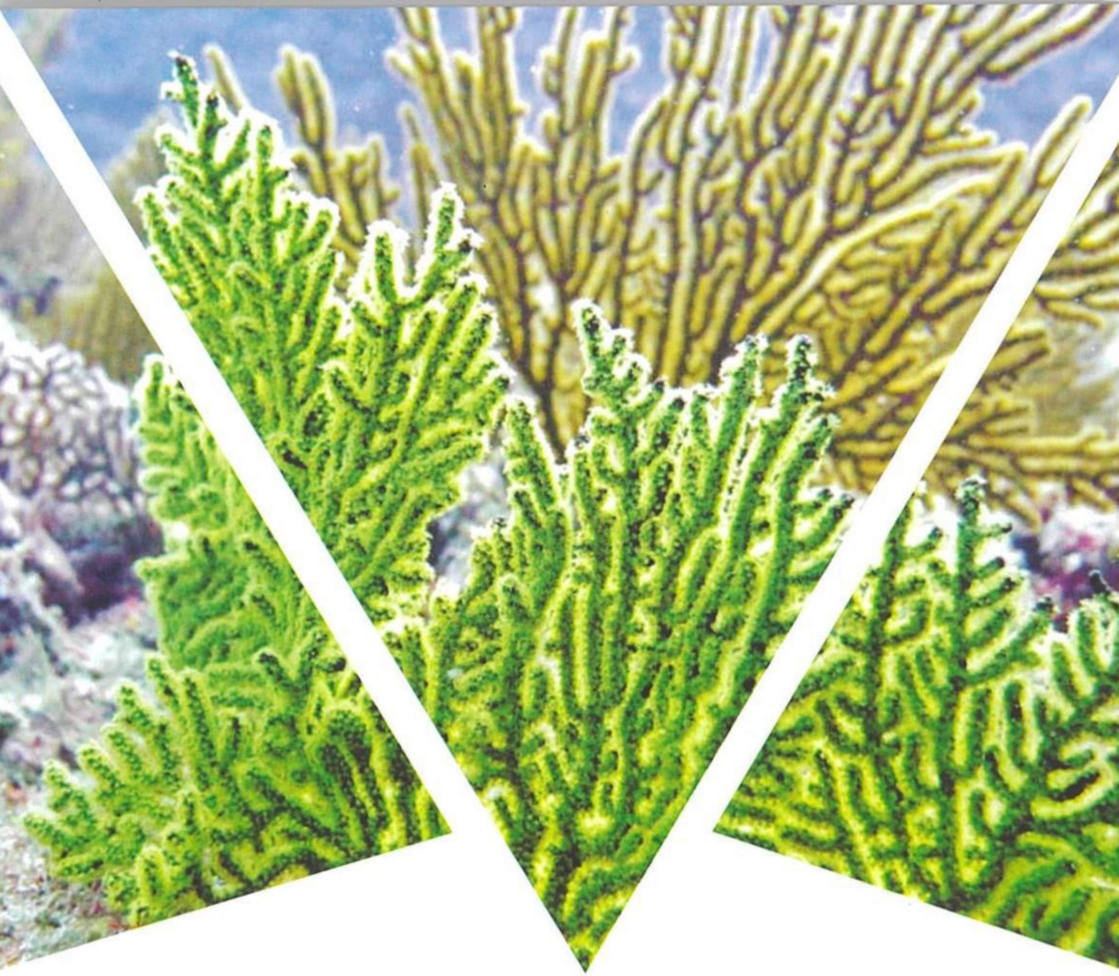

Documenting benthic invertebrate from rocky environments in the REMAPE

Secretaría de
Educación Superior,
Ciencia, Tecnología e Innovación

CENAIM - ESPOL

ESPOL

MANUAL ON DOCUMENTING BENTHIC INVERTEBRATE COMMUNITIES FROM ROCKY ENVIRONMENTS IN THE MARINE RESERVE EL PELADO, SANTA ELENA, ECUADOR

ISBN: 978-9942-36-314-5

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Proyecto Prometeo: Creación e implementación de un sistema de información marino y atlas dinámico georeferenciado de biodiversidad del ecosistema del área marina protegida El Pelado, Santa Elena, Ecuador

Proyecto: Caracterización de la biodiversidad microbiológica y de invertebrados de la reserva marina El Pelado a escalas taxonómica, metabolómica y metagenómica, para uso en salud humana y animal

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Short Title

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PROLOGUE

Marine biodiversity offers a wide range of opportunities for biotechnological development. Marine organisms constitute sources of protein, bacteria with unique properties, and biomolecules with antimicrobial or antitumor activity. Paradoxically, while marine biomes cover 70% of the planet's biodiversity, they are much less known than terrestrial biomes. The eastern tropical Pacific ecoregion (TEP) is one of the least studied zones. Ecuador located at the southern end of the TEP has a great variety of marine-coastal ecosystems. Its privileged location, next to the equatorial front, makes the Ecuadorian coast a natural laboratory suitable for studies of functional biodiversity, biodiscovery, biogeography and climate change. The fluctuating environments of the equatorial front give shape to the biodiversity of the El Pelado Marine Reserve (REMAPE), with the presence of cold and warm water organisms. Ecuador needs to create advanced interdisciplinary research capabilities on marine organisms, their microbiomes, and the sustainable use of marine resources. With support from SENESCYT, ESPOL approaches the study of the biodiversity of benthic invertebrates of REMAPE as an opportunity for scientific and technological development, with applications in health, aquaculture, and conservation. This approach requires deepening the research of the composition and the distribution of benthic communities. In Ecuador, the knowledge base on this topic is modest. This manual of marine samplings concentrated on the rocky environments of REMAPE is a useful tool for students and professionals in marine sciences and aquaculture. Its objective is to provide guidance in the creation of methods of benthic studies for ecological studies on the Ecuadorian coast characterized by turbulent and turbid waters. This manual is also a guide to simplify the collection of data on the distribution of marine organisms, in order to facilitate the initiatives of bio-discovery, repopulation and aquaculture of invertebrates in Ecuador.

ESCUELA SUPERIOR POLITÉCNICA DEL LITORAL

Cecilia A. Paredes V., Ph.D.

RECTORA

PREFACE

This manual was conceived as an integral part of a multitier and replicable model approach commenting invertebrate communities in benthic marine habitats on the continental coast of Ecuador and to make the findings widely accessible. The approach is based on field surveys using a protocol that was specifically designed for the common turbulent and turbid oceanographic conditions encountered in Santa Elena, Ecuador (zone characterized by water mixture, thermohaline gradient, and upwelling), and for the clarification of a study site's topography. A digital information platform that summarizes the findings and their interpretation for a broader audience (www.cenaim.espol.edu.ec) complements the model. The manual outlines an integrative approach towards documenting marine biodiversity and addresses students and professionals in marine sciences and aquaculture, as well as nature enthusiasts with background knowledge in marine environments and ecology. It intends to provide guidance in creating benthic survey methods for ecological studies (research and education) or to facilitate the collection of in field organism distribution data, which may complement invertebrate aquaculture and repopulation initiatives in Ecuador.

ACKNOWLEDGMENTS

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ABBREVIATIONS

CENAIM	National Center of Aquaculture and Marine Research (Centro Nacional de Acuicultura e Investigaciones Marinas)
CPCe	Coral point Count with Excel Extensions
ECC	Encrusting Calcareous Corallinacea substrate
EPP	El Pelado Platform = The area surrounding the islet
ESPOL	Escuela Superior Politécnica del Litoral
GIS	Geographic Information System
GPS	Geographic Positioning Systems
REMAPE	Reserva Marina El Pelado
SCUBA	Self Contained Underwater Breathing Apparatus

INTRODUCTION

Interest in marine studies of Ecuador's coastal habitats has been in the shadows of a plethora of research projects and publications about the Galapagos Islands. The Islands' almost binding association with Darwin's "The Origin of Species" from 1859 continues to be a catalyst in attracting researchers and an asset in marketing this internationally renowned tourism destination. In recent years, the imbalance in terms of environmental studies has shifted towards an increasing interest in the investigation of continental marine habitats. This change of perception was in part aided by the efforts governmental and national private environmental entities (*i.e.* Fundación Nazca, Equilibrio Azul, Bioélite), and through studies by students at universities with marine biology and or fisheries related programs such as the Escuela Superior Politécnica del Litoral in Guayaquil, the Universidad Estatal Peninsular de Santa Elena in Santa Elena and the Universidad Laica Eloy Alfaro in Manabí among few others. Despite a growing number of field surveys using SCUBA, the knowledge base about shallow benthic communities remains modest (Cruz *et al*. 2003), as many findings remain unpublished and poorly accessible. A notable redundancy in projects from multiple national agencies covering the same or similar topics is one of the consequences of this situation. Furthermore, the implemented survey methods vary greatly and are generally based on the monitoring protocol developed by the Fundación Charles Darwin (2002) in and for marine benthic environments of

Galapagos with vastly clearer waters than those encountered along the continental coast. Alternative survey protocols that are specifically designed for the recurrent oceanographic conditions of continental Ecuador have not very been formulated.

