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Ecological characterization of the rocky shores of Príncipe Island, Gulf of Guinea

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Dedication

I dedicate this work to my family and friends who inspire and challenge me every day to be a better researcher in marine ecology, with a special note to my friends from Príncipe Island, for whom I hope this work has a positive impact, such as their lives surely impacted mine.

Resumo

O ecossistema intertidal é um modelo ideal para o estudo da ecologia de comunidades biológicas, pois exibe uma múltipla variedade de fatores que influenciam os padrões associados à composição, abundância e distribuição de espécies. É um ambiente altamente dinâmico, variando ao longo dos ciclos ambientais como a maré, que provoca períodos alternantes de emersão e submersão, e o ciclo lunar, que induz variações da amplitude da maré. O gradiente ambiental vertical que assim se constitui submete as espécies marinhas a um intenso e progressivo stress ao longo do perfil da zona intertidal.

Sabe-se, no entanto, que os stresses associados ao gradiente vertical variam com a latitude e entre localidades. Nas latitudes mais baixas dos trópicos, a dessecação intensifica-se devido ao efeito combinado das elevadas temperaturas e a reduzida humidade relativa nas zonas intertidais expostas ao sol. Já à escala local, a diferenciação de stresses deve-se fundamentalmente à exposição a diferentes fatores ambientais, sendo os principais os gradientes de hidrodinamismo e a salinidade. Dependendo da intensidade da ação das ondas e da presença próxima de rios, os padrões de zonação e a extensão do gradiente intertidal vertical são afetados de diferentes maneiras.

As costas rochosas apresentam condições geomorfológicas que facilitam a colonização e fixação da grande maioria de espécies que vivem entre marés. Consequentemente, é neste ambiente que melhor se destacam os gradientes verticais e padrões de distribuição de espécies. A retenção de água devido à natureza geomorfológica das costas rochosas também as caracteriza pela abundância de poças de maré, criando refúgios para muitos organismos marinhos como peixes e invertebrados.

O presente trabalho visa a estudar o ecossistema intertidal da costa rochosa da ilha do Príncipe, uma das duas principais ilhas oceânicas que compõem o país insular de São Tomé e Príncipe. De formação vulcânica e fazendo parte da cadeia de vulcões extintos da Linha dos Camarões, a ilha do Príncipe situa-se no Golfo da Guiné a 350 km da costa africana. Apesar de ser uma ilha oceânica e estar relativamente distante do continente, o mar envolvente é influenciado pelas massas de água quente e salobra da pluma dos rios Congo e Níger, atingindo à superfície temperaturas entre 27°C-30°C e salinidade <33-35‰. O clima na ilha é tropical húmido equatorial com elevada precipitação anual, sendo dividido em quatro estações que alternam entre épocas secas e chuvosas. A linha de costa do Príncipe é maioritariamente de formação rochosa vulcânica, encontrando-se frequentemente intercalada por praias arenosas.

A ilha do Príncipe é uma das várias ilhas oceânicas que hoje em dia se encontra cada vez mais sujeita a variadas pressões na linha de costa, quer sejam antropogénicas (e.g. construções de infraestruturas, poluição e sobrepesca) ou associadas às alterações climáticas. As comunidades intertidais nesta região são particularmente vulneráveis ao aquecimento global, uma vez que já se encontram perto do seu limite de tolerância às altas temperaturas. Modelos climáticos já preveem para São Tomé e Príncipe um aumento de 3ºC da temperatura do ar e uma variação considerável da taxa de precipitação anual até meados deste século. Considerando o facto de que a população da ilha depende fortemente dos recursos naturais costeiros, tanto a nível económico como de subsistência, é crucial a realização de estudos que informem e monitorizem o estado dos ecossistemas costeiros.

Do ponto de vista da ecologia marinha da ilha do Príncipe, ainda existe muita pouca informação. O Atlântico Este tropical é das províncias marinhas tropicais menos conhecida, com relativamente poucos estudos feitos na zona do Golfo da Guiné e ainda menos os que se focaram nas zonas intertidais, sobretudo nas ilhas oceânicas adjacentes. A ictiofauna associada às poças de maré tropicais tem também recebido pouca atenção pelo globo, particularmente no Golfo da Guiné. Apenas um número muito reduzido de estudos abordaram os ecossistemas intertidais em costas rochosas de São Tomé e Príncipe, tratando-se principalmente de listagens de alguns grupos taxonómicos, nomeadamente algas, moluscos e peixes.

Como tal, o presente trabalho teve duas abordagens ao estudo do ecossistema intertidal das costas rochosas da ilha do Príncipe. A primeira é essencialmente descritiva, abordando a composição e os padrões de distribuição vertical das comunidades em dois ambientes intertidais distintos: um exposto a influência estuarina e o outro exposto a condições marinhas. A segunda abordagem foca-se na ictiofauna intertidal, onde se procurou determinar a importância das condições abióticas e bióticas das poças de maré na estrutura da população de peixes. A dissertação está dividida em quatro capítulos, sendo o primeiro a introdução teórica aos temas abordados, onde é apresentado o enquadramento ecológico, a contextualização da área de estudo a nível oceanográfico, climático, biogeográfico, e estudos realizados, seguido dos objetivos deste trabalho. O segundo e terceiro capítulo englobam, respetivamente, a primeira e segunda abordagem e incluem os resultados e discussão do trabalho apresentados em formato conciso de publicação científica. Por fim, o quarto capítulo apresenta as conclusões finais.

Para o estudo descritivo da costa rochosa da ilha (Capítulo 2) foi amostrada a comunidade intertidal da costa rochosa de duas regiões, Abade e Bom-Bom, e comparadas entre si. Enquanto Abade representa uma zona intertidal numa baía abrigada do vento e das ondas e influenciada por rios próximos, a costa do ilhéu Bom-Bom representa uma zona intertidal exposta ao mar, sujeita à ação das ondas e sem rios nas proximidades. No total foram amostrados 47 taxa, dos quais 17 macroalgas, uma espécie de líquene preto (*Verrucaria* sp.) e 29 taxa de macrofauna epibentónica. A zona intertidal de Bom-Bom apresentou em média maior riqueza de espécies por transecto, tanto de macroalgas como de metazoários. No entanto, a composição geral das comunidades intertidais entre as duas regiões não foi significativamente diferente. Ainda assim, a presença e ausência de espécies-chave revela a diferença entre comunidades biológicas expostas a diferentes condições ambientais. A presença de mais espécies de macroalgas e de filtradores no Bom-Bom traduz condições típicas de uma zona intertidal exposta à ondulação, com maior oxigenação e dispersão de nutrientes e partículas de alimento.

No entanto, a maior diferença encontrada entre as costas rochosas de Abade e Bom-Bom foram os padrões de distribuição e zonação das comunidades intertidais. O maior hidrodinamismo na costa do Bom-Bom permite às espécies mais em baixo no perfil de maré subsistir até à zona de humectação e acima do limite superior teórico das marés. Já em Abade, a combinação de fraca ondulação e baixa salinidade causada pela proximidade de rios impede a expansão das espécies que vivem mais em baixo no perfil de maré. Esta diferença é evidente comparando o limite superior da banda infralitoral das duas regiões, sendo em Bom-Bom 0.7 m mais alto do que em Abade. Acima deste limite, a riqueza específica baixa drasticamente. Os padrões de distribuição e zonação na costa rochosa da ilha do Príncipe apresentaram semelhanças com outras comunidades intertidais do continente no Golfo da Guiné. Assim sendo, a zona intertidal das costas rochosas do Príncipe pode ser dividida em três zonas principais, que podem variar em largura consoante as condições marinhas. Mais acima encontra-se a zona supralitoral, dominada por *Echinolittorina soroziczac* e principalmente *E. granosa*; seguida pela zona eulitoral caraterizada pelo líquene preto *Verrucaria* sp., a ostra *Saccostrea cucullata* e a craca *Chthamalus dentatus*; e por fim a zona infralitoral de maior riqueza específica, mas maioritariamente dominada por alga incrustante rosa e pelo coral mole *Palythoa caribaeorum*. O gastrópode *Neritta senegalensis* foi a espécie com a distribuição mais vasta, ocupando sempre todo o intertidal.

Para o estudo da ictiofauna intertidal (Capítulo 3), foram amostradas 60 poças em três localizações na costa norte: ilhéu Bom-Bom, Ponta Marmita e Praia Uba. Apenas poças da zona eulitoral foram amostradas, nas quais foram registados um total 746 peixes pertencentes a 18 espécies diferentes, representando 13 famílias. As cinco espécies mais abundantes foram, por ordem decrescente, *Bathygobius burtoni*, *Abudefduf taurus*, *Entomacrodus cadenati*, *Microlipophrys velifer* e *Prionurus biafraensis*, que em conjunto representam 81% do total de observações. As quatro espécies mais abundantes corresponderam às que apresentaram maior adaptabilidade a uma maior variação de condições físico-químicas nas poças de maré. Foram avistadas outras 10 espécies fora do período de amostragem em poças de maré da zona eulitoral nas mesmas costas rochosas e outras semelhantes, representando mais cinco famílias. Destas, *Lutjanus griseus* trata-se de um novo registo para São Tomé e Príncipe, sendo apenas o segundo do Atlântico Este. As espécies foram agrupadas em três categorias consoante o seu ciclo de vida nas poças de maré: residentes, residentes secundárias e transientes. As espécies residentes e algumas residentes secundárias foram as que contribuíram mais para a abundância de peixes.

Dado o contexto geográfico, climático e oceanográfico da ilha do Príncipe, os peixes demonstraram, no geral, preferência por poças de maré volumosas, com reduzida cobertura biológica (algas e coral) e maior salinidade. Poças com estas características provaram ter condições físico químicas (temperatura, salinidade e pH) mais estáveis, representando microhabitats mais favoráveis. No entanto, algumas espécies apresentaram um certo grau de especificidade de habitat, nomeadamente a presença ou ausência de cobertura de biológica. Peixes com uma natação mais ativa preferiram poças mais fundas.

Palavras-chave: ecossistema intertidal, ecologia de comunidade, poças de maré rochosas, ilha oceânica, África Ocidental Tropical.

Abstract

The present work took place in the tropical rocky shores of Príncipe Island, São Tomé and Príncipe, Gulf of Guinea. This work comprises a theoretical framework (chapter 1), two scientific articles (chapter 2 and 3) which includes the results and discussion, and the final remarks (chapter 4). In Chapter 2, two rocky shores, representing each an estuarine-influenced and a marine environment, were sampled to describe and compare distribution patterns of intertidal biological assemblages. A total of 47 taxa were documented, comprising 17 macroalgae, one marine lichen, and 29 epibenthic macrofauna taxa. Bom-Bom rocky shore presented higher species richness, and although not significantly different compared to Abade, the presence of key species suggests a noteworthy difference between the two biological communities. The biggest difference, however, was reflected in the zonation patterns between the two shores. The upper limits of the zones in Bom-Bom are uplifted compared to Abade due the effect of stronger wave action and a broader splash zone. In Chapter 3, rock pools from three locations were sampled to describe the composition, abundance and distribution of fish assemblages and their relation to the pools physical structure, water mass and biological parameters. A total of 18 species were observed during sampling and grouped into three categories according to their life cycle in the tide pools: residents, secondary residents and transients. The residents *Bathygobius burtoni*, *Entomacrodus cadenati* and *Microlipophrys velifer* and the secondary residents *Abudefduf taurus* and *Prionurus biafraensis* were the most abundant species, representing together 81% of the total number of fish recorded during this study. Overall, larger rock pools with minimal biological cover and higher salinity supported higher fish abundance and species richness. However, some species presented a degree of habitat specificity, such as the absence or presence of biological cover and the pools depth.

Key words: intertidal ecosystem, community ecology, rock pools, oceanic island, Tropical West Africa.

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CHAPTER 1

General Introduction

Intertidal Ecosystems

Rocky Shores

Intertidal habitats are highly dynamic environments that are constantly changing in space and particularly in time through environmental cycles. The fact that tides vary hourly and change cyclically according to lunar rhythms influence the presence or absence of water and the effects of the waves on a predictable cycle (Horn *et al.*, 1999), turning the intertidal habitats often into a hostile environment for the marine species living in it.

Intertidal environments can have various forms, varying from sandy beaches to mudflats, coral reefs, steep cliffs and rocky shores, all of which support different species compositions and community structures. The average size of the particles that form a shore reflects in part the geological past of the area (Raffaelli and Hawkins, 1999), but also the exposure of the shoreline to wave action and currents. Thus, the tendency of smaller particles to accumulate and be a characterizing feature of shores in sheltered conditions. However, rock-type habitats can occur at both ends of the exposure gradient (Raffaelli and Hawkins, 1999).

Rocky shores present a particular set of conditions that facilitate the attachment of certain biological communities. This habitat can have highly irregular geomorphology, with structures like fissures, crevices, holes, gullies and pools, which provide a more secure anchorage for macroalgae, and animals compared to shores composed of moving sediment like sandy beaches and mudflats. As a result, the intertidal gradient is ultimately more obvious in the rocky shores (Raffaelli and Hawkins, 1999).

Throughout the vertical gradient, the higher the marine algae and organisms are positioned, the more they will be stressed by longer periods exposed to air (emersion). During emersion, marine species are vulnerable to relative humidity variations, precipitation and evaporation, which can affect the salinity of rock pools and rock surfaces, and air temperatures, which are much more variable than the sea temperature (Raffaelli and Hawkins, 1999).

However, the stresses associated with the vertical gradient, which translates into increased time spent emerged, are known to change with latitude (Raffaelli and Hawkins, 1999). In regions at low latitudes, such as the tropics where extreme heat occurs, physiological stresses are more extreme high on the shore compared to middle latitudes. In rocky shores of tropical areas, greater desiccation becomes one of the main limiting factors to survival, due to the combined effects of heat and low relative humidity.

Thus, due to geographic patchiness and dispersal ability, the composition of intertidal communities also differs with latitude and across continents (Gibson and Yoshiyama, 1999; White *et al.*, 2015). However, intertidal community patterns also change between regions and within locations, largely driven by the physical and chemical factors of the environment (Cox *et al.* 2011; Gibson and Yoshiyama, 1999; White *et al.*, 2015).

Within a region, rocky shores can be found in a variety of environments that, depending on the physical and chemical factors, modulate the community composition and distribution (Cox *et al.* 2011; Gibson and Yoshiyama, 1999; Horn *et al.*, 1999; White *et al.*, 2015). Two of the most prominent environmental gradients, at the local level, are recognized to be exposure (to wave action and currents) and salinity. Depending on the intensity of wave action and the presence of rivers nearby, zonation patterns and the extent of the vertical intertidal gradient are affected in different ways (Raffaelli and Hawkins, 1999).

Wave action can have a great effect on the biological community of a rocky shore, in such a way that allows species to survive meters above the theoretical tidal limit, causing an uplifted zonation compared to sheltered shores (Raffaelli and Hawkins, 1999). Therefore, the exposed, turbulent, wave-beating rocky intertidal habitats are usually highly productive (Leigh *et al.*, 1987) and occupied by a diverse biological community, rich in plant and animal life, mostly invertebrates (Horn *et al.*, 1999). In extreme exposed situations, when a shore is facing very strong breakers, only the species capable of maintaining attached to the rock can occur. Sheltered shores, on the other hand, can create a different set of environmental factors where offshore topographic features, such as barriers and reefs, dissipate wave action (Raffaelli and Hawkins, 1999).

Salinity, on the other hand, affects the species composition based more directly on the gradients set by chemical factors, depending on if the rocky shore is near estuaries or other sources of freshwater inflow, or if it is surrounded by a strictly marine environment with no freshwater inflow influence. Consequently, it also affects indirectly the species composition due to the competition between brackish species and truly marine species throughout both ends of the salinity gradient.

The intertidal zone of rocky shores presents an ideal model system for studies focusing on community ecology since it displays variations of multiple environmental variables (White *et al.*, 2015). Such studies generally consist in the description of the patterns associated with the composition, abundance and distribution of species assemblages throughout different gradients and develop an understanding of the environmental variables that cause these patterns to occur (Begon *et al.*, 1986; Diamond and Case 1986; White *et al.*, 2015). These variables can include physical (i.e. topography), chemical (i.e. variations in oxygen, salinity, temperature) and biological (i.e. predation, competition, habitat selection) interactions.

Rocky shore communities are generally simple, and distribution and zonation patterns can be relatively obvious. The sequential distribution of individual species, or a set of species assemblages, throughout different tidal levels is mostly dictated by the physiological tolerance of such species in emerged conditions and how dominant they can be in occupying the rocky substrate (Raffaelli and Hawkins, 1999). The universal three-zone scheme of zonation (low, mid and upper shore) proposed by Stephenson and Stephenson (1949, 1972) and Lewis (1964) is used globally by most classical descriptive works.

Nowadays, studies on intertidal rocky shores assemblages are increasingly more important and appropriate to monitor changes driven by climate change and global-scale anthropogenic impacts effects due to their ecological characteristics and accessibility (Cruz-Motta *et al.*, 2010; Helmuth *et al.*, 2002; Hoegh-Guldberg, 1999).

Rock pools

Rock pools are a common feature of rocky shores worldwide, adding important niche space and microhabitats to coastal habitats (Firth *et al.*, 2014). Water is retained in holes and depressions during low tide, which can usually serve as a refuge for many marine organisms that are highly water-dependent, (Martins *et al.*, 2007; Zander *et al.*, 1999), attracting high diversity and abundance of various organisms such as fishes and other invertebrates (White *et al.*, 2015).

Despite rock pools being a more stable environment compared to the freely draining open rock surface, these are still subject to large fluctuations in environmental conditions during the emersion period. In low water, rock pools become isolated from the sea and thus prone to large and rapid fluctuations in the water physicochemical properties such as temperature, salinity, oxygen, carbon dioxide and pH (Huggett and Griffiths, 1986; Horn *et al.*, 1999; Martins *et al.*, 2007).

The combination of the tidal cycle and time of day can lead to rapid and considerable physical and chemical changes in isolated tidal pools (Horn *et al.*, 1999). During daytime at low tides, insolation and evaporation can rapidly change temperature and salinity. Oxygen levels fluctuate according to the tide and diurnal cycle, rising during the day due to photosynthesis, and dropping at night when only respiration occurs. At the same time, pH also oscillates with oxygen and carbon dioxide concentration. During the day, higher oxygen levels produced by photosynthesis processes turn rock pools alkaline, whereas carbon dioxide released by respiration and accumulated during the night decreases the pH of the rock pools (Huggett and Griffiths, 1986; Horn *et al.*, 1999; Martins *et al.*, 2007).

Rock pools are extremely heterogeneous microhabitats. They can have numerous shapes and volumes, are randomly distributed along the rocky shore vertical and horizontal gradients and are highly variable in time (i.e. tidal, diurnal and seasonal-induced fluctuations) and space (from geographic to local scales) (Martins *et al.*, 2007; White *et al.*, 2015).

Many coastal fishes use rock pools at some point in their life cycle to forage, take refuge from predators or as nursery areas (Horn *et al.*, 1999). As a result, several fish species have developed behavioural, physiologic and morphologic adaptations to better endure the drastic environmental fluctuations of these habitats (Zander *et al.*, 1999). Thus, many behavioural patterns of fishes found in rock pools can be interpreted as adaptations for the survival of the individual or protecting its offspring from unfavourable conditions during the low tide (Gibson, 1986).

The different gradients of biotic and abiotic factors can produce consistent patterns in the composition, distribution and abundance of fish populations (Gibson, 1982; Norton and Cook, 1999; Zander *et al.*, 1999). Thus, an important part of the study of intertidal fishes is to find the relationships between their distribution, physiology and behaviour and the environmental variables of the rock pools (Gibson, 1999a).

However, rock pools have received much less attention compared to studies of emergent rock (Martins *et al.*, 2007). Little is still known about the processes determining the structure of their biological assemblages, possibly due to the heterogeneity of rock pools, which difficulties the replication of experimental studies (Martins *et al.*, 2007; Metaxas and Scheibling, 1993; Underwood and Skilleter, 1996). Of the relatively few studies existing on rock pools, tropical rock pools have received even less attention (Gibson, 1982; Mahon and Mahon, 1994; Martins *et al.*, 2007), particularly on the west coast of Africa and adjacent islands (Zander *et al.*, 1999).

Study area

Tropical Eastern Atlantic and the Gulf of Guinea

The coastal biodiversity of the tropical Atlantic can be divided into four primary provinces: Brazil, the “Greater” Caribbean, the Mid-Atlantic Ridge, and the Eastern Atlantic (Briggs, 1974; Floeter *et al.*, 2008). The Tropical Eastern Atlantic or Tropical West Africa is located in a narrow latitudinal range, in between the cold Canary Current in the north and the cold Benguela Current in the south, with also the presence of strong upwellings of the coast of Mauritania, Namibia and, seasonally, in the northern Gulf of Guinea (Reid, 2011). This province is characterised by heavy and seasonal rainfall that is enough to reduce considerably the surface salinity levels to 30‰, increasing turbidity over extensive areas, particularly around the mouths of the Niger and Congo rivers, affecting also the more distant islands (Reid, 2011). The tidal range is small, especially near the equatorial line.

Within the biogeographic province of the Tropical Eastern Atlantic, three main regions are discernible: the Western Continental Africa, Cabo Verde archipelago and islands in the Gulf of Guinea: Ano Bom, São Tomé, Príncipe and Bioko (which is considered a continental island) (Floeter *et al.*, 2008).

Due to historical biogeographic reasons, the Tropical Eastern Atlantic has the lowest biological diversity of the main tropical marine regions of the globe (Reid, 2011). A significant portion of the Eastern Atlantic biota consists of “immigrant species from the more diverse tropical western Atlantic province and temperate Europe” (Reid, 2011). However, local endemism rates of the Tropical Eastern Atlantic, such as in the Gulf of Guinea, are high probably due to the current and historical geographic isolation from other Atlantic areas (Maia *et al.*, 2018b). Rates of endemism can be 30% for reef fish (Floeter *et al.*, 2008), 96% for gastropods (Peters *et al.*, 2013, 2015; Polidoro *et al.*, 2017), and 18% for hard corals (Polidoro *et al.*, 2017; Maia, 2018; Maia *et al.*, 2018b).

São Tomé and Príncipe

The Democratic Republic of São Tomé e Príncipe is a small insular country comprised of two main islands, São Tomé and Príncipe, and several islets. The archipelago is of volcanic formation and is part of a chain of extinct volcanoes that extends throughout 1,600 km in the Gulf of Guinea, known as the Cameroon Volcanic Line (Fig. 1.1). It also includes the islands of Annobón to the southwest and Bioko to the northeast and ending in Mount Cameroon on the mainland. São Tomé and Príncipe islands are located in equatorial latitudes (between the meridians 1044’N and 0001’S, and the parallels of 6028’E and 7028’E) about 440 km to the south and 220 km to the west, respectively, of the coasts of Nigeria and Gabon.

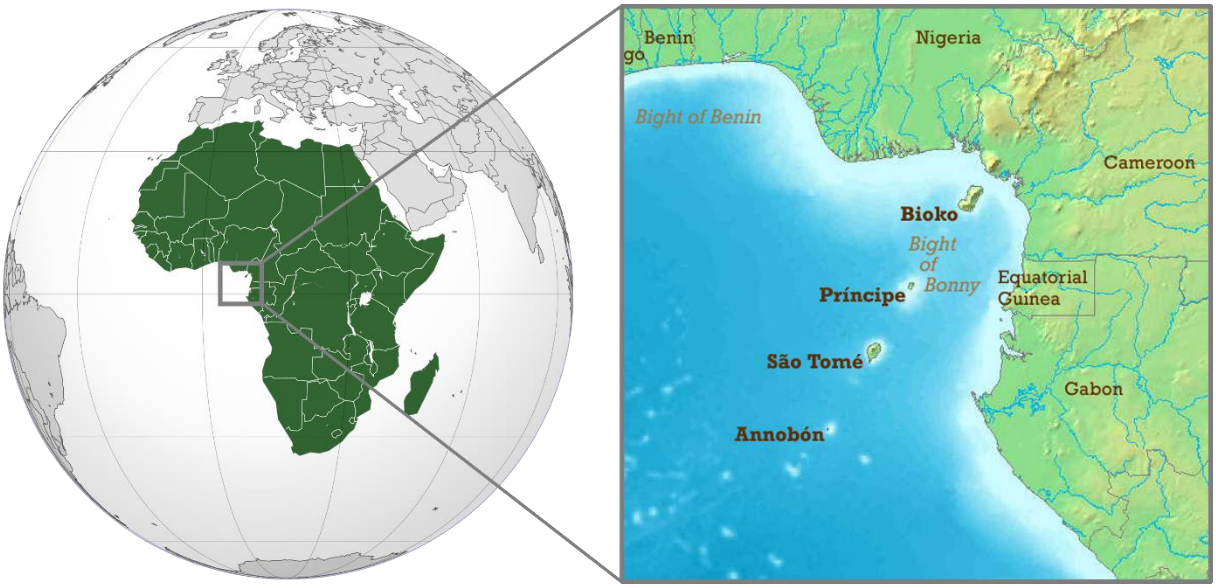

Figure 1.1. The archipelago of São Tomé and Príncipe is located in the Gulf of Guinea, in Western Africa. The two islands are part of a chain of extinct volcanoes that extends throughout 1,600 km, known as the Cameroon Volcanic Line, which also includes the islands of Annobón to the southwest and Bioko to the northeast and ending in Mount Cameroon on the mainland. Map do the right is an English modified version of the image by Janneman (2005).

Although the archipelago is considerably distant to the coast of the mainland of the Gulf of Guinea and therefore surrounded by an oceanic environment with, comparatively, higher salinity and lower temperatures (Krakstad *et al.*, 2010), both islands are still influenced by water masses from the plume of the Congo and Niger rivers with low salinity and high temperature (Krakstad *et al.*, 2010; Measey *et al.* 2007; Reid, 2011; Steentoft, 1965). Being closer to the mainland, Príncipe Island is bathed by a warmer (27°C-30°C) and less saline (<33-35‰) water mass (Krakstad *et al.*, 2010). Whereas São Tomé, that lies further south than Príncipe, is surrounded by cool, (22-28°C) more saline (34-36‰) waters originated from the south (Krakstad *et al.*, 2010; Steentoft, 1965). The islands have an equatorial humid tropical climate that can be divided into four seasons that alternate between rainier and dryer periods, with the average annual rainfall ranging from 2000-3000 mm (NBSAP II, 2015; Chou *et al.*, 2020). The rainy seasons represent the two main precipitation peaks throughout the year, with the highest one occurring between October and November, and a secondary peak around April and May (Chou *et al.*, 2020). These are interspersed by the dry seasons, with the more pronounced one ("gravana") occurring between June and September, and the lesser one between January and March (Chou *et al.*, 2020; Pisoni *et al.*, 2015; Steentoft, 1965; Vaz and Oliveira, 2007). Air temperatures vary from 18-21 ºC minimums to 30-35 ºC maximums (Herrero-Barrencua *et al.*, 2017), with an annual average of 26 ºC, and the air relative humidity is very high, reaching more than 90% at higher altitudes (NBSAP II, 2015).

São Tomé and Príncipe islands were never connected to the mainland, resulting in significant differentiation of terrestrial fauna and flora and high rates of endemism (Bonfim and Carvalho, 2009; Maia, 2018). However, marine endemism levels are low due to high oceanographic connectivity to the mainland African coast through the chain of islands (Floeter *et al.*, 2008; Maia *et al.*, 2018; Wirtz, 2003). Furthermore, a high proportion of the eastern Atlantic biota consists of immigrant species from more diverse tropical western Atlantic provinces (Brasil and Caribbean) and subtropical and temperate Europe (Floeter *et al.*, 2008; Herrero-Barrencua *et al.*, 2017; Maia *et al.*, 2018; Reid, 2011).

Príncipe Island

Príncipe Island is the second oldest of the Gulf of Guinea at 31 million years old and the smallest of the São Tomé and Principe archipelago, with a surface area of 142 km^2 (Fig. 1.2), but with three times the submersed area compared to São Tomé Island (Krakstad *et al.*, 2010). Príncipe island is largely vegetated with tropical forest and has several small islets of varying sizes (Ilhéu Bom- Bom, Pedra da Galé, Ilhéus dos Mosteiros, Tinhosas, Boné do Jóquei). In 2006 was created in Príncipe Island the Natural Park Obô, and in 2011 it was included in the World Biosphere Reserve of UNESCO inaugurated in São Tomé and Príncipe, based solely on its terrestrial fauna and flora and diversity of habitats (Abreu *et al.*, 2017). The natural park occupies the southern part of the island, with practically all inhabitants living in the northern part, mainly in the capital Santo António. Príncipe is populated by about 8000 people that inhabit mostly along the coast and make their living mainly from natural resources near or on the coasts, complemented by a small ecotourism sector (Abreu *et al.*, 2017).

Tidal ranges are relatively small in Príncipe Island, not increasing by more than 1.9 m in extreme spring tides (0.2 m to 2.1 m from hydrographic zero), and 0.4 m in neap tides (1.0 m to 1.4 m from hydrographic zero) (IH, 2019).

A significant part of the coastline of the island is of rocky formation, often interspersed with sandy beaches. In this study, rocky shores of four locations were sampled, in Abade, Bom-Bom islet, Ponta Marmita and Praia Uba (Fig. 1.2). They were chosen for reasons of logistics and ease of access, with none of them being covered by the natural park. However, rocky shores are practically unexploited by the local population, whether its fauna or flora, so there are virtually no differences in the anthropogenic pressure.

