. 2012). They also differ from the other groups because they generally have a larger set of morphological characters, displaying a complex skeletal structure and vast array of different spicule types that provide a wealth of information for the taxonomy of the group (Dohrmann et al. 2008)

Most sponges fall into the Class Demospongiae, which has an estimated 7 263 species world-wide

(Hooper & Van Soest, 2002; Van Soest et al. 2012; Redmond et al. 2013; Morrow & Cardenas, 2015; Van Soest et al. 2019). They account for 90% of all known recent sponges, though many more are known or suspected to exist (Hooper & Van Soest, 2002; Van Soest et al. 2012). Representatives can be found in all aquatic environments (fresh and salt water), and they occur from the margins of the land to the deepest ocean trenches, and in all polar, tropical and temperate seas. In addition to silica or siliceous spicules, Demospongiae produce an organic skeleton of spongin which forms an intricate web of flexible threads. When spicules are present, megascleres are monaxons or tetraxons and microscleres are asterose, meniscoid or monaxon spicules. The family Cladorhizidae, order Poecilosclerida (Desmospongiae) is the only carnivorous sponge family, lacking the filter-feeding (aquiferous) architecture and choanocyte cells considered to be diagnostic of the Porifera (Hooper & Van Soest, 2002; Van Soest et al. 2012; Wörheide et al. 2012; Hestetun et al. 2016). These typically deep-sea sponges can trap, envelop, and digest prey items, representing a unique evolutionary strategy within the phylum Porifera (Hestetun et al. 2016).

The class Homocleromorpha is a small group of marine sponges consisting of less than 100 described extant species (Wörheide et al. 2012; Van Soest et al. 2012, Redmond et al. 2013; Cruz-Barazza et al. 2014; Van Soest et al. 2019). The monophyly of this group is well accepted on the basis of their general organization and the shared features of their cytology and embryology (Muricy & Diaz 2002; Cruz-Barazza et al. 2014). The Homoscleromorpha further differ from other sponges by their exclusive cinctoblastula larvae and the presence of flagellated exopinacocytes (Boury-Esnault et al. 1990, 2003). The classification of the Homoscleromorpha has changed considerably over the years, with its ranking elevated from Suborder to Order, Subclass and Class (Topsent, 1895; Dendy, 1905; Lévi, 1973; Gazave et al. 2010, 2012). This was mainly due to the shared presence of siliceous tetractinal-like calthrops (Wörheide et al. 2012). These changes reflected the increasing knowledge of their biology and the discovery of new exclusive morphological characters within the phylum.

Taxonomy

The presence of a skeleton, its composition (calcareous or siliceous) and its nature (form, size and orientation) still forms the basis of current sponge taxonomic methods. Sponges of the Subclasses Verongimorpha (Order Verongida) and Keratose (Orders Dictyoceratida and Dendroceratida) lack spicules and are identified primarily by the presence and orientation of primary and secondary spongin fibres, as well as by the amount and disposition of foreign debris (such as sand) within their tissues. The external body shape (encrusting, tubular, fan shape or globular) varies considerably between species, and gross morphology is often used in association with internal architecture to classify and identify sponges. Other characteristics such as odour, colour, consistency, the presence of mucus and the number and size of oscules may also be as characters in sponge classification. However, these latter characteristics can be influenced by the environment which makes their use in identification difficult (Bergquist, 1978; Bavestrello et al. 1993;

Kerr & Kelly-Borges, 1994).

Although the skeleton is more constant and thus a better character for use in studies of taxonomy, sponges can make fools of taxonomists, as some species appear to incorporate bits of another's skeleton into their own bodies. The incorporated spicules are easy to spot, a bigger problem is some species losing diagnostic groups Despite some recent advances in sponge taxonomy using biochemical, genetic analyses and chemo-taxonomic techniques (Kelly-Borges et al. 1994; Kerr and Kelly-Borges, 1994; Koigoora, Pallela, Reddy et al. 2013), sponge taxonomy and systematics remain unstable. Unfortunately, this confusion has not been greatly reduced by studies on the evolution of reproductive life histories (Lévi, 1956, 1973; Bergquist, 1978; Fell, 1993; Fromont, 1994), the structure and function of sponge cells (Simpson, 1968, 1984; Vos et al. 1990), and their ecology (Bergquist, 1978; Alvarez et al. 1990; Diaz et al. 1990; Schmahl, 1990; Wulff, 1994).

Aims

South Africa has a unique coastline, influenced at the macroscale by the cold Benguela Current on the west coast and the warm Agulhas Current on the east and south coasts (Goschen, 2015). More locally, the physical environment around the coast is affected by the terrestrial topography and by the width and orientation of the continental shelf and itstheir interaction with prevailing wind fields; by coastal and shelf-edge upwelling and inshore jet currents and by eddies, rings and filaments (Lutjeharms, 2000). The region encompasses numerous biogeographic provinces, and a multitude of habitats that reflect changes in salinity, substratum and depth. As a consequence, the diversity of marine life is extensive (Blanke et al. 2009).

Although approximately 400 sponge species have been recorded from South Africa to date, many have been wrongly identified and need taxonomic revision (Ridley & Dendy, 1887; Kirkpatrick 1902, 1903a & b; Stephens 1915; Burton 1926, 1931, 1933a & b, 1936; Lévi, 1963, 1967). Further, approximately 80% of South Africa's Exclusive Economic Zone (EEZ) is considered to be under sampled. Continental South Africa has a coastline of some 3,650 km and an Exclusive Economic Zone (EEZ) of just over 1 million km^2. Waters in the EEZ extend to a depth of 5 700 m, with more than 65% deeper than 2 000 m (Griffiths et al. 2010). Most of the region's sponge samples have been collected from depths shallower than 500 m, with the largest concentration of collection from shallow hard reefs less than 40 m. The slope, bathyal and abyssal zones remain almost completely unexplored (Samaai, pers. comm.). Considering that South Africa is widely recognized as a region of high biological diversity and is widely considered to be the third most diverse country in terms of terrestrial diversity, marine species diversity is predicted to be as high due to the high number of marine habitats and ecosystems and unique coastline surrounded by three oceans (Griffith et al. 2010).

One of the major tasks in sponge biology is still to document the diversity of living species and the aims of the thesis are to: identify and describe the sponge fauna from the Agulhas Bank hard reef complex, in an

effort to document the species composition and assess their diversity in relation to latitudinal and bathymetric distribution patterns. Furthermore, we will assess whether the species composition found on Alphard Banks conforms to the inshore reefs of the Agulhas Bank.

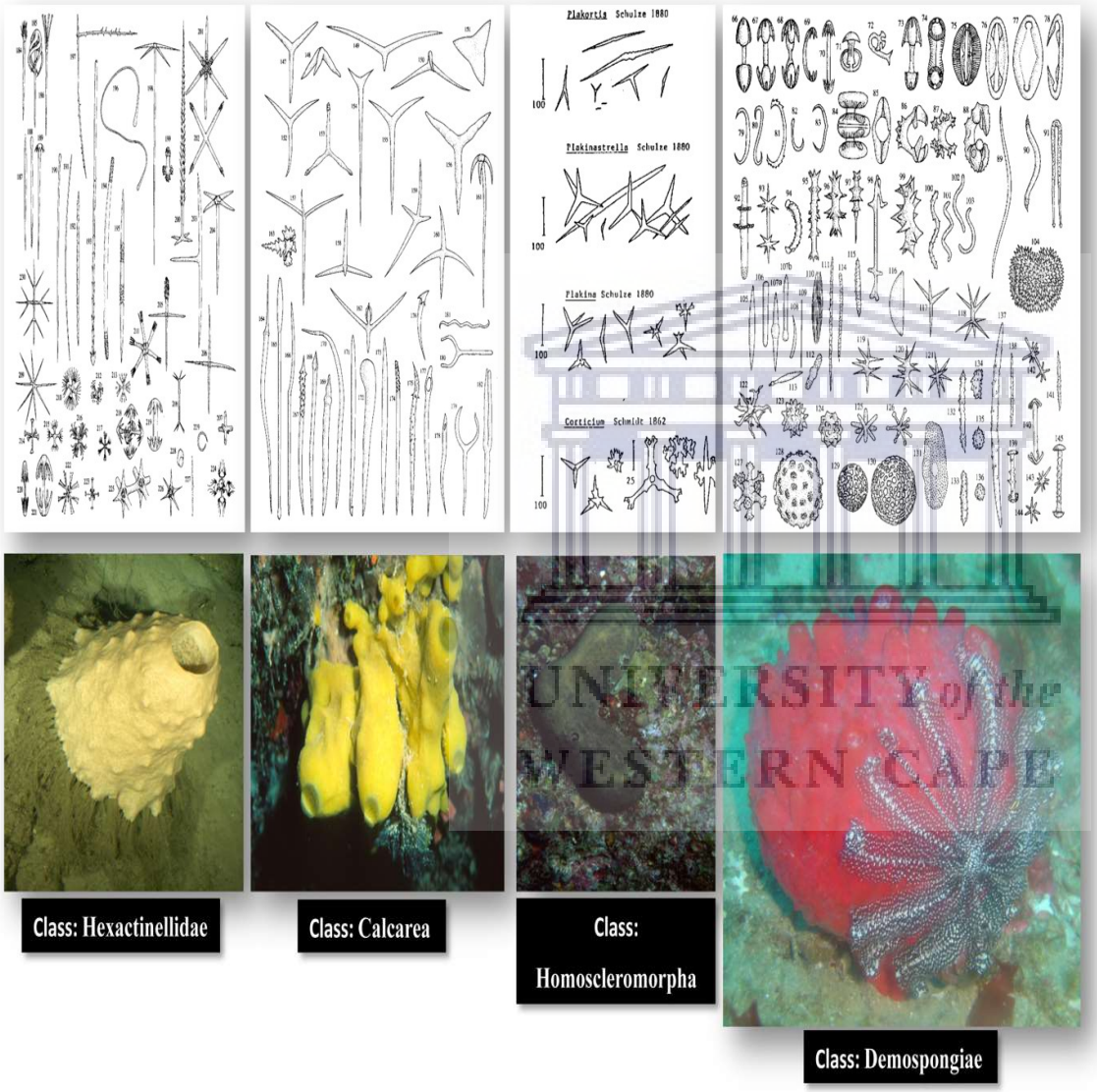

Figure 1: Spicule plates representing the four different Classes of the phylum Porifera Hexactinellida, Calcarea, Homoscleromorpha and Desmospongiae. (Samaai. 2012).

CHAPTER 2

LITERATURE REVIEW

Marine biological research in South Africa began in the early 1700s, and most of the early work was taxonomic, focusing on algae and marine invertebrates. Although the South African Museum was opened in Cape Town in 1828, most of the descriptions of South African marine life during the 19th century was undertaken by scientists from the Northern Hemisphere such as F. Krauss, J.H. Wahlberg and J.D. Gilchrist. These collectors and naturalist travelled widely around the coast, but invariably returned northwards to publish and present their findings (Gibbons, 1995). Andrew Smith, who was the first director of the South African Museum, published the first thorough ~~good~~ descriptions of marine invertebrates in the daily newspaper (Day, 1977). But even he eventually returned to England with his collections of crabs and fishes. Despite the emphasis on taxonomy, little of it was directed towards sponges, with crustaceans, molluscs and fishes being the main focus (Day, 1977).

South Africa has a rich biodiversity and some 13 000 species of free-living marine animals have been recorded or described (Gibbons et al. 1999; Griffiths et al. 2010) since the first expeditions to the Southern Seas in the early 1800s ("Challenger", "Valdivia" expeditions). Notwithstanding the above, many taxa still remain poorly documented (Gibbons et al. 1999; Griffiths et al. 2010; Metobole et al. 2017).

South African coastal and offshore waters have received significant attention from sponge researchers in the last hundred years, as evident from the extensive literature from this region: Esper (1797) described the first sponges from the Cape of Good Hope about 200 years ago. Studies of sponges in the late 1800s and early 1900s focused primarily on the fauna of unconsolidated deep-water sediments along the south and east coasts off South Africa (Van Soest, 2012). Although the lists that were compiled were comprehensive at the time., ~~there were no detailed descriptions of the species were provided~~. In the 40-odd years from 1847 to 1887, Carter published no less than 125 papers on sponges, and these included studies on species found around South Africa: a far greater number than any other author. These papers treated nearly 800 distinct species in 200 distinct genera. The greater portion of the works by Carter was purely systematic, but he has also contributed to our understanding of the anatomy and embryology of this group.

Vosmaer (1880) completed two volumes on sponge taxonomy, and named some 70 species from South Africa. The material he examined was housed in ~~came from~~ the Leyden Museum of Natural History, which contained species collected from the Cape of Good Hope and from other areas in the Atlantic Ocean. He described *Amphilectus caesper*, *Desmacidon (Myxilla?) elastica* and *Clathria lobata* from the Cape of

Good Hope; *Chondrochealia virgata, Esperia conlaremii* and *Clathria anchorata* from the Atlantic Ocean and *Clathria pena* from west Africa in the Atlantic O.

Ridley and Dendy (1883) made their sponge collections during the "Challenger" Expedition, and described 54 genera and approximately 100 species. Although many of the species they described were from the Pacific and Indo-Pacific region as well as from Antarctica, they nevertheless made a valuable contribution to our knowledge of the South African sponge fauna (Ridley & Dendy, 1883). They described ten species from South Africa: *Raspailia flagellformis, Raspailia rigida, Clathria Lobata, Coelosphaera navicelligerum, Desmacidon ramosa, Lissodendoryx digitata, Isodictya conulosa, Isodictya grandis* and one species belonging to the genus *Haliclona* (Ridley & Dendy, 1883). Baer (1905) provided a list of 24 sponge species from Zanzibar, which also included some species found off the Cape of Good Hope the Cape of Good Hope, and of these only one had also been reported by Ridley and Dendy (1887).

Kirkpatrick's (1902, 1903) reports on the Gilchrist collection provide the most complete account of South African sponges. Gilchrist~~Kirkpatrick~~'s collection comprised approximately 50 species (28 new species), the bulk of which was collected on the eastern boundaries (Natal coast) of South Africa. A Comparison of Gilchrist~~Kirkpatrick~~'s collection to those of both Ridley and Dendy (1887) and Carter (1876, 1879, 1881, 1883, 1885), indicates only six species in common. These include *Tetilla casula* Carter, *Clathria typica* Carter, *Higginsia bidentifera* Ridley and Dendy, *Desmacidon ramosum* Ridley and Dendy, *Desmacidon grande* Ridley and Dendy, and *Hamacantha esperioides* Ridley and Dendy. The collection of sponges made by Stephens (1915) ("Scotia" Expedition) during an ecological survey from False Bay to Saldanha Bay on the west coast, contained 37 new species, and expanded the range for a number of other species that had previously been recorded for South Africa. This collection, although small, contributed much to the knowledge of the South African west coast sponges, because it was confined to a short stretch of coastline. A comparison of Stephens' collection to that of Kirkpatrick reveals not a single species in common, although five genera were the same (Stephens,1915). This might be due to the fact that the "Gilchrist sponges" (Kirkpatrick, 1902) were taken over a shorter period of time in South African waters and in deeper waters than the sponges collected by the "Scotia" expedition (Stephens, 1915).

Burton initiated a survey to collect sponges along the South African west coast during the period 1926-1936. He wrote various reports that focused on particular orders and was the first to survey for lithistid sponges along the coast. In his 1926 publication, Burton described 21 species of "Myxospongida" and "Astrotetraxonida" and also included descriptions or reports of specimens from the British, Natal and Durban museums, as well as from Professor Dendy's Port Phillip (South Australia) collection (Burton, 1926). Although Burton also made an extensive collection of the sponge fauna from the Atlantic seaboard of the African continent during the Danish Expedition (1945-1946), this did not include South Africa. The total number of species recorded in this report, from the central west African coast was 65, of which nine were endemic to tropical West Africa, and five were new species. Previously, he had recorded 23 species on the Atlantic seaboard of Europe and by comparing the one to the other he concluded that half the species

recorded from tropical W Africa occurred also in the Mediterranean. These species included *Leuconia rudifera* Polèjaeff, *Tethya aurantium* Pallas, *Suberites carnosus* (Johnston), *Haliclona angulata* (Bowerbank) and *Myxilla rosacea* (Lieberkühn). Although there is much in common between the west African fauna and that of the Mediterranean (Burton, 1956), he also found that elements of the more northerly species of England occur also off west Africa.

All these expeditions collected large quantities of sponges and many of the new species that were described for the west coast were not found on the Natal Coast (Lévi, 1963). In total, the "Gilchrist" (Kirkpatrick, 1902-03), "Challenger" (Ridley and Dendy, 1883) and "Scotia" expeditions (Stephens, 1915) collected a total of 180 Demospongiae, 16 Calcarea and 6 Hexactinellida (Lévi, 1963) from around South Africa.

More recently, comprehensive works covering Southern African sponges have been produced by Lévi on the orders Poecilosclerida (Lévi, 1963) and Astrophorida (Lévi, 1967) (Class Demospongiae), and by Borojevic (1967) on the class Calcarea. Day (1974) provided the first species list and ecological notes for sponges in the False Bay region of South Africa. Although the immediately aforementioned authors dealt mainly with coastal species (water shallower than 100 m) from around the entire coast, the majority of samples were in fact collected in the Indian Ocean. Uriz (1984, 1985, 1988), focused her attention on the Namibian deep-water sponge fauna or more generally between 29`S and 04`E off western Africa.

The most recent work on regional sponges has been produced by Samaai (2002), Samaai et al, (2004) and Samaai & Gibbons (2005). Approximately 45 new species and two new genera have been described for South Africa (Samaai & Gibbons, 2005; Samaai et al. 2003; Samaai et al. 2004), with new species being discovered at an increasing rate.