At a time when the aquaculture industry of Ecuador is seeking diversification in the commercially viable species (De La Roche 2015) and trawling shrimp fisheries have been halted as of December 2012, a better understanding of the species composition and distribution of native benthic communities appears valuable. Yet there is still a paucity of information on the natural distribution of benthic species in general. This also holds true for species that have already been heavily overfished and for which culturing techniques, *e.g.* the pectinid bivalve *Spondylus* spp. (Mackensen *et al*. 2011) and reseeding strategies, *e.g.* the sea cucumber *Isostichopus fuscus*, are currently being developed in Ecuador. In the case of highly camouflaged species such as *Spondylus* sp., the inclusion of experienced fishermen is instrumental in estimating current stocks in the wild. However, to date the anecdotal knowledge and skills of local fishermen have not been incorporated into systematic field surveys.

The aim of this study was to devise a model approach that addresses the abovementioned shortcomings by adjusting field surveys to common environmental and logistic limitations and to lessen redundancies in future studies on marine invertebrate communities by facilitating access to the information generated. It was carried out in the Marine Reserve El Pelado (REMAPE, for its acronym in Spanish), situated in province of Santa Elena, Ecuador. The following principal objectives were pursued during the project summarized herein.

OBJECTIVES

The first objective was to clarify the geographic attributes and site names of the area studied. Mobile devices with satellite-based geographic positioning systems (GPS) have augmented the ease of access to a specific point of interest or a monitoring station. However, the absence of bathymetric maps at scales relevant to a particular field survey, lessens the prospector interpreting findings on the distribution of substrate types, and consequently also of benthic organisms within a topographic context. Unanswered questions may include: "How common or representative is a certain type of substrate or habitat within the area examined." Furthermore, local site names change over time and this complicates the comparison of reports using different site names. Therefore, an approach combining site coordinates, topographic features and cardinal directions was sought to locate and name study sites.

Secondly, a survey protocol was to be designed and fitted to the local oceanographic conditions and logistic limitations. The protocol should allow an efficient way to capture qualitative and quantitative data on substrate types and the sessile as well as mobile invertebrates found on them. Criteria for the design included the identification of conspicuous species that can be incorporated into a future monitoring protocol, based on their relative ease of identification in situ by skilled divers with basic training in biological sciences. The organism must thus be identifiable macroscopically and possess robust external taxonomic traits. Necessary materials and instruments must be locally available or acquirable with a modest budget.

And thirdly, a public internet information platform which interprets the findings and contains a centralized bibliography on the topic was to be created. The internet platform was to be structured in a way so that descriptions and interpretations of benthic marine communities of REMAPE are accessible via the selection of a georeferenced particular site (geographic approach) as well as via a particular habitat type or organism (ecological approach).

STUDY AREA

Location and dimensions

The REMAPE is situated in province of Santa Elena, Ecuador, and lines the coast between the northern limits of Valdivia and the river mouth at Palmar to its south (Figure 1). It extends approximately 5 km in a westerly direction from Punta Brava near Ayangue and encompasses 13.005 ha of marine habitats and 96 ha of terrestrial areas (Government of Ecuador 2016) (Area between -80.7326 y -80.8233 latitude and between -1.8993 and -2.0258 longitude). Declared as marine reserve in 2012, the REMAPE is one of 11 areas that have so far been assigned varying degrees of protection along the continental coast of Ecuador (Government of Ecuador 2016).

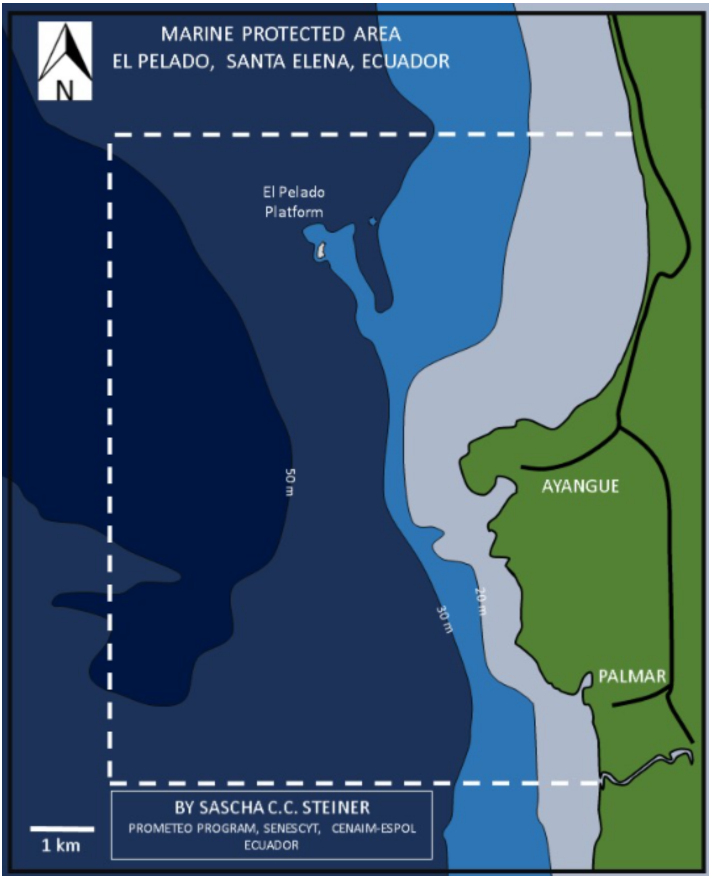


Figure 1. Delineation of the REMAPE, based on bathymetric chart IOA 1051.

Oceanographic setting

With regards to tropical surface waters, the REMAPE lies within the range of Eastern Pacific seasonal oscillations of the equatorial front, which separates the southward flowing Panama Flow and the northward flowing Peru Current. The reserve is also under the influence of the southward-flowing annual El Niño current, which originates as the western branch of the Panamá Bight gyre (Glynn

2003, based on Fiedler 1992 and Strub *et al*. 1998). Mean annual water temperature in 2015 was 26.5°C at the surface and 22.4°C in 30 m depth (S. Sonnenholzner, unpublished data). Due to the persisting winds and their intensity (Appendix 1 y 2) the western margins of the islet and its surrounding mounds have a windward exposure, whereas the eastern margins are in a leeward position with calmer waters for most of the year. Tidal amplitudes reach up to 2.3 m.

Hints from Geology

Pleistocene marine bioclastic terraces (Tablazos) characterize the coast in the vicinity of Ayangue and southward up to Palmar, while older (Miocene) shales and siltstones mark the northern shores of the REMAPE (National Geological Map of Ecuador, 1982). It is possible that El Pelado and the four additional rocky mounds are, at least superficially, composed of the same type of strata visible around Ayangue, in the cliffs between Punta Brava and Punta Del Teco. The strata in the area surrounding the “El Pelado” Islet (EPP) are tilted downward in an east-southeasterly direction and vertically fragmented into large slabs. This geological feature is typical of the topography of the area in that the steep drops in terrain are generally found on the western and northwestern margins of each of the rocky mounds. Such drops commonly include overhangs and small caves formed by the erosion of softer sediment layers. In contrast, gently inclined planes (parallel to the strata horizons) characterize the east-south easterly facing seafloor.

The implications of this particular topography are that the northern and northwestern margins provide ample habitats suitable for sciaphile (shade-loving) organisms (*e.g.* ahermatypic stony corals), while flattened surfaces facing east south-east provide well illuminated stable substrates for photoautotrophic organisms as filamentous turf algae and the associated grazers (*e.g. Diadema mexicanum* Agassiz, 1863). This topographic setting thus predetermines the distribution of habitat types described in the next section. And in a purely logistical sense, the topography is an orientation aid when navigating the terrain on SCUBA.

Along shore, rocky substrates are not distributed in such a structured manner, as they are primarily found within or near the breaker zone of swells and are exposed to stronger mechanical forces.

Clarifications on the geographic setting. Sites names

In order to establish the clearest possible image of the topography of the study area (first objective), existing nautical charts were compared, while technical reports and publications from the area were screened for maps and source maps. This process was accompanied by reconnaissance dives and snorkels to obtain an overview of the topography and the benthic communities found on these. With visibilities rarely exceeding 5 meters, the rocky reefs are often not visible from the sea surface. Fishermen were thus consulted about the distribution of rocky and sandy areas, and the presence of epibenthic macroalgal and invertebrate communities. In addition, geological maps of the region were consulted for a better understanding of the general make up of sediments.