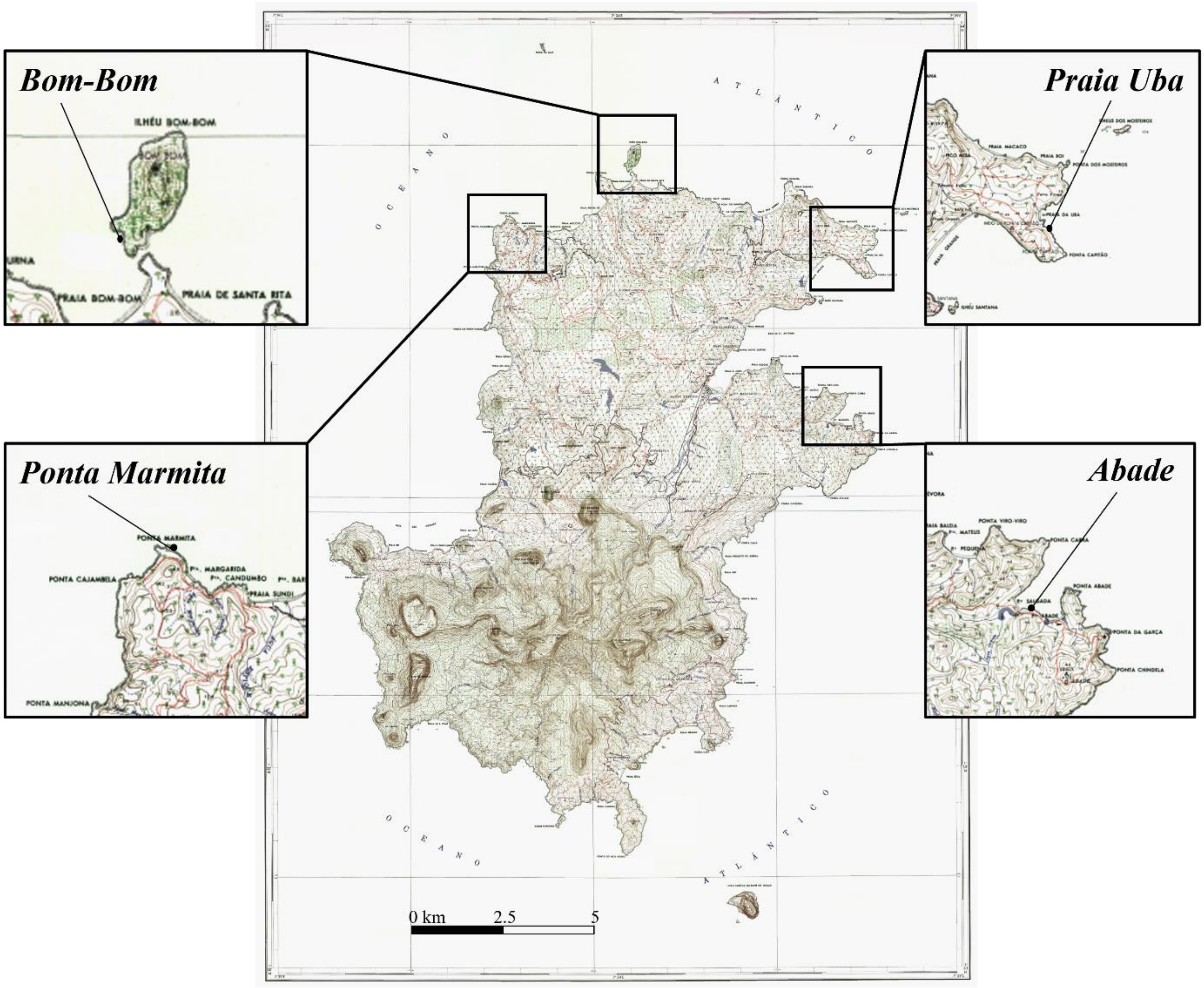

Figure 1.2. Príncipe Island has an area of 142 km^2 with a significant part of the coastline of rocky formation, often interspersed with sandy beaches. Four regions throughout the island were sampled for the different studies that comprise this work. For the descriptive study on the intertidal communities, sampling took place in Abade bay and Bom -Bom islet. As for the study on rock pools and fish assemblages, sampling took place in Bom-Bom islet, Ponta Marmita and Praia Uba. Image constructed from a 1961 map.

Abade

Abade is a small village located 3 km south-eastward of the capital Santo António, positioned inside a small and sheltered bay that is flanked by hills and with a narrow mouth that shields the bay from the prevailing winds from the north and east, and main currents (Herrero-Barrencua *et al.*, 2017). Thus, the bay is characterised by overall weak wave action. Furthermore, the bay is relatively shallow with a maximum depth of 17 meters (Herrero-Barrencua *et al.*, 2017). Three rivers also flow into the bay giving substantial freshwater input to the sea. The largest river, Água Grande, has enough flow to hold a mangrove ecosystem (Cravo *et al.*, submitted; Haroun *et al.*, 2018; Herrero-Barrencua *et al.*, 2017). Due to the existence of these characteristics, Abade bay is characterised as an estuarine environment, with fluctuations in salinity throughout the year, depending on the season (Chou *et al.*, 2020; Herrero-

Barrencua *et al.*, 2017). The portion of the rocky shore sampled in Abade is in the innermost part of the bay, between Praia Salgada and the Abade fishing village, and was used for describing the intertidal communities.

Bom-Bom islet

The Bom-Bom islet is located close to the shore on the north coast of the island and is currently occupied by a small touristic resort. Bom-Bom islet is one of the three locations studied for this work with exposed rocky shores to the sea, thus is prone to be impacted by breakers formed by the wind many kilometres away, characteristic of oceanic islands (Raffaelli and Hawkins, 1999). Also, is susceptible to wave action due to the rapid decrease in depth of the surrounding seabed. The islet's shore is practically all of rock formation with no freshwater streams. Therefore, the islet's intertidal zone is exposed to a marine environment with strong water movement and virtually identical salinity to the open surrounding sea. Sampling was carried out on the west side of the islet and was used for both the intertidal community description and the study on fish assemblage of rock pools.

Ponta Marmita

Ponta Marmita is a cape located in the northeast part of Príncipe Island. Like Bom-Bom islet, Ponta Marmita is also exposed to sea conditions, which means is prone to be impacted by breakers that come from the open ocean. Its coast is formed by a rocky shore, with some portions comprised of boulders and loose rocks. It also has an insignificant influence of freshwater streams on the salinity of coastal waters. There is also a nearby touristic resort but has no apparent impact on coastal and intertidal biological communities.

Praia Uba

Praia Uba is actually a small sandy beach flanked by extensive rocky shore platforms where rock pools were sampled for this work. It is the third location characterised by being exposed to a marine environment with strong wave action and minimal freshwater influence by nearby streams. It is also the least accessible location and further away from human presence.

Work context

Previous studies in São Tomé and Príncipe

Intertidal zones have been one of the most intensively studied marine environments throughout the globe. However, the biogeographic province of the Tropical Eastern Atlantic is still poorly studied, being the least studied of the tropical regions, despite the relatively low diversity in the region (Floeter *et al.*, 2008; Reid, 2011). Some studies were conducted on the rocky shores of West Africa, yet most works are outdated and were carried out either north (e.g. Senegal and Canaries) or south (e.g. Angola and South Africa) of the Gulf of Guinea, with very few being made in the equatorial region of the gulf (e.g. Bassindale, 1961; Buchanan, 1958; Gauld and Buchana, 1959; John and Lawson, 1991; Lawson, 1955, 1956, 1957, 1966; Lawson and John, 1982). Rock pools have also received little attention in West Africa (Gibson, 1982; Mahon and Mahon, 1994; Martins *et al.*, 2007), particularly on the islands of the Gulf of Guinea (Zander *et al.*, 1999).

There is very limited information regarding rocky shores of oceanic islands, including São Tomé and Príncipe, since their coastal ecosystems constitute one of the least explored areas worldwide, with many of its marine organisms requiring further studies (Floeter *et al.*, 2005; Floeter *et al.*, 2006). Only a limited number of studies have addressed intertidal ecosystems of São Tomé and Príncipe, including rocky shores, namely comprised of new records of species in the archipelago of different taxonomic groups. Steentoft (1965) conducted a major work on algae throughout the archipelago, where were described 101 species, of which some are found in the intertidal zone. Corals found in the intertidal were also mentioned in previous studies, namely *Siderastrea radians* (Maia, 2018) and *Palythoa caribaeorum* (Wirtz and d'Acoz, 2008), among other cnidarians (Bollen, 2017). Molluscs are one of the most well-studied groups of São Tomé and Príncipe coastal ecosystems (>250 species) and with one of the highest marine endemic rates in the archipelago (Maia, 2018). Several mollusc species can be found in the intertidal, including Polyplacophora (Dell' Angelo *et al.*, 2014; Fernandes and Rolan, 1993), Bivalvia (Fernandes and Rolan, 1993), Gastropoda (Fernandes and Rolan, 1993; Herrero-Barrencua, 2014; Reid, 2011), and Cephalopoda, namely *Octopus vulgaris* (Krakstad *et al.*, 2010). Studies relating crustaceans found in rocky shores are fewer, restricted essentially to the Grapsidae family (Fransen, 2014, Wirtz, 2003, 2018). Echinoderm species reported in previous studies include the sea-star *Oreaster clavatus* (Bollen, 2017) and several sea-urchins (Bollen, 2017; Wirtz and d'Acoz, 2008).

Fishes constitute the most well-studied group of São Tomé and Príncipe marine biodiversity. Some of the first works on biological marine diversity are of ichthyological characterization of the islands, carried out by Baltazar Osorio (1891, 1892, 1893a,b), which identified 124 species of fish distributed in 54 families. Although the marine ichthyofauna of São Tomé and Príncipe is one of the least studied in the world, it has recently attracted more scientific interest (Tuya *et al.*, 2017). Previous studies have focused mainly on the open and coastal sea, and more recently on mangrove ecosystems. However, only Afonso *et al.* (1999) and Wirtz *et al.* (2007) have reported some sightings of fish in intertidal rocky pools, but with no detailed information. These include *Echidna peli, Rypticus subbifrenatus, Abudefduf taurus, Entomacrodus cadenati, Hypleurochilus aequipinnis, Microlipophrys velifer, Ophioblennius atlanticus, Scartella cristata, Labrisomus nuchipinnis* and *Bathygobius burtoni*. Of these, it is known that the blenniids *E. cadenati, M. velifer, O. atlanticus* and *S. cristata* are resident species of the intertidal zone along the Tropical Eastern Atlantic (Zander *et al.*, 1999). More recently, only the latest publication of the field guide “Coastal marine fishes of São Tomé and Príncipe” (Vasco-Rodriges *et al.*, 2018) mentioned, although briefly, fishes that can be found in intertidal rock pools. Nevertheless, studies that focus on whole fish assemblages is non-existent and very little is known about intertidal fishes of other families (John and Lawson, 1991; Wirtz, 1980; Zander *et al.*, 1999).

As to all studies previously mentioned, these were primarily checklists and did not provide additional information on distribution patterns, nor on the overall intertidal communities and their interactions with environmental factors. Furthermore, although some of the studies explored Príncipe Island coast, knowledge of rocky shore communities in the island is practically non-existent.

Climate changes and anthropogenic threats

Global climate change, in particular ocean warming, imposes increasingly more pressure on the environmental conditions of rocky shores and their biological communities (Horn *et al.*, 1999). Marine species of tropical regions are expected to be particularly affected by climate changes and ocean warming, especially those living in the intertidal environment since they are prone to be more stenothermic, which means an increase in temperature can drive some of them above their lethal limit (Raffaelli and Hawkins, 1999).

A study by Chou *et al.* (2020) has reported future projections of climate change for São Tomé and Príncipe, predicting an increase in temperature, that could exceed 3°C in the middle of the 21st century, as well as in heavy precipitation rates. This means that the reality of possible significant impacts on biological communities of Príncipe rocky shores is almost certain and imminent, which will likely translate into changes of distribution and zonation patterns, and biological abundance and diversity, like in many other tropical shores (Horn *et al.*, 1999; Raffaelli and Hawkins, 1999; Vinagre *et al.*, 2018).

Studies that address this issue have become increasingly more important nowadays to monitor global climate changes. Thus, describing intertidal communities and zonation patterns in different environments (e.g. estuarine and marine) of the Príncipe Island may provide a baseline reference for future studies that could assess the impacts of global climate changes in these environments.

The fishing practice has been around in Príncipe Island since the beginning of Portuguese colonization, both from a subsistence point of view, as well as from an economic and sporting point of view (Abreu, 2013; Horemans *et al.*, 1994; Tous, 2015). However, these activities have intensified in the past decades by industrial fishing vessels coming from Europe and Asia, while small scale artisanal fishing remained a crucial local source of subsistence and income and a determining factor for the country's food security (Krakstad *et al.*, 2010; Maia *et al.*, 2018). Due to the different anthropogenic threats sources that the island has been suffering and the high endemism representative of the Gulf of Guinea, Príncipe Island is considered a hotspot of marine biodiversity (Roberts *et al.*, 2002). Knowing that tropical shores around the world act as nursery areas for numerous fish species that eventually recruit to coastal fisheries (Gibson, 1999b), it is possible that Príncipe Island rocky coast has also a similar value, both ecological, economic and subsistence at a local level.

CobioNet

The work developed in the present dissertation is part of the project CobioNet - *Coastal biodiversity* and *food security in peri-urban Sub-Saharan Africa: assessment, capacity building*, and *regional networking in contrasting Indian* and *Atlantic Oceans*, funded by Fundação para a Ciência e a Tecnologia and Aga Khan Development Network. The project main objectives are the assessment of biodiversity and ecosystem condition and their relation to the stability of resources, enhance ecosystem rehabilitation processes, establish biodiversity online repositories, and direct impacts on scientific and local coastal communities through networking, capacity building and dissemination actions.

Objectives

This work aims to fill the knowledge gap existent in intertidal communities of rocky shores of oceanic islands of the Gulf of Guinea, in particular Príncipe Island.

The specific objectives are:

(i) To describe and compare distribution patterns of intertidal biological assemblages in rocky shores in an estuarine-influenced and marine environments of Principe Island;
(ii) describe distributional records and assess distributions in the context of regional biogeography;
(iii) determine the importance of rock pool dimensions (depth, surface area and volume), within-pool water conditions (temperature, salinity and pH), and biological community cover (algal, coral and total), in controlling fish assemblage structure;
(iv) assess the variables which contributed more for each species abundance;
(v) describe species distribution depending on the water properties.

Methodology approach

This dissertation is divided into four chapters. The general introduction (Chapter 1) contextualizes the theme presented in this work. Chapter 2 is in the format of a scientific article with the title "Biodiversity distribution patterns on intertidal rocky shores of Príncipe Island (Gulf of Guinea)", comprised of the first two objectives where it is described the distributional and zonation patterns of biological assemblages of two rocky shores, in Abade bay and Bom-Bom islet. Chapter 3, also in the format of a scientific article, has the title of "Patterns of fish distribution in rock pools from tropical intertidal rocky shores at Príncipe Island (Gulf of Guinea)", where it was investigated rock pool fish assemblages at three locations, Bom-Bom islet, Ponta Marmita and Praia Uba, to describe the species composition and abundance in relation to abiotic and biotic factors, comprising the remaining three objectives. Chapter 4 presents the final remarks taken about this work, including self-criticism and suggestions for future works in the region.

Algae and organisms were collected for identification and conservation purposes and stored in museums. Some were deposited on the first natural history collections of Príncipe Unesco Biosphere Reserve, whereas others were deposited at the National Museum of Natural History and Science (MUHNAC).

References

Abreu, A.D. (2013). Príncipe Island's Biosphere Reserve (Democratic Republic of São Tomé and Príncipe): A Living Laboratory for Sustainable Development. In: Clusener-Godt RP-S and M (ed) AfriMAB Biosphere reserves in Sub-Sahan Africa: Showcasing sustainable development, First. Department of Environmental Affairs and UNESCO, South Africa. 284–302. ISBN: 978-0-620-57141-8

Abreu, A.D., Asensio, M.T., Cervera, J.L., Cosme, M., Fuentes, I., García-Mendoza, A., Gómez, P., Haroun, R., Herrero-Barrencua, A., Matilde, E., Niccolini, M., Otero-Ferrer, F., Tuya, F. and Viera-Rodríguez, M.A. (2017). Final Report Bio-Príncipe 2016. IU-ECOAQUA, Universidad de Las Palmas de Gran Canaria, Spain. 28 pp.

Afonso, P.F., Porteiro, F.M., Santos, R.S., Barreiros, J.P., Worms, J. and Wirtz, P. (1999). Coastal marine fishes of São Tomé Island (Gulf of Guinea). *Arquipélago, Life* and *Marine Sciences*, 17A: 65-92. Ponta Delgada. ISSN 0873–4704.

Bassindale, R. (1961). On the Marine Fauna of Ghana. *Proceedings of the Zoological Society of London*, *137*(4), 481–510. https://doi.org/10.1111/j.1469-7998.1961.tb06086.x

Begon M., Harper J.L., Townsend C.R. (1986). Ecology: Individuals, Populations, and Communities. Sinauer Associates Inc., Boston: 97–104.

Bollen, A. (2017). Compilation marine work - Marine and coastal species and ecosystems of Principe. Draft version 20 May 2017. FFI and FPT.

Buchanan, J.B. (1958). The Bottom Fauna Communities Across the Continental Shelf Off Accra, Ghana (Gold Coast). *Proceedings of the Zoological Society of London*, *130*(1), 1–56. https://doi.org/10.1111/j.1096-3642.1958.tb00562.x

Briggs, J.C. (1974). Marine zoogeography. McGraw-Hill, New York.

Bonfim, F. and Carvalho, S. (2009). Fourth national report on the biodiversity (1st draft). Directorate General of the Environment, Ministry of Natural Resources, Energy and Environment, Democratic Republic of Sao Tome and Principe. 93 pp.

Chou, S.C., Lyra, A.A., Gomes, J. L., Rodriguez, D.A., Martins, M.A., Resende, N.C, Tavares, P.S., Dereczynski, C.P., Pilotto, I.L., Martins, A. M., Carvalho, L.F.A., Onofre, J.L.L., Major, I., Penhor, M., and Santana, A. (2020). Downscaling projections of climate change in Sao Tome and Principe Islands, Africa. *Climate Dynamics*, *54*(9–10), 4021–4042. https://doi.org/10.1007/s00382-020-05212-7

Cox, T.E., Baumgartner E., Philippoff, J., Boyle K.S. (2011) Spatial and vertical patterns in the tidepool fish assemblage on the island of O‘ahu. *Environmental Biology of Fishes*, 90, 329–342.

Cravo, M., Almeida, A.J., Lima, H., Azevedo e Silva, J., Bandeira, S., Machava, V. and Paula, J. (submitted). Fish nursery role by a small mangrove at Príncipe Island, Gulf of Guinea. Submitted to *Frontiers in Marine Science*.

Cruz-Motta, J.J., Miloslavich, P., Palomo, G., Iken, K., Konar, B., Pohle, G., Trott, T., Benedetti-Cecchi, L., Herrera, C., Hernández, A., Sardi, A., Bueno, A., Castillo, J., Klein, E., Guerra-Castro, E., Gobin, J., Gómez, D. I., Riosmena-Rodríguez, R., Mead, A., Bigatti, G., Knowlton, A., and Shirayama, Y. (2010). Patterns of spatial variation of assemblages associated with intertidal rocky shores: a global perspective. *PLoS one,* 5(12), e14354. 10 pp.

Dell’ Angelo, B., Schwabe, E., Gori, S., Sosso, M., and Bonfitto, A. (2014). Chitons (Mollusca, Polyplacophora) from São Tomé and Príncipe Islands. *African Invertebrates 55*(2), 171–200.

Diamond J., Case T.J. (1986). *Community Ecology.* Harper and Row, New York: 665 pp.

Fernandes, F., and Rolan, E. (1993). Moluscos marinos de São Tomé y Principe: actualizacion bibliografica y nuevas aportaciones (Marine molluscs of São Tomé and Principe: bibliographic actualization and new contributions). *Iberus*, *11*(1), 31–47.

Firth, L.B., Schofield, M., White, F.J., Skov, M.W., Hawkins, S.J. (2014). Biodiversity in intertidal rock pools: informing engineering criteria for artificial habitat enhancement in the built environment. *Marine Environmental Research*, *102*, 122-130. Doi: 10.1016/j.marenvres.2014.03.016

Floeter, S. R., Behrens, M. D., Ferreira, C. E. L., Paddack, M. J., and Horn, M. H. (2005). Geographical gradients of marine herbivorous fishes: Patterns and processes. *Marine Biology, 147,* 1435–1447. https://doi. org/10.1007/s00227-005-0027-0

Floeter, S.R., Halpern, B.S., and Ferreira, C.E.L. (2006). Effects of fishing and protection on Brazilian reef fishes. *Biological Conservation, 128*(3), 391–402.

Floeter, S. R., Rocha, L.A., Robertson, D.R., Joyeux, J.C., Smith-Vaniz, W.F., Wirtz, P., Edwards, A.J., Barreiros, J.P., Ferreira, C.E.L., Gasparini, J.L., Brito, A., Falcón, J.M., Bowen, B.W., and Bernardi, G. (2008). Atlantic reef fish biogeography and evolution. *Journal of Biogeography, 35*(1), 22–47. https://doi.org/10.1111/j.1365-2699.2007.01790.x

Fransen C.H.J.M. (2014). True crabs. Pp 249–358 in *The living marine resources of the Western Central Atlantic. Volume 1 (Introduction, crustaceans, chitons* and *cephalopods),* (ed. Carpenter, K.E.), FAO Species Identification Guide for Fishery Purposes and American Society of Ichthyologists and Herpetologists Special Publication No. 5. Rome, Italy.

Gauld, D.T., and Buchana, J.B. (1959). The principal features of the rock shore fauna in Ghana. *Oikos, 46*, 121–132.

Gibson, R.N. (1982). Recent studies on the biology of intertidal fishes. *Oceanography and Marine Biology: An Annual Review, 20*, 902.

Gibson, R.N. (1986). Intertidal Teleosts: Life in a Fluctuating Environment. Pp. 388–408 in *The Behaviour of Teleost Fishes* (ed. Pitcher, T.J.), Springer, MA. Boston, USA.

Gibson, R.N. (1999a). Methods for Studying Intertidal Fishes. Pp. 7-25 in *Intertidal Fishes: Life in Two Worlds* (eds. Horn, M.H., Martin, K.L.M. and Chotkowski, M.A.), Academic Press. San Diego, USA.

Gibson, R.N. (1999b). Movement and Homing in Intertidal Fishes. Pp. 97-125 in *Intertidal Fishes: Life in Two Worlds* (eds. Horn, M.H., Martin, K.L.M. and Chotkowski, M.A.), Academic Press. San Diego, USA.

Gibson R.N. and Yoshiyama R.M. (1999). Intertidal fish communities. Pp. 264–296 *Intertidal Fishes: Life in Two Worlds* (eds. Horn, M.H., Martin, K.L.M. and Chotkowski, M.A.), Academic Press. San Diego, USA: 264–296.

Haroun, R., Herrero-Barrencua, A., and Abreu, A.D. (2018). Mangrove Habitats in São Tomé and Príncipe (Gulf of Guinea, Africa): Conservation and Management Status. *Coastal Research Library*, *25*, 589–605. https://doi.org/10.1007/978-3-319-73016-5_27

Helmuth, B., Harley, C.D., Halpin, P.M., O'Donnell, M., Hofmann, G.E., and Blanchette, C.A. (2002). Climate change and latitudinal patterns of intertidal thermal stress. *Science, 298*(5595), 1015–1017.

Herrero-Barrencua, A., Asensio, M., Cervera, J.L., Cosme, M., Abreu, D.A., Fuertes, I., Haroun, R., Otero-Ferrer, F., Suarez, F.J., Tuya, F. (2017a). Sea slugs (Mollusca: Heterobranchia) form Príncipe Island (Gulf of Guines): Unravelling a largely unknown fauna. Conference paper.

Herrero-Barrencua, A., Haroun, R. and Abreu, A.D. (2017b). *Caracterización preliminar de los manglares de la Isla de Príncipe (São Tomé e Príncipe)*. IU-ECOAQUA, Universidad de Las Palmas de Gran Canaria. 70 pp.

Hoegh-Guldberg, O. (1999). Climate change, coral bleaching and the future of the world's coral reefs. *Marine and Freshwater Reshearch, 50*(8), 839–866.

Horemans, B., Gallène, J. and Njock, J. (1994). Revista sectorial da pesca artesanal a São Tomé e Príncipe. Programa de Desenvolvimento Integrado das Pescas Artesanais na África Ocidental (DIPA). 32 pp + anexos. http://www.fao.org/3/an097PT/an097PT.pdf

Horn, M.H., Martin, K.L.M. and Chotkowski, M.A. (1999). Introduction. Pp. 1–6 in *Intertidal Fishes: Life in Two Worlds* (eds. Horn, M.H., Martin, K.L.M. and Chotkowski, M.A.), Academic Press. San Diego, USA.

Huggett, J., Griffiths, C.L. (1986). Some relationships between elevation, physicochemical variables and biota of intertidal rock pools. *Marine Ecology Progress Series*, *29*(2), 189–197.

IH (2019). Tabela de previsões de marés da Baía de S. António (Ilha do Príncipe) para Janeiro, Fevereiro e Março de 2020. Instituto Hidrográfico.

John, D.M. and Lawson, G.W. (1991). Littoral ecosystems of tropical western Africa, in *Intertidal and Littoral Ecosystems*, (eds AC. Mathieson and P.H. Nienhuis), Elsevier, Amsterdam, 297–323.

Krakstad, J.O., Alvheim, O., Lopes, J. and Iwamoto, T. (2010). Surveys of the Fish Resources of São Tomé and Príncipe: Survey of the demersal resources (Issue May). 66 pp.

Leigh, E.G., Paine, R. T., Quinn, J.F., and Suchanek, T.H. (1987). Wave energy and intertidal productivity. *Proceedings of the National Academy of Sciences, 84*(5), 1314–1318.

Lawson, G.W. (1955). Rocky shore zonation in the British Cameroons. *Journal of the West African Science Association, 1*, 78–88.

Lawson, G. W. (1956). Rocky Shore Zonation on the Gold Coast. *The Journal of Ecology, 44*(1), 153–170. https://doi.org/10.2307/2257159

Lawson, G.W. (1957). Seasonal Variation of Intertidal Zonation on the Coast of Ghana in Relation to Tidal Factors. *British Ecological Society, 45*(3), 831–860. http://www.jstor.org/stable/2256960

Lawson, G.W. (1966). The littoral ecology of West Africa. *Oceanography and Marine Biology, 4*, 405–448.

Lawson, G.W. and John, D.M. (1982). The marine algae and coastal environment of tropical West Africa. *Beihefte zur Nova Hedwigia, 70*, 1–455.

Lewis, J.R. (1964). The Ecology of Rocky Shores. English Universities Press, London. 323 pp.

Maia, H.A. (2018). Padrões espaciais e temporais das comunidades recifais nas Ilhas De São Tome e Principe, Golfo de Guine, Africa. PhD Thesis, 192 pp.

Maia, H.A., Morais, R.A., Siqueira, A.C., Hanazaki, N., Floeter, S.R., and Bender, M.G. (2018a). Shifting baselines among traditional fishers in São Tomé and Príncipe islands, Gulf of Guinea. *Ocean and Coastal Management*, *154*(August 2017), 133–142. https://doi.org/10.1016/j.ocecoaman.2018.01.006

Maia, Hugulay A., Morais, R. A., Quimbayo, J. P., Dias, M. S., Sampaio, C. L. S., Horta, P. A., Ferreira, C. E. L., and Floeter, S. R. (2018b). Spatial patterns and drivers of fish and benthic reef communities at São Tomé Island, Tropical Eastern Atlantic. *Marine Ecology*, *39*(6), 16 pp. https://doi.org/10.1111/maec.12520

Mahon, R. and Mahon, S.D. (1994). Structure and resilience of a tidepool fish assemblage at Barbados. *Environmental Biology of Fishes*, 41(1–4): 171-190. https://doi.org/10.1007/BF02197843

Martins, G.M., Hawkins, S. J., Thompson, R.C., and Jenkins, S.R. (2007). Community structure and functioning in intertidal rock pools: effects of pool size and shore height at different successional stages. *Marine Ecology Progress Series, 329*, 43–55.

Measey, G.J., Vences, M., Drewes, R.C., Chiari, Y., Melo, M. and Bourles, B. (2007). Freshwater paths across the ocean: molecular phylogeny of the frog *Ptychadena newtoni* gives insights into amphibian colonization of oceanic islands. *Journal of Biogeography, 34*, 7–20.

Metaxas, A., Scheibling, R.E. (1993). Community structure and organization of tidepools. *Marine*

Ecology Progress Series, 98, 187–198

NBSAP II (2015). National Biodiversity Strategy and Action Plan 2015–2020, National Commission for Monitoring and Evaluation, Ministry of Infrastructure. Natural Resources and Environment, São Tomé and Príncipe, 120 pp.

Norton, S.F. and Cook, A.E. (1999). Predation by Fishes in the Intertidal. Pp. 223-263 in *Intertidal Fishes: Life in Two Worlds* (eds. Horn, M.H., Martin, K.L.M. and Chotkowski, M.A.), Academic Press. San Diego, USA.