Currently, the number of described sponges from South Africa (defined here as extending from Oranjemund on the West Coast to Richards Bay on the East Coast), is 343 species (Samaai, pers. comm.) based solely on morphological characters. This is low compared to other marine invertebrates such as molluscs, annelids and cnidarians (Gibbons et al. 1999; Griffiths et al. 2010), and suggests that the South African sponge biodiversity is far from fully described. This lack of knowledge of marine sponge diversity threatens our ability to conserve, manage and utilize this natural resource sustainably.

The aims of this thesis are thus twofold:

1) To identify and describe the shallow water sponge fauna of the Agulhas Bank hard reef system, in order to begin to document the sponge diversity on the Agulhas Bank.
2) To make a preliminary analysis of the distribution of hard reef sponges along the continental shelf of South Africa, as well as inshore and offshore reefs that make up part of the Agulhas Bank Complex

CHAPTER 3

MATERIALS AND METHODS

Agulhas Bank

Following Sink et al. (2012), the South African Ecological Economic Zone (EEZ) can be divided into four shelf-ecoregions (Benguela, Agulhas, Natal and Delagoa) and two off-shelf ecoregions (Southeast Atlantic and Southwest Indian oceans) (Figure 2A). The Benguela ecoregion is situated along the west coast of South Africa, and is characterised by the cold, nutrient waters of the Benguela Current, that lead to high levels of productivity and industrial-scale fisheries (Lutjeharms, 2000). The Agulhas ecoregion spans the south coast of South Africa, and encompasses the Agulhas Bank, over which waters of the Agulhas and Benguela currents interact (Goschen, 2015). Although upwelling may occur at isolated capes and peninsulas, and at the edge of the shelf, water circulation is comparatively sluggish and the system displays pronounced seasonality in productivity (Probyn, 1994). The Natal and Delagoa ecoregions encompass the east coast of South Africa, with the latter situated to the north of the former. Both are characterised by warm, nutrient-poor water from the fast, south-flowing Agulhas Current, and both are relatively narrow in offshore extent (Lutjeharms, 2000).

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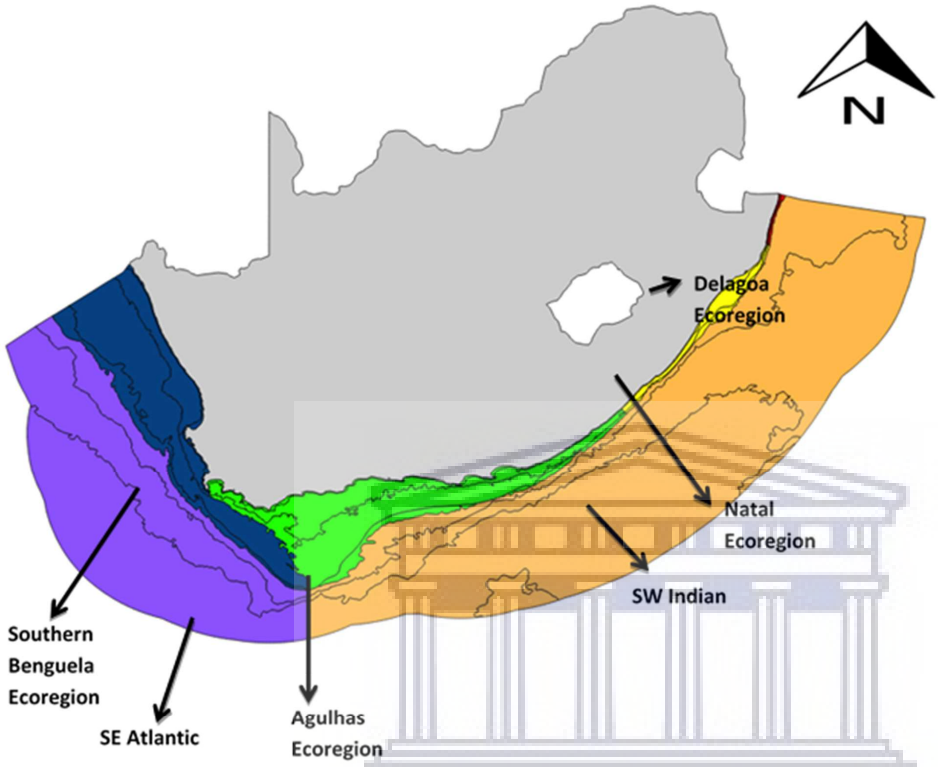

Figure 2A: A map representing the different ecoregions where sponge samples used for this study were collected during various field trips and cruises from the year 2010–2016.

The Agulhas Bank is a broad, shallow part of the continental shelf that extends 250 km south of Cape Agulhas and tapers eastwards to the coast at East London, before dropping off sharply (Hutchings et al, 2002). It supports a cluster of slender, volcanic pinnacles that extend to depths of 0–200 m below the surface (Götz et al. 2014). More than half of the seabed at the edge of the shelf in the Agulhas Ecoregion comprises hard substrata, primarily associated with the reef systems and sea mounts. The highly variable seabed topography creates a high level of diversity in benthic and pelagic habitats (Gotz et al 2014; Smale et al, 1993), which in turn supports a high marine biodiversity (Hutchings et al.2002). The relatively sluggish circulation over the Agulhas Bank, coupled with its high level of seasonal productivity further enhance biodiversity, and the region not only hosts numerous endemic species but burgeoning fisheries. The latter support a range of fisheries (demersal, pelagic, recreational), whilst the historic productivity have created oil reserves.

Our knowledge of diversity over the Agulhas Bank is woefully incomplete (Morris, 2017). This is especially true in the area of the deep reefs, which cannot be accessed using shallow-water platforms and by diving. Indeed, they are regarded as one of the three most threatened marine habitats within the country (Sink et al. 2011).

Study Sites

Samples were collected from eight inshore and offshore hard reefs within the Agulhas ecoregion: 6 Mile Bank, 12 Mile Bank, 45 Mile Bank, 72 Mile Bank, Alphard Bank, Martha`s Reef, Struisbaai and Tsitsikamma (Figure 2A). Most of the samples were collected by dredge being dragged across the ocean floor at a speed of 0.5- 1.5mph using the *RV Ellen Kuzwayo* over the period 2005–2009. The dredge was lowered to the seabed and towed at a speed of approximately 1 knot for a period of 10 minutes. On retrieval, samples were sorted into major taxa, individually photographed, preserved in 96% ethanol, and fully labelled. The inshore reefs (Martha`s Reef, Struisbaai and Tsitsikamma) are located along a coastline ~~of 150 km distance,~~ and are considerably variable in size, profile and depth range. Most sampling stations were between 10 and 40 m deep and within 5 km of the shore. Martha's Reef is situated 6 km offshore and stretches about 10 km along the outside of the off- shore border of the De Hoop Marine Protected Area. The reef ~~profile is high~~ with depths ranging from 15 to 40 m. The 6 M~~m~~ile and 12 Mile Banks are located about 10 and 21 km off Cape Agulhas, the latter being approximately circular in shape with a diameter of about 10 km. Sample depths on 12 Mile Bank between 30 and 70 m were recorded. The reef profile is generally low with a few pinnacles.

Situated around 65 km offshore of the coast at De Hoop, Alphard Bank is a cluster of slender pinnacles covering an oval area of up to 12 km across (Figure 2C). Because of their volcanic origin (Dingle & Gentle, 1972), the pinnacles are cone-shaped and rise from around 100 m to as shallow as 15 m below the surface. The 45 Mile Bank is a large (about 35 km across), low-profile (70 to 85 m deep) reef

area, approximately 80 km offshore. The farthest offshore (110 km) and deepest (average depth around 80 m) reef surveyed, 72 Mile Bank is a high-profile reef with depths ranging from 60 to 100 m. The oval bank stretches up to 20 km across. Figure 2B provides a map of study area.

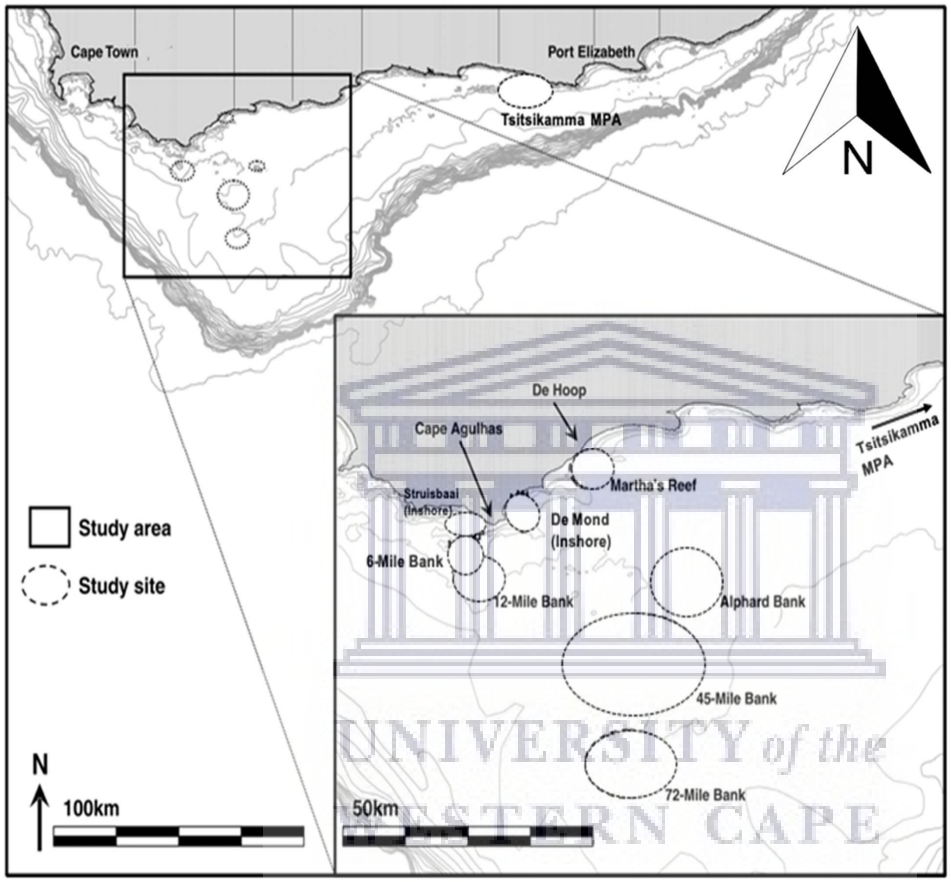

Figure 2B: Map of study site

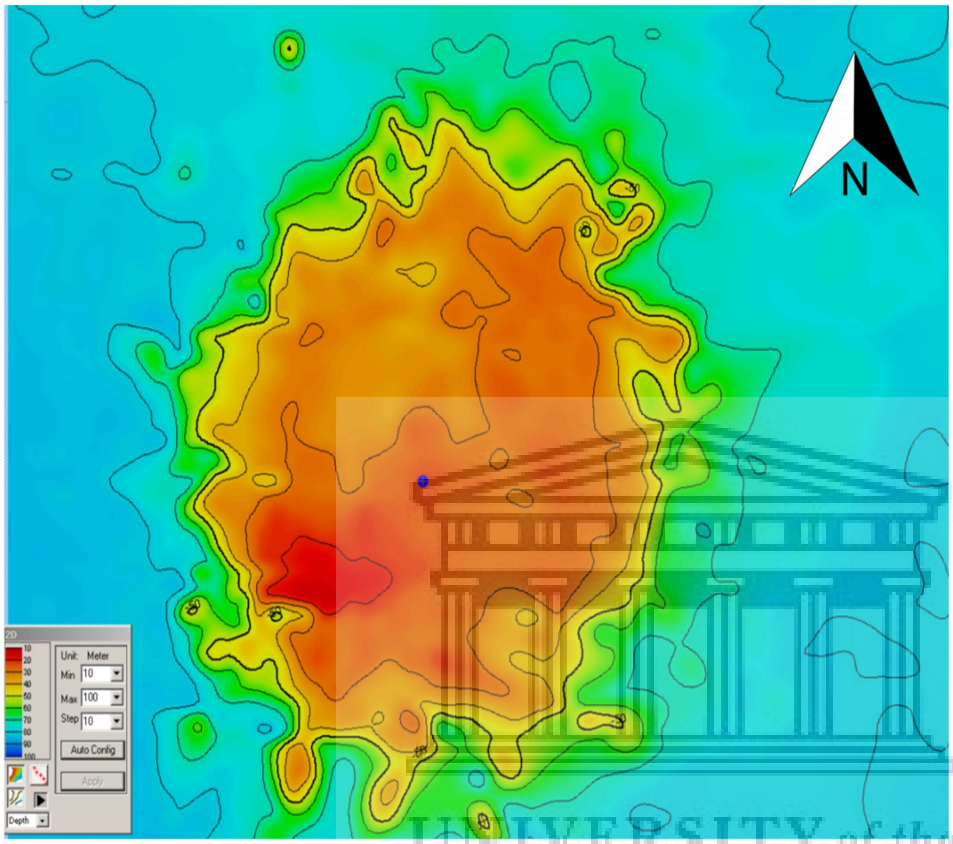

Figure 2C:3D Map of Agulhas Bank

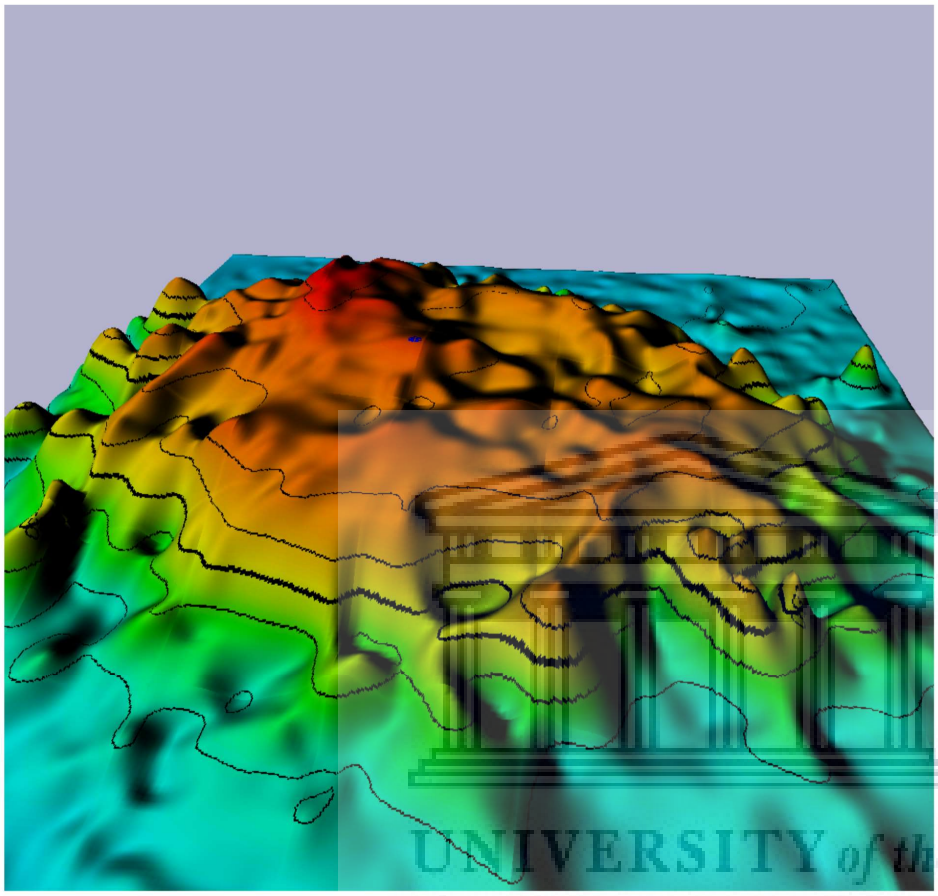

Figure 2D: 3D Maps of Agulhas bank

Taxonomic procedures

Spicule preparation

A section (3 mm^3) of each specimen that contained choanosome and ectosome was cut from the voucher specimen, and placed in an Eppendorf tube. The spicules were isolated from the section in a fume cupboard by digesting the sponge tissue in 100% nitric acid. Material was then washed three times (twice with distilled water and once with 70% ethanol) prior to microscopic examination. Between each rinse the material was centrifuged for 1–2 min. at 3000 rpm. Clean spicule samples were then stored in 96~~100~~% ethanol at room temperature.

For examination purposes, the spicules were re-suspended and pipetted onto a microscopic slide. The ethanol was evaporated off on a heated tray at 40°C. After the slides were completely dry, a few drops of Entellen were added and a cover slip put in place. The slides were then placed in an oven at 50°C for approximately one hour, or until the mountant had hardened.

Scanning electron microscopy (SEM) was carried out on the spicule preparations obtained by the same procedures described above. Small, but significant variations in the skeletal characteristics that would confer specific identity in some cases are very difficult to observe when examined through the light microscope exclusively (Fromont and Bergquist, 1985). A piece of film negative was attached to a stud with epoxy glue, and left to dry overnight. One or two drops of the clean spicule sample were placed on the stud (taking care not to overcrowd the stud) which was left to dry for one hour in an oven at 35 °C to remove the ethanol. The studs were then splutter-coated with gold and examined in a Hitatchi T3000 Scanning Electron Microscope.

Skeletal structure

In order to examine the skeletal arrangement of the sponge, a perpendicular section (approximately 5 mm^3) of tissue was cut from the voucher material and embedded in paraffin wax after it had been processed automatically through a series of dehydrating and embedding agents (Table 1). Histological sections of approximately 75 µm thickness were cut using a microtome. The wax was removed from the section by washing it in xylene under a fume cupboard. Sections were placed and mounted on microscopic slides and viewed under a Carl Zeiss microscope at various magnifications. Diagnostic features for each specimen were photographed at the appropriate level of magnification.