By overlaying the nautical charts with the sonar-topography image from the Subsecretaría de Gestión Marino Costera, Ministerio del Ambiente, it became clear that four distinct areas (mounds) ranging from minimum depths of 0 – 15 m surround the islet (Figure 2). The nautical charts IOA 1051 (1:25000) for Valdivia to Monte Verde and IOA 105111 (1:7500) for Bahia de Ayangue, were used to draw the coarse isobaths within the REMAPE. A sonar-topography image of the islet El Pelado and its neighboring terrain by the Subsecretaria de Gestión Marino Costera, Ministerio del Ambiente, in combination with satellite images by Google from November 2012, served as base for tracing bathymetric isobaths of that area. This revealed that what is referred to the “El Pelado” Islet is in fact the emergent part or islet of a single submerged peninsula delineated by the 30 m isobath.

During exploratory dives, it was noted that all mounds were composed of stratified rocky substrates and that each mound was surrounded by soft sediments. Rocky formations extended down to a maximum depth of approximately 25 m.

Up to 10 dives sites surrounding the Islet (Figure 2), many of which are equated with separate reefs or “bajos”, have been reported. To avoid misconceptions, it worth to specify, the area surrounding the islet is referred to as the El Pelado Platform (EPP), and its surrounding rocky mounds were labeled based on their cardinal direction from the islet (*i.e.* NW for northwest of the islet) and listed in Appendix 3.

Areas beyond 25 m depth were poorly explored and it remains uncertain whether other rocky mound sexist in the vicinity of the EPP. One isolated additional rocky mound that has been recorded is known as Bajo Tello. It lies in approximately 32 m depth rising to 25 m. It is located to the east of the islet (1°55’687’’S 80°46’976’’W) and is only depicted in a single nautical chart (IOA 1051 for Valdivia to Monteverde), yet absent in others. No incongruences in bathymetry or topography were evident among charts covering the nearshore sublittoral habitats around Ayangue, which were predominantly sandy.

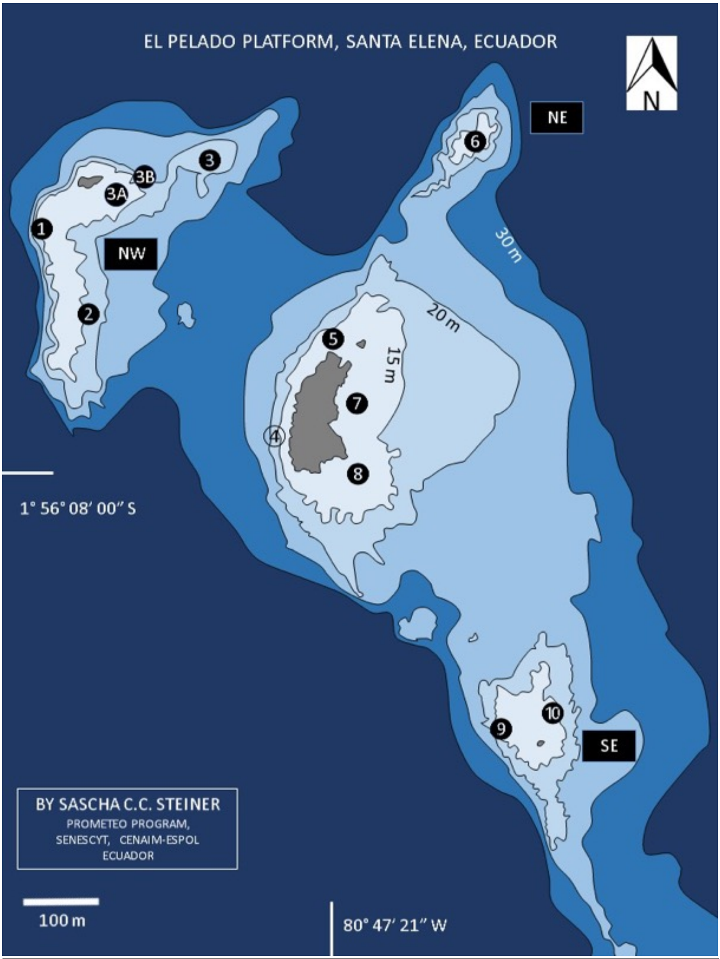


Figure 2. Bathymetric contour of the three rocky mounds surrounding El Pelado Islet, based on radar-topographic images by Subsecretaría de Gestión Marino Costera, Ministerio del Ambiente.

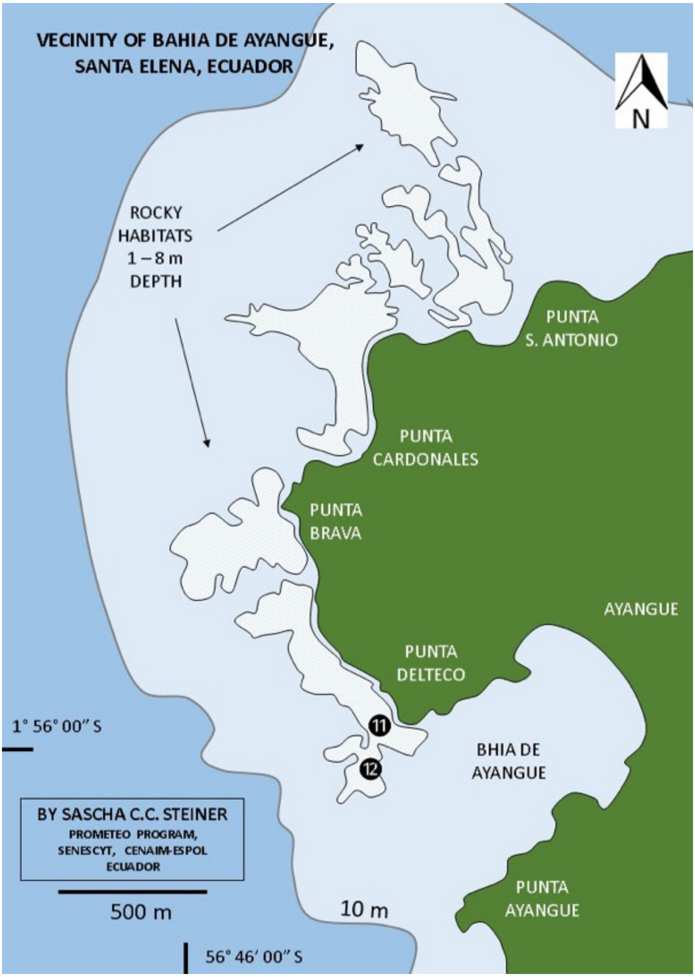


Figure 3. Coastal areas in the vicinity of Ayangue.

Resource uses in the Ayangue community

The fishing village Ayangue is the main point of access to REMAPE for the growing recreational dive industry (Cardenas and Triviño 2013) in the area. To date, snorkeling and sightseeing excursions are the most common tourist activities having modest impacts of benthic communities. However, numbers of SCUBA divers and potential for direct impacts (breakage of stony corals) is increasing. To minimize anchor damage to benthic communities, permanent buoys have been installed at the popular dive sites surrounding the "Islote El Pelado".

Another current touristic activity is represented by the whale watching tours, that bring tourists to observe the *Megaptera novaeangliae* in the whale's season (from June to the end of September). This activity started in the last 10 years and it is bringing economic benefits to the Ayangue community.

Fishing activities in and around El Pelado can be dated back to the Valdivia culture which emerged in 3500 BC and was linked to regional trade (Bauer 2007, Martín 2001). More recently, with the stop of the industrial-scale trawling in 2012 (De La Roche 2015), fishing practices concentrate around artisanal bated line-fishing, seine net fishing along the sandy shores, compressor-supported harpooning and collection in areas with rocky substrate. The collection of corals and mollusk shells as curiousity boomed in the 1980s and 1990s (Glynn 2003) and has since then declined. Despite regulations limiting fishing activities in the REMAPE, enforcement and compliance appear lax as a variety of fishing practices are observable on a daily basis, and marine curious are still available in Ayangue and many other coastal communities.

Conservation actions in the area

An organism of high interest in Ecuador is the Spondylus, due to its commercial importance that caused its overexplotation. In

2009 by petition of the Salango community, Manabí Province, the subsecretary of fishery resources under the Ministry of Agriculture, Livestock, Aquaculture and Fisheries (MAGAP), decreed a fishing closure of *Spondylus* spp. (Acuerdo Ministerial 136) and banned their harvesting, transport, and consumption (Viceministerio de Acuacultura y Pesca 2009). Given the concern about the declining stocks of natural populations, there have been research-oriented studies to support the eventual development of spondylid aquaculture and stock enhancement initiatives (Lodeiros *et al*. 2016). In addition, CENAIM-ESPOL and MAGAP are developing a repopulation program of Spondylus limbatus based on hatchery-produced juveniles, allowing the seeding of the former natural population at El Pelado, a well-known *Spondylus* spp. bed.