Osório, B. (1891). Estudos ichthyologicos ácerca da fauna dos domínios portugueses da África. 3ª Nota: Peixes maritimos das ilhas de S. Thomé, do Princípe e ilho das Rolas. *Jornal de Sciencias Mathematicas, Physicas e Naturaes,* 2ª Série 2 (6), 97–139.

Osório, B. (1892). Estudos ichthyologicos ácerca da fauna dos dominios portuguezes da Africa. 3ª Nota: Peixes maritimos das ilhas de S. Thomé, do Principe e ilho das Rolas (Continuação). *Jornal de Sciencias Mathematicas, Physicas e Naturaes*, 2ª Série 2 (7), 205–209.

Osório, B. (1893a). Estudos ichthyologicos ácerca da fauna dos dominios portuguezes da Africa. *Jornal de Sciencias Mathematicas, Physicas e Naturaes*, 2ª Série 3 (10), 136–140.

Osório, B. (1893b). Estudos ichthyologicos ácerca da fauna dos dominios portuguezes da Africa. 3ª Nota: Peixes maritimos das ilhas de S. Thomé, do Principe e ilho das Rolas (Continuação). *Jornal de Sciencias Mathematicas, Physicas e Naturaes*, 2ª Série 3 (11), 173–182.

Peters, H., O'Leary, B.C., Hawkins, J.P., Carpenter, K.E., and Roberts, C.M. (2013). Conus: First comprehensive conservation Red List assessment of a marine gastropod mollusc genus. *PLoS one*, *8*(12), e83353. doi:10.1371/journal.pone.0083353

Peters, H., O'Leary, B.C., Hawkins, J.P. and Roberts, C.M. (2015). Identifying species at extinction risk using global models of anthropogenic impact. *Global change biology*, *21*(2), 618–628.

Pisoni, T., Lima, R.F. de, Brito, A.C., Chainho, P., Félix, P.M., Caçador, I. and Carvalho, A. (2015). *Planos de gestão participativa para dois sítios de mangal na Ilha de S . Tomé: Praia das Conchas e Malanza.* Abordagem ecossistémica integrada para a conservação e gestão da biodiversidade na zona tampão dos parques naturais obô de São Tomé e Príncipe, 79 pp.

Polidoro, B.A., Carpenter, K.E., Ralph, G.M., Strongin, K., Harvey, M., Arnold, R., Buchanan, J R., Camara, K.M.A., Collette, B.B., Comeros-Raynal, M.T., De Bruyne, G., Gon, O., Harold, A.S., Harwell, H., Hulley, P.A., Iwamoto, T., Knudsen, S.W., Lewembe, J.D.; Linardich, C., Lindeman, K.C., Monteiro, V., Munroe, T., Nunoo, F.K.E., Pollock, C.M., Poss, S., Russell, B., Sayer, C., Sidibe, A., Smith-Vaniz, W., Stump, E., Sylla, M., Tito De Morais, L., Vié, J.C.; Williams, A., (2017). The status of marine biodiversity in the Eastern Central Atlantic (West and Central Africa). *Aquatic Conservation: Marine and Freshwater Ecosystems, 27,* 1021–1034. https://doi.org/10.1002/aqc.2744

Raffaelli, D. and Hawkins, S. (1999). *Intertidal Ecology*. 2nd edition, 356 p. Kluwer Academic Publishers. https://doi.org/10.2307/1352485

Reid, D.G. (2011). The genus *Echinolittorina* Habe, 1956 (Gastropoda: Littorinidae) in the Eastern Atlantic Ocean and Mediterranean Sea. *Zootaxa, 2974*(1), 65 pp. https://doi.org/10.11646/zootaxa.2974.1.1

Roberts, C.M., McClean, C.J., Veron, J.E.N., Hawkins, J.P., Allen, G.R., McAllister, D. E., Mittermeier, C.G., Schueler, F.W., Spalding, M., Wells, F., Vynne, C. and Werner, T.B., (2002). Marine biodiversity hotspots and conservation priorities for tropical reefs. *Science, 295*, 1280–1284.

Steentoft, B.Y.M. (1965). A revision of the marine algae of São Tomé and Príncipe (Gulf of Guinea). *The Journal of the Linnean Society (Botany)*, *60*(382), 99–146. https://doi.org/10.1017/cbo9780511779954.007

Stephenson, T.A. and Stephenson, A. (1949). The universal features of zonation between tide marks on rocky coasts. *Journal of Ecology, 37*, 289–305.

Stephenson, T.A. and Stephenson, A. (1972). *Life between tidemarks on rocky shores*. W. H. Freeman, San Francisco, 425 pp.

Tous, P. (2015). *Consulta para a avaliação dos recursos de peixes demersais*. Relatório do Projeto de Abordagem Ecosistémica Integrada para a Conservação e a Gestão da Biodiversidade em São Tomé e Principe (FIDA/PAPAFPA/GEF). Ministério da Economia e Cooperação Internacional.

Tuya, F., Herrero-Barrencua, A., Bosch, N.E., Abreu, A.D. and Haroun, R. (2017). Reef fish at a remote tropical island (Principe Island, Gulf of Guinea): Disentangling taxonomic, functional and phylogenetic diversity patterns with depth. *Marine* and *Freshwater Research*, 69(3): 395–402. https://doi.org/10.1071/MF17233

Underwood, A.J. and Skilleter, G.A. (1996). Effects of patch-size on the structure of assemblages in rock pools. *Journal of Experimental Marine Biology and Ecology*, *197*(1), 63–90.

Vasco-Rodrigues, N., Bertoncini, A. and Fontes, J. (2018). *Peixes marinhos costeiros de São Tomé e Príncipe* (Coastal marine fishes of São Tomé and Príncipe). Portugal: Flying Sharks. 123 pp. ISBN: 978-989-208568.

Vaz, H., and Oliveira, F. (2007). Relatótio Nacional do Estado Geral da Biodiversidade de S. Tomé e Príncipe. Direção Geral do Ambiente, Ministério de Recursos Naturais e Meio Ambiente. 115 pp.

Vinagre, C., Mendonça, V., Cereja, R., Abreu-Afonso, F., Dias, M., Mizrahi, D., and Flores, A.A.V. (2018). Ecological traps in shallow coastal waters-Potential effect of heat-waves in tropical and temperate organisms. *PLoS one*, *13*(2), 1–17. https://doi.org/10.1371/journal.pone.0192700

White, G.E., Hose, G.C. and Brown, C. (2015). Influence of rock-pool characteristics on the distribution and abundance of inter-tidal fishes. *Marine Ecology, 36*(4): 1332–1344. https://doi.org/10.1111/maec.12232

Wirtz, P. (1980). A revision of the Eastern-Atlantic Tripterygiidae (Pisces, Blennioidei) and notes on some West African blennioid fish. *Cybiton*, *11*, 83–101.

Wirtz, P. (2003). New records of marine invertebrates from São Tomé Island (Gulf of Guinea). *Journal of the Marine Biological Association of the United Kingdom, 83*(4), 735–736. https://doi.org/10.1017/S0025315403007720h

Wirtz, P. (2018). New records of marine invertebrates from São Tomé and Príncipe (Eastern tropical Atlantic). *Arquipélago - Life* and *Marine Sciences*, *35*, 41–46.

Wirtz, P., and d'Acoz, U. (2008). Crustaceans associated with Cnidaria, Bivalvia, Echinoidea and pisces at São Tomé and Príncipe islands.

Wirtz, P., Ferreira, C.E.L., Floeter, S.R., Fricke, R., Gasparini, J.L., Iwamoto, T., Rocha, L., and Sampaio, C.L.S. (2007). (Eastern Atlantic Ocean)— an update. *Zootaxa*, *48,* 1–48.

Zander, C.D., Nieder, J. and Martin, K. (1999). Vertical distribution patterns. Pp. 26-53 in *Intertidal Fishes: Life in Two Worlds* (eds. Horn, M.H., Martin, K.L.M. and Chotkowski, M.A.), Academic Press. San Diego, USA.

CHAPTER 2

Biodiversity distribution patterns on intertidal rocky shores of Príncipe Island (Gulf of Guinea)

Biodiversity distribution patterns on intertidal rocky shores of Príncipe Island (Gulf of Guinea)

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Abstract

For the past decades little work was made on intertidal communities of rocky shores of the west coast of Africa, particularly in the Gulf of Guinea. Furthermore, there is still a big lapse of information regarding rocky shores of the oceanic islands of the gulf, with virtually no study ever made. As global climate change poses increasingly more pressure on the environmental conditions of coastal ecosystems, especially in tropical regions, it is crucial to study intertidal communities and zonation patterns on vulnerable rocky shores, such as in the oceanic Príncipe Island. In this study, two rocky shores of the Príncipe Island, located in Abade and Bom-Bom, each representing respectively an estuarine-influenced and a marine environment, were sampled to describe and compare distribution patterns of intertidal biological assemblages. A total 47 taxa were documented during sampling, comprising 17 macroalgae, one marine lichen, and 29 epibenthic macrofauna taxa. Bom-Bom rocky shores had overall higher species richness, and although not significantly different compared to Abade, the presence of key species, such as more macroalgae and suspension feeder species in Bom-Bom, suggests a noteworthy difference between the biological communities of the exposed and sheltered shores. The biggest difference, however, was reflected in the zonation patterns between the two shores. The difference in zonation is noticeable, mainly in the infralittoral fringe and low eulittoral, where environment variables have a more frequent and pronounced effect. Overall, the rocky coast of Príncipe Island can be divided into three basic zones: the littoral fringe, dominated by littorinids of the genus *Echinolittorina*; the eulittoral, characterised by the black lichen *Verrucaria* sp., the oyster *Saccostrea cucullata* and the barnacle *Chthamalus dentatus*; and lower on the coast, the infralittoral fringe, largely dominated by the soft coral *Palythoa* caribaeorum and the coralline alga *Lithophyllum incrustans*. The distribution and zonation patterns presented some similarities with other rocky coasts of the mainland of the Gulf of Guinea.

Keywords: Tropical, West Africa, species richness, zonation, brackish water, wave action

Introduction

There is a vast number of studies relating to intertidal communities and distribution patterns in rocky shores globally. However, most works in West Africa are old and were carried out either north (e.g. Senegal and Canaries) or south (e.g. Angola and South Africa) of the Gulf of Guinea and very few were made in the equatorial region of the gulf (e.g. Bassindale, 1961; Buchanan, 1958; Gauld and Buchana, 1959; John and Lawson, 1991; Lawson, 1955, 1956, 1957, 1966; Lawson and John, 1982). Furthermore, very limited information exists regarding rocky shores of oceanic islands. A limited number of studies in São Tomé and Príncipe have addressed intertidal ecosystems, including rocky shores. These studies, however, were primarily checklists, namely of algae (Steentoft, 1965), molluscs (Fernandes and Rolan, 1993) and fishes (Afonso *et al.*, 1999; Wirtz *et al.*, 2007). These did not provide additional information on distribution patterns, nor on the overall intertidal communities and their interactions with environmental variables.

Rocky shore communities are generally simple, and distribution and zonation patterns can be easily described. Species distribution throughout different tidal levels is mostly linked to their physiological tolerance in emerged conditions and the biological interactions between each other (competition, herbivory and predation) (Louthan *et al.*, 2015; Raffaelli and Hawkins, 1999; Tomanek and Helmuth, 2002). Most classical descriptive works include the universal three-zone scheme of zonation (low, mid and upper shore) proposed by Stephenson and Stephenson (1949, 1972) and Lewis (1964). However, the stresses associated with the vertical gradient, which translates into increased time spent emerged, are known to change with latitude (Louthan *et al.*, 2015; Raffaelli and Hawkins, 1999). In regions at low latitudes, such as the tropics where extreme heat occurs, physiological stresses are more extreme high on the shore compared to middle latitudes (Raffaelli and Hawkins, 1999). The universal features of zonation are recognizable on tropical rocky shores, despite having generally a patchy distribution of species, which sometimes makes it difficult to assess the zonation patterns of certain species (Brusca, 1980; Raffaelli and Hawkins, 1999).

Within a region, certain environmental characteristics can also modulate community distribution on rocky shores. Exposure and salinity can affect zonation patterns and the extent of the vertical intertidal gradient, depending on the intensity of wave action and the presence of rivers nearby (Raffaelli and Hawkins, 1999). Exposed shores usually have strong wave action that allows species to survive meters above the theoretical tidal limit, causing an uplifted zonation compared to sheltered shores (Raffaelli and Hawkins, 1999). Salinity, on the other hand, can affect the species composition, where brackish species compete against truly marine species. However, little work has been done on the combination of these environmental factors in tropical regions.

Studies that address these issues have become increasingly more important nowadays to monitor global climate changes. Ocean warming, in particular, further pressures biological communities in rocky shores into their lethal temperature limits and is expected that tropical regions will be significantly affected (Horn *et al.*, 1999; Raffaelli and Hawkins, 1999; Vinagre *et al.*, 2018). According to Chou *et al.* (2020), projections of climate change for São Tomé and Príncipe predict an increase in temperature exceeding 3ºC and also heavier precipitation rates by the middle of the 21st century. Describing intertidal communities and zonation patterns in the Príncipe Island may provide a baseline reference for future studies that could assess the impacts of global climate changes in these environments.

This study aims to (i) describe and compare distribution patterns of intertidal biological assemblages in rocky shores in estuarine-influenced and marine environments of Principe Island, and (ii) record distributional records and assess distributions in the context of regional biogeography.

Materials and methods

Study locations

This work was carried out in Príncipe Island, Democratic Republic of São Tomé and Príncipe (Gulf of Guinea). The island is of volcanic formation, being part of the Cameroon Volcanic Line, which includes the islands of Annobón and São Tomé to the southwest and Bioko to the northeast and ending in Mount Cameroon. Príncipe Island is approximately 200 km from the coast of the mainland of Africa and although is characterised by a more oceanic environment compared to the mainland coast of the Gulf of Guinea (Krakstad *et al.*, 2010), it is still influenced by the plume of the Congo and Niger rivers with low salinity and high temperature water masses (Krakstad *et al.*, 2010; Measey *et al.*, 2007; Reid, 2011; Steentoft, 1965). Thus the surrounding ocean attains values of temperature of 27°C-30°C and salinity of <33-35‰ (Krakstad *et al.*, 2010). Príncipe Island was never connected to the mainland, resulting in significant differentiation of fauna and flora (Bonfim and Carvalho 2009). Still, the island lays on oceanic water of low productivity, even when influenced by the plume of the Congo and Niger rivers in wet seasons (Measey *et al.*, 2007, Reid, 2011). Of the four major tropical marine regions found globally, the eastern Atlantic has the lowest biological diversity (Reid, 2011).

The archipelago has a southern equatorial type of climate (Steentoft, 1965), with the average annual rainfall ranging from 2000-3000 mm (NBSAP II, 2015; Chou *et al.*, 2020), and with four climatic seasons that alternate between rainier and dryer periods. The major precipitation peak occurs between October and November, and a secondary peak around April and May (Chou *et al.*, 2020). These are interspersed by the less pronounced dry season that occurs between January and March (Chou *et al.*, 2020). The driest season ("gravana") occurs between June and September (Chou *et al.*, 2020; Pisoni *et al.*, 2015; Steentoft, 1965; Vaz and Oliveira, 2007). Air temperatures vary from 18-21 ºC minimums to 30-35 ºC maximums (Herrero-Barrencua *et al.*, 2017), with an annual average of 26 ºC (NBSAP II, 2015). The air relative humidity is very high, reaching more than 90% at higher altitudes (NBSAP II, 2015). Tidal ranges in the Gulf of Guinea are relatively small, not increasing by more than 1.9 m in extreme spring tides (0.2 m to 2.1 m from hydrographic zero), and 0.4 m in neap tides (1.0 m to 1.4 m from hydrographic zero) in Príncipe Island (IH, 2019).

Príncipe Island has a surface area of 142 km2 and is populated by about 8000 people that make their living mainly from natural resources near or on the coasts, complemented by a small ecotourism sector (Abreu *et al.*, 2017). A significant part of the coastline of the island is of rocky formation, often interspersed with sandy beaches. However, rocky shores are practically unexploited by the local population, whether its fauna or flora, with some minor exceptions like the crabs *Grapsus adscensionis* and *Johngarthia weileri* (JAS, personal observations).

Sampling was made at the rocky shores of Abade bay and Bom-Bom islet (Fig. 2.1). These locations were chosen for their relatively good access, for their extensive rocky shore and for presenting distinctive environment characteristics relevant for the present study.

Abade is located inside a small bay on the east coast of Príncipe Island, flanked by hills and with a narrow mouth, shielding the bay from wind from the north and east, and currents (Herrero-Barrencua *et al.*, 2017). The sea is mostly calm inside the bay, with very little wave action and substantial freshwater input from three watercourses. The largest of them, the Água Grande river, has enough flow to hold a mangrove ecosystem (Cravo *et al.*, submitted; Haroun *et al.*, 2018; Herrero-Barrencua *et al.*, 2017). These characteristics combined make this study area characterised as an estuarine environment with fluctuations in salinity throughout the year, depending on the season (Chou *et al.*, 2020; Herrero-

Barrencua *et al.*, 2017). The Abade's sampling site is in the innermost part of the bay, between Praia Salgada and the Abade's fishing village.

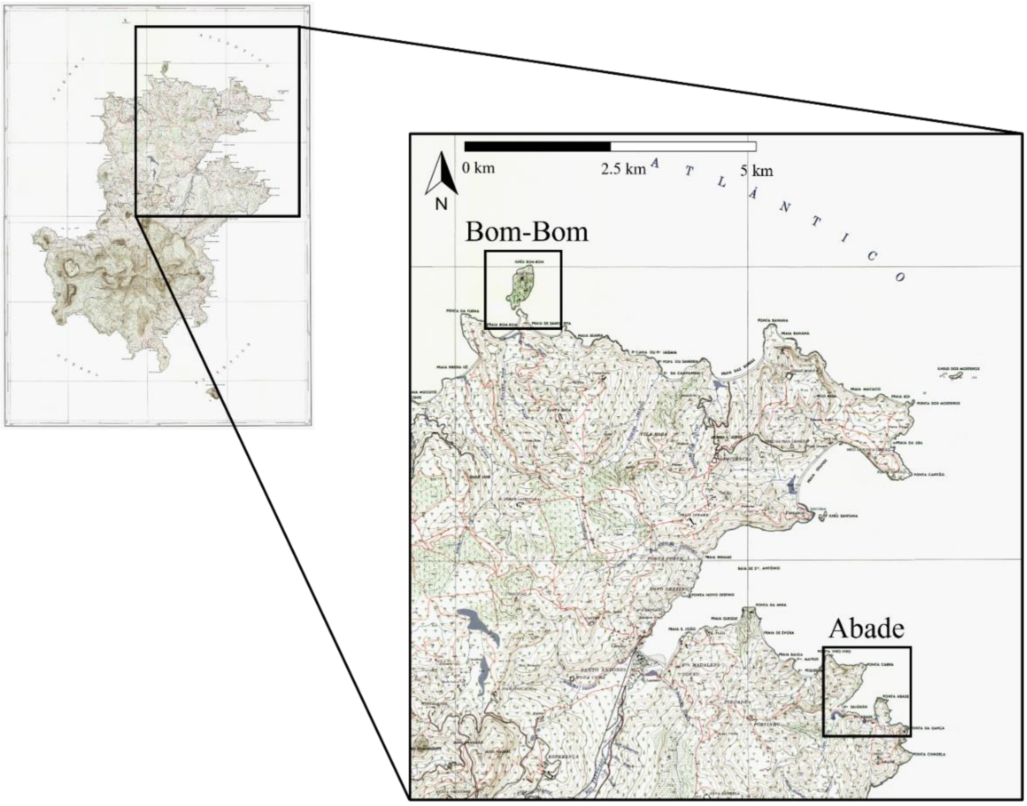

Figure 2.1. Position of the rocky shores of Abade bay and Bom-Bom islet. Abade is located 3 km south-eastward of the capital Santo António, and Bom-Bom 8 km north-westward.

The Bom-Bom islet is located in the north of the island and very close to the coast, with an exposed shore to the sea. The islet's shore is more susceptible to wave action due to the rapid decrease in depth of the surrounding seabed and more likely to be struck by breakers formed by the wind many kilometres away, which is usual in oceanic islands (Raffaelli and Hawkins, 1999). It also does not have rivers close enough or with enough flow to decrease considerably the salinity near the study area. Therefore, the Bom-Bom intertidal zone is characterised by being exposed to a marine environment, with strong water movement and virtually identical salinity to the open surrounding sea. Sampling was carried out on the west side of the islet.

Although there was some anthropogenic presence near the studied rocky shores, the human impacts are very low as there is virtually no use of these environments by local populations. Pollution was also not an issue in the study locations.

Sampling

Six transects were sampled during low spring tide between January 2020 and February 2020, which corresponds to the weak dry season (Chou *et al.*, 2020). In this period, low precipitation reduced the probability of the rock surface being wet and influencing mobile species distribution during sampling.

Three transects of 20 m long and 1.5 m wide, perpendicular to the coastline, were sampled at each location.

The rock profile from the sea-level at low water was made for each transect, using water levellers at each consecutive metre. Profile heights were later adjusted to hydrographic zero. At each metre, macro-flora and fauna were quantified using the quadrat method (Hawkins and Jones, 1992) with 50x50 cm and 49-point interceptions. Sessile species and water were quantified in percentage cover based on the point intersections, whereas mobile species were counted individually. Three contiguous quadrat replicates were sampled in each metre, at the middle, right and left.

Ideally, transects should be placed on freely draining open rock, for comparison and uniformity purposes. However, due to the irregular geomorphology of the island volcanic rocky shores, water was often found retained in crevices and cavities, making it almost impossible to avoid rock pools, big or small. So, if a quadrat was placed in a pool, crevice or some other microhabitat, it was not rejected. All sampling quadrats were recorded with a camera (GoPro Hero 7) for posterior analysis if needed.

The identification of flora and fauna species was carried out *in situ* by trained observers whenever possible. In case of taxonomic uncertainty, the alga or specimen was photographed, collected and when possible, brought to the laboratory for posterior identification. Algae species were identified using the *AlgaeBase* and the existing bibliography for the region (Steentoft, 2012). Invertebrates were identified using the dispersed available information, such as Reid (2011), Oyenekan and Olufowobi (1987), and the few comprehensive works for a few taxa, such as Manning and Holthuis (1981) for brachyuran crabs. Fishes were identified using Wirtz *et al.* (2007) and FAO (2016). Specimens from the sampling were deposited in the biodiversity collections of Principe Island Biosphere Reserve.

Data analysis

Non-parametric Mann-Whitney test was used to assess the dissimilarity in species richness between regions, as the data groups (Total biodiversity, “Algae + Verrucaria sp.”, “Sessile organisms” and “Mobile organisms”) were not homoscedastic. To assess the relationship between species richness and height from average sea level, the biological data was logarithmic transformed, and a Pearson correlation was made for each sampling region.

Results

Community composition

In total, 47 taxa were documented during sampling in the two rocky shores, comprising 17 macroalgae, one marine lichen, and 29 epibenthic macrofauna taxa (Table 2.1). In total, the most representative and diversified algae group was the Rhodophyta, and of the metazoans were the Mollusca and Arthropoda. Despite fish species not being usually considered in this type of study, some were considered. The species *Bathygobius burtoni*, *Entomacrodus cadenati* and *Microlipophrys velifer* showed to be intertidal resident species and representative of the Príncipe Island rocky coast biodiversity (Azevedo e Silva *et al.*, in prep).

The transects presented different average species richness between the two locations (Abade = 24.33 ± 3.93; Bom-Bom = 32.33 ± 3.38), however not significantly different (*p*-value = 0.40). Bom-Bom transects had higher average species richness for both algae and metazoans when compared to Abade (Fig. 2.2), but again not significantly different (Algae + *Verrucaria* sp. *p*-value = 0.4; Sessile organisms *p*-value = 0.4; Mobile organisms *p*-value = 0.2).

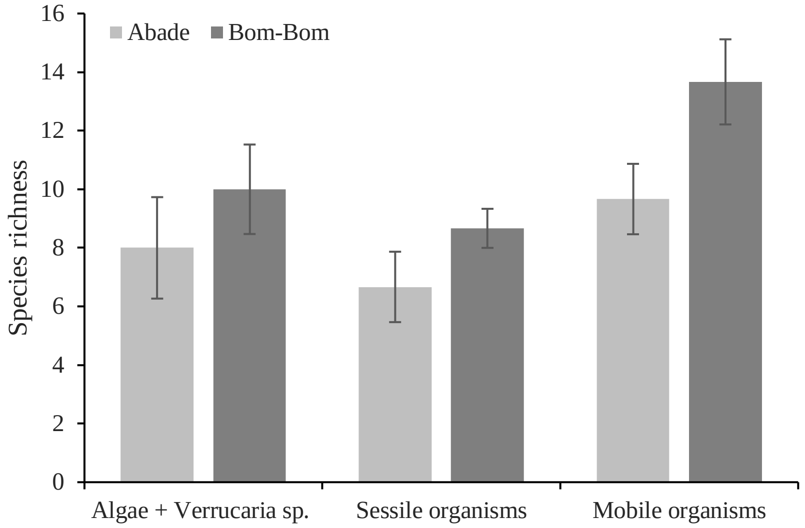

Figure 2.2. Average species richness per major type of organisms on transects in Abade and Bom-Bom.

The total community composition of each study location differed in some species (Table 2.1), although some species that were not present during the transect sampling, were recorded on posterior observations. These comprise of *Valonia ventricosa*, *Gelidiella* sp., Corallinales ND1, Corallinales ND2, *Coenobita rubescens*, *Grapsus adscensionis*, *Stramonita haemastoma*, *Dolabrifera dolabrifera* and Ophiuroidea n.d..

Still, some of the more abundant species revealed to be limited to just the location in which they were found. Bom-Bom rocky shore presented more exclusive species than Abade's. The algae *Padina boryana*, *Centroceras clavulatum*, *Ceramium* sp. and *Polysiphonia* sp., a non-determined anemone (Actiniidae ND2) and the giant barnacle *Megabalanus tintinnabulum* occurred only in Bom-Bom. Only one exclusive species from Abade was found, the small brown algae *Bostrychia* sp., occupying muddy rock surfaces.

Patterns of distribution

Water cover fluctuated along the transects (Fig. 2.4 and 2.7), depending mostly, at a larger scale, on the inclination of the rock profile, with the creation of rock pools on more irregular portions of the shore. The few exceptions were mainly due to irregularities such as holes and crevices in the rock. Based on the geomorphology heterogeneity and water retention patterns, we can summarize the micro-habitats into a few primary types: (i) the freely draining open rock surface, which can be either wet or dry, depending on how far it is from the wave splash; (ii) rock pools, that can have different shapes and depths, and different physical conditions; and (iii) crevices, which provide shade and damp micro-habitat.

Table 2.1. Taxonomic list of species sampled in the rocky shores of Príncipe Island, comparing the community composition at the two study locations, Abade bay and Bom-Bom islet. Species marked with "✓" means they were observed during sampling, whereas species marked with "(✓)" means they were observed outside the sampling period.