For SEM, a disc shaped piece of sponge (approximately 5 mm^3) was cut diagonally so that the interior and ectosome were visible. It was placed in a petri dish in a fume cupboard where a little concentrated nitric acid was carefully added to it from a pipette. The acid dissolved the mesohyl, and the rate of dissolution were controlled by the addition of distilled water. The section was rinsed in distilled water and then dried in

100% ethanol. A standard disc shaped piece of plastic which is sticky on both sides was loosely attached to a stud and the disc shaped piece of sponge was attached loosely to the plastic disc which was then placed in an environmental chamber.

Table 1: Procedure for automated histological processing sponges. For sponges stored at 70% ethanol cycle as follows.

Compound	Duration
80% ethanol	1h
95% ethanol	1h
100% ethanol	1h
100% ethanol	1h
clearing agent (Histosol)	1h
clearing agent plus 20% wax	1h
clearing agent plus 50% wax	1h
wax	1h

Microscopes

The Carl Zeiss Axioskope 40 microscope was used for both observing skeletal structure and spicule assortment. The Carl Zeiss Mrc5 camera was used to take pictures on the computer or create a live feed. The Zen program by Zeiss was later used for spicule measurements. Sponge spicules also require the use of a Scanning Electron Microscope that picks up the small differences in spicules and size classes, properties. Studs were placed inside the Scanning Electron Microscope (Hitatchi TM4000) and as much information as possible was captured, by means of photographs, spicule assortment and size classes. The camera had to calibrated to accommodate mega- as well as microscleres.

Morphometric analysis

Spicules were measured using a Carl Zeiss Axioskope 40 microscope. Twenty spicules, of each spicule category, were measured for all specimens. Line drawings of spicules were made with a calibrated camera lucida, and photographs were taken of the diagnostic skeletal characters. The species descriptions provide the spicule sizes measured in this study along with those reported previously in the literature. Spicule dimensions are given as mean length (range of length measurements) x mean width (range of width

measurements) followed by the number of spicule measurements taken. All material has been deposited in the Iziko Museum in Cape Town along with Holotype for new species. Museum and register numbers are cited in the text.

Analysis of species distribution amongst offshore reefs

In order to explore similarities in the composition of the sponge fauna amongst the different offshore reefs and banks examined here, species records were first pooled across samples per reef/bank. Owing to differences in sampling effort per reef/bank, which complicated clear analysis of richness and diversity across the region, species were simply recorded as present:absent. The similarity in community composition between the different reefs/banks was then computed using the Bray-Curtis Index, and the resultant matrix was visualised using non-metric multidimensional scaling (nmMDS): all analyses being conducted using Primer 7 software following Clarke and Gorley (2008). Data were also re-analysed pooling across species within genera, and relationships between the patterns observed by the two levels of hierarchical classification were determined using correlation analysis (Clark, 2001). The 2Stage routine (Clark, 2015) in Primer 7 was also used to test for the correlation between the similarity matrices generated between different reefs/banks by species and genera. In order to explore which of the measured environmental and sampling variables might contribute to the structure of the similarity matrix, a distance based linear model (DistLM) was constructed following (Botwe, 2015). For each bank/reef, the predictor variables were the number of samples (N) collected, the mid depth sampled (m), the distance from shore (nm) and the latitude and longitude – the latter two in decimal degrees: all predictors were $\log_{10}(x+1)$ transformed and then normalized. Analyses were repeated at both the genus and species level, and models were visualized using dbRA plots (Clark, 2015).

CHAPTER 4

RESULTS

A total of 362 sponge specimens were collected from the hard reefs on the Agulhas Bank, comprising 111 taxa (Appendix 1: Supplementary Table 1). There are representatives of 68 genera, 40 families, 16 orders and 3 classes. No Hexactinellida were collected. The collection includes 17 species new to science and represents geographical range extensions for approximately 70 species. 4 new species are described in the thesis Although 111 species were collected, only 11 species of Demospongiae (Phylum Porifera) are described here in full. The balance of the material will be the subject of future detailed work, and are summarized subsequently to the systematic account, which follows Morrow & Cárdenas (2015).

Systematics – Species descriptions

Order Tetractinellida Marshall, 1876

Family Neopoltidae Sollas, 1888

Genus *Homophymia* Vacelet & Vasseur, 1971

Definition. Fan-shaped or clavate Neopeltidae with monocrepid pseudotetraclones, smooth monocrepid pseudophyl- lotriaenes as ectosomal megascleres, and a single form of amphiaster microsclere (Kelly 2000 & Kelly, 2007).

***Homophymia rugopellais* sp. nov.**

(Figure 3; Table 2)

Etymology. Species was named *rugopellais*, Latin words for rough skin being rugopellais. Specimen is textured like skin with goosebumps

Material examined. TS 1577, 72 Mile Bank Agulhas Bank, South Africa (35,7901ºS; 20,6439ºE), depth of 39-42m, 19/3/05 by dredge, collected by T. Samaai, depth 39–42 m on board the RV Ellen Khuswayo, dredge. TS 1446, Alphard Bank Agulhas Bank, South Africa (35,9185ºS; 19,8988ºE), 25/03/05 by dredge, collected by T. Samaai, depth 68 m on board the RV Ellen Khuswayo.

Locality. 72 Mile Bank & Alphard's Bank, Agulhas Bank offshore hard reefs.

Description. TS 1557 is a Massive semi globular sponge, 10.5 cm high x 13.5 cm wide x 8.5 cm thick, that bulges on the apex of the sponge and encrusted by a thin sponge with turrets. Specimen TS 1446 is 8 cm high x 9 cm wide x 6.5 cm thick. Apex is depressed, forming a translucent oscular sieve. Surface undulating but smooth, sandpapery to the touch with oscules not readily visible on the apex of the sponge; ostia evenly scattered, 0.1 mm diameter. Texture skin-like being stony, hard and not compressible, the sponge tears hard and difficult to break. Colour in life, bulge on apex is dark brown ~~in bulge on top~~ remainder of sponge ~~and~~ being light brown/mustard; in preservative, dark brown.

Skeleton. Choanosomal skeleton consists of huge smooth monocrepid interlocking desmas and tracts of large oxeas that radiate towards the apical surface. Amphiasters are abundant throughout the choanosome and the ectosomal region. The cladome partially zygose with neighbouring 'cladomes' and desmas forming an interlocking structure. Some of these cladomes are claw like and lock in foreign euasters; of a *Geodia* sp. sponge growing mutually with *Homophymia* pellis n.sp The Ectosomal skeleton has tracts of large oxeas fanning out at the surface.

Spicules. Megascleres. Desmas monocrepides, huge smooth ramose pseudotetra- clones. The rhabd is irregular in shape and thickness, is sinuous, and the cladome is furcate with sinuous branches, often clawing eusaters from a mutual sponge, 353.42 µm (240 µm–422.4 µm), 10x, N=50. Oxeas, large and straight, 358.67 µm (259.2 µm–537 µm), 10x, N=50.

Remarks. The only known *Homophymia* species described from the Western Indian Ocean (WIO) is *Homophymia lamellose* Vacelet & Vasseur, 1971, collected from Madagascar (Vacelet & Vasseur, 1971). The genus *Homophymia* is well defined (Kelly, 2000, 2003) with currently two additional species described from the New Zealand (*H. stipitate* Kelly, 2000) and from Norfolk Ridge and New Caledonia (H. *pollubrum* Schlacher-Hoenlinger, Pisera & Hooper, 2005). This is the first record of the genus in South Africa.

Homophymia lamellosa (see Table 2) is contoured and blade-shaped or folded lamellate fan with the largest specimen approximately 8 cm high x 10 cm wide x 2 cm thick. The oscules are 1 to 2 mm in diameter and located at the top of a digiform expansion of the blades. *Homophymia pollubrum* is spherical with an apical depression and a short stalk, whereas H. *stipitata* is parsnip-shaped (stipitate) with an irregular globular body that bulges in places. *Homophymia* ***r****ugopell*~~*a*~~*is*\ **sp. nov.** is globular and encrusted by a light brown sponge with turrets (*Haliclona* sp.) and the top of the sponge is infused by a dark brown skin like *Geodia* sp.

The species collected from Alphard Banks is well differentiated from the Western/Central Indo-Pacific *Homophymia* in being massive globular shaped, having oscula organized on the apex and having a symbiotic mutual relationship with a species of *Haliclona* and *Geodia*; the morphology of which is dissimilar to that of *H. stipitata* and *H. pollubrum* with penicillate body and only a slight expansion of the apical region.

Based on the above, the species from Agulhas Bank is considered new to science. Specimen also has oxeas and no phyllotrianes

Key diagnostic characters.

- Globular with a light brown and apical dark brown depression.
- Choanosome covered with pseudotriaenes with an extremely sinuous cladome and short irregular rhabd.
- Desmas are quite large and smooth with sizes ranging from 240 µm–422.4 µm.
- Two types of amphiaster.
- ~~Mutual relationship with Haliclona and geodia species~~
- Euasters clawed by cladomes

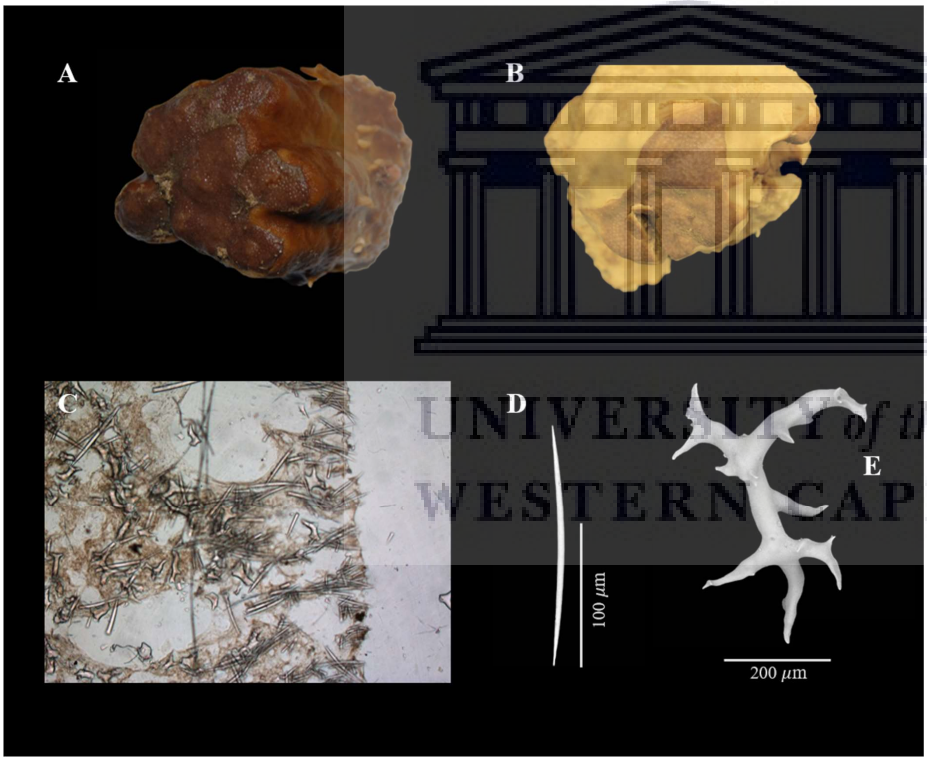

Figure 3: A – *In Situ*. B – *Ex Situ*. C – Skeletal Structure. D – Oxea. E – Desmas

Table 2: Morphological characters of species of South African and WIO *Homophymia* species

Species	Morphology	Colour	Location	Spicule Types
Homophymia rugopellais sp. nov	Semi massive Globular Evenly distributed ostia Texture –skin like	Light brown sponge with turrets Dark brown in places	Alphard Bank, Western Cape	Desmas Oxeas Amphiasters
Homophymia Lamellosa **(Vacelet& Vasseur, 1971)**	Contoured. Blade-shaped	Sponge is brown, Encrusted with light brown sponge	Western and Northern Mada-gascar	Desmas Amphiasters Phyllotrianes
*Homophymia Pollubrum***(Schlachter-Hoenlinger, Pisera and Hooper, 2005)**	Vase shaped with Thick walls and round opening. Attached to substratum with simple base	Sponge is Dirty white o beige	New Caledonia	Dicranoclones Spirasters Rays type 1 Rays type 2
Homophymia Stipitata **(Kelly, 2000)**	Specimen is double stalked Globular Apex Bulbous ti subglobular body	Light golden brown	New Zeeland	Choanosomal Desmas Amphiasters Strongyloxeas Pseudotriaenes

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Order Tethyida Morrow & Cardenas, 2015

Family Hemiasterellidae Lendenfeld, 1889

Genus *Hemiasterella* Carter, 1879

Definition. Hemiasterellidae with vasiform, cup-shaped or massive growth forms lacking axial condensation, with plumo-reticulate choanosomal skeleton; euasters with thick acanthose strongylote rays, sometimes calthrops-like.

***Hemiasterella vasiformis* (Kirkpatrick, 1903)**

(Figure 4)

Synonymy.

Hemiastrella vasiformis (Burton, 1931), pg. 353, Pl. XXIII, fig. 11.

Kalastrella vasiformis (Kirkpatrick, 1903), pg. 238, pl. v, fig. 3.

Hemiastrella vasifomis (Topsent, 1919), pg. 7.

Material examined.TS 1385, Alphard's Bank Agulhas Bank, South Africa, (35,0590º S; 19,9186º E), depth of 50m ,~~specimen was collected on~~ 26/3/05 ~~by dredge at a depth of 50 m on~~on board the *RV Ellen Khuswayo*,dredge voyage 35. TS 1432, 12 Mile Bank Agulhas Bank, South Africa (35,0303º S; 20,9203º E), depth of 39-42m, 28/3/05, collected by means of demersal longline ~~at a depth of 39–42 m~~ on board the RV Ellen Khuswayo, voyage 35. TS 1456, Alphard's Bank, Agulhas Bank South Africa (35,0303º S; 20,9203º E), depth of 39-42m, 28/3/05, collected by means of demersal longline at a depth ~~of 39–42 m~~ on board the RV Ellen Khuswayo. TS 1576, Alphard's Bank Agulhas Bank South Africa (35,0590º S; 20,8765º E),depth of 65m, 26/3/05, collected by means of demersal longline ~~at a depth of 65 m~~ on board the RV Ellen Khuswayo, voyage 35.

Comparative material examined. SAMC–A24732 (cross-reference TS 846 & SAF 3-Sod44), Gotham reef, Sodwana Bay (27.4916°S; 32.7022°E) depth of 34m, South Africa, 05/11/03, collected by T. Samaai, ~~depth 34 m.~~

Description. Massive, frilly, fan shaped sponge with a slightly thickened base, 130 x 70 x 20 mm in diameter. Surface smooth with oscules flush to surface, 1 mm diameter, and ostia also visible. Texture firm, rubbery to the touch, slightly compressibility and difficult to tear. Colour in situ is pinkish-purple and gradually turning red red with a white patchy base; in preservative-yellow. Spicules not visible.

Skeleton. The Choanosomal skeleton consists of bands of oxeas that are relatively loose, without axial compression, the bands are ~~which is~~ linked from the base through the deeper choanosomal region. Towards the ectosomal, the ill-defined columns~~anastomosing~~ bands become plumo-reticulate. The ectosomal skeleton is well developed and densely packed with oxyasters. No other spicules are found in the ectosome.

Spicules. Megascleres. Oxea, 1224 µm (921.6–1574.6), 5x, n=50. Strongyleoxeas, smooth, fusiform and medially curved, 964µm (825.6–1440), 5x, n=50. **Microscleres**. Oxyasters, ~~smooth with~~ spines on the tips, 31.68µm (21.6–40.8) 40x, n=50.

Substratum, depth range and ecology. Found on rocky reef at a depth between 39–65 m.

Geographical distribution. The species is confined to the South African coastline. South African East Coast - Tugela Banks, Aliwal shoal, Durban (Natal Bioregion) and Sodwana Bay (Delagoa bioregion). South African south coast – Agulhas Bank (Agulhas ecoregion)

Remarks. This species is easily recognizable by its bright colouration that fades towards the stem. All samples look similar in terms of colouration. The species was first described by Kirkpatrick (1903) from the Tugela Banks and later by Burton (1931) from Durban, South Africa. The current specimens conform closely in morphology, colouration, spiculation and skeletal features to the descriptions of Kirkpatrick (1902) and Samaai et al. (2019).

Key diagnostic characters.