Furthermore, it is worth mentioning that The Fishery National Institute started a research-oriented study to evaluate the resource and found an average abundance of 0.02 individuals in 100 m², showing no sign of stock recovery of traditional beds located in Santa Elena Province (Vice-ministerio de Acuacultura y Pesca 2010). Currently, the National Center of Aquaculture and Marine Research (CENAIM) of Escuela Superior Politécnica del Litoral (ESPOL), in a more exhaustive prospection in 2015 at EPP, found an average abundance of 3 individuals in 100 m² (Sacha Steiner, CENAIM-ESPOL, personal communication, 2016), suggesting an incipient recovery.

METHODS

Based on the new maps that were generated (Figures. 1, 2 and 3) 12 sites of interest were chosen for qualitative and quantitative surveys (Anexo 3). Ten sites around the "Islote El Pelado"(Figure 2) and two along the coast (Figure 3).The site selection was preceded by a stratified-random process by superimposing a 50 x 50 m plots onto the generated maps and by randomly drawing plots within each area (islet and coast) that meet the criteria: maximum depth of 16 m, rocky substrate, distinct exposures (windward vs. leeward) and ease of access (mooring) where possible.

We found a few coastal sites with rocky substrates, mostly near Ayangue. The sites were in shallow and highly turbulent areas, and were thus qualitatively examined while snorkeling, one site was surveyed quantitatively using SCUBA.

Due to the broad taxonomic approach in the second objective of the study, as well as the logistic and environmental limitations encountered, various trials were carried out to establish an adequate survey method (sample size) and protocol to quantify benthic invertebrates and the substrate types on which they were found. A detailed account of this process that led to the findings presented here is described below.

In sum, qualitative as well a quantitative data on the presence of benthic invertebrates were collected during the daytime across four 40 m belt transects per site. Nocturnally active organisms are thus underrepresented in the survey. Initial surveys based solely on in situ quantification of invertebrates were too time-consuming for the broad range of organisms considered. Consequently, trails with video and photo-transects were carried out with the aim of maximizing the information gathered per dive. Individual video transects covering 20 m and photo-series transects composed of 20 images, each detailing 120 cm^2, and 2.4 m^2 per transect, proved most effective given the dive-inherent time limitations and the common turbidity levels (1-5 m) in the area.

The above-mentioned survey methods were integrated into a single protocol for three divers and carried out on each of the four belt transects per site. The principal points of this protocol were: (a) Invertebrates that tend to be located in crevices during the day, like the sea urchin *Centrostephanus coronatus* (Verril 1867) or that are heavily overgrown by epibionts such as the gastropod *Vasum caestus* (Broderip 1833), were quantified in situ. (b) Larger sessile organism's such as octocorals (*i.e. Muricea purpurea*, Verrill 1864) were quantified using video transects (c). Transects based on photo series (named photo-transects from here on forth) were used to record and subsequently quantify substrate composition.

The protocol was carried out by two divers who surveyed two transects per dive in depths of up to 16 m. One diver was assigned to the in-situ quantification of "cryptic and camouflaged" species (point a), the second diver to the photo- and video-transects (points b and c). A third diver, was assigned to solely quantify highly camouflaged species. In this study, the target was Spondylus, as genus that has received increased attention by aquaculture. This third diver needed to have many years of experience in the recognizing and collecting of the species. Throughout the surveys, specimens of the species seen were collected in an effort to build a reference collection and in some cases for taxonomic validation. The identification of invertebrates was based on external features described in field guides (Hickman 1998, 2008, Hickman and Finet 1999, Rivera and Martínez 2011) and taxonomic keys (Cairns and Kitahara 2012), and the examination of sclerites in case of Alcyonacea (Breedy and Guzman 2015).

No trials were necessary in preparation for a public internet platform describing the benthic communities of REMAPE. Following Steiner and Molina (2012), the findings were sorted by study site (geographic approach) and by habitat type (ecological approach) (http://www.cenaim.espol.edu.ec/es/node/453).

Transect length and materials

In Ecuador, the most widely used protocol is the monitoring protocol of the Fundación Charles Darwin (2002) targeting the abundance of sessile invertebrates along 100 m^2 belt transect and their benthic cover within five 0.25 m^2 quadrates, per transect. The transect line used as reference is described as the commonly used PVC measuring tapes. The protocol includes fish surveys, which were not considered in the current study.

A transect length of 50 m was considered inappropriate due to the rather turbulent waters in the study area, and the poor visibility that complicated the communication between various divers along the transect. Fifty meters also often exceeded the area available at a particular depth on smaller rocky formations. A shorter transects (20 m) with more replicates per site, proved to be more efficient. Furthermore, it appeared beneficial to work with a metal chain as transect (Hill and Wilkinson 2004) rather than a PVC measuring tape in turbulent conditions for two reasons.

First, the chain transect can be placed to contour the seafloor, thus eliminating discrepancies between transect length and actual area served. Such discrepancies are commonly encountered in structural complex rocky environments. Although rugosity can be addressed with a varietal of estimates or categorizations, these bare inaccuracies that may undermine the comparison of data from different researchers and projects. Secondly, a chain is much more stable than a PVC tape in agitated waters. However, two nuisances inherent in using a chain needed to be minimized: the fact that chains can cause damage and injures to fragile sessile organisms (*i.e* ramous stony corals), and that it is usually in a corroded state, leaving dive slates or data sheets consistently smeared during the handling in a dive. The so called “lead core rope” combines the benefits of handling a rope and the negatively buoyant and contouring feature of a chain. However, this specific type of rope is not readily available in many regions of the world including Ecuador.

In order to combine the above-mentioned benefits of lines and chains, a weighted line was constructed by securing lead weights of approx. 36 gr, every 50 cm on a 20 m transect line of 5 mm in diameter (Figure 4, close up and in field). Both lines and lead weight were available at local fishing supply stores. The lead weights may also serve as reference points for the point-intercept method. Additional survey points can be added with markers (on white rope) or with zip ties. For the sake of better orientation under water. Small PVC platelets were added to the line at 2 m intervals.

Figure 4. Left: close-up of transect line with lead weights (every 50 cm); Right: rolled up transect line.

Substrate quality and time constraints in situ

During exploratory dives in the preparation for the project we encountered a variety of substrate types that were part of conspicuous horizontal biological zones, on rocky terrain, between 0 and 20 m. Therefore, the classification and quantification of substrate types was an essential objective as benthic communities are intimately shaped by the type of substrate that is available, whether live or dead, an aspect that had not been incorporated in the FDC protocol from 2001. Ignoring the classification of substrate types eliminates many clues in the interpretation of the community structure encountered. However, the quantification of substrate types proved to be quite time consuming and this represented a logistic constraint.

During a series of trials, the maximum surface area that could be captured in a digital image, and that permitted the recognition of substrate types and small invertebrates was sought. Given the common turbidity levels, an area of 30 x 30 cm was the largest area that could be recorded at a resolution that allowed the subsequent successful identification of substrate types and organisms on a computer screen, when photographs were taken without stabilizing tripod. With added stability, a slightly larger area could be captured and provided the needed clarity within the image. In order to maximize the quantifiable area per images, rectangular format 4:3 was chosen. In the end a quatripod with an image capturing area of 40 x 30 cm was constructed for the camera Canon Power Shot D20, using the underwater housing WP DC45 (Figure 5 and 6). Such a method would also allow us to train divers with more limited biological knowledge in an unequivocal application of this method.

Figure 5. Quatripod with an image capturing area of 40 x 30.

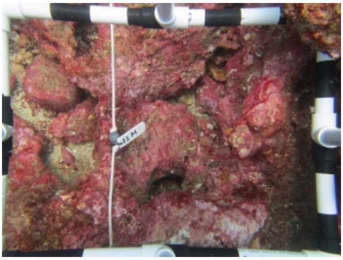


Figure 6. Sample image made with quatripod.

Time constraints during in situ quantification of invertebrates, large sessile invertebrates

Among the large invertebrates, some taxa were particularly abundant, namely Octocorals in the Alyconacea. Their quantification also can be extremely time consuming and represented a further logistic constraint.

Video transects that captured the area of 20 m x 1 m on either side of the transect line proved to be adequate for the identification of Alcyonacea. To do so, the diver kept to one side of the transect (*i.e*. the right side), and swam first from 0 to 20 m along the line recording first the area from 0 to 20 m, and then the area from 20 to 0 m on the other side of the transect during the return swim. Each swim covering between 0 m and 20 m was filmed in two to three minutes. However, several trials were necessary to practice maintaining a course and stability in agitated waters, and to handle the camera in such a way that species could later be identified. A T-bar was used to delineate the area to be filmed along the transect (1 m^2) (Figure 7). More detailed description of the process is given in the protocol described below.