	Taxonomy			**Location**	
	Phylum	**Family**	**Species**	**Abade**	**Bom-Bom**
Chromista	Ochrophyta	Acinetosporaceae	*Feldmannia mitchelliae* (Harvey) H.-S.Kim 2010	✓	✓
		Dictyotaceae	*Dictyota* sp. J.V.Lamour., 1809	✓	✓
			Lobophora variegata (J.V.Lamouroux) Womersley ex E.C.Oliveira 1977	✓	✓
			Padina boryana Thivy, 1966		✓
Plantae	Chlorophyta	Chlorophyta n.d.	Chlorophyta ND1	✓	✓
		Valoniaceae	*Valonia ventricosa* J.Agardh, 1887	✓	(✓)
	Rhodophyta	Gelidiellaceae	*Gelidiella* sp. Feldmann and G.Hamel, 1934	(✓)	✓
		Ceramiaceae	*Centroceras clavulatu*m (C.Agardh) Montagne, 1846		✓
			Ceramium sp. Roth, 1797		✓
		Rhodomelaceae	*Bostrychia* sp. Montagne, 1842	✓	
			Laurencia obtusa (Hudson) J.V.Lamouroux 1813	✓	✓
			Polysiphonia sp. Greville, 1823		✓
		Corallinales n.d.	Corallinales ND1	(✓)	✓
			Corallinales ND2	✓	(✓)
		Lithophyllaceae	*Lithophyllum incrustans*	✓	✓
			Lithophyllaceae n.d.	✓	✓
	Ochrophyta	Phaeophyceae n.d.	Phaeophyceae n.d.	✓	✓
Fungi	Ascomycota	Verrucariaceae	*Verrucaria* sp. H.A.Schrader, 1794	✓	✓
Metazoa	Cnidaria	Siderastreidae	*Siderastrea radians* (Pallas, 1766)	✓	✓
		Sphenopidae	*Palythoa caribaeorum* Duchassaing and Michelotti, 1860	✓	✓
		Actiniidae	Actiniidae ND1	✓	✓
			Actiniidae ND2		✓
			Actiniidae ND3	✓	✓
	Annelida	Serpullidae	Serpullidae n.d.	✓	✓
	Arthropoda	Balanidae	*Megabalanus tintinnabulum* (Linnaeus, 1758)		✓
		Chthamalidae	*Chthamalus dentatus* Krauss, 1848	✓	✓
		Coenobitidae	*Coenobita rubescens* (Greeff, 1884)	✓	(✓)
		Diogenidae	Diogenidae n.d.	✓	✓
		Grapsidae	*Grapsus adscensionis* (Osbeck, 1765)	(✓)	✓
			Pachygrapsus gracilis (de Saussure, 1857)	✓	✓
		Xanthidae	*Platypodiella picta* (A. Milne-Edwards, 1869)	✓	✓
		Decapoda n.d.	Brachyura n.d. (Megalopa)		✓
	Mollusca	Ostreidae	*Saccostrea cucullata* (Born, 1778)	✓	✓
			Striostrea denticulata (Born, 1778)	✓	✓
		Mytilidae	*Mytilus* sp. Linnaeus, 1798	✓	✓
		Littorinidae	*Echinolittorina granosa* (Philippi, 1845)	✓	✓
			Echinolittorina soroziczac D. Reid, 2011	✓	✓
		Neritidae	*Nerita senegalensis* Gmelin, 1791	✓	✓
		Muricidae	*Thais nodosa* (Linnaeus, 1758)	✓	✓
			Stramonita haemastoma (Linnaeus, 1767)	✓	(✓)

Metazoa	Mollusca	Fissurellidae	*Fissurella* sp. Bruguière, 1789	✓	✓
		Aplysiidae	*Dolabrifera dolabrifera* (Rang, 1828)	(✓)	✓
	Echinodermata	Echinometridae	*Echinometra lucunter* (Linnaeus, 1758)	✓	✓
		Ophiuroidea n.d.	Ophiuroidea n.d.	(✓)	✓
	Chordata	Gobiidae	*Bathygobius burtoni* (O'Shaughnessy, 1875)	✓	✓
		Blenniidae	*Entomacrodus cadenati* Springer, 1967	✓	✓
			Microlipophrys velifer (Norman, 1935)	✓	✓
		Total species	**47**	**35**	**42**

The variation in species assemblages can be seen throughout both horizontal (variability of rocky shore profile including water retention) and vertical gradients (zonation in relation to tidal height). Abade transects were occupied from the start by a mix of species with different distribution ranges (Fig. 2.3). The more strictly water-dependent species occurred in the lower part of the transects, occupying a relative narrow infralittoral fringe. These were bounded to the less frequently emerged open rock surfaces lower in the shore, to permanent water reservoirs (e.g. rock pools), and in some cases, to more damp crevices. Most of these species were distributed from the water line to mostly below 0.5 m high, which corresponds to the mean low water of spring tides (MLWS) (Fig. 2.5). These include the algae *Dictyota* sp., *Bostrychia* sp., *Laurencia obtusa*, Corallinales ND2, *Lithophyllum incrustans*, Phaeophyceae n.d., the cnidarians *Palythoa caribaeorum* and Actiniidae ND3, the crab *Platypodiella picta*, the molluscs *Striostrea denticulata*, *Mytilus* sp., *Stramonita haemastoma*, *Fissurella* sp., *N. senegalensis* eggs, the sea-urchin *Echinometra lucunter* and the blenny *Entomacrodus cadenati*. The alga *Bostrychia* sp. dominated where more muddy surfaces occurred.

The other highly water-dependent species occurred mostly above 1 m high (Fig. 2.5) mainly in rock pools and occasionally in crevices. These include the algae *Feldmannia mitchelliae*, *Lobophora variegata*, Chlorophyta ND1, *Valonia ventricosa*, Lithophyllaceae n.d., the cnidarians *Siderastrea radians* and Actiniidae ND1, the spirorbid tube worm Serpullidae n.d., the hermit crab Diogenidae n.d., the gastropod *Thais nodosa*, and the fishes *Bathygobius burtoni* and *Microlipophrys velifer*.

The gastropod *Nerita senegalensis* also preferred wet environments, as most of its abundance varies proportionally to water cover, gradually reducing with the increasing of height and distance to the sea (Fig. 2.3). However, it can be moderately desiccation-tolerant since it appeared high on the shore and in places with very low water to none. Both *N. senegalensis* and *Mytilus* sp. distributed from water line up to just above the extreme high water of spring tides (EHWS), having the widest vertical distribution in Abade's rocky shores (Figure 2.5). The crab *Pachygrapsus gracilis* ranged between wet and dry environments and with no particular zone confinement, having a broad vertical distribution (Figure 2.5).

Growing in bare rock and usually seen emerged during low water is the encrusting black lichen *Verrucaria* sp., the small barnacle *Chthamalus dentatus* and the oyster *S. cucullata*. *Verrucaria* sp. has the largest cumulative cover, thus occupying the most area in Abade's rocky coast, and ranged from 0.5 m to 1.5 m high approximately (Fig. 2.5). *Chthamalus dentatus* and *S. cucullata* have a slightly uplifted vertical distribution, ranging from 0.7 to 2 m high (Fig. 2.5).

The littorinids *Echinolittorina soroziczac* and *E. granosa* are distributed further up the shore, occurring predominantly above 1.5 m high (Fig. 2.5) and seem to have an inverse relation with water cover (Fig. 2.3 and 2.4). *Echinolittorina granosa* dominates overall in the upper half of the Abade rocky shore and always reached the end of the transects (Fig. 2.3). At the top end of transect #3, the hermit-crab

Coenobita rubescens was sighted in the border between the intertidal volcanic rock and the terrestrial soil and sand where palm trees grow (Fig. 2.3).

All transects in both locations had an average slope increasing landwards, except for Bom-Bom transect #1, which between metres 8 to 16 had a significant negative slope, reaching 0.44 m of height (Fig. 2.6). As a result, species composition and distribution data from this transect deviate from the average.

Besides Bom-Bom having a relatively different flora and fauna when compared to Abade, both in species richness and composition, it also presents considerable differences in the overall species distribution patterns. Bom-Bom transects have proportionally more species near sea level, with three-quarters of total species occurring at least below MLWS (Fig. 2.8). The vast majority were highly water-dependent species that appeared rarely emerged on freely draining open rock surfaces or submerged in rock pools and crevices. These include all algae except for Corallinales n.d., Lithophyllaceae n.d. and *F. mitchelliae*, the cnidarians *P. caribaeorum* and Actiniidae ND1, the tube-worm Serpullidae n.d., the arthropods *M. tintinnabulum*, Diodenidae n.d. and *Platypodiella picta*, the molluscs *S. denticulata*, *T. nodosa*, *Fissurella* sp. and *N. senegalensis* eggs, the echinoderms *E. lucunter* and Ophiuroidea n.d., and the fishes *E. cadenati* and *Microlipophrys velifer*. They reached various heights; however, there is an overall rise of their upper limit compared to the similar water-dependent species assemblage in Abade.

More sessile species occupy the freely draining open rock at the beginning of the transects (Fig. 2.6) such as *C. clavulatum*, the mix of *Ceramium* sp. and *Polysiphonia* sp., Lithophyllacea ND1, Serpullidae n.d., *M. tintinnabulum*, and the small barnacle *C. dentatus*. Excluding the small barnacle, the rest were gradually replaced by the black lichen *Verrucaria* sp. and the oyster *S. cucullata*.

Slow mobile species that can attach firmly to the rock, such as *Fissurela* sp. and *E. lucunter*, are generally more abundant near the sea and are gradually replaced by mobile organisms less capable of attachment, like Diogenidae n.d., small crabs, *N. senegalensis* and *T. nodosa* (Fig. 2.6). The crabs *Grapsus adscensionis* and *Pachygrapsus gracilis*, being well adapted to holding on to rocks, were also common in the break zone and throughout the lower shore. *Grapsus adscensionis* is very elusive and fast, hence few individuals were counted despite being abundant on the studied rocky shores. Nevertheless, its known vertical distribution data is according to what was observed in the field, where individuals were seen grazing on algae like *C. clavulatum* and the mix of *Ceramium* sp. and *Polysiphonia* sp..

Other water-dependant species were only found upper in the shore in rock pools, such as *L. variegata*, *Gelidiella* sp., *S. radians*, Actiniidae ND3, *Dolabrifera dolabrifera* and *B. burtoni*. Many of these occurrences are from the second half of transect #1 (Fig. 2.6), hence their low vertical position (Fig. 2.8). *Nerita senegalensis* has also a wide-ranging distribution, occupying various environments, in favour of wetter areas (Fig. 2.7), unless when the rock was over-occupied with more dominant species, such as sea urchins and coral (Fig. 2.6).

The small littorinids *E. soroziczac* and *E. granosa* appear higher on the shore and dominate the upper end of the rocky coast (Fig. 2.6) above 1.5 m height or mean high water of neap tides (MHNT) (Fig. 2.8). Individuals found in transect #1 were an exception, being found at lower heights than 0.76 m because they were farther from the sea (Fig. 2.6).

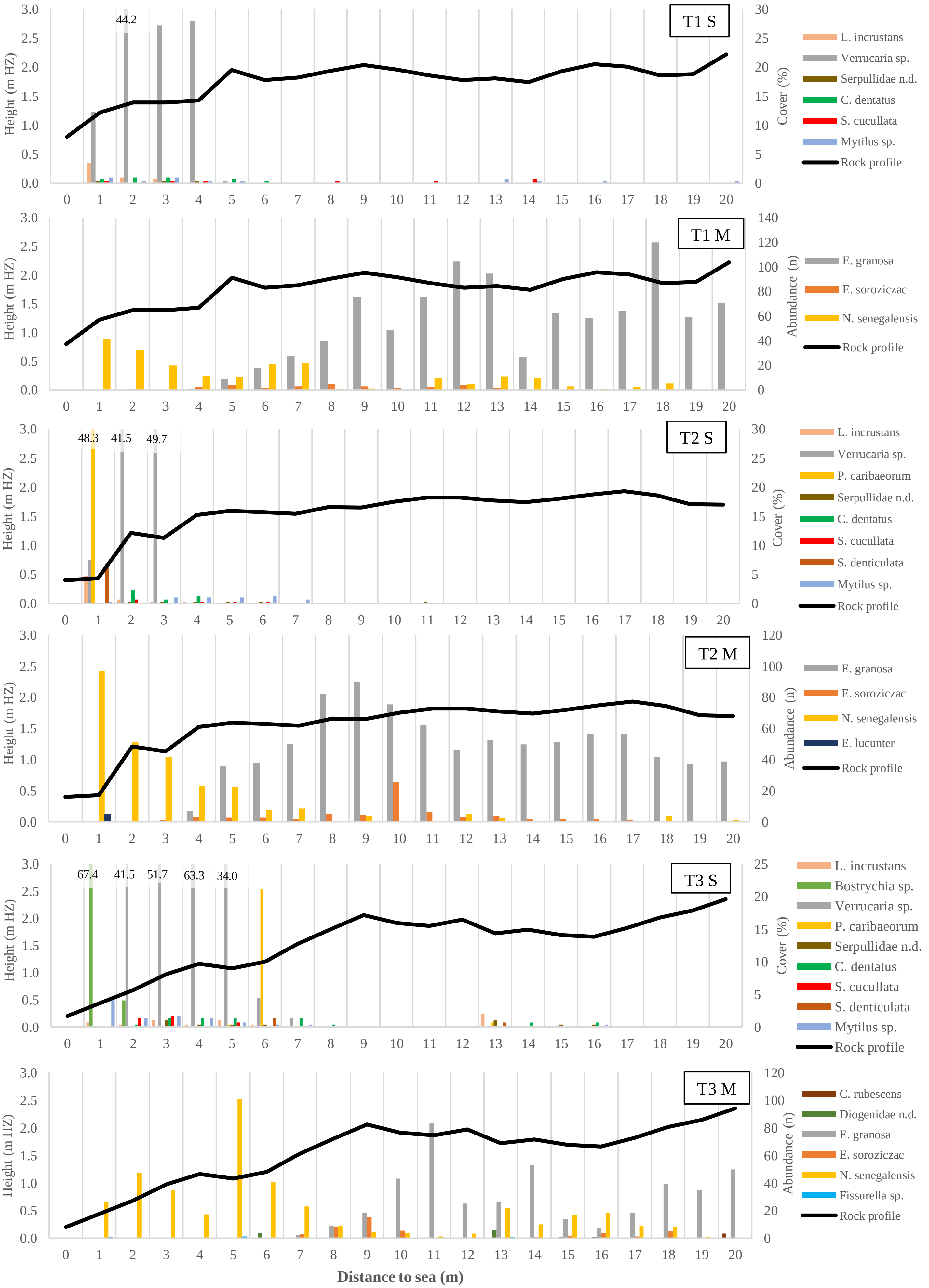

Figure 2.3. Distribution of the most representative sessile (S) and mobile (M) species found in Abade's transects, along each transect rock profile. Height is referred to the Hydrographic Zero (HZ).

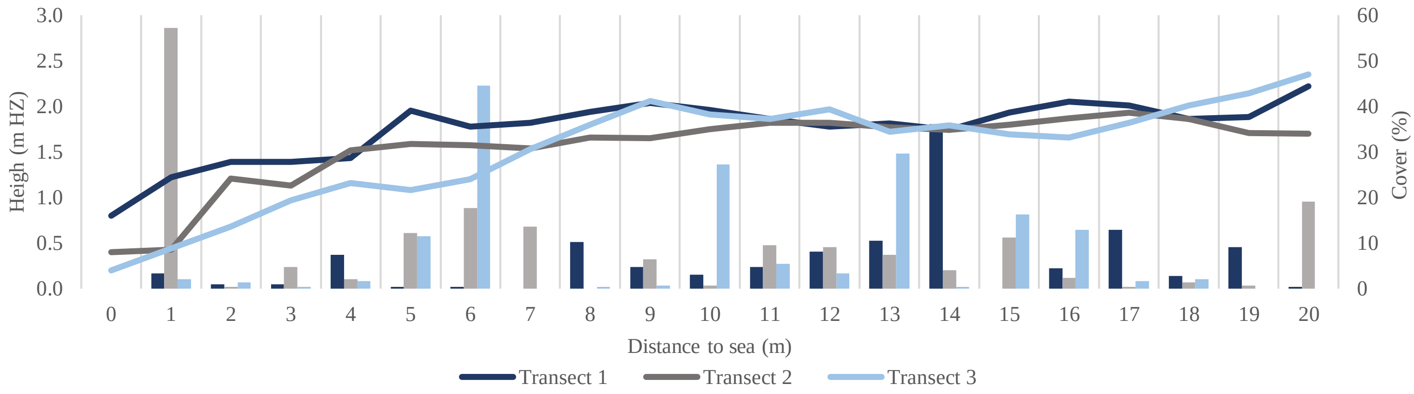

Figure 2.4. Water accumulation along the three transects sampled in Abade's rocky shore. Sea-level height at low-water for transect number 1 is 0.8 m, for transect number 2 is 0.4 m, and for transect number 3 is 0.2 m. Height is referred to the Hydrographic Zero (HZ).

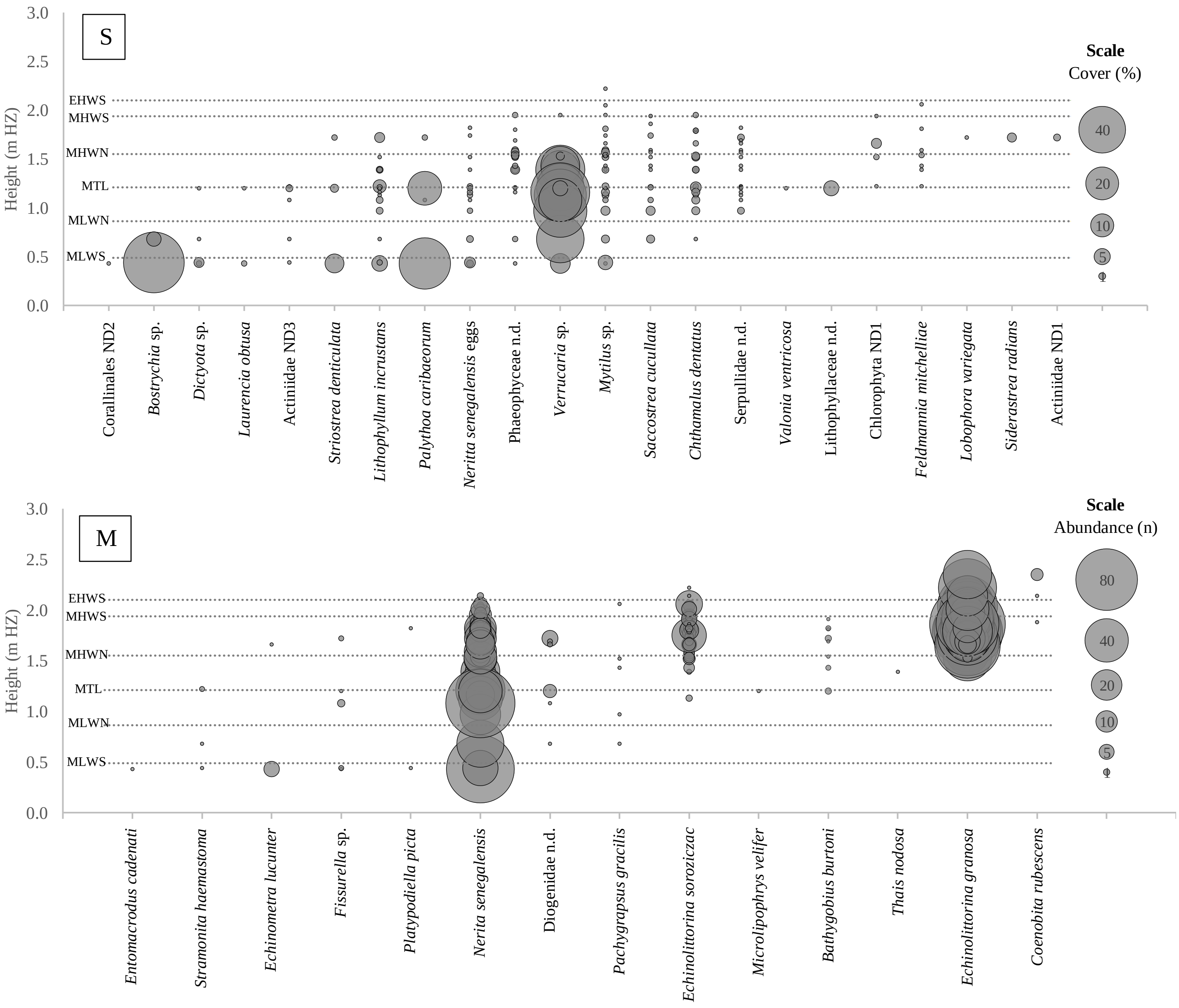

Figure 2.5. Vertical distribution along the height from Hydrographic Zero (HZ) of the average percentage cover of all sessile species (S) and average abundance of all mobile species (M) documented in Abade's transects. Tidal levels are represented for comparison context purposes.

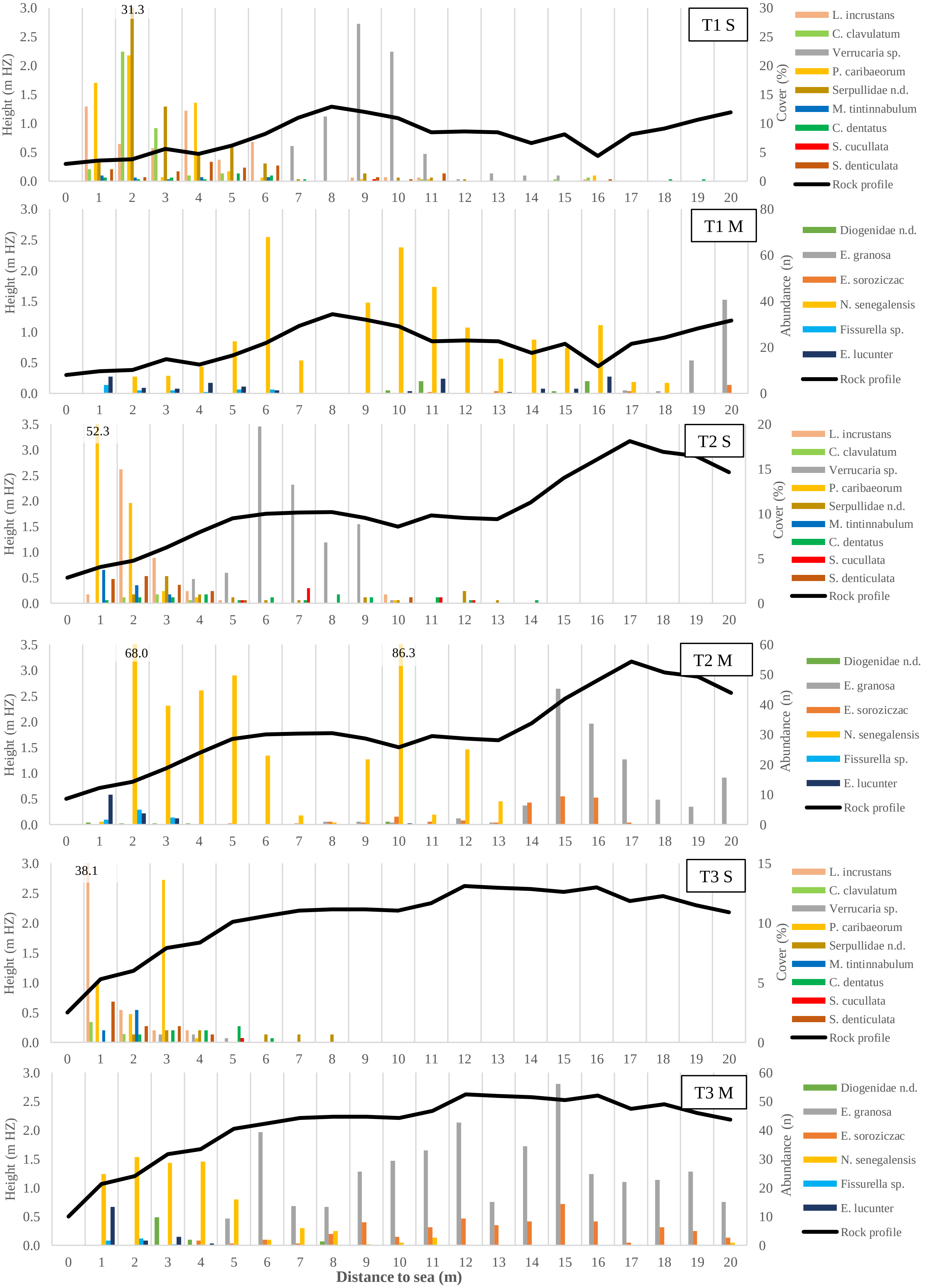

Figure 2.6. Distribution of the most representative sessile (S) and mobile (M) species found in Bom-Bom's transects, along each transect rock profile. Height is referred to the Hydrographic Zero (HZ).

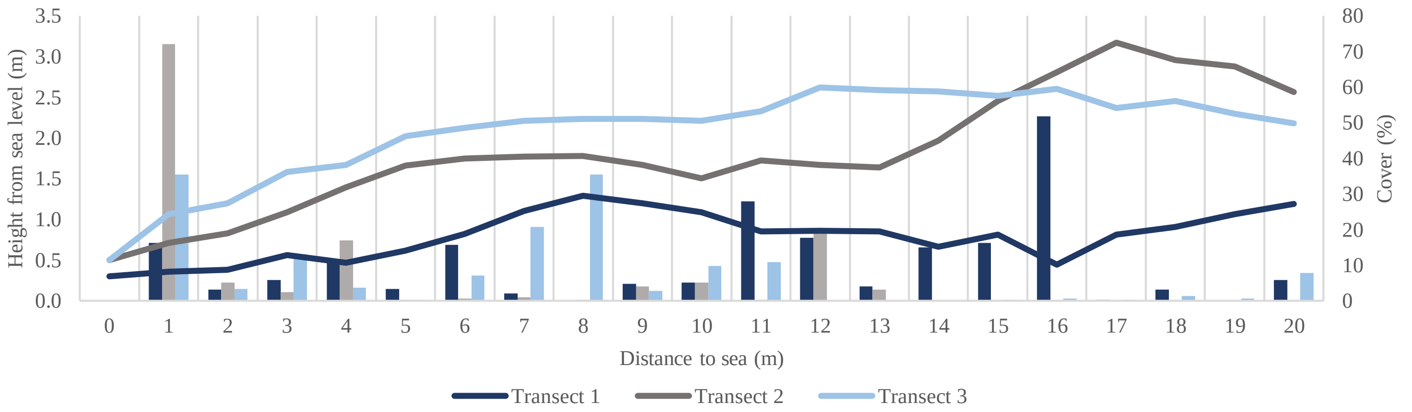

Figure 2.7. Water accumulation along the three transects sampled in Bom-Bom's rocky shore. Sea-level height at low-water for transect number 1 is 0.3 m, for transect number 2 is 0.5 m, and for transect number 3 is 0.5 m. Height is referred to the Hydrographic Zero (HZ).

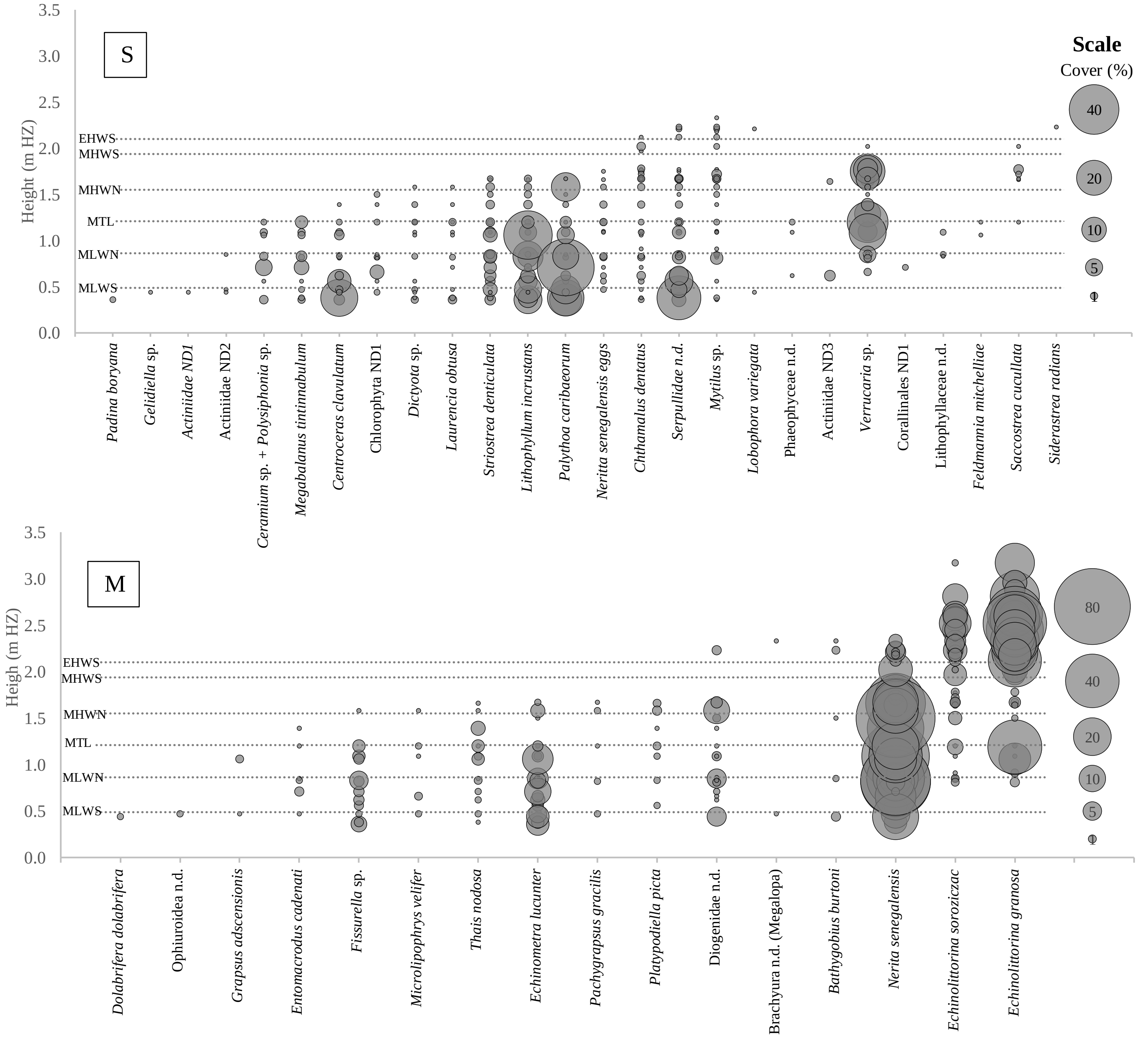

Figure 2.8. Vertical distribution along the height from Hydrographic Zero (HZ) of the average percentage cover of all sessile species (S) and average abundance of all mobile species (M) documented in Bom-Bom's transects. Tidal levels are represented for comparison context purposes.

Species richness showed to be significantly and inversely correlated to heigh from average sea level, both in Abade (ρ = - 0.696; *p*-value = 6,473E-10) and Bom-Bom (ρ = - 0.711; *p*-value = 1,884E-10), as evident in Figure 2.9. It is also noticeable a difference between both regressions of species richness in the initial portion of the vertical gradient, which gradually decreases with height until interception near meter 3, with the trendline of Bom-Bom standing overall above Abade's.