- Sponge massive, cup- to multi-lamellate vase shaped.
- Colour pinkish-purple red.
- Ectosomal skeleton densely packed with oxyasters

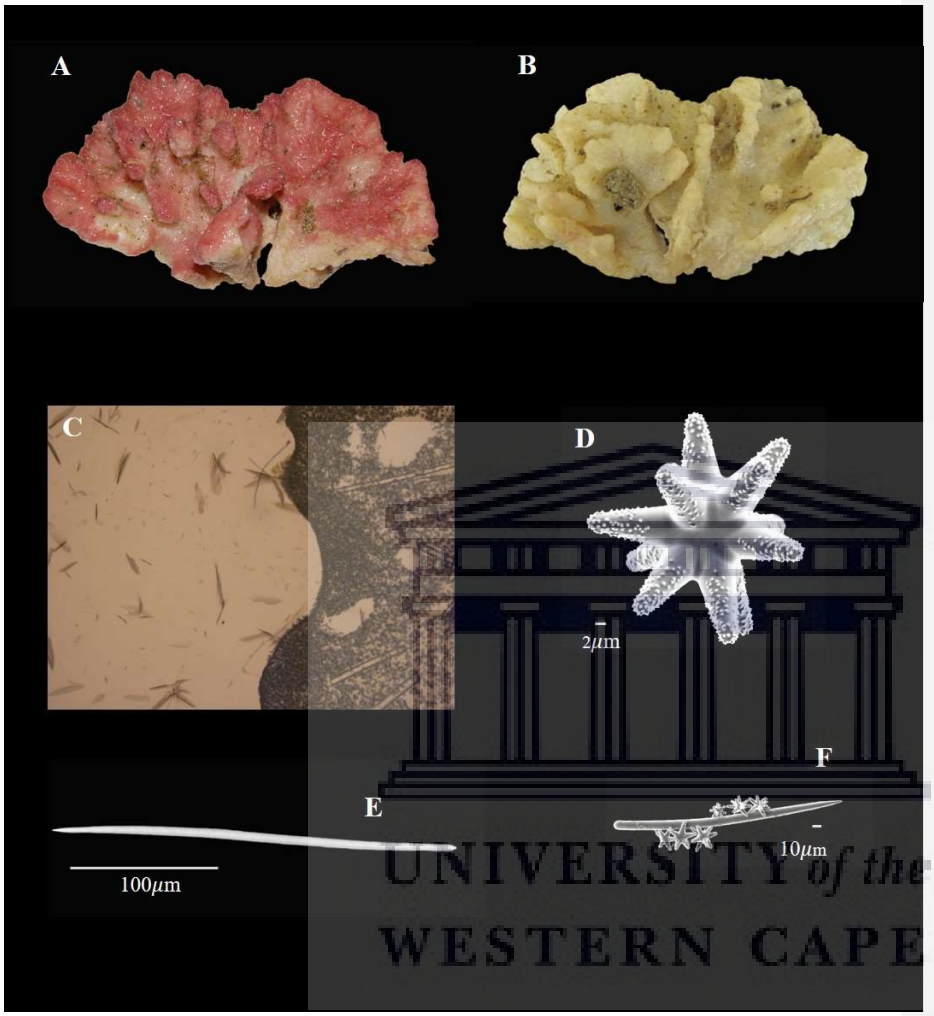

Figure 4: A – *In Situ*. B – *Ex Situ*. C – Skeletal Structure. D – Oxyaster. E – Srongyleoxea. F – Oxea.

Order Poecilosclerida Topsent, 1928

Family Guitarridae Dendy, 1924

Genus *Guitarra* Carter, 1874

Definition. Mostly thickly encrusting sponge. *Guitarra* has a complex arrangement of oxeas or styles. Megascleres of one size class, often at the sponge surface. Microscleres often Placochelae, and palmate acanthoisochelae or bipocilla-like isochelae. Very often these microscleres are accompanied by sigmas, spiny isochelae and other spicules that resemble isochelae.

***Guitarra panema* sp. nov.**

(Figure 5; Table 3)

Etymology. Species is called *panem*, panem being the latin word for bread, original specimen strongly resembles a bread roll.

Material examined. TS 1507, Alphard's Bank Agulhas Bank, South Africa, (35,0374º S; 20,8660º E), depth of 34m,24/3/05 by means of scuba dive ~~at a depth of 34m~~ on board the *RV Ellen Khuswayo*, voyage number 35. Cross referenced with TS 1498, 12 Mile Bank Agulhas Bank, South Africa, (35, 0186º S; 19, 9157º E~~),~~), depth of 29m , 27/3/05. Collected by means of scuba diving ~~at a depth of 29m~~ on board the *RV Ellen Khuswayo,* voyage number 35. TS 1527, Alphard's Bank Agulhas Bank, South Africa, (35, 0389º S; 20, 8652º E), depth of 28m, 24/3/05. Collected by means of scuba ~~diving at a depth of 28m~~ on board the Ellen Khuswayo, voyage number 35. TS 1703, Martha`s Reef Agulhas Bank, South Africa (35, 0342º S; 20, 8660º E) depth of 29m, 10/8/05. Collected ~~by means of dredge at a depth of 29m~~ on board the Ellen Khuswayo, voyage number 35 by means of dredge. TS 1732, Alphard's Bank Agulhas Bank South Africa, (35,0342º S; 20, 86º E06), 8/8/05. Collected by means of scuba diving at a depth of 26-30m on board the Ellen Khuswayo, voyage number 35. TS 1749, 45 Mile Reef Agulhas Bank South Africa (35,9185 S; 19, 8988º E), 25m depth , 8/8/05. Collected by means of scuba diving ~~at a depth of 25m~~ on board the Ellen Khuswayo, voyage number 35.

Comparative material examined. SAM-H4938 (Ts 189), South Paw, Oudekraal (33°56'S, 17°21'E), depth 17–20 m, collected by P. Coetzee, 23 April 1996. Ts 281, Coral Gardens, Oudekraal (33°59'S, 17°23'E), depth 10 m, collected by T. Samaai, 16 June 1997.

Locality. Agulhas Bank - Alphards Bank, 12 Mile Bank, 45 Mile Bank and Martha's Reef.

Description. Thickly encrusting sponge, 70 x 45 mm x 20 mm in diameter. Surface smooth, slightly hispid under microscope, with slightly elevated protuberances. Oscules randomly scattered on upper surface, not raised, 0.5 mm diameter in between protuberances. Not visible in preserved specimen. Texture firm,

compressible and tough. Colour in life dark brown to reddish, choanosome light brown; in ethanol colour fades to brown.

Skeleton. The choanosomal skeleton is composed of bundles or bands of ascending multispicular tracts of aniso-oxeas, united by intertwined bundles that become oblique on the surface, sometimes tangential or palisade. The bands~~It~~ run~~s~~ perpendicular to sponge surface, protruding to sponge exterior. Choanosomal multispicular tracts form bundles of 120–210 ~~um~~ µm in thickness. Single megascleres are scattered through choanosome.Microscleres are wide spread throughout sponge. Ectosome, megascleres form groups, 80–100 µm in diameter and at the base and 200–320 µm being ~~tangential.~~

Spicules. Megascleres. Anisoxeas being fusiform on one side, straight or slightly curved of uniform size, often obtuse at the top of the thick half, 345.6 (240–349.6) µm x 12 (10–12) µm, 10x, n=50. Microscleres. Placochelae of two sizes: (I) Large 46.8 (45.6–4 8) µm, (II) small 34 (28.8–38.4) µm, 40x, n=50. Spined sigmoid isochelae, punctuate with more or less accentuated curvature, 12 (12) µm, 40x, n=50.

Substratum, depth range and ecology. Specimens found on hard substratum between 10-40 m in depth

Remarks. ~~Carballo and Uriz (1998) synomomised *Guitarra fimbriata var. indica* Dendy, 1916 *sensu* Lèvi (1963) with *Guitarra flamenco* Carballo & Uriz, 1998. Samaai & Gibbons (2005) described *Guitarra flamenca* from Ouderkraal on the south west coast of South Africa based on the presence and structure of the spined isochelae. *Guitarra flamenca* is differentiated from other species by possessing peculiar bipocilla-like spiny isochelae in two size categories and in the forms of placochelae. The spined isochelae of *G. flamenco* differs from the spined isochelae in *Guitarra indica* (Dendy, 1916). The placochelae are also smaller than in *G. flamenca* (see Table 3), and *G.flamenca* lacks the stout sigmoid-like spined isochelae and large size category of the placochelae,~~ as found in *G. fimbriata* ~~(Berquist & Fromont, 1988; Carballo and Uriz, 1998). *Guitarra fimbriata* is restricted to the Northen Hemisphere (Boury-Esnault et al. 1993; Van Soest et al. 2019~~). *Guitarra* sp.1 differs from *G. flamenco* in the absence of the bipochelae and presence of the sigmoid spined isochelae. Dendy (1916) described the presence of the microscleres in *G. indica* as follows: "placochelae of the typical *Guitarra* form but with the shaft very abruptly constricted in the middle. Numerous smaller forms occur with less sharp constriction in the middle of the shaft, also numerous very slender forms of various sizes without fimbriae or with very feebly developed fimbria". Based on this, *G. indica* is clearly different from the *Giurarra Panema* sp. nov and the species described by Lévi (1963), her, could soon be considered synonymous for the Agulhas Bank species.

Based on the above, the species from Agulhas Bank is considered new to science having sigmoid spiny isochelae and anisoxeas.

Key diagnostic characters.

- Thickly encrusting sponge
- Small sigmoid spiny isochelae and anisoxeas

- Dark brown to reddish colouration

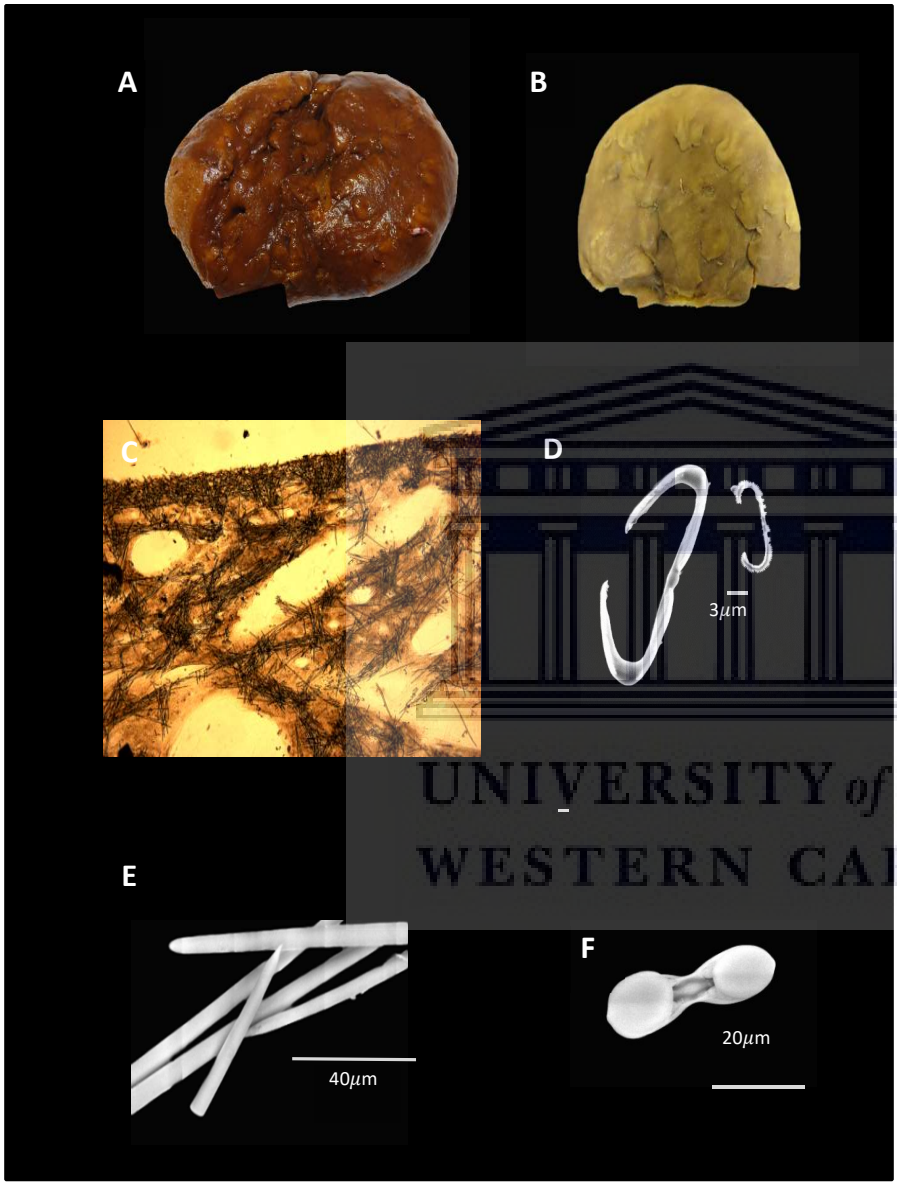

Figure 5: ***A*** **– In** ***situ. B – Ex Situ. C*** **– Skeletal Structure** ***D – Spiny Isochelae E -Anisoxea F – Placohelae in*** **two size classes**

Table 3. Spicule dimensions of *Guitarra* species and specimens

Species	Type Location	Aniso-oxea	Styles	Placochelae (Large)	Placochelae (Small)	Bipocilla like Isochelae	Spiny Isochelae
***Guitarra fimbriata* Carter, 1874 as measured by Burton 1929**	Celtic Seas (Northern Hemisphere)	310 x 5		90-10	40-50		10-11
***Guitarra flamenca(* Carballo & Uriz, 1998)**	Shark Island, Namibia (Southern Hemisphere)		252-380 x12	74-95	3	11-18	
***Guitarra flamenca* Carballo & Uriz, 1998 sensu Samaai & Gibbons, 2005)**	South Paw, Oudekraal, (Southern Hemisphere)	253-337x 12	69-90	55-69	18		
***Guitarra indica (* Dendy, 1916**	Western India (Northern Hemisphere)		266 x7	41	No measurements given	10	
***Guitarra fimbriata var. indica* (Dendy, 1916 sensu Lèvi 1963)**	Agulhas Bank (Southern Hemisphere)	200-250x2-8		40-50	30	10-15	
***Guitarra panema* sp.nov**	Agulhas Bank (Southern Hemisphere)	204-349		45.6-48	28.8-38.4		12

Order Tethyida Morrow & Cárdenas, 2015

Family Tethyidae Gray, 1848

Genus Tethya Lamarck, 1815

Definitions. Tethyidae with a spherical, sometimes hemispherical body with a well-developed cortex, Main skeleton formed by strongyloxea bundles radiating from the center of the sponge and bristling, generally flattened, sometimes conical, tubercles on the surface. Main megascleres are usually strongyloxeas, interstitial (auxiliary) megascleres are often styles. Megasters are spherasters or oxyspherasters. Microsters are tylasters, strongylasters or oxyasters, normally with spined rays, with polyrhabds in some species (Sarà., 2002).

Tethya oxyspherasterar **sp. nov.**

(Figure 6; Table 4)

Etymology. Species is names *oxyspheraster* as it is set apart by the two size categories of oxyspheraster compared to the other South African species who only have one.

Material examined. TS 1708, 12 Mile Bank Agulhas Bank, South Africa (35,0303º S; 20,9203º E), depth of 39-56m,-12/8/05 by means of dredge at a depth of 39–56 m on board the RV Ellen Khuswayo by means of dredge, voyage number 46.

Comparative material examined. *Tethya rubra* (Samaai & Gibbons., 2005), SAM-4900 (fragment BMNH 1997.5.12.124) (Ts 180), South Paw, Oudekraal (33°56'S, 17°21'E), depth 12 m, collected by P. Coetzee, 23 April 1996.

Locality. 12 Mile Bank, Agulhas Bank, South Africa

Description. Globular to sub-spherical sponge, 60 × 50 × 25 mm diameter, attach to substratum and spicules protruding. Surface hispid and tuberculate, with few oscules visible, 1–2 mm in diameter. Well-pronounced cortex, ~5 mm thick. Texture firm, fibrous and velvety to the touch, not compressible. Colour in life, reddish-orange externally with orange choanosome; in preservative, externally pale-orange and internally yellow-beige. A very distinct core is visible *in situ* with a slight colour differentiation.

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Skeleton. Specimen has radial tracts of strongyleoxeas starting at the base and fanning out from the choanosome out towards tuberculate surface. Choanosome, cortex and surroundings often filled with auxiliary megascleres as well as microsclere. Cortex is 1640 µm – 1800 µm wide.

Spicules. Megascleres. Anisostrongyloxea, smooth, straight, thickest centrally with proximal end strongylote, distally hastate: (I) Large size, 734.59 µm (614.4–8 44.4) x 48 µm. 10 x, n=25. (II) Medium size, 384.8 µm (268.8–480) × 32 µm. 10x, n=25. **Microscleres**. (I)Large Oxyspheraster,50.1 (38.4–60), 40x, n=25; (II) Medium Oxyspheraster, 35.42 (24–50.4), 40x, n=25; Micraster, 7.37 (2.4-14.4), 10x, n=25

Substratum, depth range and ecology. Specimen was found on hard substratum at a depth of 12 m. Only one specimen.

Remarks. *Tethya* contain two knows species from South Africa; *Tethya magna* Kirkpatrick, 1903 and *Tethya samaaii* Ribeiro & Muricy, 2011 from Natal and Ouderkraal respectively. *Tethya oxyspeheraster* sp.1 differs from the other South African species having **two** size catogories of oxyspheraster and one size category of micraster. *Tethya oxyspheraster* **sp. nov.** does not have any tylasters or spheraster present. Based on the above, *Tethya* sp. is considered a new species. Morphologically the species also differs from T. Aurantium in terms of colour and shape

Key diagnostic characters.