The identification process was based on previous trials in identifying organisms with external morphological features. In the case of Alcyonacea these included the branching pattern of the colony and coloration of the extended polyps. Branching patterns are best studies and established with samples of dried specimens, compiled previous to the quantitative surveys.

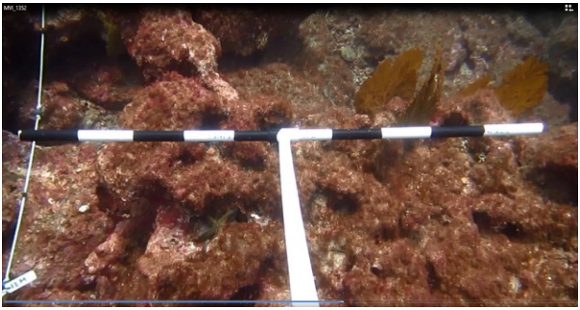


Figure 7. Caption of 1 m wide T-bar used to delineate area to be filmed along the transect.

In suboptimal visibility conditions, recording times were longer as it was necessary to include close ups for individual specimens to allow their later identifications. Nonetheless, this method was very time efficient in the field, leaving the quantification for time outside of the water. In dense and highly intertwined Alcyonaria assemblages, this method cannot be properly applicable because individual colonies are not clearly discernable.

Small, cryptic, nocturnal and highly camouflaged invertebrates

A large variety of invertebrates are small and cannot be properly identified in photo or video transects described above. Others may be identifiable in terms of their size but tend to be hidden in recesses (bivalves) and crevices, or so heavily overgrown by epibionts (gastropods) that they are not recognizable in photos or videos. And yet others are nocturnal (urchins) spending daylight hours under ledges or nooks within the terrain.

For all of these cases, the in situ examination by a diver that looks into all nooks and crannies remains inevitable. This is an intrinsically time-consuming feat if these particular organisms are to be described or quantified. So, there is no alternative, however, the above-mentioned activities are highly efficient with regards to time spent in the water and can be executed by a single diver freeing up the second diver, specifically for the small, cryptic and highly camouflaged species.

Proposed protocol

Based on the adjustments above and the experiences during the benthic surveys at the "Islote El Pelado", the following survey protocol for documenting benthic invertebrate communities is proposed. The protocol is designed for a minimum of two divers, yet it is recommended that three divers are involved.

Step 1. Exploratory dives / snorkels in the study area

To gain a general understanding of the topography, bathymetry and benthic composition of a particular site, reconnaissance dives are necessary. Considering depths of no more than 15 m, it is suggested that at least one SCUBA dive should be planned for every 4500 m^2 to be examined. Approximate estimates can easily be obtained using measurement tools available in Google Earth.

Snorkeling may allow for larger areas to be covered for an initial appraisal.

Step 2. Site selection

Sites are commonly selected in a stratified manner (Lang *et al*. 2010). Based on step 1, sites of interest (for detailed surveys) within the study area can be randomly selected, again the specific objectives of the study need to be adhered to. In the present study, sites with representative (abiotic and biotic characteristics, *e.g.* substrate type, inclination) benthic cover were selected.

Step 3. Placement of transect

Staring points of transects are haphazardly selected within the ecological zone examined. The transect line is to be rolled out in such a manner as to incur minimal changes in depth between the starting and ending point. While unreeling the transect divers should ensure that line has sufficient slack to properly line the contour of the seafloor. This is best done by 2 divers, one that unreels the line and the second that ensures a linear and adhering placement along de seafloor. If a third diver is available, he/she could be to specifically be in charge of unreeling and reeling in the transect line, as this does inquire some time effort. Meanwhile others can already start surveying the transect, especially the person with the most time consuming task, namely the assessment of small and cryptic organisms.

Step 4. Photo-transect (substrate types / sessile organisms)

Diver 1 places the quatripod at every meter on alternate sides of along the transect line, and records an image of the seafloor. For example, at 0, 1 and 3 m etc. on the right and 2, 4, and 6 m etc. on the left (Figure 8). The number and position of images to be recorded per transect may be varied depending on the specific needs of the project at hand. Examples of common substrate types at EPP were: Encrusting Calcareous Corallinacea (ECC), Balanomorph Barnacles (with ECC), filamentous turf algae, filamentous turf algae with sediment accumulations, and shill. It should be noted that bare rock per se didn't exist as substrate type, as all stable surfaces were covered my epileptic growth.

Step 5. Video-transect macro invertebrates

After completing the photo-transect from the 0 m to the 20 m mark (Step 4), Diver 1 removes the camera from the quadripod and swims from 20 m towards 0 m along one side of the transect (*i.e*. right side) recording and covering 1 m to the right of the transect. (Figure 8). In calm waters, with moderate visibility up to 5 m, the 20 m swim should take no less than 2 minutes. In more turbulent waters, and poor visibility (1-3 m), the swim speed needs to be reduced, and intermitted close-ups of individual species are necessary to allow for their later on screen identification. In the event of periodic wave sets, it is recommended to interrupt the video during the onset of such sets and to continue the recording when the set has passed. It is important to always record the beginning and end of each transect line, to later be able to properly place and label the transects when video files are saved. Once the first 20 m have been recorded, Diver 1 returns from 0 m toward 20 m while recording the area within 1 m of the opposite side of the transect line. When this transect is completed, Diver 1 has completed his/her part in the transect. He /she may proceed to a second transect if already delayed, or utilize the time to record detailed images of organisms, to gradually build an image cumulative depository of the organisms encountered.

Step 6. Cryptic and camouflaged invertebrates

At the point when Diver 1 initiates his surveys (step 4), diver 2 commences the in-situ search and quantification of small, hidden or camouflaged invertebrates. It may be advisable that Divers 1 and 2 start on opposite ends of the transect. Diver 2 should also survey one side of the transect first and then the other. It is inevitable that Divers 1 and 2 cross each other's paths, but this does not represent a hindrance as long as it is agreed upon how to act and react in that situation.

For the quantification of organisms that generally grow in clusters in hundreds (ahermatypic Scleractinia) an estimated needs to be considered, defined and then applied.

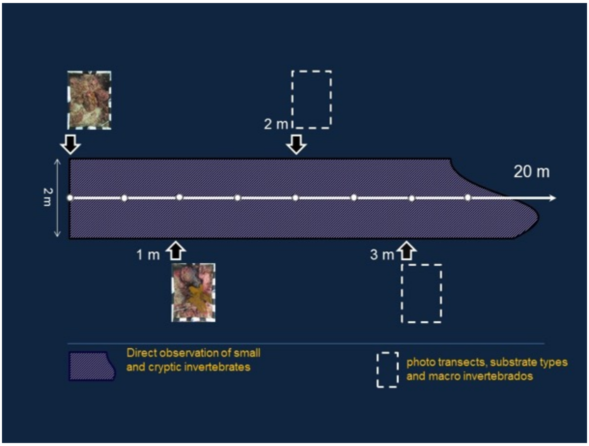


Figure 8. Spatial alignment of in situ, video and Photo transects. (Change this for BW image).

Transect replicates

In-depth up to 15 m, four transects 20 m could be surveyed by 2 - 3 divers as described above. The necessary number of transects that adequately capture the representatives species composition at the study site will have to be determined on the base of the studies objectives and support on literature sources.

Species selection for Protocol

We suggest that a reduced number of species from the groups described in this document (Cnidaria, Mollusca, and Echinodermata) are to be selected for monitoring proposed (see below). To do so experts in each of the invertebrate groups are to be consulted to determine which species are best fit for monitoring purposes when only a few per group are to be selected.

Data recording

While photos and videos transects are recorded in a digital format during the survey, data from the cryptic transects must first be noted on slates. For the latter, it is recommended that the slates are prepared in a tabular manner that allows a quick record of the organisms identified. Photocopies or manual copies are to be created of these initial records and stored as evidence. Later digital version must be generated to allow subsequent analyses. All digital files (photo and video files) are to be copied and stored in at least 2 separate storing devices.

Substrate composition per image (photo transect) may be evaluated using CPCe (Kohler and Gill, 2006). The quantification of individual substrate types can be done using the software, or by using the software partially and recording data in separate sheet. Make sure to cite all software used properly in any document or publication resulting from the analysis. The following tables were used to document and quantify the field data (Figure 9).

Benthic Survey Data Sheet SUBSTRATES, El Pelado, Santa Elena, ECUADOR

Date: Area Name: Site Name:

Recorder(s):

	Image Number																			
TRANSECT																				
FOTO/FRAME	F1	F2	F3	F4	F5	F6	F7	F8	F9	F10	F11	F12	F13	F14	F15	F16	F17	F18	F19	F20
CATEGORY																				
Encrusting Corallinacea																				
Corallinacea over Cirripedia																				
Filament. algae w sediments																				
Filament. algae on rock																				
Pebbles																				
Sediment																				
Shill / conchilla																				
Rubble																				
Organims																				

Figure 9. Photo transects: Composition of substrates, bases on 10 random points per frame (F) for each of the 20 images per transect were recorded in the following format.