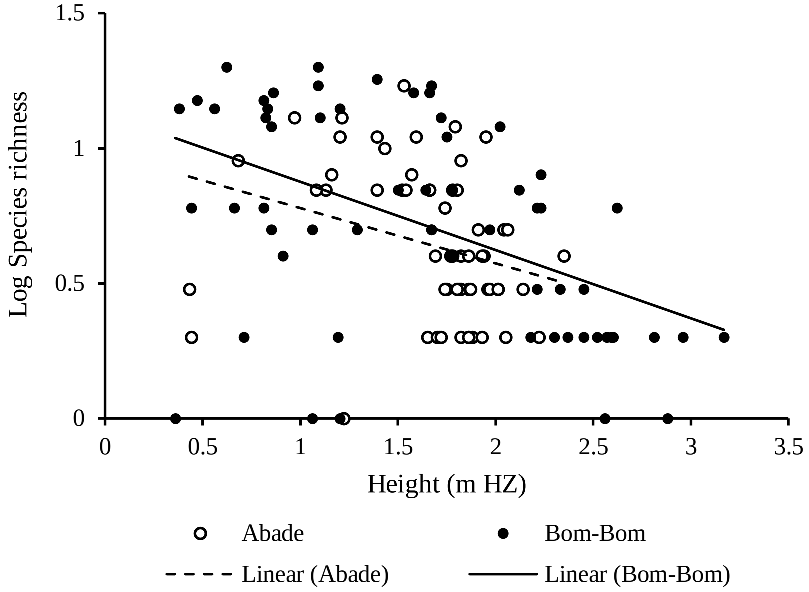

Figure 2.9. Logarithmized species richness at Abade and Bom-Bom in function of the height from the Hydrographic Zero (HZ).

General zonation patterns

An intertidal zonation scheme was determined based on vertical distribution patterns of different species assemblages (Fig. 2.10). Three major zones were clearly discernible based on the dominant biological assemblages, although some species occurred in more than one zone and the boundaries were blurred in places. In this study, the three major zones are referred to as the sublittoral fringe (low shore), the eulittoral zone (mid shore) and the littoral fringe (high shore), adopting terms used by Lewis (1964). To be noted that Lewis' scheme is based on the relative positions of the main community types that are observed along the vertical gradient. Although the upper limits of distribution of many species can be associated with major high and low tidal levels (Raffaelli and Hawkins, 1999), the zones were not defined directly from these but rather associated with based on the vertical distribution patterns of the main species.

The sublittoral fringe corresponds to the zone with the most species and differs the most in species composition between locations. It is characterised and delineated by a profusion of highly water-dependent species that are only periodically emerged during spring tides. The encrusting pink alga *L. incrustans*, the yellow coral *P. caribaeorum*, the oyster *S. denticulata*, and the sea-urchin *E. lucunter* are common representative species of the sublittoral in both shores. The sublittoral upper limit at Abade is determined by the range of these species and the alga *Bostrychia* sp., corresponding to the mean low water of spring tides (MLWS). This zone differs considerably in the more exposed shore of Bom-Bom. It features a more co-dominated assemblage, which comprises the same common sublittoral species as well as the Bom-Bom characterising species *C. clavulatum*, *Ceramium* sp., *Polysiphonia* sp., and *M. tintinnabulum*. The distribution in which these species occur extends up to the mean sea level (MSL), making Bom-Bom's sublittoral considerably more extensive.

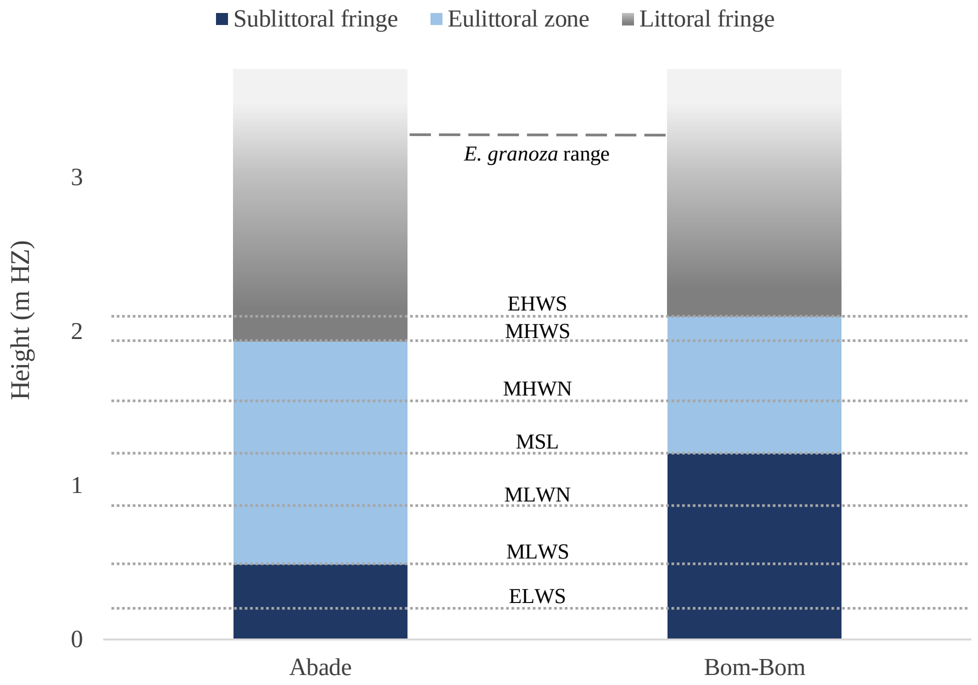

Figure 2.10. Intertidal zonation of the Abade sheltered rocky shore and the Bom-Bom exposed rocky shore. Regional tidal levels from the Hydrographic Zero (HZ) are represented for comparison: extreme low water of spring tides (ELWS) at 0.20 m; mean low water of spring tides (MLWS) at 0.49 m; mean low water of neap tides (MLWN) at 0.86 m; mean sea level (MSL), at 1.20 m; mean high water of neap tides (MHWN) at 1.55 m; mean high water of spring tides (MHWS) at 1.94 m; and extreme high water of spring tides (EHWS) at 2.10 m. The upper limit of the littoral fringe corresponds to the range of the littorinid *Echinolittorina granosa.*

The eulittoral zone is occupied by a variety of species with different distribution and attachment strategies, and likely varying tolerances to physical stresses. The zone's lower limit is well marked with the appearance of the black lichen *Verrucaria* sp. in the open rock surface, which vastly dominate the low eulittoral, and the oyster *S. cuccullata*. The eulittoral lower limit is significantly lower in Abade, setting a wider eulittoral zone for the sheltered coast. *Chthamalus dentatus* and *S. cucullata* occupy the freely draining open rock surface and establish the upper limit of the zone, which varies just slightly between both locations. In Abade, it corresponds to the mean high water of spring tides (MHWS) and in Bom-Bom to the extreme high water of spring tides (EHWS). Species like *N. senegalensis*, Serpullidae n.d., *Mytilus* sp. and Phaeophyceae n.d. also characterise this zone, inhabiting continuously from the sublittoral and into the eulittoral, with preference of humid or shaded crevices.

The littoral fringe is dominated by the littorinids *E. soroziczac* and mainly by *E. granosa* which define the zone's upper limit. Both gastropods were recorded from the mid of the eulittoral zone, with only *E. soroziczac* distribution varying significantly between regions, wherein Abade extends until just above EHWS, and in Bom-Bom above 3m high. The lower limit it's defined by the absence of the sessile species which established the eulittoral upper limit. The theoretical approximate upper limit for each region is, however, uncertain due to the lack of data to determine *E. granosa* maximum range.

Discussion

Community composition

Although the species richness of different taxonomic groups did not indicate a significant difference between the two study locations, some conspicuous and dominant species showed to be exclusive of just one location. This indicates there is a noteworthy distinction between the biological communities of both rocky shores. The differences are suggested to be mainly due to the wave action (stronger at Bom-Bom) and the influx of freshwater (higher in Abade). Overall, it was observed higher species richness in Bom-Bom compared to Abade.

Wave action has a profound effect on the biological characteristics of rocky shores, being mostly beneficial (Raffaelli and Hawkins, 1999). Waves disperse more particle food in the water, making Bom-Bom shore more favourable to suspension feeders (McQuaid and Branch, 1985; Raffaelli and Hawkins, 1999), thus having the unique occurrence of the anemone Actiniidae ND2 and the barnacle *M. tintinnabulum*. There is also more supply of oxygen and nutrients for both flora and fauna (Raffaelli and Hawkins, 1999), allowing the existence of more species from the first trophic level, particularly algae such as *Padina boryana*, *Centroceras clavulatum*, *Ceramium* sp. and *Polysiphonia* sp.. Consequently, a more complex food chain establishes, composed of algae feeders like crabs, limpets, sea snails, sea slugs, sea urchins and herbivorous fishes, and finally more predators. In addition, an exposed shore is more prone to have a stronger post-larvae supply brought by water movement, and it was observed crab post-larvae (megalopa) only in Bom-Bom islet and other rocky coasts around the island with similar exposed conditions (authors' personal observations).

In Abade, the large amount of suspended debris carried by rivers, like fine sediment and organic matter, that are deposited into the bay, combined with the overall poor water movement, results in anoxic zones and turbid water (Herrero-Barrencua *et al.*, 2017), limiting oxygen supply to intertidal flora and fauna. Even during the dry seasons, where there is substantially less freshwater input, the weak water movement in the bay is not enough to significantly increase the oxygen levels, although there is a lack of data to support this scenario. Furthermore, there is the salinity stress that restricts many marine species to survive on Abade's rocky shore. According to Raffaelli and Hawkins (1999) "Only those that can tolerate lower and often fluctuating salinities can endure the less saline periods. Species richness is low, but populations may attain very high densities and biomasses", like in the case of *Echinolittorina granosa*. Even so, a considerable number of marine species occur at the estuarine influenced Abade embayment. The exclusive and abundant small brown algae *Bostrychia* sp. is found associated with muddy rock surfaces; this species also occurs in local brackish water environments such as local mangroves (authors' personal observations).

Only sessile species presented relevant differences between locations. Mobile organisms, unlike sessile species, can distance themselves from physiological and biological stresses, and seek refuge if needed. Therefore, are generally more adaptable to environment fluctuations and have better chances of surviving.

Additionally, to the sampling period, some unidentified individuals of Polyplacophora where found in both shores under boulders in rock pools. Dell' Angelo *et al.* (2014) have described some species of this class for São Tomé and Príncipe islands, of wich the species *Lepidochitona rolani* was reported in the intertidal environment.

Overall, although no statistically significant differences were found in terms of species composition, some key species revealed that there is a notable difference between the intertidal communities of the exposed and sheltered shores, as for instance the presence of more algae and suspension feeder species on the more exposed shore and muddy associated alga on the sheltered shore.

Patterns of distribution

In tropical areas, desiccation becomes an extreme stress factor due to the combined effects of heat and low relative humidity in the rocky shores (Louthan et al., 2015; Raffaelli and Hawkins, 1999; Williams, 1993). Therefore, it is most likely that species upper limits are more directly set by physical factors, especially for sessile species, although limited information is available from tropical shores (Williams, 1993). The same physical conditions that affect species composition also determine, to some extent, the vertical and horizontal gradients and subsequentially the patterns of species distribution. Thus, acknowledging first the water accumulation patterns is vital to better understand species distribution patterns.

Wave action enables species to live higher on the coast in the splash zone, particularly in the tropics, where organisms are often exposed to extreme heat (Raffaelli and Hawkins, 1999). Being wave action more prominent at Bom-Bom shore, and with a much lower freshwater influence, low shore communities spread out as far as tides and waves allow it. Whereas in Abade, the low salinity water inflow and the absence of wave splashes seem to hamper the expansion of low shore communities, resulting in a narrower sublittoral fringe. This is evident when comparing the width of the sublittoral fringe at each location, which have a 0.7 m difference between the respective upper limits. A similar difference was also found between exposed and sheltered rocky coasts in Ghana (Gauld and Buchana, 1959). Lithothamnia was a constant feature that clearly marked the upper limit of the sublittoral fringe both in Príncipe Island and in all the West African shores (Gauld and Buchana, 1959). The distribution of the echinoid *E. lucunter* was also linked to the upper limit of the sublittoral fringe in the Gulf Guinea (Lawson, 1956).

Lower limits are, however, unlikely to be related directly to physical factors, but rather to biological ones such as competition (Connell, 1961a,b), grazing and predation (Louthan et al., 2015; Raffaelli and Hawkins, 1999; Tomanek and Helmuth, 2002). With extreme desiccation, the liveable space becomes the limiting resource, making the competition for space the primary bottleneck. Low-shore sessile species are the most dominant, usually for being much larger and growing more rapidly than species higher on the shore (Stephenson and Stephenson, 1949; Southward, 1958; Lewis, 1964), such as the zoanthid *P. caribaeorum* (Guilhem *et al.*, 2020) and the coralline encrusting red algae *L. incrustans* (Edyvean and Ford, 1987). Thus, there is little available space on the rock surface in the sublittoral fringe, which sets the lower limit for most species found in the eulittoral zone. In certain cases, similar to that of Abade, the usual dominant species do not occur abundantly in the subtidal due to the low salinity, allowing mid-shore species, such as *Verrucaria* sp. and *S. cucullata*, to be able to grow free of competition downwards (Wallentinus, 1991), hence the broader eulittoral zone in Abade.

Mobile organisms, on the other hand, have more dynamic upper and lower limits which reflect the behaviour and physiological tolerances of different species and single individuals (Raffaelli and Hawkins, 1999). The dynamic boundaries are, in this case, in accordance with both the physical conditions and biological interactions that set upper and lower limits respectively. However much of this discussion is theoretical, and hard evidence, as Raffaelli and Hawkins (1999) point out, is seldom seen. In Bom-Bom, the main grazers such as grapsid crabs (Grapsidae), the limpet *Fissurella* sp. and the sea-urchin *E. lucunter*, are more abundant in the sublittoral fringe, since it is characterised by short turf-forming algae such as *Centroceras clavulatum, Ceramium* sp., *Laurencia obtusa,* and *Dictyota* sp..

In Abade, grazer organisms are more scattered throughout the coast since they forage mostly in algae from rock pools. The upper limit of the sublittoral fringe of rocky coasts in mainland in the Gulf Guinea was also linked to the distribution of the echinoid *E. lucunter* (Lawson, 1956).

The fast drainage to the sea and high exposure to intense solar radiation are constant factors in the tropical rocky shores. Combined, these make the dry open rock surface the most common habitat in Príncipe's rocky shores during low tide, mainly in the eulittoral and littoral zones. A black band of *Verrucaria* sp. covers most of the low eulittoral. Usually, *Verrucaria* lichens are a common worldwide feature of the littoral fringe (Boaventura *et al.*, 2002; Raffaelli and Hawkins, 1999). But because of extreme desiccation, it is probably forced to occupied lower areas that are emersed for considerably less time. Very few organisms can occupy such extreme conditions, where high temperatures and intense solar radiation exposure limits survival considerably. Only *C. dentatus* and *S. cucullata* were found also inhabiting the eulittoral exposed rock surface, although presenting minimal cover.

Rocky shores are rarely smooth, and the basaltic coast Príncipe Island is no exception. Its volcanic rock formation is characterised by numerous fissures, crevices, gullies and pools, which provide vital refuges from extreme physical stresses at increasingly higher grounds (Williams, 1993). Rock pools occur in various shapes and sizes and were often the major exceptions to the upper limits of many lower shore species. They generally enhance physical conditions in such a way that sometimes enables species richness to be nearly as high in the eulittoral as in the low sublittoral, thus the occasionally substantial abundance of sublittoral dominant species in the higher shore. Rock pools can be also the exception to the biological factors that determine the lower limits of intertidal zones, being sometimes determined by physical factors instead of biological factors. In Bom-Bom, the rock pools have the particular characteristic of hosting species that cannot settle in the open rock surface due to wave action, such as the hermit-crab Diogenidae n.d., other small crabs and fishes.

In Abade, rock pools gain particular importance for marine intertidal species that live in a low salinity environment. During low water periods, pools provide higher salinity conditions through evaporation of the retained water (Horn *et al.*, 1999), offering a more bearable microhabitat for many marine species to cope with at a certain vertical range. This may explain why some species have uplifted lower-limits of distribution in Abade compared to Bom-Bom, such as the tube-worm species (Serpullidae) and the goby *B. burtoni*, appearing in rock pools above MLWN in Abade. But because of insufficient wave action locally, water above MHWS becomes too saline to tolerate.

Fissures and crevices are usually shaded and either very wet or damp, resulting in considerable small-scale variation in physical conditions. Nevertheless, these microhabitats are still too stressful for many marine species. In the eulittoral zone and littoral fringe, tube worms, mussels and gastropods, mainly *N. senegalensis,* dominate these microhabitats if sufficient water is retained during emersion time. The mussel *Mytilus* sp. can occur in extreme wave exposure (Hiscock, 1983) and reduced-salinity environments (Russell *et al.*, 1983; Hawkins *et al.*, 1992), and was found from the sublittoral to the low littoral fringe on both studied shores, having the widest vertical distribution of all sessile species. *Nerita senegalensis* is also known to be very tolerant to fluctuations in temperature and salinity and can withstand significant wave action and desiccation conditions (Oyenekan and Olufowobi, 1987). Found from the sublittoral to the low littoral fringe in both shores, submerged and emerged, it is the most adaptable and broadly distributed species of all rocky shore communities in Principe Island.

Although fissures and crevices independently represent a small proportional area, in total correspond to a significant and continuous microhabitat along the shore rock surface. Thus, the species that inhabit them have virtually an uninterrupted presence in their respective range of distribution, more so for mobile organisms than sessile species. Sessile organisms like Serpullidae n.d., *C. dentatus* and *Mytilus* sp. are significantly small to be able to colonize a wide spectrum of the coast, within their physiological tolerance.

At progressively higher levels on the shore, crevices are exposed to air for increasingly longer periods of time. Despite the high relative humidity of the air on the island, extreme desiccation still poses a challenge (Raffaelli and Hawkins, 1999; Williams, 1993). Slightly damp and shaded, crevices on the high shore provide the only refuge from extreme air temperatures, where only a few gastropods species can endure. *Nerita senegalensis* is still able to inhabit these hardship environments up to just above EHWS. A considerable gain in size of upper shore individuals, that were mostly emerged, was noticed in field observations, although not quantified. This is probably due to the increasing capability with the ageing of concealing a bigger volume of seawater within the shell, which is use for respiration during exposure (Oyenekan and Olufowobi, 1987).

However, as conditions get less suitable for *N. senegalensis*, the species is gradually replaced by the more desiccation-tolerant littorinids *E. granosa* and *E. soroziczac*, who dominate much of the upper shore. Littorinids are recognised as being a characteristic group of the littoral fringe worldwide (Boaventura *et al.*, 2002; Miller and Denny, 2011; Raffaelli and Hawkins, 1999; Reid, 2011), and in particular those from the genus *Echinollitorina* are the more dominant on rocky shores in tropical and warm-temperate latitudes of the global oceans (Reid, 2011). The two species presented very similar vertical distributions comparatively to each other, although *E. granosa* occupied a higher shore level than *E. soroziczac*. A similar arrangement was described in the continental coast of the Gulf of Guinea, with *E. granosa* and *E. pulchella* (littorinid of the same subspecies of *E. soroziczac* (*Amerolittorina*)). Whenever these two species were found together, *E. granosa* occupied a higher level (Gauld and Buchana, 1959; Lawson, 1956; Lawson, 1966; Lawson and John, 1982; John and Lawson, 1991). *Echinollitorina granosa* appeared in great abundance in both exposed and sheltered shores, as previously noticed by Gauld and Buchana (1959), but in much larger numbers in Abade because of the preference for sheltered coasts (Bassindale, 1961; Collignon, 1960) and coastlines influenced by river outflows. Additionally, a wider eulittoral zone with a lower low limit had probably granted more space for colonisation by the littorinid. *Echinollitorina granosa* maximum upper limit description was limited by the sampling methodology, but individuals were sighted reaching up to 3 m above MHWS in the Gulf of Guinea (Bassindale, 1961). Its observed distribution from the upper eulittoral and in the littoral fringe is according with previous records in the Gulf of Guinea (Bassindale, 1961; Gauld and Buchana, 1959; Lawson, 1956; Vermeij, 1972, 1973). For example, in comparison to other rocky shores studied in the mainland of the Gulf of Guinea, the clearly observable littorinid and balanoid zones also marked the littoral fringe and provide an upper limit to the midlittoral zone (Gauld and Buchana, 1959).

Overall, because intertidal species in this region are particularly vulnerable due to harsh physical conditions, mainly extreme desiccation, the upper limits, mainly of sessile species, appear to be predominantly defined by the direct action of physical factors (tides, wave-action). While lower limits are defined more directly by biological factors, particularly competition for space in the infralittoral fringe and low eulittoral zone. As a result, the rocky coast is characterised by a highly crowded and patched infralittoral fringe dominated by more water-dependent species, followed by an eulittoral zone and littoral fringe with a much lower species density and richness, and largely dominated by highly desiccation-tolerant species. The infralittoral is largely dominated by the soft coral *P. caribaeorum*, the coralline alga *L. incrustans*. The eulittoral is characterised by the black lichen *Verrucaria* sp., the oyster *S. cucullata* and the barnacle *C. dentatus*. A band of black lichen is located lower compared to regions of other latitudes. And the littoral fringe is predominantly dominated by littorinids *E. granosa* and *E. soroziczac*. The universal zonation system of three zones proposed by Stephenson and Stephenson

(1949) and Lewis (1964) can be applied in the intertidal communities of the tropical rocky coast of Príncipe Island.Príncipe Island has at least two distinct marine community structures that reflect essentially two common, but different rocky shores environments found around the island, namely those exposed to the marine environment and those influenced by rivers and with natural barriers. The difference in the zonation and species distribution patterns is noticeable, mainly in the infralittoral fringe and low eulittoral, where environment variables have a more frequent and pronounced effect.

Some sampling constraints of the present study limit our interpretation of the distribution patterns observed, and much remains to be done. In particular no data was collected from other seasons, which could reveal seasonal variation in species composition and abundance, especially in Abade where there is greater seasonality induced by the precipitation cycles along seasons.

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References

Abreu, A.D., Asensio, M.T., Cervera, J.L., Cosme, M., Fuentes, I., García-Mendoza, A., Gómez, P., Haroun, R., Herrero Barrencua, A., Matilde, E., Niccolini, M., Otero-Ferrer, F., Tuya, F. and Viera-Rodríguez, M.A. (2017). Final Report Bio-Príncipe 2016. IU-ECOAQUA, Universidad de Las Palmas de Gran Canaria, Spain. 28 pp.

Afonso, P.F., Porteiro, F.M., Santos, R.S., Barreiros, J.P., Worms, J. and Wirtz, P. (1999). Coastal marine fishes of São Tomé Island (Gulf of Guinea). *Arquipélago, Life* and *Marine Sciences,* 17A: 65–92. Ponta Delgada. ISSN 0873-4704.

Azevedo e Silva, J., Almeida, A.J., Cravo, M., Pais, M.P., Santos, Y. and Paula, J. (in prep.). Patterns of fish distribution in rock pools from tropical intertidal rocky shores at Príncipe Island (Gulf of Guinea).

Bassindale, R. (1961). On the Marine Fauna of Ghana. *Proceedings of the Zoological Society of London, 137*(4), 481–510. https://doi.org/10.1111/j.1469-7998.1961.tb06086.x

Boaventura, D., Ré, P., Da Fonseca, L.C. and Hawkins, S.J. (2002). Intertidal rocky shore communities of the continental Portuguese coast: Analysis of distribution patterns. *Marine Ecology, 23*(1), 69–90. https://doi.org/10.1046/j.1439-0485.2002.02758.x

Bonfim, F. and Carvalho, S. (2009). Fourth national report on the biodiversity (1st draft). Directorate General of the Environment, Ministry of Natural Resources, Energy and Environment, Democratic Republic of Sao Tome and Principe. 93 pp.

Brusca, R.C. (1980). *Common intertidal invertebrates of the Gulf of California* (Vol. 2). University of Arizona Press, Tucson. 513 pp.

Buchanan, J.B. (1958). The Bottom Fauna Communities Across the Continental Shelf Off Accra, Ghana (Gold Coast). *Proceedings of the Zoological Society of London*, *130*(1), 1–56. https://doi.org/10.1111/j.1096-3642.1958.tb00562.x

Chou, S.C., Lyra, A.A., Gomes, J. L., Rodriguez, D.A., Martins, M.A., Resende, N.C, Tavares, P.S., Dereczynski, C.P., Pilotto, I.L., Martins, A. M., Carvalho, L.F.A., Onofre, J.L.L., Major, I., Penhor, M. and Santana, A. (2020). Downscaling projections of climate change in Sao Tome and Principe Islands, Africa. *Climate Dynamics*, *54*(9–10), 4021–4042. https://doi.org/10.1007/s00382-020-05212-7

Collignon, J. (1960). Observations faunistiques et écologiques sur les Mollusques testacés de la baie de Pointe-Noire (Moyen-Congo). *Bulletin de l'I. F. A. N., 22*(2), 411–464.

Connell, J.H. (1961a). The influence of intra-specific competition and other factors on the distribution of the barnacle *Chthamalus stellatus*. *Ecology, 42*, 710–723.

Connell, J.H. (1961b). Effects on competition, predation by *Thais lapillus*, and other factors on natural populations of the barnacle *Balanus balanoides*. *Ecological monographs, 31*(1), 61–104.

Cravo, M., Almeida, A.J., Lima, H., Azevedo e Silva, J., Bandeira, S., Machava, V. and Paula, J. (submitted). Fish nursery role by a small mangrove at Príncipe Island, Gulf of Guinea. Submitted to *Frontiers in Marine Science*.

Dell' Angelo, B., Schwabe, E., Gori, S., Sosso, M. and Bonfitto, A. (2014). Chitons (Mollusca, Polyplacophora) from São Tomé and Príncipe Islands. *African Invertebrates, 55*(2), 171–200.

Edyvean, R.G.J. and Ford, H. (1987). Growth rates of *Lithophyllum incrustans* (Corallinales, rhodophyta) from southwest Wales. *British Phycological Journal, 22*(2), 139–146. https://doi.org/10.1080/00071618700650161

FAO (2016). *The living marine resources of the Eastern Central Atlantic: Volume 4: Bony fishes part 2 (Perciformes to Tetradontiformes)* and *Sea turtles*. FAO Species Identification Guide for Fisheries Purposes, FAO, pp. 2343–3124. Rome, Italy.

Fernandes, F. and Rolan, E. (1993). Moluscos marinos de São Tomé y Principe: actualizacion bibliografica y nuevas aportaciones (Marine molluscs of São Tomé and Principe: bibliographic actualization and new contributions). *Iberus*, *11*(1), 31–47.

Gauld, D.T. and Buchana, J.B. (1959). The principal features of the rock shore fauna in Ghana. *Oikos, 46*, 121–132.

Guilhem, I.F., Masi, B.P. and Creed, J.C. (2020). Impact of invasive *Tubastraea* spp. (cnidaria: Anthozoa) on the growth of the space dominating tropical rocky shore zoantharian *Palythoa caribaeorum* (Duchassaing and Michelotti, 1860). *Aquatic Invasions, 15*(1), 98–113. https://doi.org/10.3391/AI.2020.15.1.07

Haroun, R., Herrero-Barrencua, A. and Abreu, A.D. (2018). Mangrove Habitats in São Tomé and Príncipe (Gulf of Guinea, Africa): Conservation and Management Status. *Coastal Research Library, 25*, 589–605. https://doi.org/10.1007/978-3-319-73016-5_27

Hawkins, S.J. and Jones, H.D. (1992). *Rocky shores. Marine field course guide 1.* Immel Publishing, London. 144 pp.

Hawkins, S.J., Allen, J.R., White, K.N., Conlan, K., Hendry, K., Jones, H.D. and Russell, G. (1992). Restoring and managing disused docks: marine habitat creation and nature conservation in inner city areas, in *Proc. Symp. on Habitat Restoration, National Oceanic and Atmospheric Administration, September 1990* (ed. G. Thayer), Maryland Seagrant Publications.

Herrero-Barrencua, A., Haroun, R. and Abreu, A.D. (2017). *Caracterización preliminar de los manglares de la Isla de Príncipe (São Tomé e Príncipe)*. IU-ECOAQUA, Universidad de Las Palmas de Gran Canaria. 70 pp.

Hiscock, K. (1983). Water movement, in *Sublittoral Ecology. The Ecology of the Shallow Sublittoral Benthos* (eds R Earll and D.G. Erwin), Oxford University Press, Oxford, pp. 58-96.

Horn, M.H., Martin, K.L.M. and Chotkowski, M.A. (1999). Introduction. Pp. 1–6 in *Intertidal Fishes: Life in Two Worlds* (eds. Horn, M.H., Martin, K.L.M. and Chotkowski, M.A.), Academic Press. San Diego, USA.

IH (2019). Tabela de previsões de marés da Baía de S. António (Ilha do Príncipe) para Janeiro, Fevereiro e Março de 2020. Instituto Hidrográfico.

John, D.M. and Lawson, G.W. (1991). Littoral ecosystems of tropical western Africa, in *Intertidal and Littoral Ecosystems,* (eds AC. Mathieson and P.H. Nienhuis), Elsevier, Amsterdam, 297–323.