- Globular to sub-spherical sponge
- No tylasters or chiasters
- Two catogories of oxyspheraster

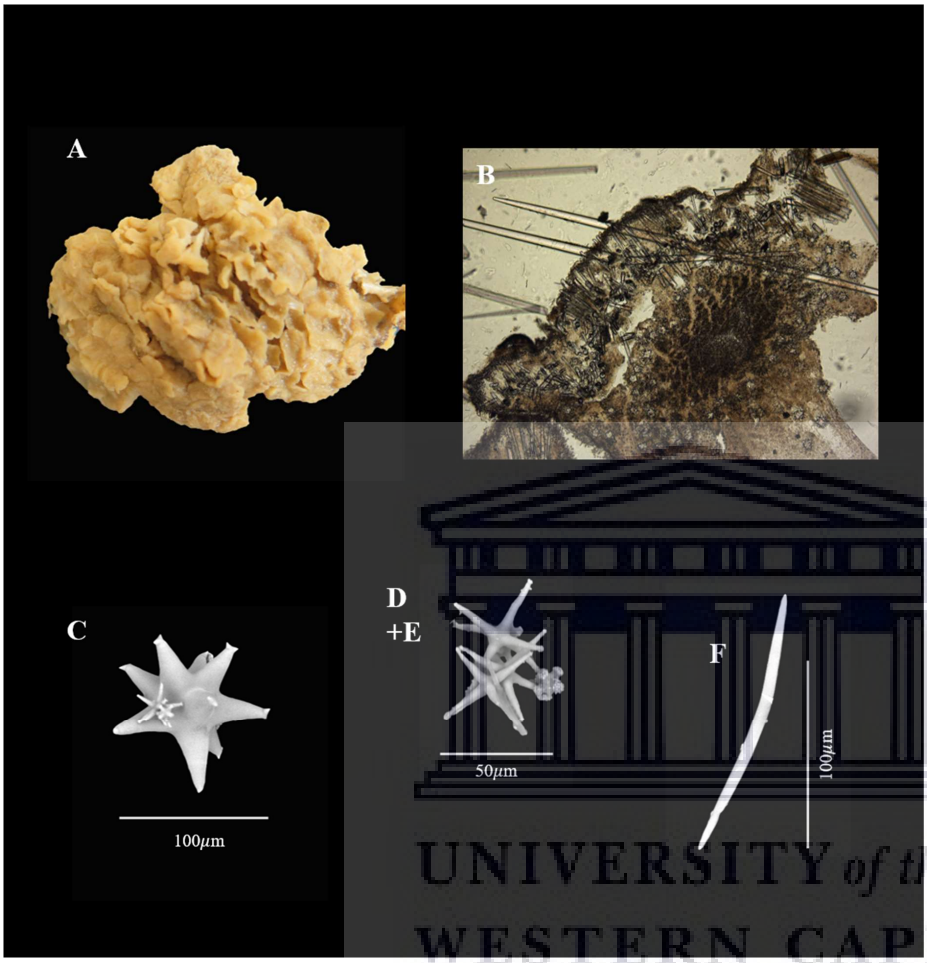

Figure 6: A – *In Situ.* B – Skeletal Structure. C – Large oxyspheraster. D – Medium oxyspheraster. E – Micraster. F- Large and medium Anisostrongyle

Table 4 Morphological characters of species of South African *Tethya*

Species	Morphology	Colour	Location	Spicule Types
***Tethya magna* (Kirkpatrick, 1903)**	Oval, spherical Polygonal plates Cortex with intercortical cavities No spicules visible	Purple-brown Cortex-silver Pith-yellow	Cone Point, KZ Natal	Strongyloxea Spheraster Asters chiasters
***Tethya samaaii* (Ribeiro & Muricy, 2011)**	Massive, spherical, sub spherical or irregular globular Rooting or attachement at base Surface mammilate, tubelate or polygonal Elevations separated by channels or porous grooves Surface hispid Texture firm, barely compressible Prominent cortex	In situ, red-orange Choanosome orange Infixation, pale orange Internally yellow to beige	South Paw, Oudekraal Coral Gardens	Strongyloxea Anisostrongyleoxea Small Anisostrongyle Tylasters Small Oxyspheraster Micro Oxyspheraster
***Tehya oxysperastera* sp. Nov**	Surface hispid and tuberculate. Few visible oscules (1-2 mm diameter). Well-pronounced cortex, ~5 mm thick. Texture firm, fibrous and velvety to the touch, not compressible.	In situ, red-orange Choanosome orange In fixation, pale orange Internally yellow to beige	12 Mile Bank	Large Anisostrongyleoxea Medium Anisostrongyleoxea Micraster Large Oxyspheraster Medium Oxyspheraster
***Tetgya Irisae*(Sorokins, ekans,yang & Cardenas, 2019)**	Surface tubercules Well developed cortex canals	Strongyleoxeas Subtylostyle Long rayed Oxyspheraster Acanthooxyspheraster	Australian Commonwealth Marine Reserve	White to grey

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Order Poecilosclerida Topsent, 1928

Family Latrunculiidae Topsent, 1922

Genus *Latrunculia* Du Bocage, 1869

Definition. Encrusting or cushion sponges with trumpet-shaped or cylindrical oscules and convex or concave areolate pore fields; colour in life typically blackish green, khaki to olive green. Choanosomal architecture consists of megascleres arranged in an irregular broad-meshed reticulation of wispy tracts that lack spongin reinforcement. Megascleres are frequently slightly irregular and wavy anisostyles, occasionally with acanthose heads. Ectosomal skeleton composed of an oblique to tangential layer of megascleres, above which, is a palisade of anisodiscorhabd microscleres with their basal portions buried in the outer ectosome. Microscleres are anisodiscorhabds (modified from Samaai et al. 2012).

***Subgenus Latrunculia (Biannulata)* (Samaai, Gibbons & Kelly, 2006)**

Definition. *Latrunculia* species in which the anisodiscorhabd microscleres have only two distinct whorls of projections around the shaft, the median and subsidiary whorls, between the undifferentiated manubrium and basal whorl, and the undifferentiated apical whorl and apex (after Samaai et al. 2012).

***Latrunculia (Biannulata) kerwathi* Samaai, Janson & Kelly, 2012**

(Figure 7)

Material examined. SAM-A24719, cross referenced with TS 1420, 45 Mile Bank South Africa (35,3494º S; 20,6081º E), 21/3/05, sample collected by dredge, depth 85 m on board the Ellen Khuswayo.

Comparative material examined. *Latrunculia* (*Biannulata*) *gotzi***,** SAM-A24718: Alphard Banks, Agulhas continental shelf, about 60 km from Cape Agulhas, Western Cape, South Africa, 35.054° S, 20.919° E, RV *Ellen Khuzwayo*, collected on SCUBA, 25 March 2009, 41 m [cross reference numbers TS 1482 (3557/03). *Latrunculia (Biannulata) Algoaensis,* SAM-A24720: Bell Buoy Reef, Algoa Bay, off Port Elizabeth Eastern Cape, South Africa, 33.7843° S; 25.8216° E, collected by Shirley Parker-Nance on SCUBA, 22 March 2010, 22–30 m [cross reference number RU-510-3]. Latrunculia *lunaviridis***,** BMNH 1996.7.3.6: Ouderkraal, Southpaw, Cape Town, South Africa, 33.8159'S, 18.6322'E, collected by P. Coetzee, University of Port Elizabeth, 25 February 1996, 17–20 m, a fragment of the type has been deposited in the South African Museum, Cape Town (SAM H-4960). Latrunculia *microacanthoxea,* BMNH 1996.7.3.1: Rheeders Bay, Tsitsikamma National Park, South Africa, collected by P. Coetzee, University of Port Elizabeth, 15 February 1995, 28 m.

Description. Thinly encrusting sponge, 15 mm long × 10 mm wide × 8 mm thick, with numerous low-lying crater shaped areolate porefields 5 mm in diameter; oscules volcano shaped and conical 1 mm wide, 0.2 mm high. surface undulating but smooth. Texture firm, slightly granular but does not breaks easily and hard to tear. Colour in life dark greenish brown, in ethanol light green.

Skeleton. The skeleton is highly cavernous with ~~an irregular tract~~ of megascleres. Tracts of mesh like grids reaching up from the choanosome of the sponge to the surface. Megascleres and microscleres are found at random ~~between the~~ choanosome and the tracts. Anisodiscorhabds forms~~s~~ a thin palisade like layer in the ectosome.

Spicules. Megascleres. Styles, smooth, slightly sinuous, centrally thickened, sparsely and finely spined along the proximal end of the shaft, often polytylote; 327 (346–394) x 11 (10–12) µm, 10x, n=50. Microscleres. Anisodiscorhabd, 51 (46–57) µm, 40x, n=50.

Substratum, depth range and ecology. Single specimen found at 85 m on the hard reef substratum at 45mile bank

Geographical distribution. 45-Mile Bank, Agulhas Bank, South Africa.

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Remarks. This species was described by Samaai et al. (2012). Only a small piece was collected from 45 Mile Bank. More surveying of the area is required to determine if 45 Mile represents a niche habitat for the species. Sponge is really small and thinly encrusting, and can be easily missed when dredging or diving.

Key diagnostic charaters

- Irregular tract of megascleres
- Size class of anisodiscorhabd

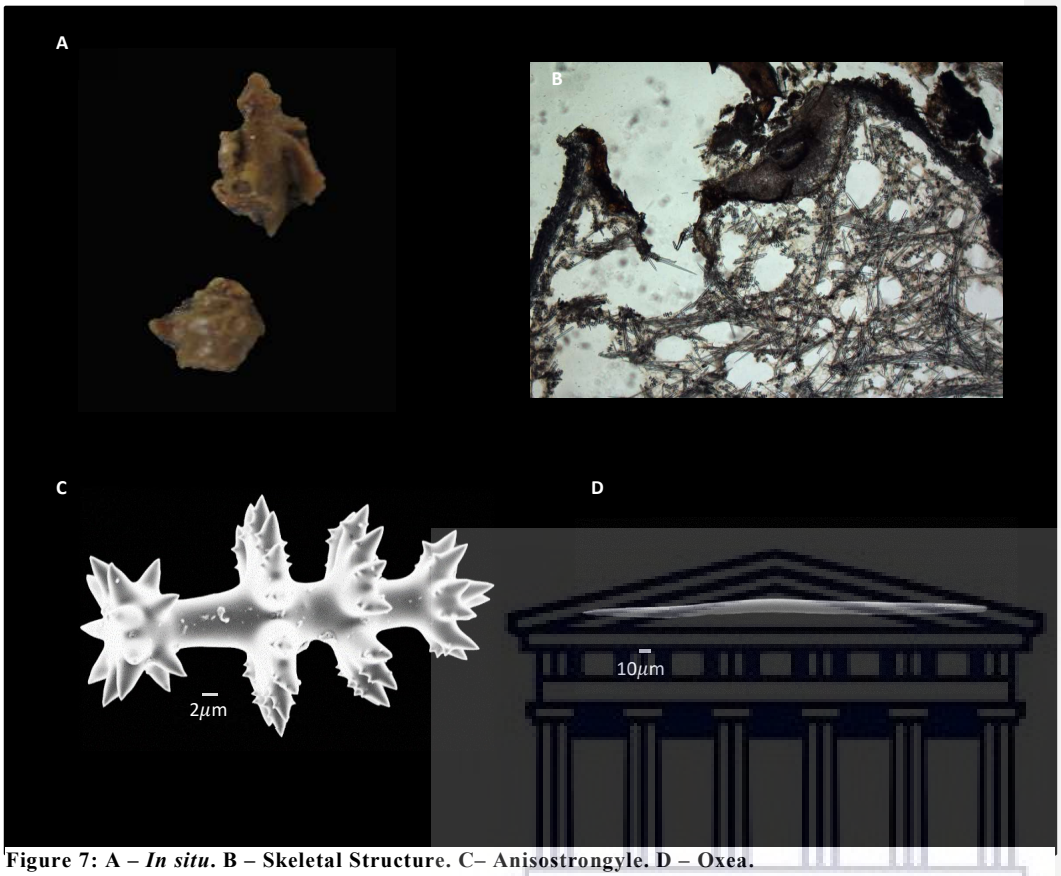

Figure 7: A – *In situ*. B – Skeletal Structure. C– Anisostrongyle. D – Oxea.

Order Poecilosclerida Topsent, 1928

Family Latrunculiidae Topsent, 1922

Genus Strongylodesma Levi, 1969

Definition. Latrunculiidae with a packed ectosome of paratangential strongyles and, in most species, a clear subectosomal band of collagenous mesohyl. Choanosomal architecture consists of megascleres arranged in an irregular, large-meshed reticulation formed by insubstantial or heavy tracts of megascleres, which are smooth or terminally spined strongyles, in a single, broad, size category, many of which have characteristic 'shepherd's crook' modifications. Species contain pyrroloquinoline alkaloids such as batzellins, isobatzellins, discorhabdins and their derivatives (Samaai et al. 2009).

***Strongylodesma agulhasiensis* sp. nov.**

(Figure 8; Table 5)

Etymology. ~~Most *Strongylodesma* species only have one single dischorahb or strongyle. Most also look the same morphologically. Species thus called *Strongylodesma agulhasiensis* as reference to where it was found.~~

Material examined. TS 1551, Alphard Bank, Agulhas Bank, South Africa (35,0374º S; 20,8660º E), depth of 27m on 24/3/05 b y means of~~,~~ SCUBA, collected by T Samaai, ~~depth 27 m,~~ on board the Ellen Khuswayo.

Locality. Alphard Bank, Agulhas Bank, South Africa

Description. Semi-spherical, surface smooth with numerous, randomly scattered cylindrical or volcano-shaped, thin-lipped oscules, 3–4 mm high and 2–3 mm wide, with internal dividing membranes. Areolate porefields smooth, fungiform, 1 mm high and 2–6 mm wide. Sand particles sometimes present on the surface of the sponge. Texture resilient, slightly compressible. Colour in life greenish-brown, interior brown; in preservative dark leather brown. The specimen is 45 mm long x 25 mm wide x 15 mm thick.

Skeleton. The choanosomal skeleton consists of a dense, meandering, irregular reticulation of wispy tracts of megascleres approximately 80–100 µm thick. In the deeper choanosome, the tracts are ill-defined but diverge towards the surface where they become more robust and plumose, tracts now 200–250 µm wide. Interstitial megascleres are common. The sub-ectosome is a dense paratangential feltwork of anisostrongyles approximately 320 µm deep, these protrude beyond the surface in a haphazard manner.

Spicules. Megascleres. Anisostrongyles, smooth, 348 (307–403) x 7.32 (7.2–9.6) µm. 10x, n= 50.

Substratum, depth range and ecology.Found on shallow hard sub stratum of 27 m.

Remarks. *Stronylodesma* has 3 prominent species in South Africa – *S. tsitsikammaensis, S. algoaensis* and *also S. aliwaliensis*. A fourth species is now added with *Strongylodesma agulhasiensis* **sp. nov.**: further investigation is needed to determine depth and habitat preference of species. For comparative differences see Table 5. The *Strongylodesma* sp. is considered a new species. Its different from the other known South African species in terms of colouration, spiculation and the size classes of the Anisostrongyle

Key diagnostic characters

- Distinct volcano shaped oscules
- Aerolate porefield
- Distinct old leather colour in ethanol
- Sand particles present on exterior

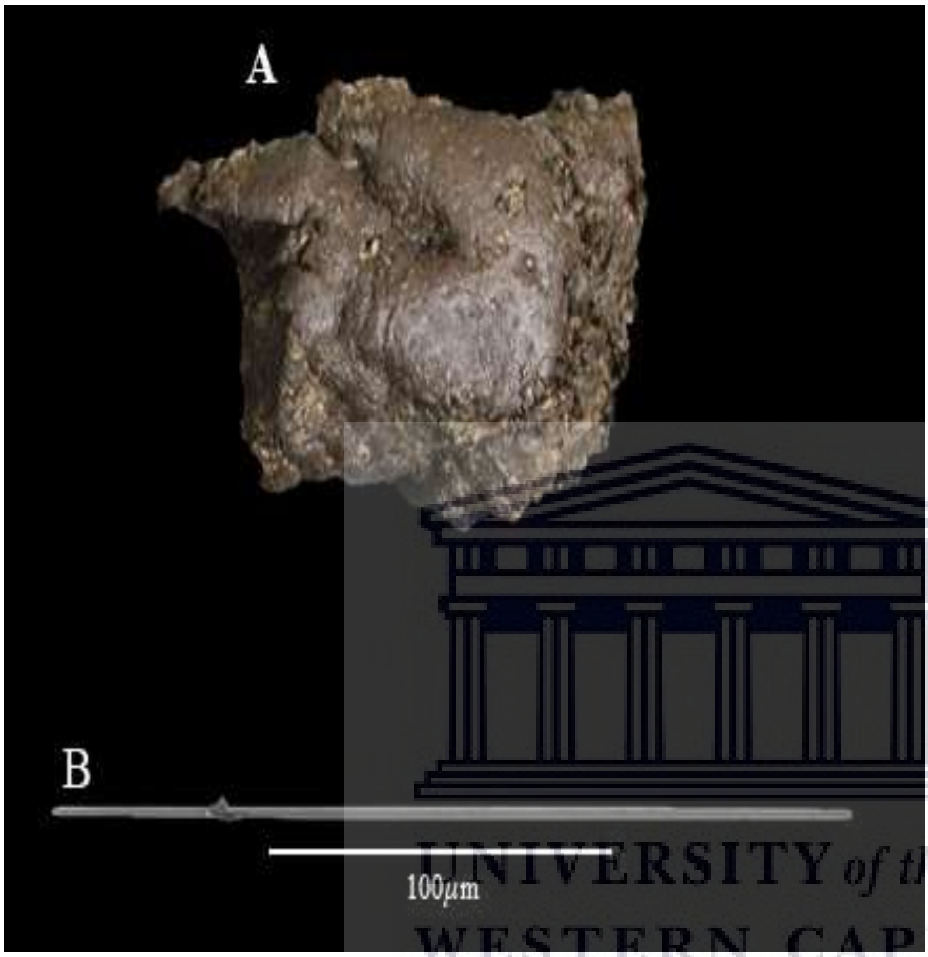

Figure 8: A- ***In situ*****. B – Anisostrongyle**

Table 5. Morphological characters of species of South African *Strongylodesma*

Species	Morphology	Colour	Location	Spicule Types
Strongylodesma tsitsikammaensis	Texture soft to medium No spicules visable Mushroom like papilla Oscules visable	Light green	Alphards Bank, Western Cape 12mile Bank, Western Cape 45mile bank, Western Cape	Anisostrongyle
Strongylodesma algoaensis	Firm, hard sponge Surface undulating but smooth Bigger oscules than *Tsitsikammaensis*	Dark brown to black	Alphards Bank, Western Cape Algoa bay, Eastern Cape	Anisostrongyle with distinct axial canal
Strongylodesma Aliewaliensis	Semispherical Very tough Convoluted tracts	Olive green	Aliwal Shoal, KZ Natal	Smooth anisostrongyles
***Strongylodesma agulhasensis* sp. nov.**	Cilindricalal Oscules Aerolate Porefields Surface smooth	Greenish- brown	Alphards Bank, Western Cape	Anisostrongyle s
***Strongylodesma Areolata* (Levi, 1969)**	Spherical or semi spherical Elevated, volcano like oscules	Brown- red brown	Vema Seamount, South Atlantic	Spined Strongyles
***Strongylodesma Tongaensis* (Samaai& Kelly, 2009)**	Encursting sponge No oscules Visible	Dark green	Tonga	Smooth strongyles

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Order Poecilosclerida Topsent, 1928

Family Myxilliidae Dendy, 1922

Genus *Myxilla* Schmidt, 1862

Definition. *Myxilla* has ectosomal tylote tornotes with soined apices and a choanosomal reticulation of styles, either smooth or spined in nature. Microscleres contain anchorate chelae with three teeth (Van Soest, 2012).