Substrate data was subsequently presents compiled to a single value of each category per transect and incorporated into a single sheet with the in-situ quantifications of individual species, and those derived from video transects as follows (Figure 10):

Benthic Survey Data Compilha, ECUADOR

Date:

Recorders:

SITE AND TRANSECT	2T3	2T4	3AT1	3AT2	3AT3	3AT4	3BT1	3BT2	3BT3	3BT4	5T1	5T2	5T3	5T4	Cont.
SUBSTRATE CATEGORIES															One column per transect
1															One row per species
2															
3 ...															
SPECIES															
1															
2															
3 ...															

Figure 10. Compilation of quantification of substrate categories and invertebrate species per transect.

Substrate-habitat types

For those who have explored tropical rocky environments or coral reefs in the Western Pacific or the Caribbean, El Pelado's benthic communities were architecturally shaped by its rocky topography rather than the presence of large colonial sessile invertebrates such as barrel and pipe sponges or fire corals and stony corals.

With a few exceptions (Alcyonacea), sessile invertebrates were small and did not “shape” the seafloor architecturally other than in an encrusting manner. Large fleshy marcroalgae, such as *Sargassum*, which do occur along the rocky coast, were also absent. In fact, at a first glance, El Pelado and its surrounding rocky mounds are all but barren. Pelado means “peeled” and refers to the unvegetated state of the islet. Yet colloquially, this term is also applied to the barren appearance of the rocky seafloor.

With regards to benthic organisms, one pivotal ecological aspect of the islet and its surrounding rocky reefs is the absence of bare rock. Stable surfaces are covered by various groups of algae and/or invertebrates. Spatial competition can be regarded as one of the key biological facets that have shaped the presence and distribution of benthic organisms in general and sessile species in particular (Ferguson *et al*. 2013). In such scenarios, bioerosion by abrasive feeding mechanisms, (*i.e*. of herbivorous snails, sea urchins and a variety of fishes such as Scaridae) and bio-corrosion, (*i.e*. by boring sponges and bivalves) are part of the processes opening new settlement spaces.

The western slopes and vertical walls on the rocky mounds of the El Pelado, display narrow, or more compressed, bands of the habitat types described for the easterly-facing slopes. However, the turf zone, characteristic for the leeward slopes, is generally absent on the western drop offs.

East facing slopes

Among the rocky substrates on the eastward facing (gently inclined) terra in sup to 25 m depth, three principal substrate-habitat categories can be discerned based on substrate morphology and the presence of sessile organisms, and they are here referred to as the Balanomorpha, Turf, and the Muricea Zones. Their defining features are summarized in (Appendix 4).

Planar and slightly inclined rocky terrains (Substrate-Habitat Type A, Balanomorpha Zone):

The common tidal and wave amplitudes at El Pelado expose areas in 1-10 m depth to high turbulence regimes. Erect colonies of invertebrates, such as the Alcyonacea - otherwise characteristic of the area, cannot withstand the mechanical impact from the surge and so these sections are also devoid of sediment or rubble deposits. Smaller organisms such as the encrusting balanomorph Cirripedia (barnacles) have in turn colonized extensive areas and created a calcareous veneer covering vast sections of the rocky substrate. Such “secondary hard substrates” in turn are colonized by encrusting calcareous corallinacea and serve as settlement base for other sessile organisms. In the immediate vicinity of aggregations of the seas urchin *Diadema mexicanum*, a nocturnal grazer, the calcareous veneers appear particularly “clean”, whereas filamentous algal turfs are common in the absence of the urchin aggregations. The benthic cover of filamentous turf algae increased in the deeper quarter of this zone where *D. mexicanum* was less common.

Colloquially, this habitat type is often referred to as “planchón” by fishermen who focus on collecting or catching invertebrates. However, the term “planchón” is synonymously applied to the easterly facing and gently inclined planes, whose surfaces are covered by filamentous turf algae rather than barnacles (see Turf Zone below). Planchón thus refers to planar topography, rather than a particular type of organism cover, although it does imply that large erect sessile organisms protruding into the water column, such as gorgonians, are not characteristic of the area in question.

Coral reefs, another type of secondary hard substrate, are absent at El Pelado. Yet within the Balanomorpha Zone, patchy assemblages of *Pocillopora* spp. are present on the easterly slopes of the islet and its northwesterly and southwesterly neighboring rocky mounds. The most extensive formations are found in the eastern (protected) areas of the islet El Pelado and particularly characterizes sites 5 and 7.

Planar and slightly inclined rocky terrains with sediment accumulation by algae (Substrate-Habitat Type B, Turf Zone):

In the EPP aggregations of *D. mexicanum*, were not found at depths greater than seven meters at EPP. Homogeneous planes of filamentous algal turfs generally commenced at the lower distribution limit of such aggregations and extend to depths of approximately 12 m. These algal mats act as sediment traps and have formed a 5-10 cm thick algal-sediment layer covering the rocky base. Large sessile invertebrates are uncommon in this type of habitat. These points towards settlement limitations for the larvae of sessile invertebrates within this habitat type.

Structurally complex rocky environments (Substrate-Habitat Type C, Muricea Zone):

East ward facing slopes between 10 and 25 m depth were structurally more heterogeneous. They were comprised of large angular slabs of fragmented strata and rounded boulders, and harbored rubble and sediment accumulations in depressions within the terrain. Rounded rocks transported from shallower depths lined the transitional area between the base of the rocky reefs and the surrounding sediment seafloor. Alcyonacea were the most conspicuous group of sessile organisms that compartmentalize the water column in the immediate vicinity of the seafloor. One genus in particular, *Muricea* with several color morphotypes of *M. plantaginea*, outnumbered all other octocorals throughout the EPP.

The highly convoluted structural environment provides a myriad of gradients in light levels and exposures to water currents. It is not surprising therefore that this is the section in which we have found most of the invertebrates listed in Appendix 7. Rocky surfaces harbor diverse communities of primarily filamentous and encrusting calcareous algae, and sessile invertebrates. In addition, the accumulated rubble and sediments (primarily mollusk and echinoderm shell fragments) offer a complex interstitial space for cryptic and interstitial animals (meiofauna).

As a result of the structural complexity, shade-loving (sciaphile) sessile organisms find suitable conditions for settlement. Among the conspicuous stony corals in the area were ahermatypic (non-reef building) species. The shallow end of this zone is where most *Spondylus* were found, possibly indicating a preferential habitat.

Two dome shaped rocky mounds (Sites 3 and 6) do not come close to the surface. Their apical portions lie in waters deeper than 10 m depth. Structurally and ecologically these mounds can be described as falling into the Muricea Zone, and they are also quite diverse with regards to Alcyonacea and ahermatypic Scleractinia.

Sandy seafloor surrounding the rocky mounds (Substrate-Habitat Type D):

Sandy environments were not examined in this study. However, from the reconnaissance dives it can be summarized that the sediments in the immediate surroundings of the rocky mounts are calcareous and predominantly composed of mollusk. No epipsammic macro algae (settling on sandy substrates) were seen, pointing at bioturbation and predation among the ecological limitations within euphotic depths.

Near shore rocky environments with algal-zoantharian cover (Substrate-Habitat Type E, Zoantharia Zone):

The rocky environments near Punta Delteco, where heavily overgrown by combination of filamentous algae and zoanthids, forming extensive mats in approximately 3 to 6 m depth. Together they constitute the dominant epibenthic growth, leaving little space for other sessile organisms. While filamentous algal mats can be observed into the breaker zone, zoanthids do not extend into this mechanically turbulent environment.

Selected invertebrates of continental Ecuador

Taking the mentioned methodological aspects into account, the four groups of conspicuous invertebrates (Alcyonaria, Scleractinia, Gastropoda and Echinodermata) lend themselves to be incorporated into a new monitoring protocol addressing epibenthic communities in the region. The species recorded possessed robust external and macroscopic traits that allow their identification. Combined in a monitoring protocol they represent a cross section of invertebrates with multiple substrates and feeding affinities. Approximately 80 invertebrate species were registered (Appendix 7) at the EPP.

Two of the groups were sessile cnidarian belong to the anthozoan, Orders Alcyonacea and Scleractinia, and two were mollusks (including gastropods and bivalves) and echinoderms (Asteroidea, Echinoidea).

The Alcyonaria are evidently well adapted to the local conditions. Consequently, affliction such a to certain diseases (Etnoyer *et al*. 2016) parasitic epibionts (Bo *et al*. 2012) or the absence or presence of mobile commensals as crustaceans and brittle stars (ophiuroids) are useful indicators on the health of this particular group. Coupled with the distribution of size classes the colony recruitment or reproductive success of this group could be monitored. However, octocorals remain a poorly understood group of cnidarians in terms of taxonomy and phylogeny (McFadden *et al*. 2010). Comprehensive distribution data as obtainable by the protocol applied here may render additional insights, particularly for the eastern Pacific.