Krakstad, J.O., Alvheim, O., Lopes, J. and Iwamoto, T. (2010). Surveys of the Fish Resources of São Tomé and Príncipe: Survey of the demersal resources (Issue May). 66 pp.

Lawson, G.W. (1955). Rocky shore zonation in the British Cameroons. *Journal of the West African Science Association, 1,* 78–88.

Lawson, G.W. (1956). Rocky Shore Zonation on the Gold Coast. *The Journal of Ecology, 44*(1), 153–170. https://doi.org/10.2307/2257159

Lawson, G.W. (1957). Seasonal Variation of Intertidal Zonation on the Coast of Ghana in Relation to Tidal Factors. *British Ecological Society, 45*(3), 831–860. http://www.jstor.org/stable/2256960

Lawson, G.W. (1966). The littoral ecology of West Africa. *Oceanography and Marine Biology, 4,* 405–448.

Lawson, G.W. and John, D.M. (1982). The marine algae and coastal environment of tropical West Africa. *Beihefte zur Nova Hedwigia, 70,* 1–455.

Lewis, J.R. (1964). The Ecology of Rocky Shores. English Universities Press, London. 323 pp.

Louthan, A.M., Doak, D.F. and Angert, A.L. (2015). Where and When do Species Interactions Set Range Limits?. *Trends in Ecology and Evolution, 30*(12), 780-792. http://dx.doi.org/10.1016/j.tree.2015.09.011

Manning, R.B., and Holthuis, L.B. (1981). West African Brachyuran crabs (Crustacea: Decapoda). *Smithsonian Contributions to Zoology, 306,* 1–379. https://doi.org/10.5479/si.00810282.306

McQuaid, C. and Branch, G. (1985). Trophic structure of rocky intertidal communities response to wave action and implications for energy flow. *Marine Ecology Progress Series, 22,* 153–161. https://doi.org/10.3354/meps022153

Measey, G.J., Vences, M., Drewes, R.C., Chiari, Y., Melo, M. and Bourles, B. (2007). Freshwater paths across the ocean: molecular phylogeny of the frog *Ptychadena newtoni* gives insights into amphibian colonization of oceanic islands. *Journal of Biogeography, 34*, 7–20.

Miller, L.P. and Denny, M.W. (2011). Importance of behavior and morphological traits for controlling body temperature in littorinid snails. *Biological Bulletin, 220*(3), 209–223. https://doi.org/10.1086/BBLv220n3p209

NBSAP II (2015). National Biodiversity Strategy and Action Plan 2015–2020, National Commission for Monitoring and Evaluation, Ministry of Infrastructure. Natural Resources and Environment, São Tomé and Príncipe, pp. 120.

Oyenekan, J.A., and Olufowobi, M.O. (1987). Some Aspects of the Ecology of the Mollusc *Nerita senegalensis* in Lagos Harbour, Nigeria. *Journal of Tropical Ecology, 3*(2), 183–187.

Pisoni, T., Lima, R.F. de, Brito, A.C., Chainho, P., Félix, P.M., Caçador, I. and Carvalho, A. (2015). *Planos de gestão participativa para dois sítios de mangal na Ilha de S. Tomé: Praia das Conchas e Malanza.* Abordagem ecossistémica integrada para a conservação e gestão da biodiversidade na zona tampão dos parques naturais obô de São Tomé e Príncipe, 79 pp.

Raffaelli, D. and Hawkins, S. (1999). *Intertidal Ecology*. 2nd edition, 356 p. Kluwer Academic Publishers. https://doi.org/10.2307/1352485

Russell, G., Hawkins, S.J., Evans, L.C., Jones, H.D. and Holmes, G.D. (1983). Restoration of a disused dock basin as a habitat for marine benthos and fish. *Journal of Applied Ecology, 20*, 43–58.

Reid, D.G. (2011). The genus *Echinolittorina* Habe, 1956 (Gastropoda: Littorinidae) in the Eastern Atlantic Ocean and Mediterranean Sea. *Zootaxa, 2974*(1). 65 pp. https://doi.org/10.11646/zootaxa.2974.1.1

Southward, A.J. (1959) The zonation of plants and animals on rocky seashores. *Biological Reviews, 33*(2), 137–177.

Steentoft, B.Y.M. (1965). A revision of the marine algae of São Tomé and Príncipe (Gulf of Guinea). *The Journal of the Linnean Society (Botany), 60*(382), 99–146. https://doi.org/10.1017/cbo9780511779954.007

Stephenson, T.A. and Stephenson, A. (1949). The universal features of zonation between tide marks on rocky coasts. *Journal of Ecology, 37*, 289–305.

Stephenson, T.A. and Stephenson, A. (1972). *Life between tidemarks on rocky shores*. W. H. Freeman, San Francisco, 425 pp.

Tomanek, L. and Helmuth, B. (2002). Physiological Ecology of Rocky Intertidal Organisms: A Synergy of Concepts. *Integrative and Comparative Biology, 42*(4), 771-775.

Vaz, H and Oliveira, F. (2007). Relatótio Nacional do Estado Geral da Biodiversidade de S. Tomé e Príncipe. Direção Geral do Ambiente, Ministério de Recursos Naturais e Meio Ambiente. 115 pp.

Vermeij, G.J. (1972). Intraspecific Shore-Level Size Gradients in Intertidal Molluscs. *Ecology, 53*, 693–700.

Vermeij, G.J. (1973). Morphological patterns in high-intertidal gastropods: Adaptive strategies and their limitations. *Marine Biology, 20*(4), 319–346. https://doi.org/10.1007/BF00354275

Vinagre, C., Mendonça, V., Cereja, R., Abreu-Afonso, F., Dias, M., Mizrahi, D., and Flores, A.A.V. (2018). Ecological traps in shallow coastal waters-Potential effect of heat-waves in tropical and temperate organisms. *PLoS one, 13*(2), 1–17. https://doi.org/10.1371/journal.pone.0192700

Wallentinus, I. (1991) The Baltic Sea Gradient, in *Intertidal and Littoral Ecosystems* (eds Mathieson, A.C. and Nienhuis, P.H.), Elsevier, Amsterdam, pp. 83–108.

Williams, G.A. (1993). Seasonal variation in algal species richness and abundance in the presence of molluscan herbivores on a tropical rocky shore. *Journal of Experimental Marine Biology and Ecology, 167*(2), 261–275. https://doi.org/10.1016/0022-0981(93)90034-L

Wirtz, P., Ferreira, C.E.L., Floeter, S.R., Fricke, R., Gasparini, J.L., Iwamoto, T., Rocha, L., and Sampaio, C.L.S. (2007). (Eastern Atlantic Ocean) – an update. *Zootaxa*, *48*, 1–48.

CHAPTER 3

Patterns of fish distribution in rock pools from tropical intertidal rocky shores at Príncipe Island (Gulf of Guinea)

Patterns of fish distribution in rock pools from tropical intertidal rocky shores at Príncipe Island (Gulf of Guinea)

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Abstract

Little is known about the biological communities of intertidal rocky shores and rock pools of the west coast of Africa, especially in the Gulf of Guinea. Rock pools are characteristic habitats of the intertidal zone of structurally complex rocky shores, adding important niche space to coastal fish species, an essential resource to the artisanal subsistence fisheries. Global climate change, such as ocean warming, poses increasing pressure on the environmental conditions of the rocky shores and their biological communities, particularly in tropical regions. Rock pools constitute good models for monitoring these impacts. In this study, rock pools of the Príncipe Island were sampled for describing the composition, abundance and distribution of fish assemblages and their relation to pool structure (volume, depth), water mass (temperature, salinity and pH) and biological (algal and coral cover, and biological species richness) parameters. A total of 18 species and one leptocephalus larva of unknown species, representing 13 families, were observed during sampling in the three study locations. In decreasing order, the five most abundant species were *Bathygobius burtoni*, *Abudefduf taurus*, *Entomacrodus cadenati*, *Microlipophrys velifer* and *Prionurus biafraensis*, representing together 81% of the total number of fish recorded during this study, of which the four most abundant species also proved to be the better adapted to the range of conditions found in rock pools. A new species has been recorded for the region, *Lutjanus griseus*, as well for the entire African coast south of the Canary Islands. Overall, larger rock pools with minimal biological cover and higher salinity were found to support higher fish abundance and species richness. Most species individually preferred rock pools with larger volumes, but some presented a degree of habitat specificity, such as the absence or presence of biological cover. The more active swimmer species preferred deeper pools.

Keywords: Fish assemblages, species richness, pool characteristics, S. Tomé and Príncipe, West Africa.

Introduction

Due to their accessibility, intertidal zones have been one of the most intensively studied marine environments throughout the globe, and many of these studies have included ichthyofauna. However tropical rock pools have received little attention (Gibson, 1982; Mahon and Mahon, 1994; Martins *et al.*, 2007), particularly on the west coast of Africa and adjacent islands (Zander *et al.*, 1999). In this region, some families like Clinidae, Tripterygiidae and Bleniidae were the most commonly reported in intertidal habitats in past studies, but recent work, particularly focusing on whole assemblages, is non-existent and very little is known about intertidal fishes of other families (John and Lawson, 1991; Wirtz, 1980; Zander *et al.*, 1999).

Although the marine ichthyofauna of the Gulf of Guinea is one of the least studied in the world, marine fish in São Tomé and Príncipe have recently attracted scientific interest (Tuya *et al.*, 2017). Previous studies on marine fishes of São Tomé and Príncipe have focused mainly on the open and coastal sea, and recently on mangrove ecosystems (Cravo *et al.*; Herrero-Barrencua *et al.*, 2017). Afonso *et al.* (1999) and Wirtz *et al.* (2007) documented limited sightings of fish in intertidal rocky pools, but with no detailed information. Of these, it is known that the blenniids *Entomacrodus cadenati*, *Microlipophrys velifer*, *Ophioblennius atlanticus* and *Scartella cristata* are resident species of the intertidal zone and widely distributed throughout the west coast of Africa (Zander *et al.* 1999).

Rock pools are characteristic habitats found on the intertidal zone of rocky shores, adding important niche space to coastal habitats (Firth *et al.*, 2014). During low tide, water is retained in holes and depressions that serve as refuge for many invertebrates and fishes (Martins *et al.*, 2007; Zander *et al.*, 1999), especially in tropical rocky shores, where extreme sun exposure can lead to high temperatures and desiccation on the open rock surface (Raffaelli and Hawkins, 1999). This makes rock pools as microhabitats that attract high diversity and abundance of various organisms (White *et al.*, 2015). Nonetheless, rock pool organisms are subject to stressful environmental conditions during the low water period. When isolated from the sea, rock pools become prone to large and rapid fluctuations in the water physicochemical properties such as temperature, salinity, oxygen, carbon dioxide and pH (Huggett and Griffiths, 1986; Horn *et al.*, 1999; Martins *et al.*, 2007). Additionally, rock pools exist in numerous shapes and volumes, are randomly distributed along the rocky shore vertical and horizontal gradients and are highly variable in time (i.e. tidal, diurnal and seasonal-induced fluctuations) and space (from geographic to local scales) (Martins *et al.*, 2007; White *et al.*, 2015). They also become more exposed to marine and terrestrial predators, such as fishes and birds (Gibson, 1986; Vinagre *et al.*, 2018; White *et al.*, 2015). Consequently, several fish species have developed behavioural, physiologic and morphologic adaptations to better endure the drastic environmental fluctuations of these habitats (Zander *et al.*, 1999).

Many coastal fishes use rock pools at some point in their life cycle to forage, take refuge from predators or as nursery areas (Horn *et al.*, 1999), and thus rock pools represent a key habitat for supporting coastal fish species diversity (White *et al.*, 2015). The fishes can be grouped into three categories based on how they use rock pools and the proportion of their life cycle spent within them (Griffiths, 2003; Mahon and Mahon, 1994; White *et al.*, 2015), namely primary residents, secondary residents and transients. Residents spend almost their entire juvenile and adult lives in the intertidal. They are generally small, benthic fishes that are physiologically, morphologically and behaviourally adapted for intertidal life (White and Brown, 2013). Secondary residents use the intertidal primarily as a nursery area, after which they migrate into the subtidal and off-shore habitats as they mature (Norton and Cook, 1999; White and Brown, 2013). A third group, transients, usually comprise a small fraction of the species in rock pools

(Mahon and Mahon, 1994), occupying them on only certain occasions to forage or by accident, when trapped at low tide (Griffiths, 2003; Horn *et al.*, 1999; White *et al.*, 2015).

Throughout the tide cycles, intertidal fishes need to find and occupy a favourable rock pool during emersion time to increase the chances of survival. Thus, many behavioural patterns of fishes found in rock pools can be interpreted as adaptations for protecting either the individual or its offspring from unfavourable conditions during the low tide (Gibson, 1986). Consequently, the different gradients of biotic and abiotic factors can produce consistent patterns in the structure, distribution and abundance of fish assemblages (Gibson, 1982; Norton and Cook, 1999; Zander *et al.*, 1999). Thus, an important part of the study of intertidal fishes is to find the relationships between their distribution, physiology and behaviour and the environmental variables of the rock pools (Gibson, 1999a).

Apart from the intrinsic scientific interest of intertidal fishes and their physiological behaviour, the intertidal and adjacent subtidal zones are also nursery areas for numerous fish species that eventually recruit to coastal fisheries (Gibson, 1999b). The degradation and drastic modification of such areas have, therefore, consequences for both the economy and quality of life of many coastal and island countries (Gibson, 1999b), such as São Tomé and Príncipe, where small-scale coastal fisheries are crucial to the domestic trade and coastal food resources (Abreu, 2013; Horemans *et al.*, 1994; Tous, 2015). Global climate change, in particular ocean warming, further pressures the environmental conditions on the rocky shores and their biological communities (Horn *et al.*, 1999). The projections of climate change for São Tomé and Príncipe predict an increase in temperature, that could exceed 3°C in the middle of the 21st century, as well as in heavy precipitation rates (Chou *et al.*, 2020). Impacts of increased temperature on biological communities in tropical waters are expected to be significant because the species are more stenothermic and an increase in temperature can drive some of them above their lethal limit (Raffaelli and Hawkins, 1999), and rock pools constitute good models for assessing these impacts (Vinagre *et al.*, 2018).

In this study, we investigated rock pool fish assemblages at Príncipe Island, in the eastern tropical Atlantic, with the objective of describing the species composition and abundance in relation to abiotic and biotic factors. We aimed to (i) determine the importance of rock pool dimension (depth, surface area and volume), within-pool water conditions (temperature, salinity and pH), and algal, coral and total biological community cover, in controlling fish assemblage structure; (ii) assess the variables which contributed more for each species abundance; and (iii) describe species distribution depending on the water properties.

Materials and methods

Study sites and rock pool sampling

Sampling took place in three locations, Bom-Bom islet, Ponta Marmita and Praia Uba, located on the north coast of Príncipe Island, Democratic Republic of São Tomé and Príncipe (Gulf of Guinea) (Fig. 3.1). These locations were chosen for their extensive and exposed rocky shores, numerous rock pools and their relative ease of access.

A total of 20 permanent rock pools were sampled at each of the three locations, between January and February 2020. Only rock pools from the eulittoral zone (mid-zone) were sampled to allow comparison between the different rocky shores. The lower limit of the eulittoral zone is delimited by a noticeably and abrupt absence of the coralline alga *Lithophyllum incrustans* and the zoanthid *Palythoa caribaeorum* in the freely draining open rock surface, followed by its replacement by the black lichen *Verrucaria* sp., and the oyster *Saccostrea cucullata* which defined the upper limit of the eulittoral.

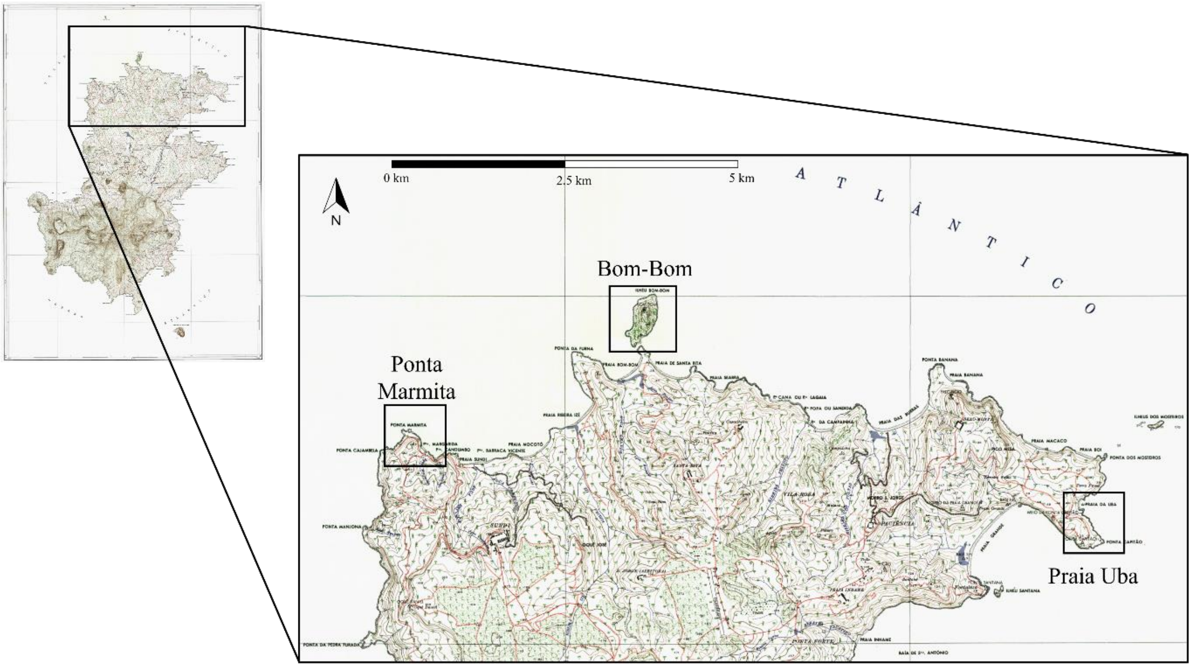

Figure 3.1. Location of the rocky shore sampling sites in Príncipe Island: Bom-Bom Islet, Ponta Marmita, and Praia Uba.

Sampling was performed only close to peak low tide so that splashes from waves would not influence the water conditions of the rock pools. Freshwater drainage from land or pools connected to other adjacent pools were avoided to keep conditions uniform. Therefore, only saltwater rock pools isolated during emersion were sampled. Salinity, temperature and pH were registered in each pool with a YSI Multiparameter 350i probe.

To determine the area of the rock pools, a scaled photo was taken directly above each pool, as parallel and horizontally as possible. Rock pools were only considered if they fitted in full in the photo frame of the camera (GoPro Hero 7 Black in "Linear mode"). The area was then calculated using the software ImageJ^{TM}. To calculate the water volume, average depth was calculated from ten random depth measures. Maximum depth was also recorded.

Fish and biological community sampling

Due to the lack of obstructions such as large algae, fish were clearly visible and direct observations were deemed adequate to census fish populations (Gibson, 1999a). Sampling consisted of firstly identifying the visible fish species in the rock pool and their respective abundance with minimal disturbance, so fishes would not hide on crevices. Besides ichthyofauna, the presence of the different algae and other animal species were also documented. The percentage cover of algae and coral was recorded and estimated to the nearest 10% by eye for the whole rock pool. If necessary, occasional boulders were moved to account for possible hidden fish and other organisms. To ensure the correct species identification, a final inspection was made with a waterproof camera filming the bottom, ledges and holes of the rock pool to allow a better point of view for later examination.

Observations in the field were carried out by trained observers. Species identifications were confirmed both *in situ* and on recorded videos using existing bibliography for the region (Afonso *et al.*, 1999; FAO, 2016a, 2016b; Vasco-Rodrigues *et al.*, 2018; Wirtz *et al.*, 2007). Taxonomy was updated according to Nelson (2006).

Data treatment

Firstly, draftsman plots using Pearson correlations were made between the environmental variables to determine high correlated pairs ($p > 0.9$) and eliminate any possible redundant variables. The remaining ones were normalised, and a resemblance matrix was constructed using Euclidean distance. Rock pool variables were then compared among the three study sites using principal coordinates analysis (PCO) and permutational multivariate analysis of variance (PERMANOVA) with pairwise tests.

Fish assemblage data were logarithmised, and a resemblance matrix was constructed using the "zero-adjusted" Bray–Curtis similarity coefficient (Clarke *et al.*, 2006). Rock pool fish assemblages were also compared among the three study sites using PCO and PERMANOVA with pairwise tests. In this PCO, the vectors of the environmental variables correlated >0.3 and the main fish species correlated >0.4 were overlapped for comparison. Analysis of variance (ANOVA) was used to compare the mean species richness and total abundance among sampling locations. Because the location effect was not found to be a main driver on fish assemblage, data from all three sites were pooled for further analysis.

Based on the resemblance matrix of fish samples, a distance-based linear model (DISTLM) was used in combination with a distance-based redundancy analysis (dbRDA) for visualizing results (Anderson *et al.*, 2008), to identify which of the environmental variables accounted for the best significant part of the variation in fish assemblage composition, while also assessing the relationship of all variables individually with marginal tests. The selection criterion and procedure were, respectively, Akaike information criterion for small sample sizes (AICc) and BEST, which selects the best possible model.

The effect of rock pool environmental variables (excluding pH and temperature) on the abundance of individual species was analysed using multiple linear regressions with stepwise variable selection. Regressions were only conducted for species that occurred in more than three pools and were represented by more than ten individuals.

All analyses were performed using the software PRIMER-E^{TM} v. 6.1.11, PERMANOVA+ v. 1.0.1 and IBM SPSS^{TM} Statistics v. 26.0.

Results

Fish assemblages

A total of 746 fish from 18 species and one unknown leptocephalus species, representing 13 families, were observed during sampling in the three study locations (Table 3.1). In decreasing order, the five most abundant species were *Bathygobius burtoni*, *Abudefduf taurus*, *E. cadenati*, *M. velifer* and *Prionurus biafraensis*, which together represented 81% of the total number of fish recorded during this study.

Sampled rock pool communities were composed predominantly of resident fishes, making up to 64% of total individuals, but only 16% of total species. Secondary residents, in contrast, were 34% of total individuals but 58% of total species. Finally, transient fishes represented only 2% of total abundance but still 26% of total species.

Ten more species were sighted in field observations outside the sampling period in the eulittoral zone of the same rocky shores and other similar ones throughout the island, adding five more families (Table 3.2). These were *Muraena melanotis*, *Sardinella* cf. *aurita*, *Mugil curema*, *Epinephelus adscensionis*, *Lutjanus griseus*, *Spicara nigricauda, Cirrhitus atlanticus*, *Hypleurochilus* sp., *O. atlanticus* and *S. cristata*.

Lutjanus griseus is a newly confirmed species for the archipelago. The specimen (Fig. 3.2) was found in a rock pool in Bom-Bom with considerable volume (>5000 L), and with a maximum depth of >50 cm. Underwater footage allowed us to confirm the identity of the species by its colour pattern, according to Allen (1985) and Anderson (2002). It had a pinkish-grey colouration with the centre of the scales on the back and sides darker than the edges, giving the appearance of parallel rows along the body. It also showed two characteristics of young specimens, such as an oblique dark stripe running from the snout through the eye and towards the anterior base of the dorsal fin, and a whitish blue line on the cheek below the eye. Being a small specimen (<15 cm), it suggests that the species does have a considerable part of its life cycle in the East Atlantic Ocean. Previous records on the archipelago were inconclusive as to the identification and confirmation of the species (Afonso *et al.*, 1999; Fowler, 1936), with the only confirmed occurrence in the East Atlantic Ocean being in the Canary Islands (Falcón *et al.*, 2018).

Figure 3.2. Young specimen of *Lutjanus griseus* found in a rock pool along the eulittoral zone of Bom-Bom rocky shore.

Table 3.1. Fish species documented in 60 rock pools from three locations in Príncipe Island, Bom-Bom islet (BB), Ponta Marmita (PM) and Praia Uba (PU). R – resident, SR – secondary resident, T – transient.

Family	**Species**	**Local common name**	**Residency status**	**Number observed**	**Location**	**Rock pool**
Anguilliformes n.d.	Leptocephalus larva	-	T	1	PM	32
Muraenidae	*Echidna peli* (Kaup, 1856)	Moreia-salpicada	SR	1	PM	32
Atherinidae	*Atherina lopeziana* (Rossignol and Blache, 1961)	-	T	12	BB, PU	6, 60
Serranidae	*Rypticus subbifrenatus* (Bloch and Schneider, 1801)	Peixe-sabão	SR	2	BB, PM	16, 32
Epinephelidae	*Cephalopholis nigri* (Günther, 1859)	Cota-uê	T	1	PM	30
Apogonidae	*Apogon imberbis* (Linnaeus, 1758)	Cardinal	T	1	BB	10
Carangidae	*Caranx hippos* (Linnaeus, 1766)	Corcovado	SR	3	BB	17, 19
Pomacentridae	*Abudefduf saxatilis* (Linnaeus, 1758)	Txim-txim	SR	29	BB, PM	15, 32
Pomacentridae	*Abudefduf taurus* (Müller and Troschel, 1848)	Txim-txim -preto	SR	129	BB, PM, PU	5, 6, 8, 9, 11, 15, 17, 18, 19, 23, 28, 31, 32, 33, 34, 36, 39, 44, 45, 46, 48, 49, 50, 58, 59
Pomacentridae	*Microspathodon frontatus* Emery, 1970	Txim-txim	SR	27	BB, PM, PU	5, 6, 15, 19, 28, 29, 60
Pomacentridae	*Stegastes imbricatus* Jenyns, 1840	Txim-txim	SR	39	BB, PM, PU	5, 6, 9, 15, 17, 18, 19, 27, 28, 29, 31, 34, 50, 60
Labridae	*Thalassoma newtoni* (Osório, 1891)	Dodongo	SR	1	PM	29
Blenniidae	*Entomacrodus cadenati* Springer, 1967	-	R	97	BB, PM, PU	1, 2, 3, 4, 5, 6, 8, 9, 11, 12, 15, 18, 23, 28, 29, 31, 35, 37, 41, 42, 43, 45, 46, 50, 51, 52, 53, 54, 56, 57, 58, 59, 60
Blenniidae	*Microlipophrys velifer* (Norman, 1935)	-	R	86	BB, PM, PU	4, 10, 16, 17, 19, 20, 21, 23, 24, 28, 29, 31, 32, 36, 38, 39, 40, 41, 42, 43, 45, 49, 51, 52, 53, 55, 57, 58, 59
Labrisomidae	*Labrisomus nuchipinnis* (Quoy and Gaimard, 1824)	-	SR	20	BB, PM, PU	6, 9, 11, 12, 17, 19, 35, 37, 49, 56, 60
Gobiidae	*Bathygobius burtoni* (O'Shaghnessy, 1875)	Xarroco/Batu	R	245	BB, PM, PU	5, 6, 7, 8, 9, 10, 11, 15, 16, 17, 18, 19, 20, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 42, 44, 45, 46, 47, 48, 50, 51, 57, 59, 60
Acanthuridae	*Acanthurus monroviae* Steindachner, 1876	Lâmina	SR	3	BB, PM	5, 15, 32
Acanthuridae	*Prionurus biafraensis* (Blache and Rossignol, 1962)	Cabo-verde	SR	48	BB, PM, PU	15, 17, 19, 28, 29, 30, 32, 36, 45, 49, 56, 60
Monacanthidae	*Cantherhines pullus* (Ranzani, 1842)	Peixe-asno -magro	T	1	BB	5

Table 3.2. Additional fish species observed outside the sampling period in rock pools of the eulittoral zone in several rocky shores around the island.

Family	Species	Local common name	Residency status
Muraenidae	*Muraena melanotis* (Kaup, 1859)	Moreia-mapinta	SR
Clupeidae	*Sardinella* cf. *aurita* (Valenciennes, 1847)	Sardinha/Longo	T
Mugilidae	*Mugil curema* (Valenciennes, 1836)	Tainha	SR
Epinephelidae	*Epinephelus adscensionis* (Osbeck, 1765)	Garoupa	SR
Lutjanidae	*Lutjanus griseus* (Duméril, 1861)	Corvina	SR
Sparidae	*Spicara nigricauda* (Norman, 1931)	-	T
Cirrhitidae	*Cirrhitus atlanticus* Osório, 1893	Cazebudo	SR
Blenniidae	*Hypleurochilus aequipinnis* (Günther, 1861)	-	R
Blenniidae	*Ophioblennius atlanticus* (Valenciennes, 1836)	-	SR
Blenniidae	*Scartella cristata* (Linnaeus, 1758)	-	R

Sampling sites comparison

Sampled fish assemblages varied between the three locations (Fig. 3.3). Some species appeared in all three locations, such as *A. taurus*, *Microspathodon frontatus*, *Stegastes imbricatus*, *E. cadenati*, *M. velifer*, *L. nuchipinnis*, *B. burtoni* and *P. biafraensis* (Fig. 3.3). Others were found in only two locations, like *Atherina lopeziana* that was found in Bom-Bom and Praia Uba, and *Rypticus subbifrenatus*, *Abudefduf saxatilis* and *Acanthurus monroviae* that were found in Bom-Bom and Ponta Marmita (Fig. 3.4). Lastly, some were found on only one rocky shore. *Apogon imberbis*, *Caranx hippos* and *Cantherhines pullus* were found in Bom-Bom, and the leptocephalus (Anguilliformes n.d.), *Echidna peli*, *Cephalopholis nigri* and *Thalassoma newtoni* in Ponta Marmita (Fig. 3.4). As a result, Bom-Bom presented the highest average species richness and abundance per rock pool (Fig. 3.3), although some of the species recorded in only one or two sites were also found in the other locations in field observations outside the sampling period.