Subgenus *Myxilla* (*Myxilla*) Schmidt, 1862

Definition. Myxilla with isotropic skeleton, it contains acanthostyles of a single size class (Van Soest, 2002).

***Myxilla (Myxilla) simplex* (Baer, 1906)**

(Figure 9)

Synonymy.

Dendoryx simplex Baer, 1906: pg 22, Taf. II Fig. 7, Taf. V Fig. 20-25.

Lissodendoryx (Lissodendoryx) simplex (Baer, 1906): pg 22, Taf. II Fig. 7, Taf. V Fig. 20-25.

Myxilla simplex Stephens, 1915: 447. Burton 1936: 141; 1956: 130. Lévi 1963: 40. Uriz 1987: 61; 1988: 165.

Material examined. TS 1487, 45 Mile Bank Agulhas Bank, South Africa (35,3213º S; 20,8660º E), specimen was collected on 23/3/05 by dredge at a depth of 70 m on board the RV Ellen Khuswayo, voyage number 35.

Other material examined. SAM-H4925 (fragment BMNH 1997.5.12.27) (Ts 254), Oudekraal (33°59'S, 18°22'E), depth 6–8 metres, collected by T. Samaai, Z. Toeffie and G. Isaacs, 02/07/97.

Description. Thickly encrusting sponge, sometime hemispherical, 50 x 30 x 20 mm in diameter. Surface undulating but smooth, minutely hispid and covered in slime. Texture firm but soft, brittle and tear easily. Ostia not visible; oscules randomly scattered over the surface, 1 mm diameter. Colour in life shimmery orange-yellow, in ethanol grey. Sponge is covered in polychaets and hydroids.

Skeleton. Choanosome skeleton a more or less well-defined reneroid network of acanthostyles, with meshes uni-, bi- or poly- spicular. Primary and secondary fibres not clearly distinguishable. Ectosome with a dense, tangential layer of tornotes. Microscleres scattered throughout.

Spicules. **Megascleres.** Acanthostyles, proximally curved, tip abrupt, 219.79 µm (192 µm–240 µm), 40x, n=50. Tornotes (oxeas), 160.18 µm (122.4 µm–208.8 µm). 40x, n=50. **Microscleres.** Sigmata C and S shape, 17.22 µm (9.6 µm–21.6 µm), 40x, n = 50. Anchorate isochelae, 16.66 µm (9.6 µm–31.2 µm), 40x, n=50.

Substratum, depth range and ecology. On rocky substrata, epiphytic and epizootic. Associated with foliose and encrusting algae, and other sponges. Depth range 0–232 m.

Geographical distribution. The species has a perceived wide distribution ranging from Namibia to the Gulf of Aden. On the West Coast of South Africa up to Namibia where the species commonly occurs, it can be found at a depth of up to 277 m; in this case it was 70m.

Remarks. The present material conforms well to *Myxilla* (*Myxilla*) *simplex*, which has been described by Baer (1906), Lévi (1963), Uriz (1988) and Samaai and Gibbons (2005) from the west coast of South Africa. The specimen examined by Uriz (1988) off Namibia had more robust megascleres than those described here, while these are stronger than those of the littoral specimen described by Lévi (1963) and similar to the shallow water form described by Samaai and Gibbons (2005). It would therefore appear that water depth might have some role to play in this spicule nature (as *Crambe; chelastra &* Lévi, 1960), and cautions against discriminating species on the basis of spicule measurements alone. This species is one of the most common sponges along the west coast of South Africa, and its habit of growing in close association with other sponges may lead to the inclusion of abundant foreign material (chiefly other spicule debris). The difference is sizes with regards to water depth may require more investigation, yet it confirms to all the other requirements t0o conform to *Myxilla (Myxilla) simplex*

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Key diagnostic characters

- Both C and S shaped Chelae
- Chelae at 2 different size classes
- Occurs at depths up to 230 meters
- Yellow – orange colour

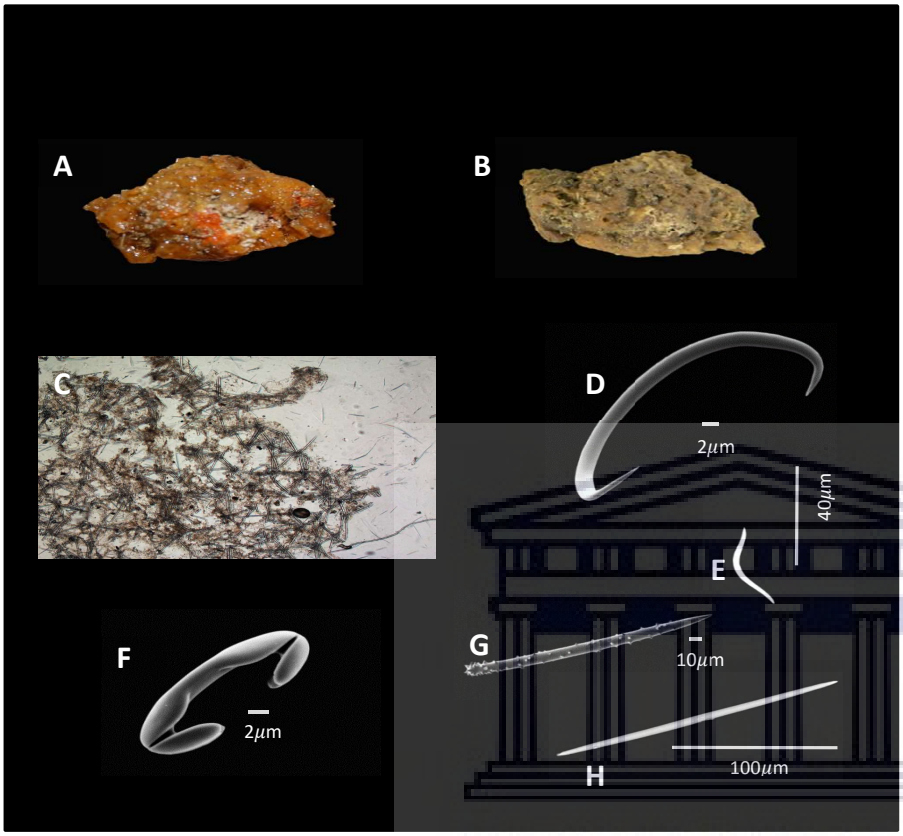

Figure 9: A - *In Situ*. B - *Ex Situ.* C - Skeletal Structure. D - Chelae E - Toxa F – Palmate Isochelae. G - Acanthostyles. H - Tornotes

Order Poecilosclerida Topsent ,1928

Family Microcionidae Carter, 1875

Subfamily Ophlitaspongiinae Laubenfeld, 1936

Genus *Echinoclathria* Carter 1885

Definition. Ophlitaspongiinae with the choanosomal skeleton consisting of a relatively homogeneous renieroid spongin fibre reticulation cored by smaller, smooth principal spicules and echinated by the same

spicules, and a vestigial radial extra-axial skeleton composed of larger principal spicules forming plumose brushes on the external surface (Hooper, 2002).

***Echinoclathria dichotoma* (Lévi, 1963)**

(Figure 10)

Synonomy.

Echinoclathria dichotoma (Lévi, 1963) Figs 4A, 35A–F
Ophlitaspongia dichotoma Lévi, 1963: 59.

Material examined. TS 1689, 12 Mile Bank Agulhas Bank South Africa, (35,0303º S; 19,9186º E) depth of 70m,~~12 Mile Bank Agulhas Bank South Africa,~~ specimen was collected on 23/3/05 ~~by dredge at a depth o~~f ~~70 m~~ on board the RV Ellen Khuswayo, voyage 35 by means of dredge~~.~~

Other material examined. SAM-H4913 (fragment BMNH 1997.5.12.112) (Ts 214), Oudekraal (33°59'S, 18°22'E), depth 15 m, collected by P. Coetzee, 22/04/1996 1996. BMNH 1997.5.12.84 (fragment Ts 488), Coral Gardens, Oudekraal (33°59'S, 17°23'E), depth 15 m, collected by G. Isaacs, Z. Toeffie and M.J. Gibbons, 03/02/1997. BMNH 1997.5.12.91 (fragment Ts 266), Justin's Cave, Oudekraal (33°58'S, 18°20'E), depth 15 m, collected by G. Isaacs, Z. Toeffie and M.J. Gibbons, T. Samaai, 10/04/1997.

Description. Arborescent and digitate; branches thick, cylindrical, 8 cm diameter, with rounded apical margins. Sponge dimensions 6 cm x 7 cm; oscules small, randomly scattered on lateral margins, 1–2 mm diameter; ostia 0.5 mm diameter flush with surface. Surface undulating but smooth, microscopically hispid. Texture firm, tough and resilient with medium compression. Colour *in situ* orange, in ethanol beige/yellow. No spicules are visible on the specimen.

Skeleton. Choanosomal skeleton irregularly reneroid, with slightly compressed, irregularly isodictyal axis, and with plumo- reticulate extra-axial regions. Axial skeleton not differentiated into primary and secondary transverse components; spongin fibres relatively thick 48-63 µm with 1–2 principal styles per tract, this produces a ~~ing~~ nearly regular reneroid skeleton, the~~whereas~~ longitudinal primary fibres are plumose, arborescent, producing radial tracts which diverge regularly forming plumoreticulate tracts (142–175 µm thick) there are ~~and~~ protruding ~~through~~ fibres in peripheral skeleton. Ectosome with principal styles, arranged peripherally to form a vestigial, plumose or radial, extra-axial skeleton; styles protrude through surface to distance of 200 µm, singly or in brushes. Subectosome distinct from ectosome, tangentially arranged layer of auxiliary styles in pauci or multi- spicular tracts.

Spicules. **Megascleres**. Styles short, thick, smooth, slightly curved proximally, tip abrupt, 282.24 µm (230.4 µm–364.8 µm), 10x, n=50. **Microscleres**. Toxas in two size categories, 92.53 µm (57.6 µm–133 µm), 40x, n=50.

Substrate, depth range and ecology. On rocky substrata, overgrowing other sponges, hydroids and algae. Depth range 15–110 m.

Geographical distribution. West and South coast of South Africa.

Remarks. Common sponge along the South African coastline. Species very closely conforms to Holotype by means of skeleton arrangement, spicules ~~as well as~~ colouration. Specimen is a fragment. ~~Upright nature of sponge allows for easy breakage when dredge is dragged over or across~~. Agulhas Bank Complex consists of reefs and mounds, which should present favourable conditions for the species. Specimen is slightly less "fuzzy" than other records. Reasoning for that will require further investigation. The specimen does fulfill the requirments and tick the boxed to be categorized as *Echinoclathria Dichotoma*

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Key diagnostic characters

- Deep red colouring
- Finger like growth
- "fuzzy" to the touch

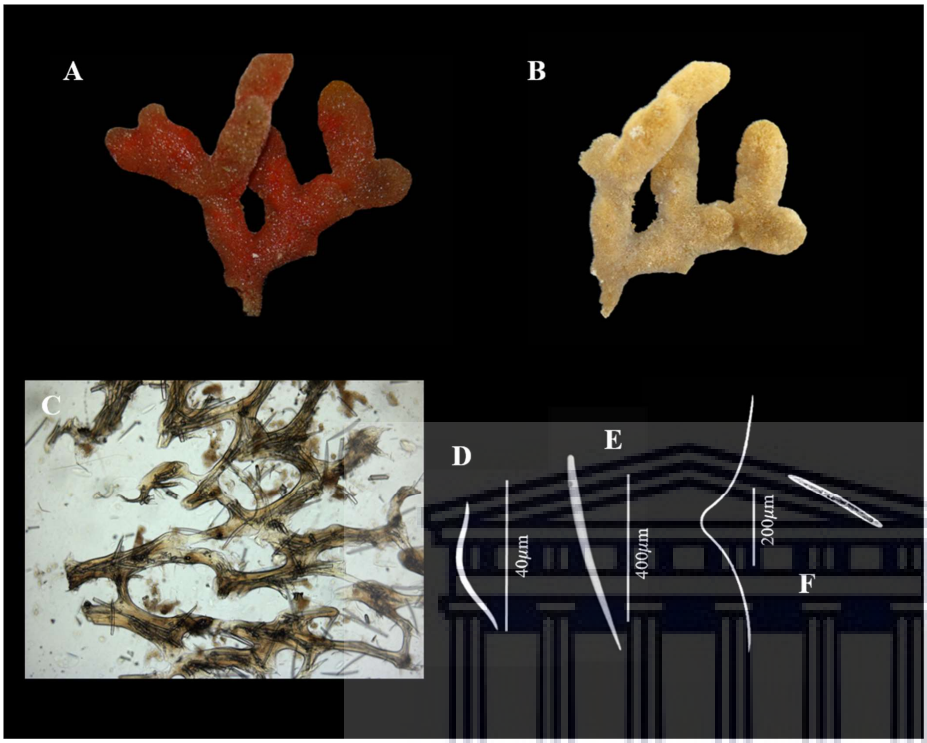

Figure 10. A – *In Situ*. B – *Ex Situ*. C – Skeletal Structure. D – Small Toxa. E – Style. F – Toxa

Order Poecilosclerida Topsent, 1928

Family Microcionidae Carter ,1875

Genus Antho Gray, 1867

Definition. Ophlitaspongiinae with choanosomal skeleton modified to a basal or axial renieroid reticulation of acanthose or occasionally smooth styles and/or strongyles, overlaying a plumose(-reticulate) subectosomal skeleton of smooth principal styles, with or without echinating spicules (Hooper, 2002).

Subgenus *Antho* (*Acarnia*), Gray, 1867

Definition. *Antho* with predominantly (acantho)tylostrongyles forming the renieroid skeleton, less often acanthostyles, and a special category of echinating acanthostyles overlap the main skeleton (Hooper., 2002).

***Antho (Acarnia) arboriaensis* sp.nov.**

(Figure 11; Table 6)

Etymology. Original specimen looked like a Christmas tree, latin word for tree is Arbor, hence the name *Antho (Acarnia) arboriaensis*.

Material examined. TS 1557, Alphards Bank Agulhas Bank, South Africa, (35,0374º S; 20,9203º E) at a depth of 46m on, specimen was collected 24/3/05 by dredge at a depth of 46 m on board the RV Ellen Khuswayo, voyage number 35.

Locality. Alphards Bank, Agulhas Bank, South Africa

Description. Sponge dimentions areThinly encrusting sponges, 75 mm high x 50 mm wide x 10 mm thick. Surface undulating, dimpled, but smooth and hispid. Oscules not visible; Ostia 1 mm in diameter clustered on the surface. Texture is soft, barely compressible, tears & breaks easily. Colour in life red orange, choanosome brighter orange; in ethanol beige. Epifauna present.

Skeleton. Choanosome with a basal isotropic reneroid reticulate skeleton of acanthostrongyles, in groups of 3–7; a secondary, dendritic plumose skeleton of smooth or slightly spined primary subtylostyles present, arising perpendicularly from the base. Ectosome heavily collagenous, with paucispicular plumose brushes of large, smooth styles andor acanthostyles that protrude through the surface, and arise from multispicular tracts of smaller, terminally spined styles in subectosome. Auxiliary megascleres of subectosome also scattered in deeper choanosome. Microscleres scattered throughout.

Spicules. Megascleres. Styles, thick, smooth, slightly curved proximally, 199.2 µm (184.8 µm–220.8 µm), 40x, n=50. Auxilliary styles, terminally spined, curved, proximally thin, andtornote tip abrupt, 166.29 µm (139.2 µm–206.4 µm), 40x, n=50. Acanthostrongyle, 287.78 µm (120 µm–177.6 µm), 40x, n=50. **Microscleres**. Toxas. 57.71 µm (31.2 µm –76.8 µm), 40x, n=50. Palmate isochelea, 12.71 µm (7.2 µm–19.2 µm), 40 x, n=50.

Substrate, depth range and ecology. *Antho* (*Acarnia*) arboriasp.1 was first recorded from hard flat reefs at a depth of 46 m. The substrate was unknown but epifauna in the form of polychaetes was noted.