Within the Alcyonacea, the Plexauridae were highly characteristic for the EPP in general and for the Muricea Zone in particular.

Gorgonidae were far less representative, but their external defining features were conducive to an in-situ identification. In addition, a single representative Clavularidae, namely the invasive *Carijoa riseii* (Duchassaing & Michelotti, 1860), was also abundant, particularly in waters up to 10 m depth.

Colonial hermatypic Scleractinia are a common component of benthic monitoring protocols in tropical settings (Lang *et al*. 2010, Hodgeson *et al*. 2006). As an important component of coral reefs their status is one indicator used to evaluate the health of coral reefs, and in consequence the diversity they harbor.

Among the Scleractinia, ahermatypic species had a wider distribution than hermatypic species. In fact, hermatypic corals had a spotty distribution, limited to leeward sites and included a single assemblage of approximately 10 colonies of the massive species Pavona clavus, and several clusters of ramous stony corals of at least two Pocillopora spp. At the EPP reef corals are not abundant and have not formed coral reefs sensu stricto. However, the ahermatypic Scleractinia are abundant. While they do not render themselves for monitoring studies of coral bleaching episodes, as they are azooxanthellate, they may serve as calcareous representative of stony coral in studies addressing ocean acidification and its impact on calcareous organisms. However, some species lack distinctive macroscopic traits that allow an in situ identification and therefore a few selected species may be incorporated into a monitoring protocol for a broad range of users.

Gastropods and bivalves were generally heavily overgrown and thus difficult to see and identify in situ. A few species were regularly detected and are thus considered herein. Mollusks have historically been important in Ecuador as food source, but the shells have been an essential material artefacts created for social, commercial or religious purposes. Currently, studies on cultivation of several bivalve species are underway, as are evaluations of possible sanctuaries for the most threatened representatives. The incorporation of these species into a survey protocol, with the inclusion of experienced fishermen, promises to render vital information on the current stock of these species in the wild.

An organism of commercial importance is *Spondylus*. Although modern fishing of spondylus started around the end of 1970s, most natural populations are overfished and local populations have been depleted in many regions. They are now subjected to special regulations for their harvesting, in particular in Mexico and Ecuador (Lodeiros *et al*. 2016). In 2009 by petition of the Salango community, Manabí Province, the subsecretary of fishery resources under the Ministry of Agriculture, Livestock, Aquaculture and Fisheries (MAGAP), decreed a fishing closure of Spondylus spp. (Acuerdo Ministerial 136) and banned their harvesting, transport, and consumption (Viceministerio de Acuacultura y Pesca 2009).

Lastly, most asteroids and echinoids encountered were easily identifiable in situ. The easy identification and the comparatives distributions of echinoderms at El Pelado make them good candidates for a regional monitoring protocol. Stratified bathymetric distribution (Sonnenholzner *et al*. 2013) may be a useful feature in monitoring the influence of temperature anomalies on mobile benthic communities. The mentioned species also encompasses grazers, corallivores and predators that are part of different trophic levels.

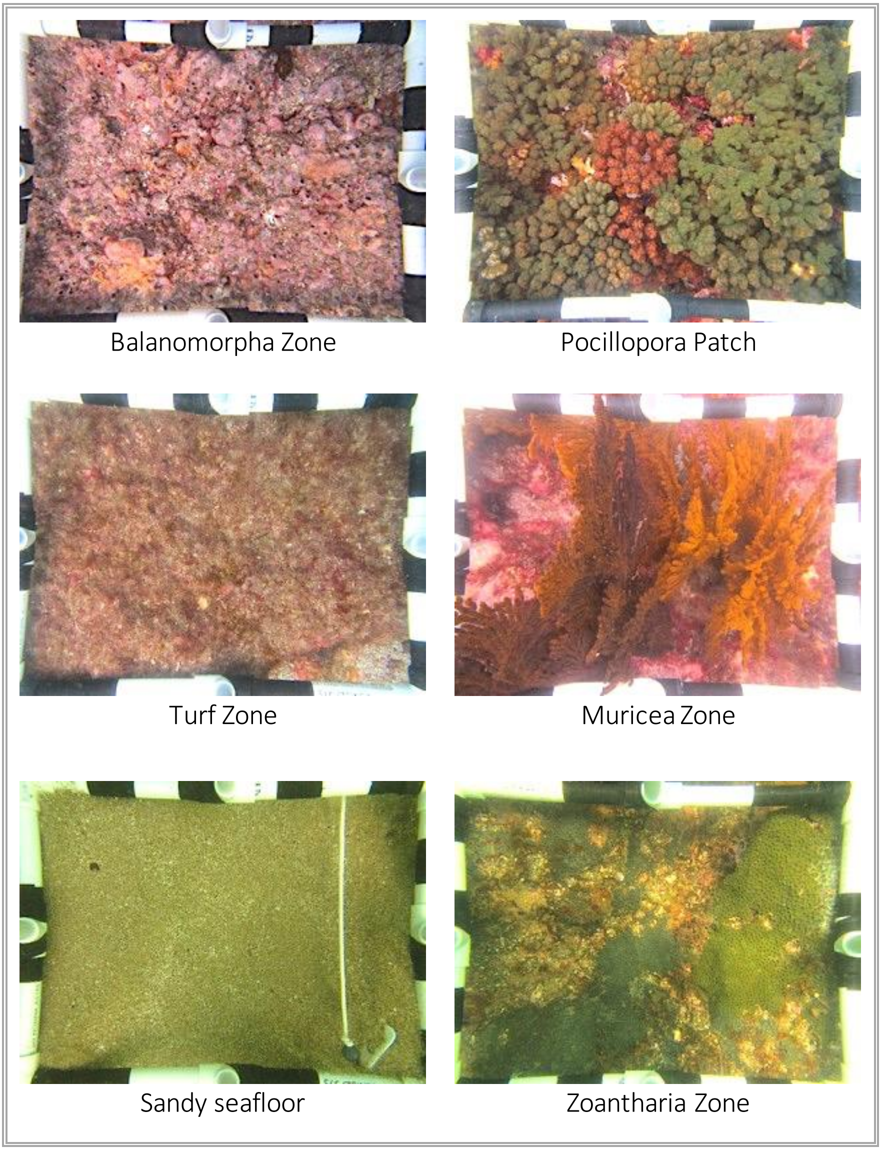


Plate 1. Substrate details covering benthos plots of 40 x 30 cm.

Pacifigorgia firma

Pacifigorgia irene

Pacifigorgia rubicunda

Heterogorgia hickmani

Leptogorgia alba

Leptogorgia pumila

Plate 2. Alcyonacea 1.

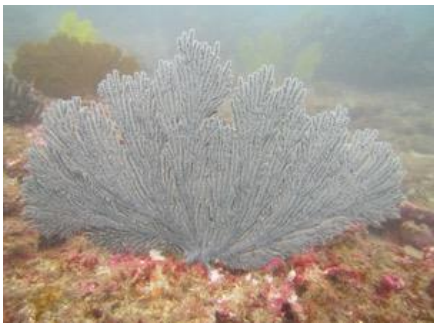
Leptogorgia taboguilla

Muricea austera

Muricea fruticosa

Muricea plantaginea

Muricea plantaginea



Muricea plantaginea

Plate 3. Alcyonacea 2.

Muricea purpurea

Muricea squarrosa

Muricea aspera

Psammogorgia arbuscula

Unidentified

Carijoa riisei

Plate 4. Alcyonacea 3.

Tubastraea coccinea

Tubastraea coccinea

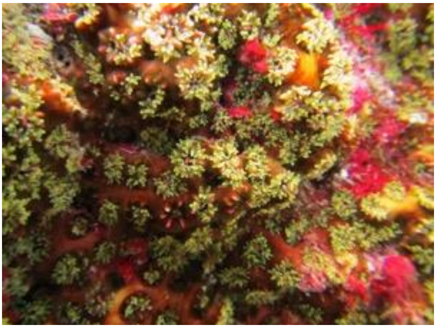
Cladopsammia eguchi

Cladopsammia eguchi

Cladopsammia gracilis

Cladopsammia gracilis

Plate 5. Scleractinia. Ahermatipic corals 1.



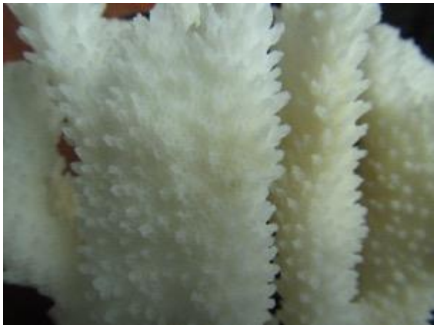
Phyllangia consagensis *Phyllangia consagensis*

Astrangia browni *Astrangia browni*

Oulangia bradleyi *Oulangia bradleyi*

Plate 6. Scleractinia. Ahermatipic corals 2.