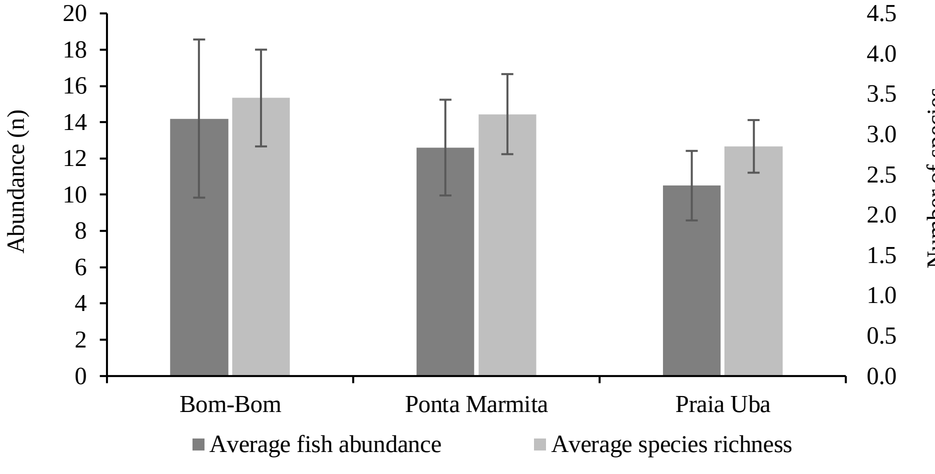

Figure 3.3. Average fish abundance (±SE) and average species richness (±SE) per rock pool in each location, Bom-Bom (3.45±0.6 and 14.2±4.36), Ponta Marmita (3.25±0.5 and 12.6±2.64) and Praia Uba (2.85±0.33 and 10.5±1.92).

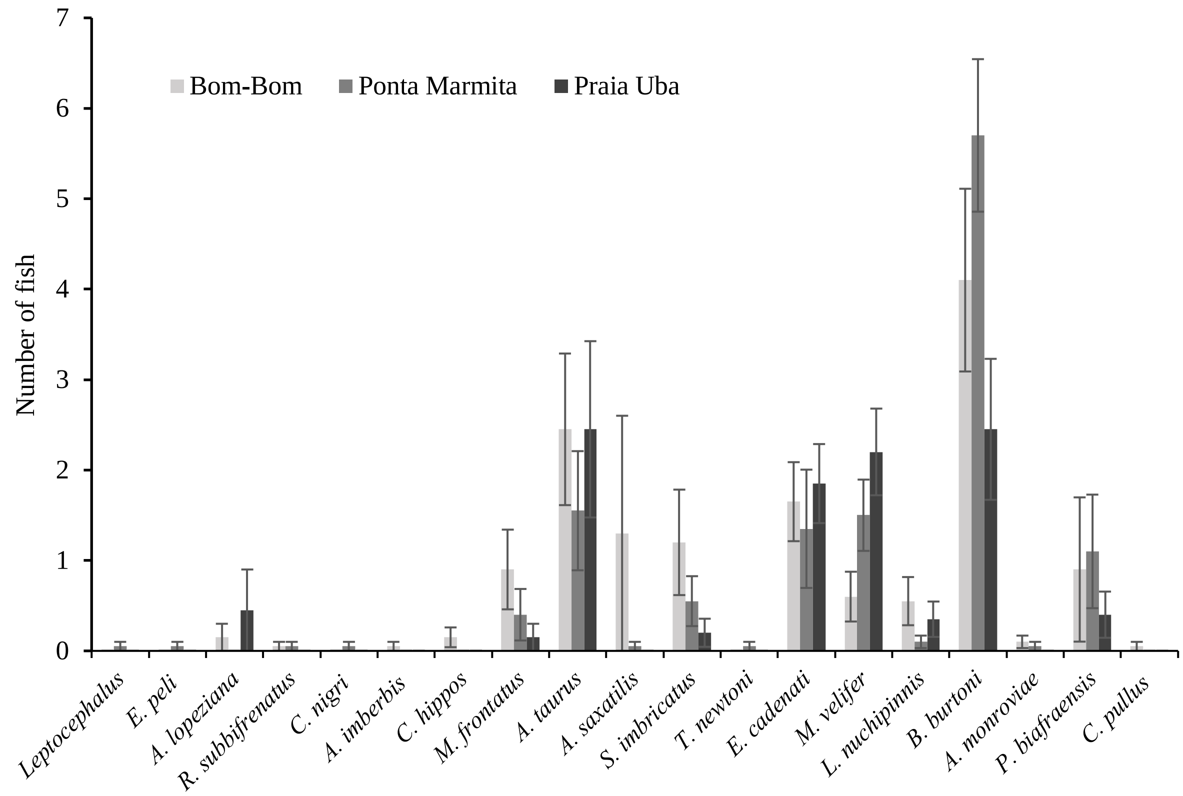

Figure 3.4. Average (±SE) number of fish of each species observed in rock pools of the eulittoral zone of the three locations, Bom-Bom, Ponta Marmita and Praia Uba.

According to the permutational multivariate analysis of variance (PERMANOVA), there was an overall significant difference in the fish assemblage between locations (p = 0.0059, Pseudo-F = 2.918), except between Bom-Bom and Praia Uba (p = 0.1914). The PCO of fish data (Fig. 3.5a) shows a slight clumping of pools from the same location, particularly those of Ponta Marmita. However, being the three rocky shores very similar in geomorphology, exposure and orientation to the sea, the location effect was not considered as the main cause of the differences in fish assemblages, but rather the influence of environmental variables (PERMANOVA, p = 0.001, Pseudo-F = 5.5045), as suggested by the PCO plot of environmental data (Fig. 3.5b). PERMANOVA pairwise tests also revealed differences between all locations. Furthermore, there were no significant differences among the three study locations in species richness (F = 0.392, p = 0.677) and total abundance (F = 0.348, p = 0.707), or in any pair of study sites. Based on these results, data from all three locations were pooled for further analysis.

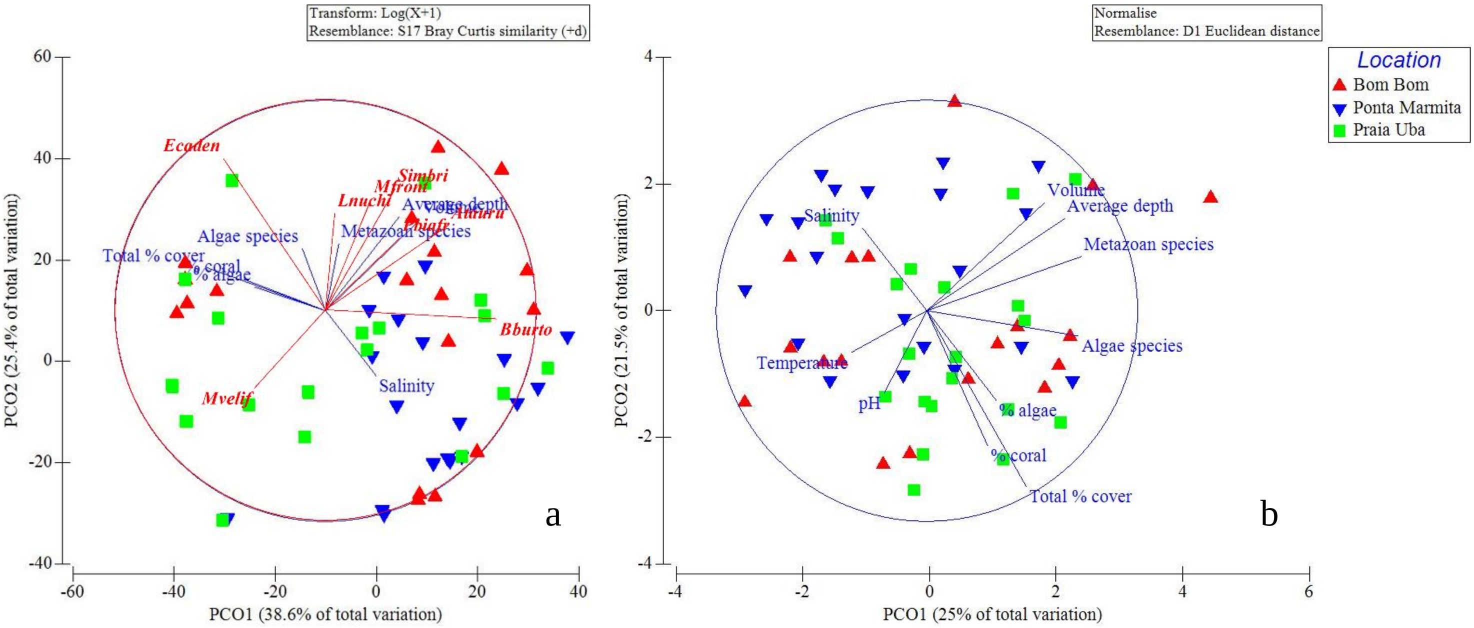

Figure 3.5. Principal coordinates ordination (PCO) plots of fish assemblages to the left (a) and rock pools environmental variables (b) at each location: (▲) Bom-Bom islet, (▼) Ponta Marmita and (■) Praia Uba. (a) environmental vectors inside the circle represent their effect on fish assemblages, and the fish species vectors show in which pools these species tend to be found. (b) vectors represent the effect on the characteristics of the rock pools. The longer the line, the more influential is the variable. The fish species name codes correspond to the following: *Ataurus - Abudefduf taurus*; *Mfront - Microspathodon frontatus*; *Simbri – Stegastes imbricatus*; *Ecaden - Entomacrodus cadenati*; *Mvelif - Microlipophrys velifer*; *Lnuchi - Labrisomus nuchipinnis*; *Bburto - Bathygobius burtoni*; *Pbiafr - Prionurus biafraensis*.

Relationship between rock pool characteristics and fish assemblages

Sixty permanent rock pools were sampled, with a range of areas (from 0.08 to ~8 m²), volumes (from 9.8 to ~3180 L), and depths from 10 to 75 cm deep. The physicochemical properties of the water in the rock pools (temperature, pH and salinity) fluctuated more in smaller volumes and oscillated somewhat less as volume increased (Fig. 3.6).

Algal cover varied from 0% to 90% and was mainly composed of incrusting coralline pink algae, with some presence of other small red and brown algae. Coral cover varied from 0% to 90% and was predominantly comprised of the zoanthid *P. caribaeorum*. Other sessile organisms were also abundant, like sponges, anemones, tube-worms (Serpullidae n.d.), oysters (*Striostrea denticulate*) and mussels (*Mytilus* sp.), but rarely exceeding a 10% cover. In terms of mobile organisms, the most frequent and abundant were the sea-urchin *Echinometra lucunter*, a hermit-crab (Diogenidae n.d.), and the gastropod *Nerita senegalensis*. Crabs from the family Grapsidae and other small crabs, sea-slugs of the genus *Dolabrifera* and a limpet (*Fissurela* sp.) were also common, but not as abundant.

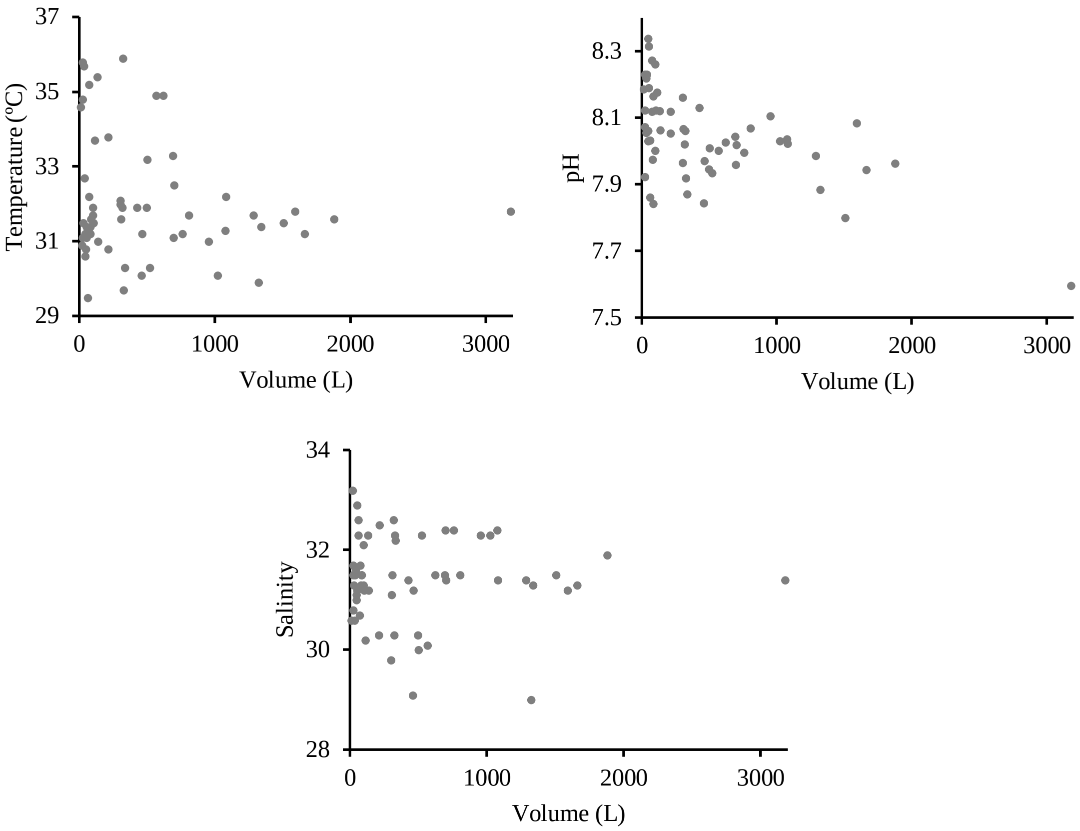

Figure 3.6. Water parameters (temperature, pH, and salinity) in function of the volume (L) of each rock pool from the three sampled rocky shores.

According to the DISTLM, the variation of fish assemblages is better explained by the combination of three environmental variables: volume, total biological community percentage cover and salinity, in order of most influential (Table 3.3). Of these significant variables, volume and salinity had a more positive influence on fish assemblage, whereas the total percentage cover had a more negative one. Of the remaining variables, the average depth, pH, coral percentage cover and algae percentage cover had also a significant impact on fish assemblage (Table 3.3). The presence of other metazoans is marginally significant.

The distance-based redundancy analysis (dbRDA) using only the 3 variables mentioned above (Fig. 3.7) accounted for 32.8% of the total variation in the data, of which 89.1% could be explained by the first two axes. Rock pools with more fish tend to be further positioned to the left in the ordination plot and those with less fish placed more to the right. There were a few pools with large numbers of fish that were somewhat smaller in volume and had a high total percentage cover (rock pools number 6, 28, 29). These pools grouped towards the bottom right corner of the ordination plot (Fig. 3.7). Pools with smaller volumes are closely grouped along a strong gradient which is parallel to the vectors of total % cover and salinity (Fig. 3.7).

The values for which were observed the highest species richness and abundance (Fig. 3.8) were respectively at the maximum depths of 35 cm and 65 cm; for salinity at 32.4 and 31.9; for temperature in 31.2ºC and 31.6ºC; and for pH at 7.996 and 7.964.

Table 3.3. Rock pool environmental variables influence on the variation of fish assemblages on the DISTLM analysis. The best predictive model (with the lowest AICc) achieved when testing all possible combinations (BEST routine) is composed of three variables: volume, total % cover and salinity, and explains 32.8% of the variance.

Marginal tests

Variable	SS (trace)	Pseudo-F	*p*	Prop.
(1) Average depth	8378.6	6.5381	0.002	0.10455
(2) Volume	12565	10.412	0.001	0.15678
(3) pH	2995.6	2.1745	0.042	0.037379
(4) Salinity	5077	3.7875	0.007	0.063349
(5) Temperature	928.98	0.65674	0.615	0.011592
(6) % coral	5147.3	3.8435	0.007	0.064226
(7) % algae	4275.2	3.1557	0.022	0.053345
(8) Total % cover	11426	9.3117	0.001	0.14257
(9) Algae species	2847.1	2.0627	0.091	0.035525
(10) Metazoan species	3282.2	2.3914	0.057	0.040954

Overall best solution

AICc	R^2	RSS	No. Variables	Selections
405.09	0.32814	53845	3	2;4;8

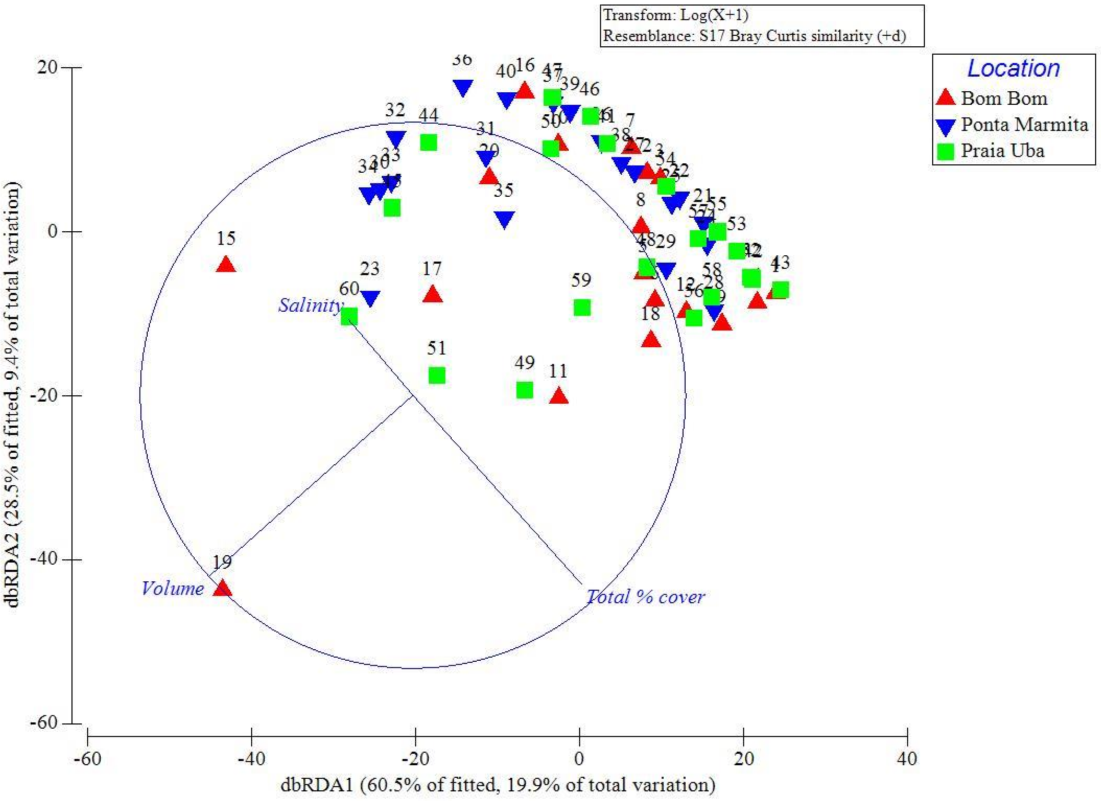

Figure 3.7. Distance-based redundancy analysis (dbRDA) ordination plot of the best DISTLM model, displaying the relationship between rock pool variables that best explain the variation of fish assemblages among pools at different sites: (▲) Bom-Bom islet, (▼) Ponta Marmita and (■) Praia Uba. Numbers refer to the identification of each rock pool. Vectors represent the correlation of variables with both axes.

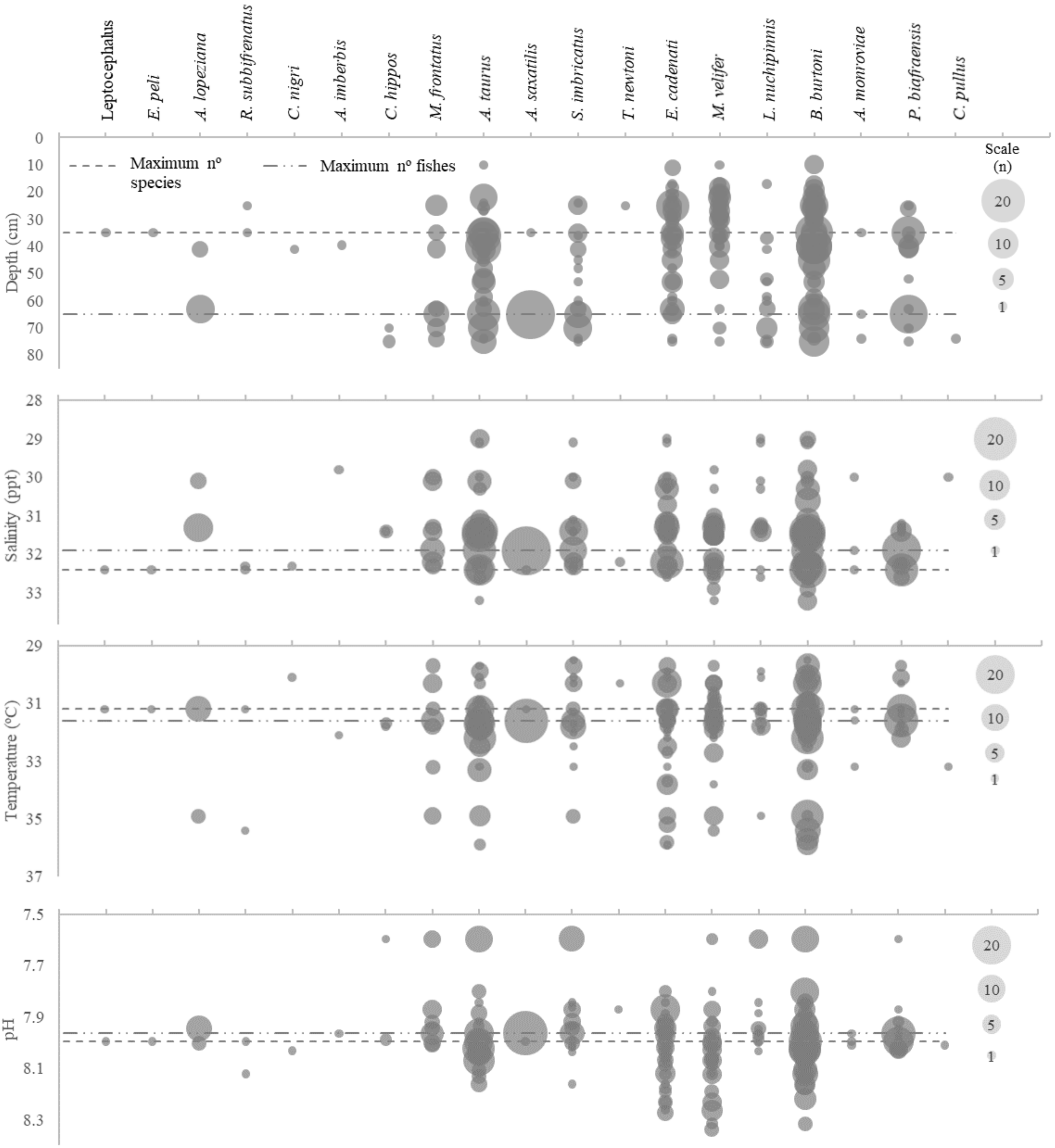

Figure 3.8. Distribution of fish species abundance and richness along the observed variation of depth, salinity, temperature and pH. The lines across the plots represent the value for which it was found the maximum number of fish and species.

The observed patterns of distribution of the richness and abundance of fish species are suggested to be linked to their physiological tolerances. Given their distribution along the different gradients, the species that showed the greatest adaptability to the available vertical space and to the fluctuations in physical-chemical properties were the residents *B. burtoni*, *E. cadenati*, *M. velifer* and the secondary resident *A. taurus*, the four more abundant species.

The step-wise regression analysis was restricted to eight species for which there were sufficient data (Table 3.4). The presence of *A. taurus*, *M. frontatus* and *S. imbricatus* was higher in rock pools with larger volumes. *Entomacrodus cadenati* was more significantly correlated with pools with higher algae cover and the presence of other organisms. *Microlipophrys velifer* presence model was not produced since no variables could significantly predict its abundance. *Labrisomus nuchipinnis* was more prone to be found in pools with larger volumes and more total cover of the bottom by both organisms and algae. *Bathygobius burtoni* and *P. biafraensis* were more significantly correlated with rock pools with larger volumes and minimal biological cover on the pools' bottom. These results can also be observed in the PCO of fish assemblages (Fig. 3.5a).

Table 3.4. Relationship between rock pool characteristics and species abundance (stepwise regression results). Variables are listed in order of addition in the forward selection model. Only rock pool characteristics that had a significant effect on species abundance are displayed in the table.

Species	**Variables**	**R^2**	**F**	***p***
Abudefduf taurus	Volume	0.377	35.032	<0.0001
Microspathodon frontatus	Volume	0.204	14.876	0.0003
Stegastes imbricatus	Volume	0.388	36.762	<0.0001
Entomacrodus cadenati	% algae	0.110	7.177	0.0100
	Other metozoans	0.188	5.442	0.0230
Microlipophrys velifer	No variables were entered into the equation			
Labrisomus nuchipinnis	Volume	0.431	45.671	<0.0001
	Total % cover	0.506	7.606	0.0078
Bathygobius burtoni	Total % cover	0.361	32.704	<0.0001
	Volume	0.537	21.686	<0.0001
Prionurus biafraensis	Volume	0.132	8.847	0.0043
	Total % cover	0.224	6.736	0.0120

Discussion

Overall, fish assemblage composition and abundance seemed mostly influenced by the volume of rock pools, total flora and fauna coverage and water salinity. Particularly, rock pools with larger volumes supported generally a higher abundance and richness of fish species. The volume of a tidepool is often a good predictor of the number of fish it contains (Gibson, 1999a; Mahon and Mahon, 1994; Prochazka and Griffiths, 1992). Besides the fact that larger volumes allow for more space (and thus more biomass), it also decreases the fluctuation of the physicochemical properties of the water (temperature, salinity, pH and oxygen saturation) throughout tidal and diurnal cycles and variations in climate conditions (Raffaelli and Hawkins, 1999). This provides more stable conditions that a higher number of fish species can tolerate. This becomes particularly important in rock pools of tropical rocky shores, as extreme exposure to the heat and sun during the dryer seasons can cause dangerous rises in water temperatures (Vinagre *et al.*, 2018). Rainfall, which is strong in the region (Chou *et al.*, 2020; Herrero-Barrencua *et al.*, 2017), and freshwater runoff, can decrease considerably the salinity of rock pools (Evans *et al.*, 1999; Raffaelli and Hawkins, 1999).

The average salinity in rock pools was somewhat lower than expected for the dry season, despite the rising temperatures during the day that could lead to high evaporation in the pools (Evans *et al.*, 1999). This may be due to the salinity of the surrounding sea being already reduced by the influence of the plume of the Congo and Niger rivers throughout the Gulf of Guinea at this time of the year (Measey *et al.* 2007; Reid, 2011). Any salinity variation in pools with similar volumes was most likely caused by different climate conditions, such as occasional rainfall before the sampling period. Considering the salinity range registered, rock pools with higher salinity offered a more ideal environment for marine animals and attracted more fishes, although some fish species showed tolerance to lower salinities. However, the range of salinity obtained is only representative of the weaker dry season in which this study took place. Thus, it is likely that salinity may play a more critical role in determining fish assemblage variations throughout the seasons (Horn *et al.*, 1999).

The amount of biological cover on the pools' bottom was the second most influencing variable and it tends to be negatively correlated to fish composition and abundance, especially in smaller volumes. It possibly reflects the effect on the pH of the water, as pH is affected by the amount of dissolved carbon dioxide and oxygen in the water (Martin and Bridges, 1999; Raffaelli and Hawkins, 1999), which in turn depends largely on the conjunction of the tidal and diurnal cycles at emersion time, and the presence of animals and plants (Truchot and Duhamel-Jouve, 1980). During the day, pools with coral and algae produce oxygen which overrides carbon dioxide released from respiration, turning alkaline during emersion time (Martin and Bridges, 1999; Raffaelli and Hawkins, 1999; Truchot and Duhamel-Jouve, 1980), as was observed in this study. At night during emersion, the pH decreases as more carbon dioxide is solubilized and there is no oxygen production, making the pools more acidic (Raffaelli and Hawkins, 1999; Truchot and Duhamel-Jouve, 1980). Thus, the more cover of algae and/or coral a pool has, the more susceptible is to large and rapid oxygen and carbon dioxide changes, and consequently very discrepant pH values. This occurs especially in small and shallow pools that can change possibly by several pH units over the course of a few hours (Bridges, 1993a, 1993b; Martin and Bridges, 1999). Large rock pools are less susceptible to these changes and have less polarized pH values (Raffaelli and Hawkins, 1999). Even those that had 80% or more of total biological cover still presented high fish abundance.