Remarks. *Antho* (*Acarnia*) *arboriaensis* has a very distinct red colour and resembles the shape of a Christmas tree. The new species closely resembles *Antho (Acarnia) kellyae* Samaai & Gibbons, *2005* in spicultion but is differentiated by having only one class of ectosomal styles instead of three. *Antho (Acarnia) prima sensu* Lévi (1963) is completely different as it only has tylotes, styles and toxas. Hooper (1996)

published the monograph on Microcionidae, comparing to the species listed, Arboria is different in terms of colouration and spiculation

Key diagnostic characters

- 1 class of ectosomal styles
- Looks like a chrismas tree

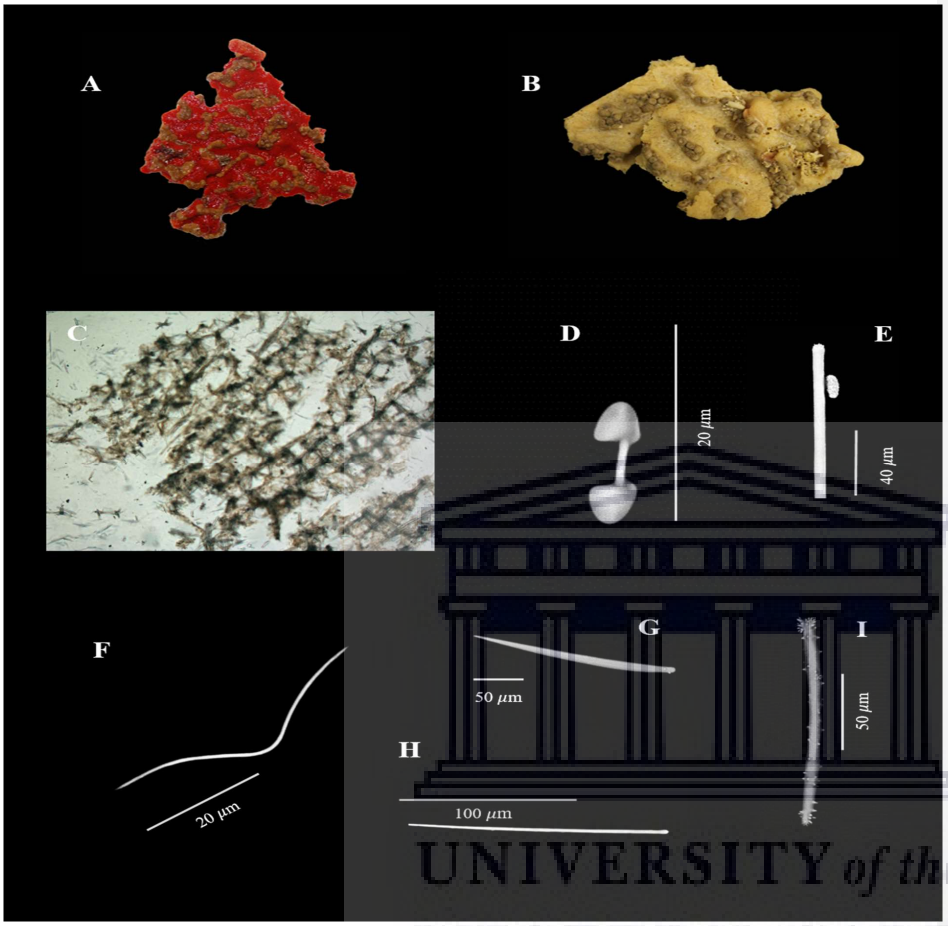

Figure 11: A – *In Situ*. B – *Ex Situ*. C – Skeletal Structure. D – Palmate Isochelae. E – Contamination. F – Toxa. G – Style. H – Auxillary Style. I – Acanthostrongyl

Table 6. Comparative table for *Antho (Acarnia)* spp.

Species	Morphology	Colour	Location	Spicule Types
Antho (Acarnia) kellyae	Encrusting, Bulbous to mammalate sponge	Bright red to orange Orange choanosome	Oudekraal	Thin Raphides, 2x Smooth Toxa Primary Isochelae

				Acanthostyles 2x Primary Styles 2x Auxillary Styles
Antho (Acarnia) arborensis sp. nov	Thinly encrusting sponge, undulating and dimpled surface	Bright red	Alphards Bank	Style Auxillary Style Acanthostyle Toxas Palmate Isochelea
***Antho (Acarnia) Bakusi* (Sim&Lee, 1998)**	Encrusting sponge Surface rough Oscules visible	Orange to red	East China sea	Styles Acabthostrongyles Acanthostyles Isochele Toxas

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Order Poecilosclerida Topsent, 1928

Family Tedaniidae Ridley & Dendy, 1866

Genus *Tedania* Gray, 1867

Definition. Tedaniidae with differentiated ectosomal and choanosomal megascleres.

Subgenus *Tedania* (*Tedania*), 1867

Definition. Smooth, relatively small styles, occasionally strongylote styles as structural megascleres and microspined tylotes as ectoso- mal megascleres. Distribution: predominantly in tropical and warm-temperate waters of all three ocean systems (Gray, 1867)

***Tedania (Tedania) scotiae* (Stephens, 1915)**

(Figure 12)

Synonomy.

Tedania scotiae Stephens, 1915: 448.

Tedania scottiae Lévi, 1963: 34. Day 1969: 21.

Tedania brondstedi Burton, 1936: 145.

Material examined. TS 477, Agulhas Bank South Africa, (35,0538º S; 20,9203º E) at a depth of 46m, specimen was collected 25/3/05 by dredge ~~at a depth of 46 m~~ on board the RV Ellen Khuswayo, voyage 35. (TS 1402,), Alphards Bank Agulhas Bank, South Africa (35,0538º S; 20,9203º E), at a depth of 46, specimen was collected on 25/3/05 by dredge ~~at a depth of 46 m on~~ board the RV Ellen Khuswayo, voyage 35. TS 1554, Alphards Bank Agulhas Bank South Africa (35,0538º S; 20,9203º E), at a depth of 46m specimen was collected on 25/3/05 ~~by dredge at a depth of 46 m~~ on board the RV Ellen Khuswayo, voyage 35. TS 1727, Alphards Bank Agulhas Bank South Africa (35,0702º S; 20,9203º E), at a depth of 35m specimen was collected on 9/8/05 by dredge ~~at a depth of 35 m~~ on board the RV Ellen Khuswayo, voyage 46. TS 1769, Alphards Bank Agulhas Bank South Africa (35,0702º S; 20,9203º E)~~,~~, specimen was collected on 9/8/05 by dredge at a depth of 39–42 m on board the RV Ellen Khuswayo, voyage 46.

Description. Thickly encrusting sponge with dimensions 50 x 40 x 10 mm. The surface smooth but undulating with soft texture with no visible spicules and oscules. Ostia small, evenly scattered, 0.1 mm in diameter; ectosome thick. Texture firm and compressible. Ostia present are 0.1mm in size and are randomly distributed on the sponge. The texture is soft and spongy, tears so-so, and is medium compressible. Colour *in situ* light orange, in ethanol pale-yellow to pink.

Skeleton. Choanosome with an irregularly tight-meshed, plumo- reticulate skeleton of smooth primary styles. Styles cemented by spongin, organized into broad primary and secondary fibres, not distinguished by size. Thick bundles of tylotes orientated at oblique angles, or perpendicularly, to primary and secondary fibres. Onychaetes scattered randomly throughout or aligned in tracts. Ectosome composed of a dense palisade of horizontally arranged tylotes that arise from choanosomal tylotes, occasionally forming plumose brushes.

Spicules. Megascleres. Styles styles smooth, slightly curved proximally, 135.13 µm (105.6 µm–153.6 µm), 40x, n=50. Tylotes smooth, with well-defined swellings at each end, 362.40 µm (259.8 µm–480 µm), 10x, n=50. **Microscleres.** Onychaetes straight, abruptly pointed proximally, tapering to a long point distally, finely spined, 40 µm (33.6 µm–52.8 µm), 40x, n=50.

Substrate, depth range and ecology. In shallow subtidal on hard rocky reef.

Geographical distribution. West and south coasts of South Africa

Remarks. The sponge conforms to the original descriptions of Stephens (1915) and Lévi (1963). Lévi (1963) described the species from a number of deep locations on the south coast of South Africa.

Key diagnostic characters

- ~~Shallow water sponge right through to deep locations~~
- Bright orange colour-slimy texture
- Choanosome with an irregularly tight-meshed, plumo- reticulate skeleton of smooth primary styles.

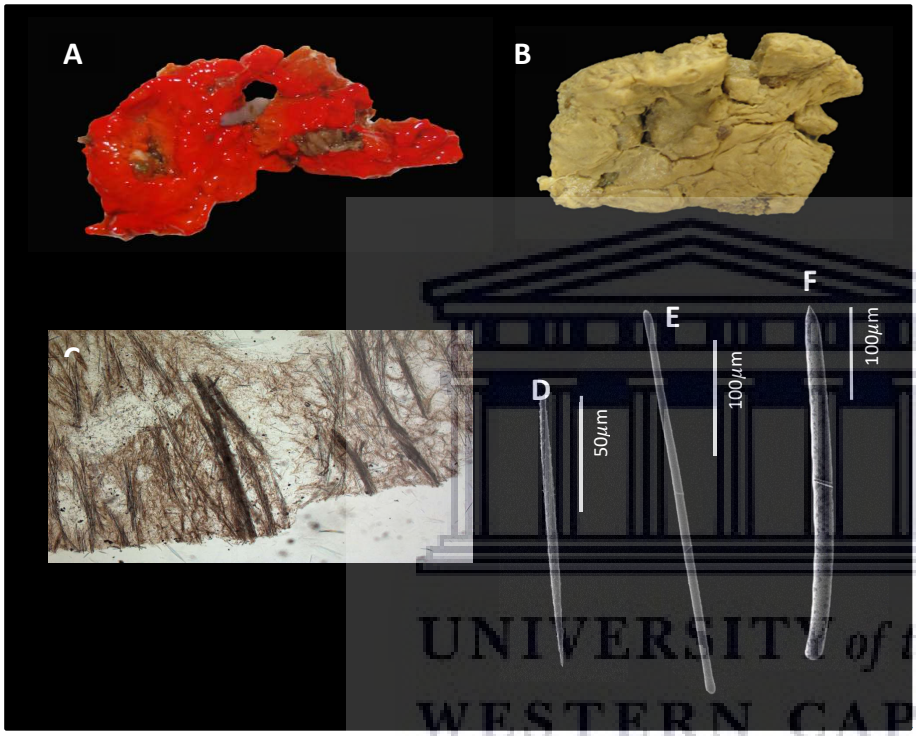

Fig 12. A – *In Situ*.B – *Ex Situ*. C – Skeletal Structure. D – Onychaete. E – Tylote. F – Style.

Order Verongiida Berquist, 1978

Family Aplysinidae Carter, 1875

Genus *Aplysina* Nardo , 1834

Definition. Aplysinidae characterized by the possession of fibers of only one kind, with foreign detritus and having a thick pith component. The fibers form a regular reticulum with large polygonal meshes and no specialized surface arrangement (Bergquist & Cook, 2002).

***Aplysina capensis* Carter, 1875**

(Figure 13)

Material examined. TS 1691, Alphard's Bank Agulhas Bank, South Africa (35,0342º S; 20,8606º E), at a depth of 26-30m ~~Alphard's Bank Agulhas Bank South Africa~~, sample was collected on 08/08/05 by SCUBA ~~at a depth of between 26–30m~~ on board the RV Ellen Khuswayo, voyage 46. TS 1742 (35, 0398º S; 20, 8647º E), at a depth of 17-25m at 45 Mile Reef Agulhas Bank South Africa, sample was collected on 08/08/05 by SCUBA ~~at a depth of 17–25m~~ on board the RV Ellen Khuswayo, voyage 46.

Description. Massive, mound-shaped sponge 40 x 55 x 35 mm in size. Surface undulating but smooth. Oscules visible ranging from between 1–2mm on the surface, randomly distributed. Texture is soft and spongy, but difficult to tear or break. Colour *in situ* purple; in ethanol purple.

Skeleton. Choanosome with a delicate and irregular network of spongin fibers. They have a bark often dark purple but maybe pink in light, Cellular cavities formed by bark are maintained between the fibers of the branches. Colour is not confined to the cells and the entire mass can appear purple. Foreign matter or bodies are very common inside sponge.

Substrate, depth range and Ecology. Specimen found in shallow waters (17-25 m) on rocky substrata. Carter (1881) stated that the species is contaminated with so many spicules from other species that it has to live in close proximity to many other sponge species.

Geographical distribution. Port Elizabeth and Alphard Bank, South Coast of South Africa.

Remarks. Specimens are known for very often having foreign spicules embedded within; they are unintentionally obtained by living in closee proximity to other sponges. Carter (1881) stated that specimens were found on hard objects. These specimens conform to Carter's (1881) original description.

Key diagnostic characters

- Shallow water specimen
- Deep purple colour
- Foreign bodies common inside sponge and incorporated into fibres

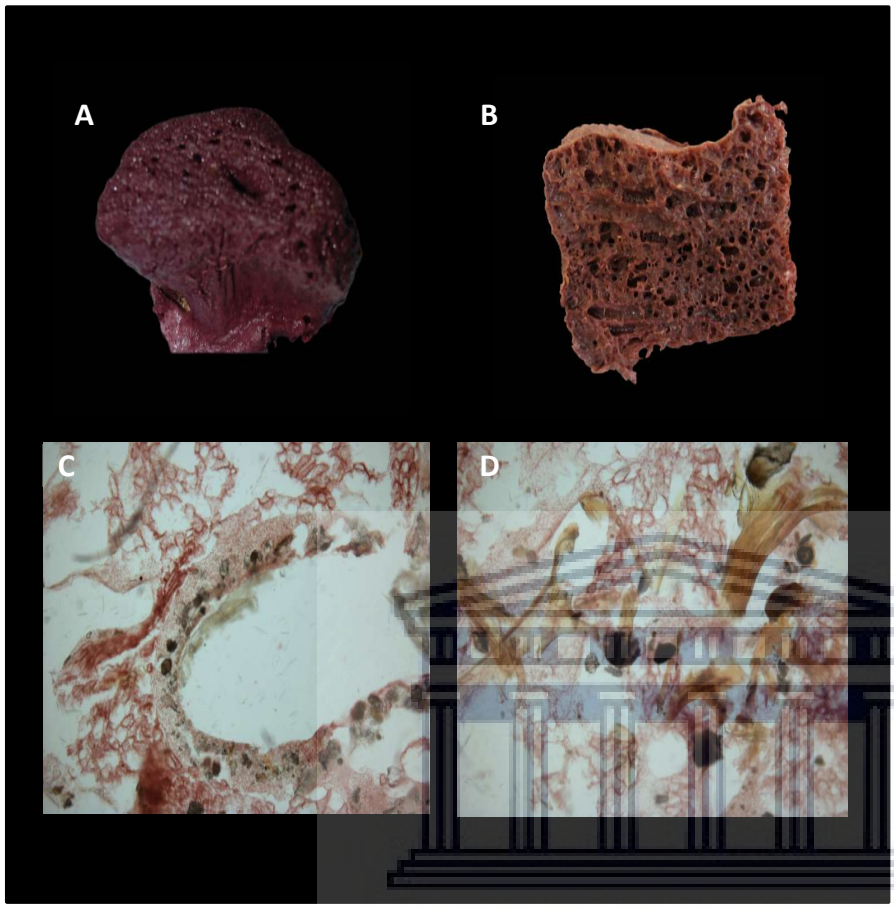

Fig 13. A – *In Situ*. B – *Ex Situ*. C – Skeletal Structure. D – Skeletal Structure.

CHAPTER 5

DISCUSSION

The sponge fauna occurring over soft sediments on the Agulhas Bank is relatively well known due to the annual research trawl surveys conducted by various South African government departments (Maduray, 2014; Atkinson and Sink, 2018), and more than 111 species have been described or are known to occur there. Although Alphard Bank, 72 Mile Bank and 45 Mile Bank form part of the newly declared Agulhas Bank MPA and are important nursery or spawning sites for a number of line-fish species (Hutchings et al. 2002), there has been a lack of directed scientific research, especially on the diversity and community structure of benthic invertebrates and habitat mapping. As a consequence, the current study represents the first extensive survey directed at the more poorly known hard reefs on the Agulhas Bank.

The dominant orders recovered here were Poecilosclerida (42 spp.), Tetractinellida (20 spp.) and Axinellida (12 spp) and together accounted for 66% of all species. The orders Suberitida and Haplosclerida were also relatively well represented with six and seven species respectively, making up a combined 11,7% of total species. The orders Biemnida, and Clionaida were each represented by two species, whilst the orders Trachycladida, Chondrillida and Dictyoceratida had but a single species: the order Polymastida had four species and Tethyida, Verongida had three species each. Both the classes Calcarea and Homoscleromorpha were represented by one order and two species. In terms of families, Ancorinidae (7 spp.), Geodiidae (7 spp.), Halichondriidae (7 spp.), Axinellidae (10 spp.) and Microcionidae (13 spp.) were dominant, and the genera *Stelletta (*5 spp*.), Tedania* and *Haliclona* (6 spp. each) and *Clathria* (9 spp.) were important, both in by number of species and frequency of occurrence in the samples. Only seven species were common to four or more study sites (*Axinella* sp.1, *Ptilocaulis* sp.1, *Geodia megastar*, *Geodia ovifractus*, *Guitarra* sp.1, *Hemiasterella vasiformis*, *Pachastrella caliculata*). When compared to Maduray (2014), the Agulhas Bank yielded a much higher species diversity than both west coast and south coast respectively. Maduray (2014) had more specimens but it's commonly known that species diversity increases from west to east coast (Samaai, 2006). The incredible diversity at Agulhas Bank can be due to the fact that the region has both cool and warm temperate waters (Samaai, 2006).

Perhaps not surprisingly, the composition of the communities recovered over hard substrata on the Agulhas Bank differ from those recovered on softer substrata. The vast majority of specimens collected were found on hard substratum.