Although dissolved oxygen was not registered, it is known to decline with increasing temperature which can lead to hypoxia during daytime during low tides in small and shallow pools without plants or coral (Martin and Bridges, 1999; White *et al.*, 2015). Smaller pools can easily gain higher temperatures, as was frequently recorded. Thus, only the fish species truly adapted for thermal and respiratory stress are capable of surviving in such pools, which are mostly residents and juveniles of secondary residents. Additionally, oxygen consumption by residents can exacerbate aquatic hypoxia in tidepools, especially in the absence of algae by day, or at night in pools with several organisms and algae (Martin and Bridges, 1999; Richards, 2011; Truchot and Duhamel-Jouve, 1980), reducing, even more, the possibility of other fish species settling.

Depth also affected considerably the abundance and richness of fish species found in rock pools, being the third most influencing variable for fish assemblages in this study. In terms of water conditions, deeper pools offer more protection from the heat and sun and allow the formation of a vertical salinity gradient under rainfall, allowing fish to shelter at the bottom in higher salinity. Whereas physically, deeper pools allow freer movement of fishes that are more active swimmers or that form schools (Lawson *et al.*, 1999), and offer more protection from predatory birds, which were frequently seen around the pools preying on trapped fish. These included four species of herons, *Egretta gularis*, *Egretta garzetta*, *Bubulcus ibis* and *Butorides striatus*, and the endemic kingfish, *Corythornis nais*.

The structural complexity of the rock pools was not documented during sampling, but structures like rock ledges, caves, crevasses and loose rocks were seen sheltering fish in some rock pools. The presence of some of these structures appears to be very important in determining the abundance and choice of habitat by intertidal fish, which may be linked to predator avoidance behaviour (White *et al.*, 2015). The higher the complexity, the higher the availability of cover for fish to hide. A similar study by White *et al.* (2015) in Sidney, Australia, revealed that the rock pools' structural complexity, as well as depth, were the main variables in determining fish presence, even more than volume. Based on field observations in this study of fish behaviour and the regularity of predatory birds around rock pools, it is likely that structural complexity also plays an important role in determining fish preference of rock pools on the island, a hypothesis that requires further study.

The vertical position of a rock pool on the rocky shore is also known to affect fish community composition (Zander *et al.*, 1999). Residents *E. cadenati*, *M. velifer* and *B. burtoni* and secondary residents *A. taurus* and *P. biafraensis* were the most abundant in the eulittoral zone. But field observations suggest that the further up in the tide profile the pools were positioned, the more they would be occupied by these species, particularly by *A. taurus* and *B. burtoni*. Whereas pools lower on the coast attained noticeably higher species richness and a more equitable proportion of residents, secondary residents and transients, likely due to the more stable water conditions. The age structure of fish assemblages is also affected by the pools' vertical position, as some studies have observed that smaller fishes are generally more abundant higher up on the intertidal zone (Zander *et al.*, 1999), demonstrating great tolerance to drastic physicochemical changes. This is probably because marine predators and interspecific competitors cannot withstand the physiochemical changes occurring at higher levels in the shore and thus there is a lower risk of predation and competition for available resources (White *et al.*, 2015). Although no individual size measures were taken in this study, field observations and underwater video footage provide a general qualitative perception of fish sizes. In the eulittoral, residents appeared both as juveniles and adults, and secondary residents and transients were predominantly and distinctively juveniles, with occasional occurrences of sub-adults and adults of damselfishes, *L. nuchipinnis*, *C. atlanticus* and moray eels, for example. Pools higher on the shore were practically comprised of just juveniles, with the exception of *B. burtoni*. Rock pools lower on the shore were seen with adult residents and noticeably more sub-adults and adults of secondary residents and transients, but still with an apparent juvenile dominance. This suggests that the rocky shores of Príncipe Island act as a nursery area for numerous species, including some with local commercial interest, such as *S. aurita*, *R. subbifrenatus*, *C. nigri*, *E. adscensionis*, *C. hippos*, *S. nigricauda*, *A. saxatilis*, *S. imbricatus*, *A. monroviae*, *P. biafraensis* and *C. pullus* (Afonso *et al.*, 1999; Direção das Pescas, 2015; Horemans *et al.*, 1994; Tous, 2015). *Lutjanus griseus* is also commercially exploited but with still no records in the local markets of São Tomé and Príncipe.

Of all the fish species observed in this study, some displayed a form of microhabitat selectivity, while others were more generalists. Volume and total biological cover played crucial roles in determining the habitat preference of several species, reinforcing the previous results obtained of general patterns in total fish assemblages.

Six out of the eight species with sufficient data for the regression analysis were more likely to be found in rock pools with larger volumes (*A. taurus*, *M. frontatus*, *S. imbricatus*, *L. nuchipinnis*, *B. burtoni* and

P. biafraensis). Even though these species proved to be well adapted to a variety of pool environments, including small and shallow ones, the larger pools are more stable and provide better continuous favourable conditions. *Entomacrodus cadenati* and *M. velifer* were, apparently, just as likely to be found in smaller and larger volumes. This can be related to the utilization of different habitats along different stages of their development, as their size increases.

In addition to the above mentioned, the other species observed in sampled rock pools were either secondary residents (*E. peli*, *R. subbifrenatus*, *C. hippos*, *A. saxatilis*, *T. newtoni* and *A. monroviae*) or transients (Leptocephalus, *A. lopeziana*, *C. nigri*, *Apogon imberbis* and *C. pullus*). These occurred in low abundance and were mostly found in larger and deeper pools, possibly because they are mostly temporary or accidental inhabitants of the intertidal zone. They were found in a narrow range of temperature, salinity and pH values, in comparison to residents and the more abundant secondary residents, suggesting that these species that thrive in open waters are more sensitive to fluctuations in water conditions. Furthermore, some of these species are pelagic and more active swimmers (*C. hippos*, *A. saxatilis*, *T. newtoni*, *A. monroviae*, *A. lopeziana*), many of them gregarious, which creates large demands for space (Lawson *et al.*, 1999; White *et al.*, 2015). Different species were seen often shoaling within pools, maybe as a mechanism of defence against predators, including *P. biafraensis* and *A. taurus*.

Predation by birds is a frequent cause of death in rock pools. For primary and secondary residents that spend a significant part of their life in tide pools, predator avoidance is a must-have skill to ensure survival on the rocky shores. A couple of strategies can come in the form of cryptic colouration and cryptic behaviour (Gibson, 1982). When reliant on their colouration, fish species may choose the rock pools in which they can best camouflage themselves. In addition to *B. burtoni* and *P. biafraensis* being more abundant in larger pools, they were also more likely to be found in pools with more bare rock, possibly for camouflage purposes. Their cryptic colouration (Miller, 2016; Rocha, 2016) matched well with the rocky black and grey bottom. They also displayed cryptic behaviour. When approached by observers, these fishes often remained still, especially *B. burtoni*. Juveniles of *P. biafraensis* sometimes laid sideways against the rock and even changed colour slightly between lighter and darker shades.

Entomacrodus cadenati, *M. velifer* (Williams and Springer, 2016a) and *L. nuchipinnis* (Williams and Springer, 2016b) also present cryptic colouration and were seen changing colouration between darker and lighter patterns, allowing them to camouflage themselves against a wide range of surroundings (rocks, coral, oysters, coralline and other algae, and sandy bottoms). Both *E. cadenati* and *L. nuchipinnis* were, in fact, significantly more likely to be found in pools with some cover of algae, coral or both. *Microlipophrys velifer* was not even correlated with any type of environment since it was seen practically in rock pools of all descriptions.

Other species also have cryptic colouration in relation to a variety of backgrounds found in the rock pools. *Abudefduf taurus*, which has a yellowish colouration with five vertical brown bars (Edwards, 2016), is camouflaged against surfaces covered by the yellow coral *P. caribaeorum*, which is very common in both the intertidal and sublittoral. Although *A. taurus* was not correlated with coral percentage cover in this study, it is important to point out that several individuals were regularly sighted in lower intertidal rock pools completely covered with the yellow coral (Fig. 3.9), as in shallow coastal waters. Eulittoral rock pools covered in coral may attain harsh water conditions, hence possibly the absence of correlation with coral percentage cover in this study.

Figure 3.9. *Abudefduf taurus* cryptic colouration against backgrounds covered by the soft yellow coral *Palythoa caribaeorum*. Photo taken in a low shore rock pool in Bom-Bom islet.

Overall, given the geographical and climatic context of Príncipe Island, fish assemblages tend to prefer rock pools with larger volumes, reduced presence of biological cover and higher salinity. However, other within-pool characteristics that were not sampled may also influence fish presence, in particular the availability of cover (i.e. rock ledges, caves, and loose rocks) which is likely to be representative of a good habitat (White *et al.*, 2015).

Primary and secondary resident species contributed the most to the fish assemblage structure, but the extent to which they do so is likely to vary vertically in the shore and seasonally, due to the variations in rock pool conditions (Gibson and Yoshiyama, 1999; Zander *et al.*, 1999). It may also change seasonally due to the arrival of juveniles following the breeding season (White *et al.*, 2015) and the migration of secondary residents to lower zones in the shore as they grow bigger (Norton and Cook, 1999).

The conspicuously small size of many residents, but particularly of secondary residents and transients, indicates that these rocky shores play a role as a nursery area for many coastal species of Príncipe Island. Future studies should focus on the structural complexity of the pools to assess the effect of certain structures on fish species abundance and composition. In addition, seasonal and vertical distribution patterns should be also assessed with more detail, taking also into account size/age structure of fish populations in order to better understand the use of these areas as nursery grounds throughout the year.

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References

Abreu, A.D. (2013). Príncipe Island's Biosphere Reserve (Democratic Republic of São Tomé and Príncipe): a living laboratory for sustainable development. Department of Environmental Affairs.

Afonso, P.F., Porteiro, F.M., Santos, R.S., Barreiros, J.P., Worms, J. and Wirtz, P. (1999). Coastal marine fishes of São Tomé island (Gulf of Guinea). *Arquipélago, Life* and *Marine Sciences, 17*A, 65–92. Ponta Delgada. ISSN 0873-4704.

Allen, G.R. (1985). An annotated and illustrated catalogue of lutjanid species known to date. FAO species catalogue. *FAO Fisheries Synopsis* (125) Vol. 6, 208 p. Rome, Italy.

Anderson, M.J., Gorley, R.N. and Clarke K.R. (2008). PERMANOVA+ for PRIMER: Guide to Software and Statistical Methods. PRIMER-E. Plymouth, UK.

Anderson, W.D.Jr. (2002). Lutjanidae. Pp. 1479-1504 in *The living marine resources of the Western Central Atlantic. Volume 3: Bony fishes part 2 (Opistognathidae to Molidae), sea turtles* and *marine mammals* (ed. Carpenter, K.E.), FAO Species Identification Guide for Fishery Purposes and American Society of Ichthyologists and Herpetologists Special Publication No. 5. Rome, Italy.

Bridges, C.R. (1993a). Adaptation of vertebrates to the intertidal environment. Adaptation of vertebrates to the intertidal environment. *The Vertebrate Gas Transport Cascade: Adaptations to the Environment* and *Mode of Life*, 12–22.

Bridges, C.R. (1993b). Ecophysiology of intertidal fish. *Fish ecophysiology*, 375–400.

Chou, S.C., de Arruda Lyra, A., Gomes, J.L., Rodriguez, D.A., Martins, M.A., Resende, N.C., Tavares, P.S., Dereczynski, C.P., Pilotto, I.L., Martins, A.M., Carvalho, L.F.A., Onofre, J.L.L., Major, I., Penhor, M. and Santana, A. (2020). Downscaling projections of climate change in Sao Tome and Príncipe Islands, Africa. *Climate Dynamics*, *54*(9-10), 4021-4042. https://doi.org/10.1007/s00382-020-05212-7

Clarke, K.R., Somerfield, P.J., and Chapman, M.G. (2006). On resemblance measures for ecological studies, including taxonomic dissimilarities and a zero-adjusted Bray–Curtis coefficient for denuded assemblages. *Journal of experimental marine biology* and *ecology*, *330*(1), 55-80.

Cravo, M., Almeida, A.J., Lima, H., Azevedo e Silva, J., Bandeira, S., Machava, V. and Paula, J. (submitted). Fish nursery role by a small mangrove at Príncipe Island, Gulf of Guinea. Submitted to *Frontiers in Marine Science.*

Direção das Pescas (2015). *Relatório do Estudo de Seguimento de Espécies Demersais da Zona Costeira de S. Tomé e Príncipe*. Report by the Fisheries Department, Ministry of Economy and International Co-Operation.

Edwards, A.J. (2016). Pomacentridae. Pp. 665-1509 in *The living marine resources of the Eastern Central Atlantic. Volume 4: Bony fishes part 2 (Perciformes to Tetradontiformes)* and *Sea turtles* (eds. Carpenter K.E. and De Angelis N.), FAO Species Identification Guide for Fishery Purposes, FAO. Rome, Italy.

Evans, D.H., Claiborne, J.B., Gregg, A. and Kormanik, G.A. (1999). Osmoregulation, Acid-Base Regulation, and Nitrogen Excretion. Pp. 79-96 in *Intertidal Fishes: Life in Two Worlds* (eds.

Horn, M.H., Martin, K.L.M. and Chotkowski, M.A.), Academic Press. San Diego, USA.

FAO (2016a). *The living marine resources of the Eastern Central Atlantic. Volume. 3: Bony fishes part 1 (Elopiformes to Scorpaeniformes)*. FAO Species Identification Guide for Fisheries Purposes, FAO, pp. 1511-2350. Rome, Italy.

FAO (2016b). *The living marine resources of the Eastern Central Atlantic: Volume 4: Bony fishes part 2 (Perciformes to Tetradontiformes)* and *Sea turtles*. FAO Species Identification Guide for Fisheries Purposes, FAO, pp. 2343-3124. Rome, Italy.

Firth, L.B., Schofield, M., White, F.J., Skov, M.W. and Hawkins, S.J. (2014). Biodiversity in intertidal rock pools: informing engineering criteria for artificial habitat enhancement in the built environment. *Marine Environmental Research*, *102*, 122-130. https://doi.org/10.1016/j.marenvres.2014.03.016

Fowler, H.W. (1936). The marine fishes of West Africa. *Bulletin of the American Museum of natural History*, 70(1-2), 1-1493.

Gibson, R.N. (1982). Recent studies on the biology of intertidal fishes. *Oceanography* and *Marine Biology: An Annual Review*, *20*, 363-414.

Gibson, R.N. (1986). Intertidal Teleosts: Life in a Fluctuating Environment. Pp. 388-408 in *The Behaviour of Teleost Fishes* (ed. Pitcher, T.J.), Springer, MA. Boston, USA.

Gibson, R.N. (1999a). Methods for Studying Intertidal Fishes. Pp. 7-25 in *Intertidal Fishes: Life in Two Worlds* (eds. Horn, M.H., Martin, K.L.M. and Chotkowski, M.A.), Academic Press. San Diego, USA.

Gibson, R.N. (1999b). Movement and Homing in Intertidal Fishes. Pp. 97-125 in *Intertidal Fishes: Life in Two Worlds* (eds. Horn, M.H., Martin, K.L.M. and Chotkowski, M.A.), Academic Press. San Diego, USA.

Gibson, R.N. and Yoshiyama, R.M. (1999). Intertidal fish communities. Pp. 264-296 in *Intertidal Fishes: Life in Two Worlds* (eds. Horn, M.H., Martin, K.L.M. and Chotkowski, M.A.), Academic Press. San Diego, USA.

Griffiths, S.P. (2003). Rockpool ichthyofaunas of temperate Australia: Species composition, residency and biogeographic patterns. *Estuarine, Coastal* and *Shelf Science*, *58*(1), 173-186. https://doi.org/10.1016/S0272-7714(03)00073-8

Herrero-Barrencua, A., Haroun, R. and Abreu, A.D. (2017). *Caracterización preliminar de los manglares de la Isla de Príncipe (São Tomé e Príncipe)*. IU-ECOAQUA, Universidad de Las Palmas de Gran Canaria. 70 pp.

Horemans, B., Gallène, J. and Njock, J. (1994). Revista sectorial da pesca artesanal a São Tomé e Príncipe. Programa de Desenvolvimento Integrado das Pescas Artesanais na África Ocidental.

Horn, M.H., Martin, K.L.M. and Chotkowski, M.A. (1999). Introduction. Pp. 1-6 in *Intertidal Fishes: Life in Two Worlds* (eds. Horn, M.H., Martin, K.L.M. and Chotkowski, M.A.), Academic Press. San Diego, USA.

Huggett, J. and Griffiths, C.L. (1986). Some relationships between elevation, physico-chemical variables and biota of intertidal rock pools. *Marine Ecology Progress Series, 29*(2), 189-197.

John, D.M. and Lawson, G.W. (1991). I.ittoral ecosystems of tropical western Africa. Pp. 297-322 in *Intertidal* and *Littoral Ecosystems* (eds. Mathieson, A.C. and Nienhuis, P.H.). Elsevier, Amsterdam.

Falcón, J.M., Brito, A., Herrera, R., Monterroso, O., Rodríguez, M., Álvarez, O., Ramos, E. and Miguel, A. (2018). New records of tropical littoral fishes from the Canary Islands as a result of two driving forces: natural expansion and introduction by oil platforms. *Revista de la Academia Canaria de Ciencias, 30*, 39-56.

Lawson, G.L., Kramer, D.L. and Hunte, W. (1999). Size-related habitat and schooling behaviour in two species of surgeonfish (*Acanthurus bahianus* and *A. coeruleus*) on a fringing reef in Barbados, West Indies. *Environmental Biology of Fishes*, *54*, 19–33.

Mahon, R. and Mahon, S.D. (1994). Structure and resilience of a tidepool fish assemblage at Barbados. *Environmental Biology of Fishes, 41*(1-4), 171–190. https://doi.org/10.1007/BF02197843

Martin, K.L.M. and Bidges, C.R. (1999). Respiration in Water and Air. Pp. 54-78 in *Intertidal Fishes: Life in Two Worlds* (eds. Horn, M.H., Martin, K.L.M. and Chotkowski, M.A.), Academic Press. San Diego, USA.

Martins, G.M., Hawkins, S.J., Thompson, R.C. and Jenkins, S.R. (2007). Community structure and functioning in intertidal rock pools : effects of pool size and shore height at different successional stages. *Marine Ecology Progress Series*, *329*, 43–55.

Measey, G.J., Vences, M., Drewes, R.C., Chiari, Y., Melo, M. and Bourles, B. (2007). Freshwater paths across the ocean: molecular phylogeny of the frog *Ptychadena newtoni* gives insights into amphibian colonization of oceanic islands. *Journal of Biogeography*, *34*, 7–20.

Miller P.J. (2016). Gobiidae. Pp. 2830-2843 in *The living marine resources of the Eastern Central Atlantic. Volume 4: Bony fishes part 2 (Perciformes to Tetradontiformes)* and *Sea turtles* (eds. Carpenter K.E. and De Angelis N.), FAO Species Identification Guide for Fishery Purposes, FAO. Rome, Italy.

Nelson, J.S. (2006). *Fishes of the World* (4th ed.). John Wiley and Sons, Inc. New Jersey, USA.

Norton, S.F. and Cook, A.E. (1999). Predation by Fishes in the Intertidal. Pp. 223-263 in *Intertidal Fishes: Life in Two Worlds* (eds. Horn, M.H., Martin, K.L.M. and Chotkowski, M.A.), Academic Press. San Diego, USA.

Raffaelli, D. and Hawkins, S. (1999). *Intertidal Ecology*. 2nd edition, 519 p. Springer Science and Business Media. Dordrecht, The Netherlands.

Reid, D.G. (2011). The genus *Echinolittorina* Habe, 1956 (Gastropoda: Littorinidae) in the Eastern Atlantic Ocean and Mediterranean Sea. *Zootaxa*, *2974*(1), 1–65. https://doi.org/10.11646/zootaxa.2974.1.1

Richards, J.G. (2011). Physiological, behavioural and biochemical adaptations of intertidal fishes to hypoxia. *Journal of Experimental Biology, 214*(2), 191–199. https://doi.org/10.1242/jeb.047951

Rocha, L.A. (2016). Acanthuridae. Pp. 2856-2862 in *The living marine resources of the Eastern Central*

Atlantic. Volume 4: Bony fishes part 2 (Perciformes to Tetradontiformes) and *Sea turtles* (eds. Carpenter K.E. and De Angelis N.), FAO Species Identification Guide for Fishery Purposes, FAO. Rome, Italy.

Tous, P. (2015). *Consulta para a avaliação dos recursos de peixes demersais*. Relatório do Projeto de Abordagem Ecosistémica Integrada para a Conservação e a Gestão da Biodiversidade em São Tomé e Principe (FIDA/PAPAFPA/GEF). Ministério da Economia e Cooperação Internacional.

Truchot, J.P. and Duhamel-Jouve, A. (1980). Oxygen and carbon dioxide in the marine intertidal environment: Diurnal and tidal changes in rockpools. *Respiration Physiology*, *39*(3), 241–254. https://doi.org/10.1016/0034-5687(80)90056-0

Tuya, F., Herrero-Barrencua, A., Bosch, N.E., Abreu, A.D. and Haroun, R. (2017). Reef fish at a remote tropical island (Principe Island, Gulf of Guinea): Disentangling taxonomic, functional and phylogenetic diversity patterns with depth. *Marine* and *Freshwater Research*, *69*(3), 395–402. https://doi.org/10.1071/MF17233

Vasco-Rodrigues, N., Bertoncini, A. and Fontes, J. (2018). *Peixes marinhos costeiros de São Tomé e Príncipe* (Coastal marine fishes of São Tomé and Príncipe). Portugal: Flying Sharks. 123 pp. ISBN: 978-989-208568.

Vinagre, C., Mendonça, V., Cereja, R., Abreu-Afonso, F., Dias, M., Mizrahi, D. and Flores, A.A.V. (2018). Ecological traps in shallow coastal waters-Potential effect of heat-waves in tropical and temperate organisms. *PLoS one*, *13*(2), 1–17. https://doi.org/10.1371/journal.pone.0192700

White, G.E. and Brown, C. (2013). Site fidelity and homing behaviour in intertidal fishes. *Marine Biology*, *160*(6), 1365–1372. https://doi.org/10.1007/s00227-013-2188-6

White, G.E., Hose, G.C. and Brown, C. (2015). Influence of rock-pool characteristics on the distribution and abundance of inter-tidal fishes. *Marine Ecology*, *36*(4), 1332–1344. https://doi.org/10.1111/maec.12232

Williams, J.T. and Springer, V.G. (2016a). Blenniidae. Pp. 2799-2806 in *The living marine resources of the Eastern Central Atlantic. Volume 4: Bony fishes part 2 (Perciformes to Tetradontiformes)* and *Sea turtles* (eds. Carpenter K.E. and De Angelis N.), FAO Species Identification Guide for Fishery Purposes, FAO. Rome, Italy.

Williams, J.T. and Springer, V.G. (2016b). Labrisomidae. Pp. 2796-2798 in *The living marine resources of the Eastern Central Atlantic. Volume 4: Bony fishes part 2 (Perciformes to Tetradontiformes)* and *Sea turtles* (eds. Carpenter K.E. and De Angelis N.), FAO Species Identification Guide for Fishery Purposes, FAO. Rome, Italy.

Wirtz, P. (1980). A revision of the Eastern-Atlantic Tripterygiidae (Pisces, Blennioidei) and notes on some West African blennioid fish. *Cybiton*, *11*, 83–101.

Wirtz, P., Ferreira, C.E.L., Floeter, S.R., Fricke, R., Gasparini, J.L., Iwamoto, T., Rocha, L. and Sampaio, C.L.S. (2007). Coastal Fishes of São Tomé and Príncipe islands, Gulf of Guinea (Eastern Atlantic Ocean) - an update. *Zootaxa*, *1523*, 1–48.

Zander, C.D., Nieder, J. and Martin, K. (1999). Vertical distribution patterns. Pp. 26-53 in *Intertidal Fishes: Life in Two Worlds* (eds. Horn, M.H., Martin, K.L.M. and Chotkowski, M.A.), Academic Press. San Diego, USA.

CHAPTER 4

Final remarks

Intertidal habitats from oceanic islands all around the world are still poorly studied, as well as the intertidal community structure of rocky shores of West Africa, particularly on the equatorial region of the Gulf of Guinea. In this work, these themes were addressed to complement the lack of available information. Given this, two studies were conducted. In the first study, two rocky shores of different locations of the island were sampled, each exposed to different environments (estuary and marine), in order to describe and compare the composition, distribution and zonation patterns of the biological community of each location. In the second study, rock pools from rocky shores of three different locations were sampled to analyse the relation between fish assemblage structure and environmental variables.

The main conclusions are:

- Zonation patterns are noticeably different between estuarine and marine exposed rocky shores of Príncipe Island with slightly different biological communities;
- Príncipe Island rocky shores have similar zonation patterns compared with other rocky coasts of the mainland of Gulf of Guinea;
- Intertidal fish assemblages of Príncipe rocky shores tend to prefer rock pools with larger volumes, reduced presence of biological cover and higher salinity;
- Fish assemblage structure was found to be mainly composed of primary and secondary resident species.

Overall, this work is in accordance with previous studies on tropical intertidal rocky shores in terms of how the main stresses affect the species distribution patterns. The intertidal biological communities of Príncipe Island showed to be very susceptible to the physical conditions in the region, where extreme desiccation is a major and determining factor for the intertidal species survival. Thus, the upper limits of algae and sessile organisms appear to be mostly defined by the direct action of physical factors, namely tides and wave action, which have a major influence on the environmental conditions. On the other hand, lower limits are more defined by biological factors, such as competition for space, herbivory and predation. As a result, the tropical island rocky shores are characterised overall by a highly crowded/patched infralittoral fringe dominated by soft coral, coralline algae and oysters, followed by a eulittoral zone and littoral fringe with a much lower species density and richness, which are largely dominated by highly desiccation-tolerant littorinids and other gastropods.

Príncipe Island has at least two distinct marine community structures that reflect essentially two common, but different rocky shores environments found around the island, namely the shores exposed to the marine environment and shores sheltered by natural barriers and influenced by rivers. Although no significant differences were found in terms of species composition, key species revealed that there is a notable difference between intertidal communities of the exposed and sheltered shores. These differences came down to the presence of more algae and suspension feeder species in the more exposed shore compared to the sheltered shore. The main difference, however, was found in the zonation and species distribution patterns, mainly in the infralittoral fringe and low eulittoral, where environment variables had a more frequent and pronounced effect.

As for the intertidal fish that inhabit the rock pools, they are also affected by the characteristic environmental conditions of the region, namely extreme sun exposure and abundance of rainfalls. Thus, fish assemblages generally preferred rock pools where conditions are more stable throughout the tide cycles, which in this case translates in larger volumes, reduced presence of biological cover, such as macroalgae and corals, and higher salinity. Because the salinity of the sea surrounding Príncipe Island is still influenced by the plume of the Niger and Congo rivers, evaporation does not lead to often extreme saline pools, which is normally the case in many other regions where precipitation is lesser. The most abundant fish species sampled were those that are seemingly tolerant to a wider range of physicochemical conditions in the rock pools, which mainly corresponded with resident species and a few secondary resident species. Some of the species found inhabiting the rock pools are of commercial interest.

Some constraints were found and much remains to be done about the intertidal environments on the rocky shores of Príncipe Island. One important flaw of this work was the lack of data from other seasons, which may reveal seasonal variation in species composition, richness and even distribution patterns. Rainy seasons in this region have high precipitation rates that not only directly affect the salinity and temperature of the rock surface and rock pools but also of the adjacent sea through the amount of freshwater released by the rivers. Furthermore, there should have been sampled more area of rocky coast to obtain a more accurate estimate of the structure of intertidal communities in the different environments, both in terms of composition and distribution patterns. Some intertidal species previously found on rocky shores of São Tomé and Príncipe, namely algae and molluscs, were either not seen or found outside the sampling period.

Future studies on rocky shores should carry out seasonal sampling and study other rocky shores of the island that present both similar and different conditions in the environmental gradients studied in this work, namely exposure to wave action and salinity. Recommendations include the great bay of Santo António as it has environmental conditions in both wave exposure and salinity gradient spectra, and both the east and south coast of the island for their exposure to the predominant winds from the southeast.

As for the ichthyofauna in rock pools, some variables also remained to be studied. Other within-pool characteristics that were not sampled may also influence fish presence, in particular the availability the bottom complexity (i.e. rock ledges, caves, and loose rocks), which can modulate biodiversity level, and oxygen availability. The vertical position of the rock pools on the shore was also not studied, which likely affects the fish assemblage structure due to the variations in rock pool conditions. Conducting a more detailed study on the structural complexity of the pools and their vertical position on the shore would provide a more accurate account of the effect of certain structures and physicochemical conditions on fish assemblage structure and which habitat species prefer. Additionally, seasonal patterns should also be assessed as the fish composition may change due to the arrival of juveniles following the breeding season or the migration of secondary residents to lower zones in the shore as they grow bigger.

There is potential for many other different approaches to future studies of fish assemblages on rock pools on the island. Future studies should investigate the following: the age structure of the intertidal fish populations; the role of the rocky shores as nursery grounds; the importance of rocky shores to the recruitment of fish to coastal fisheries; herbivory, predatory and competition behaviours in the intertidal; and whether the rock pools in this region are considered ecological traps due to the climate changes.

Like other oceanic islands, Príncipe Island is considered a natural laboratory with much potential for future studies since its marine biological communities are yet relatively unknown to science. All things considered, it is important to monitor the intertidal environments of the Príncipe Island, as well as those of other oceanic islands, due to their vulnerability to local and global-scale anthropogenic threats and climate change.