But the sponge communities recorded on the different reefs/banks here were not uniform in composition. The observed richness by species and genera at each reef/bank varied, and as to be expected ~~and~~ there was a very significant relationship between patterns of richness at the different levels of taxonomic

resolution (R^2=0.97; p <0.05; DF = 4) (Figure 14). At the species-level of analysis, the nmMDS plot shows three clear clusters comprising 6 Mile Reef and Martha's Bank, and the balance of sites, with 72 Mile Reef and 12 Mile Bank being more similar to each other than either is to a group comprising Alphard's Bank and 45 Mile Reef (Figure 15). Whilst the latter two groups are also evident if the data are analysed at the genus level, it is also clear that communities at 6 Mile Reef are more similar to the other reefs/banks than they are to those of Martha's Reef, and the 2Stage analysis reveals a correlation value of ~0.8 between the two similarity of matrices.

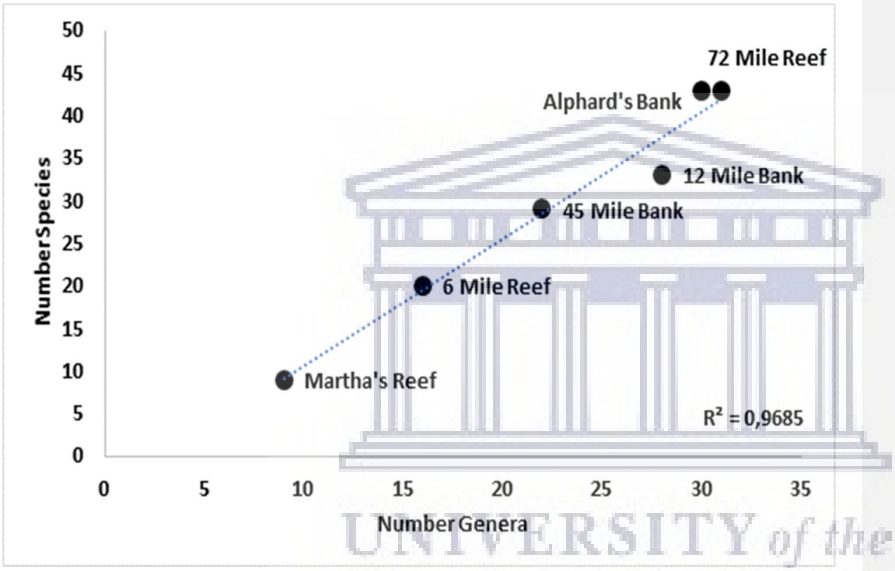

Fig 14. - Number of species and genera per site

The results of the DistLM reveal similar patterns, whether the data are analysed by species or genus, though a greater amount of total variation is explained with the latter than the former (Figure 15; Table 7). In the marginal tests it is clear that the number of samples collected at each site plays a significant role in influencing community composition, and it and longitude are the only factors selected in the sequential tests. In other words, it is very difficult to make meaningful observations regarding the communities on each bank/reef because of sampling artefacts, and the relationships between sampling effort and richness are strong. Indeed, there is no asymptote to these relationships, which suggests that the number of species to be found in the area is likely to be higher still (van Dooren, 2016).

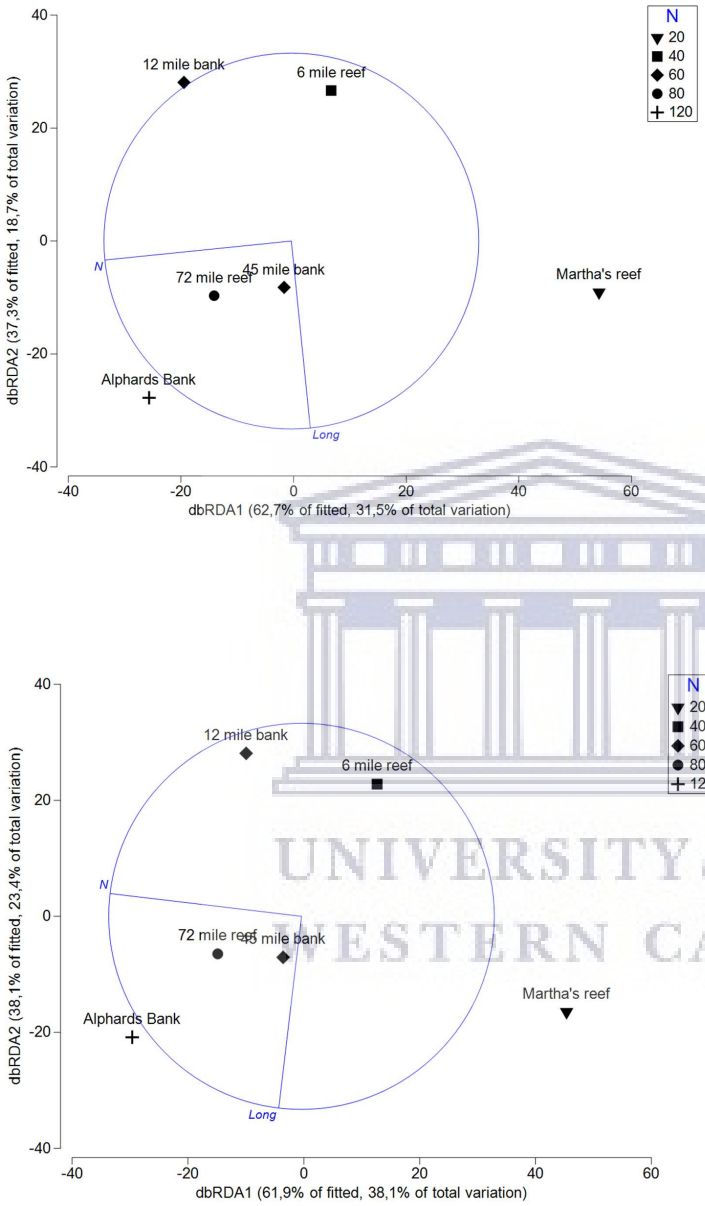

Figure 15. - DistLM plots of Agulhas Bank sites

Table 7. - 2 sets of DistLM analysis

Marginal Tests							
Variable	SS (trace)	Pseudo-F	*P*	Prop.			
Depth	2600	1,626	0,194	0,289			
N	3410,9	2,443	0,003	0,379			
Distance	3073,5	2,076	0,026	0,342			
Latitude	1983,5	1,132	0,375	0,221			
Longitude	2261,8	1,344	0,268	0,251			
Sequential Tests							
Variable	Adj R^2	SS (trace)	Pseudo-F	*P*	Prop.	Cumul.	res. df
+N	0,224	3410,9	2,443	0,001	0,379	0,379	4
+Long	0,359	2123,6	1,841	0,162	0,236	0,615	3

The results show that the sponge fauna of the hard reefs on the Agulhas Bank is diverse and heterogeneously distributed. The large number of new records and range extensions highlight a lack of knowledge and underscore the fact that there likely to be a number of still-to-be described species. Although these habitats are unseen, they are nevertheless subjected to various forms of human exploitation including intensive commercial and recreational fishing, mining, oil and gas operations and dumping (Sink et al. 2019). In South Africa, our lack of knowledge about deep hard ground habitats is problematic because these habitats are exploited to a greater extent than their shallow counterparts (Garratt et al. 1993) and as returns diminish inshore, fishing pressure is increasing in deeper habitats (Penney et al. 1999, Mann, 2000). This lack of knowledge in the face of increasing exploitation/development has long been recognised as an impediment to the management of biodiversity (Gibbons *et al.* 1999, Lombard *et al*. 2004, Griffiths et al. 2010; Sink et al. 2012; Sink *et al,* 2018), yet it is still subject to insufficient funding.

At Present South Africa is protecting a number of deep-sea benthic biodiversity and benthic fishery habitats through the declaration in 2019 of 20 new offshore MPAs, which has brought overall protection of the country's EEZ to 5% (National Gazette no 42478). MPAs are being designed using the systematic conservation planning approach (Majiet et al. 2013), which make priorities based on threats, vulnerability and uniqueness (Sink, 2011). Unfortunately, the information needs of managers considerably exceed existing knowledge, especially for the outer continental shelf and continental slope, and decisions are frequently made based on expert opinion, often using proxies for biodiversity (Sink, 2011). Hard information is

desperately needed in order to validate and support these newly erected MPAs, including the extension of the Agulhas Bank MPA.

Sponges represent a useful indicator of benthic biodiversity, because being sessile they are easily sampled and because they are generally large, they provide habitat and structure for other organisms. The data presented here suggest that the diversity of sponges on the banks and reefs of the Agulhas Bank is high, that the regions supports a number of previously unknown species with untapped metabolites, and that the diversity is likely to be higher still. Being filter feeders, sponges impact surrounding waters, but are also greatly impacted due to climate anomalies (Coma et al. 2009, Di Camillo & Cerrano, 2015).

Supplementary Table 1. Species presence and absence list

Species	12 mile bank	45 mile bank	6 mile reef	72 mile reef	Alphards Bank	Martha's reef	Still Bay	Struisbaai	Tsitsikamma MPA
Acarnus claudei	1	0	0	1	3	0	0	0	0
Amorphinopsis sp. 1	1	1	0	0	0	2	0	0	0
Ancorina nanosclera	1	0	0	0	0	0	0	0	0
Antho (Acarnia) kellyae	0	0	0	0	2	0	0	0	0
Antho (Acarnia) Arboria~~Panem~~ sp.nov	0	0	0	2	0	0	0	0	0
Antho (Acarnia) sp. 2	0	0	0	0	1	0	0	0	0
Aplysilla sp. 1	0	0	0	0	0	0	0	1	1
Aplysina capensis	0	2	0	1	4	0	0	0	0
Aplysina sp. 1	0	0	0	0	3	0	0	0	0
Aplysina sp. 2	0	0	0	0	1	0	0	0	0
Asbestopluma (Asbestopluma) sp. 1	1	1	0	1	0	0	0	0	0
Axinella natalensis	0	1	1	0	0	0	0	0	0
Axinella sp. 1	1	1	1	1	2	0	0	0	0
Axinella sp. 2	1	0	0	0	1	0	0	0	0
Axinella weltnerii	3	0	0	1	0	0	0	0	0
Biemna anisotoxa	0	0	0	2	0	0	0	0	0
Biemna megalosigma	1	0	0	0	2	0	0	0	0
Ceratopsion microxephora	1	0	0	0	0	0	0	0	0
Chondropsis isimangaliso	0	0	1	0	0	0	0	0	1
Clathria (Clathria) dayi	0	1	0	0	0	0	0	0	0
Clathria (Clathria) sp. 3	0	0	1	0	0	0	0	0	0
Clathria (Clathria) zoanthifera	0	0	0	1	0	0	0	0	0
Clathria (Isociella) sp. 1	2	0	0	1	0	0	0	0	1
Clathria (Thalysias) hooperi	0	0	0	0	1	0	0	0	0
Clathria (Thalysias) lissoclada	0	2	0	0	0	0	0	0	0
Clathria (Thalysias) nervosa	0	0	0	0	1	0	0	0	0
Clathria (Thalysias) oxitoxa	0	1	1	0	4	0	0	0	0
Clathria(Axosuberitis) sp. 1	1	0	0	0	0	0	0	0	0
Clathrina sp. 1	0	0	0	0	0	0	0	0	1
Cliona grandis	0	0	0	0	0	1	0	0	0
Desmacidon ectofibrosa	0	0	0	0	1	0	0	0	0

Dragmacidon sanguineum	0	0	1	2	0	0	0	0	0
Echinoclathria dichotoma	1	0	0	0	0	0	0	0	0
Ectyonopsis flabellata	0	0	0	1	0	0	0	0	0
Ectyonopsis pluridentata	1	0	0	0	0	0	0	0	0
Erylus ~~amorphus~~amorphous	0	1	0	1	0	0	0	0	0
Erylus gilchristi	0	2	0	0	0	0	0	0	0
Geodia ~~amorphus~~amorphous	0	0	0	1	0	0	0	0	0
Geodia ~~megaster~~megastar	0	2	1	6	1	0	0	0	0
Geodia ovifractus	2	3	3	2	1	0	0	0	0
Geodia perarmata	1	3	0	0	1	0	0	0	0
Guitarra Panema sp.nov	1	1	1	1	3	1	0	0	0
Halichondria sp. 1	0	0	1	0	0	0	0	0	0
Haliclona (Haliclona) anonyma	0	1	0	0	0	0	0	0	0
Haliclona (Haliclona) sp. 1	0	0	0	1	0	0	0	0	0
Haliclona (Haliclona) sp. 2	0	0	0	0	0	0	0	0	1
Haliclona (Reniera) ciocalyptoides	1	0	0	4	0	0	0	1	6
Haliclona(Gellius) flagillifer	0	0	2	0	0	0	0	0	0
Halisarca ectofibrosa	1	0	0	0	0	0	0	0	0
Hemiasterella vasiformis	5	0	1	1	2	0	0	0	0
Higginsia bidentifera	0	0	0	2	2	0	0	0	0
Histodermella natalensis	0	1	0	1	0	0	0	0	0
Homaxinella flagelliformis	0	1	0	0	1	0	0	0	0
Homophymia pellis n.sp	0	0	0	1	1	0	0	0	0
Hymeniacidon perlevis	0	0	0	0	0	0	0	0	3
Hymeniacidon sp. 1	0	1	0	0	1	1	0	0	0
Hyrtios sp. 1	1	0	0	1	0	0	0	0	0
Isodictya ~~elastica~~elastic	0	0	0	1	0	0	0	0	0
Isodictya multiformis	0	0	0	0	1	0	0	0	0
Latrunculia (Biannulata) gotzi	0	0	0	0	2	0	0	0	0
Latrunculia (Biannulata) kerwathi	1	0	0	0	0	0	0	0	1
Latrunculia (Biannulata) microacanthoxea	0	0	0	0	1	0	0	0	1
Leucosolenia sp. 1	0	0	0	0	4	0	0	0	0

Lissodendoryx (Lissodendoryx) ~~d~~digitata~~digitata~~digitate	0	0	0	1		0	1	0	2
Mycale (Aegogropila meridionalis	0	0	0	0	0	0	1	1	2
Myxilla (Ectyomyxilla) sp. 1	0	0	0	0	1	0	0	0	0
Myxilla (Myxilla) simplex	0	1	0	0	0	0	0	0	0
Myxilla (Myxilla) sp. 1	0	1	0	0	0	0	0	0	0
Pachastrella caliculata	0	3	1	4	11	1	0	0	0
Pachastrella sp. 1	1	0	0	0	1	0	0	0	0
Pachastrella sp. 2	0	0	0	1	0	0	0	0	0
Pachastrella sp. 3	0	0	0	0	1	0	0	0	0
Penares alatus	0	1	0	2	0	0	0	0	0
Petrosia (Strongylophora) vulcaniensis	1	0	0	2	2	0	0	0	0
Phakellia rubra	0	0	1	0	0	0	0	0	0
Phorbas bardajii	1	0	0	0	0	0	0	0	0
Phorbas clathratus	1	1	0	0	1	0	0	0	0
Phorbas fibrosus	0	0	0	1	0	0	0	0	0
Phorbas sp. 1	0	0	0	1	0	1	0	0	0
Phycopsis sp. 1	0	0	0	0	1	0	0	0	0
Plakina sp. 1	0	1	0	0	1	0	0	0	0
Polymastia atlanticus	1	0	0	0	0	0	0	0	0
Polymastia bouryesnaultae	0	0	0	0	2	0	0	0	0
Polymastia littoralis	0	0	0	0	0	0	0	0	3
Polymastia sp. 1	0	2	0	0	0	0	0	0	0
Psammocinia sp. 1	0	0	0	0	2	0	0	0	1
Pseudosuberites sp. 1	0	0	0	1	0	0	0	0	0
Ptilocaulis sp. 1	1	0	1	2	0	1	0	0	
Raspailia rigida	0	1	0	0	1	1	0	0	0
Rhabdastrella ~~p~~primitive~~primitive~~primitive	0	0	0	1	0	0	0	0	0
Sigmaxinella arborea	1	0	2	0	1	0	0	0	0
Spheciospongia vagabunda	0	0	0	0	1	0	0	0	0
Spongia (Heterofibria) smaragdus	0	0	0	2	0	0	0	0	2
Spongia (Heterofibria) sp. 1	1	0	0	0	0	0	0	0	0
Spongosorites sp. 1	0	0	1	0	0	0	0	0	0
Stelletta agulhana	0	0	1	0	0	0	0	0	0
Stelletta capensis	0	0	2	1	0	1	0	0	0
Stelletta cyathoides	0	0	0	1	0	0	0	0	0

Stelletta ~~rugosa~~rugose	1	2	0	3	0	0	0	0	0
Stelletta trisclera	1	0	0	2	0	0	0	0	0
Stellitethya sp. 1	0	1	0	0	0	0	0	0	0
Strongylodesma tsitsikammaensis	0	0	0	11	0	0	0	0	11
Strongylodesma Agulhasensis sp.nov	0	0	0	0	1	0	0	0	0
Tedania (Tedania) scotiae	0	0	0	1	7	0	0	0	0
Tedania (Tedania) sp. 1	0	0	0	1	3	0	0	0	0
Tedania (Tedania) sp. 2	0	0	0	0	2	0	0	0	0
Tedania (Tedania) sp. 3	0	0	0	1	0	0	0	0	0
Tedania (Tedania) stylonychaeta	1	0	0	0	0	0	0	0	0
Tethya Oxyspherastera sp.nov	1	0	1	0	0	0	0	0	0
Tetilla ~~casula~~casual	0	1	0	0	1	0	0	0	0
Trachycladus spinispirulifer	0	0	0	0	0	0	0	0	1
Tsitsikamma favus	0	0	0	0	0	0	0	0	1
Grand Total	**51**	**50**	**26**	**69**	**120**	**11**	**3**	**4**	**28**

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