Pavona clavus



Pocillopora capitata

Pocillopora eydouxy

Pocillopora verrucosa

Plate 7. Scleractinia. Hermatipic corals 1.

Columbella major

Conus brunneus

Pustulatirus sanguineos

Latirus sp.

Opeatostoma pseudodon

Hexaplex princeps

Plate 8. Gastropoda 1.

Hexaplex radix

Vasula melones

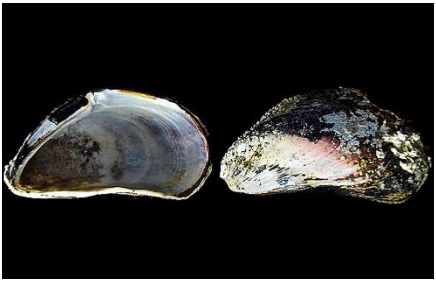
Vasula speciosa

Vasum caestus

Plate 9. Gastropoda 2.

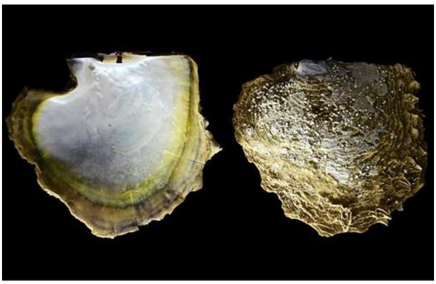
Barbatia reeveana

Chama buddiana

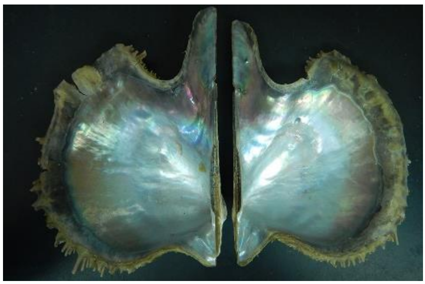


Modiolus capax

Hyotissa hyotis



Pinctada mazatlanica



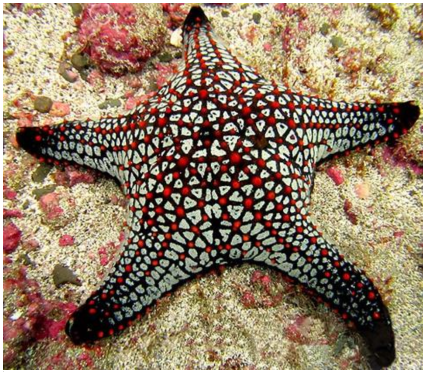
Pteria sterna

Plate 10. Bivalvia 1.

Pharia pyramidata

Phataria unifascialis

Nidorellia armata

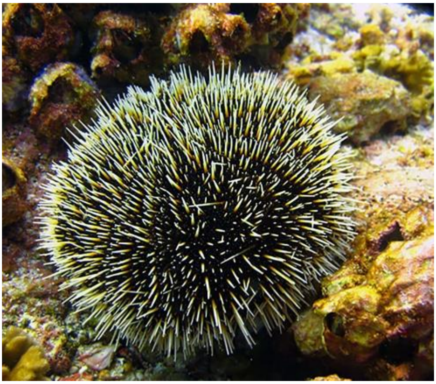


Pentacerastus cumingii

Echinometra vanbrunti

Toxopneustes roseus

Plate 11. Echinodermata 1.



Tripneustes depressus

Lytechinus semituberculatus

Diadema mexicanum

Astropyga pulvinata

Eucidaris thouarsii

Plate 12. Echinodermata 2.

Cucumaria flamma

Isostichopus fuscus

Plate 13. Echinodermata 3.

APPLICATION

Monitoring with reduced species list

The protocol can be applied in monitoring projects, focusing on the detection of changes in species composition or in the incidence of certain afflictions of the organisms considered. For monitoring purposes, it is suggested to utilize a reduced list of species with clear external traits that allow the identification of the species. This facilitates the training process of skilled divers assigned to such a task.

General inventory, complemented by random swims (larger species list)

Another application of the protocol is in combination with a general inventory of species. This can best be accomplished by random swims across a variety of habitats, during which all species found are recorded. Such surveys can also be evaluated semi-quantitatively giving the seen species certain abundance ranks" (*e.g.* common, occasional, rare). In addition, a selected list of species may be applied for a more accurate quantitative survey, using belt transects as described above.

Facilitating access to findings

The benefits of centralized information points like a website or GIS system, include the high access of the information presented and an interpretation of the information so that it is understandable and valuable to a broad audience. This website should be considered as a complementary tool for future research. For example, CENAIM used the WEB GIS platform to create a website with all the information obtained from the biodiversity inventory. (http://www.cenaim.espol.edu.ec/es/node/453). With this free inventory, students and professionals can complement their research with the information located on WEB GIS, such as dive sites, depths, and organisms.

CONCLUSION

Geographical approach to understanding benthic communities

The search and study of bathymetric maps and source materials proved to beneficial to understanding the topography and orientation of the seafloor surrounding the El Pelado islet. Through this study, we are now aware of the bathymetry of the Pelado area and the profile of the different dive sites.

Adjusting survey methods to local conditions

In situ surveys of benthic invertebrates are inherently time consuming and linked to various degrees of logistical and financial constraints. No one survey protocol fits every objective, and each geographic location brings its particular challenges. In the rocky environments of continental Ecuador, characterized by poor visibilities and turbulent sea, the described protocol approach proved to be most effective to describe marine invertebrate communities. The use of weighted transect lines rendered much needed "stability" of the transects under the commonly turbulent conditions. This is recommended for studies with similar objectives in the region, as its construction requires nominal investments. The same would apply to the use of weighted quadrats.

The most cost intensive acquisition, besides SCUBA equipment, is digital underwater cameras, yet these have become readily available and increasingly easy to handle by nonprofessionals. Photo-transects were a highly effective way to record substrate types while video-transects facilitated time-efficient records of the distribution of large sessile and mobile invertebrates. Both photo- and video-transect data can be stored for later and repeated analysis without the time constraints inherent of in situ quantifications of SCUBA. Therefore, the development of a digital image recording protocol for recording benthic composition, at least of larger organisms, is highly recommended, as costs for boats, SCUBA equipment and weather conditions are among the principal limitations in studies of this nature.

However, highly camouflaged organisms and small or cryptic invertebrates are still best identified in situ by skilled divers. By applying digital imagery survey as described above, a lot of survey time can be freed up for the more time consuming in-field assessments.

Nadon and Stirling (2005) comprehensively reviewed the benefits and limitations of commonly used techniques in the quantitative characterization of coral reefs and provide a sound starting point in deciding which method best suits the needs of a particular project. While rocky benthic habitats of continental Ecuador are rarely covered by coral reefs (Glynn 2003), the assessment of its sessile benthic invertebrate communities follow similar principles. The combination of various methods of measuring or estimating the community structure of benthic organisms is generally referred to as survey protocols. Such protocols address differing aspects of marine habitats, such as biodiversity, the condition of particular species (*i.e*. Atlantic and Gulf Rapid Reef Assessment, Reef Check), as well as, productivity (Caribbean Coastal Marine Productivity CARICOMP, 2001). Details of the mentioned protocols are found in Lang *et al*. 2010, Hodgeson *et al*. 2006 and CARICIMP (2001) respectively.

Field surveys on benthic invertebrate communities along the continental coast of Ecuador has so far focused on particular regions, similar to the current study, to document species richness and the community structures. Cumulatively, the knowledge base on these communities is growing but remains disconnected. What has been missing is a cost-encompassing monitoring initiative to evaluate the health of different sites using the same technique. Furthermore, substrate-habitat characteristics have been disregarded as essential component of community structure.

Therefore, considering the way in which this methodology fit with the Ecuadorean oceanographic conditions and the good results we obtained with this research, this protocol represents a perfect methodology to be applied all along the continental coast of Ecuador.

RECOMMENDATIONS

However, the specific objectives of a study, the environmental conditions at the survey locations, as well as logistic and financial limitations will shape the methodology applied.

Geographical approach to understanding benthic communities

The presence and distribution of habitat-substrate types could also be interpreted in combination with the structural characteristics of the study area and its exposure to tidal and wave regimes. The relation between substrate quality and exposure to oceanographic parameters on one hand, and the distribution of algae and invertebrates on the other hand, has been demonstrated in numerous studies from tropical to temperate regions (Diaz *et al*. 2004).

It is, therefore, recommended that a concerted effort is made to understand the geographic features of an area in which qualitative and quantitative assessments of benthic communities are undertaken. In regions that have been extensively explored this procedure may not incur excessive efforts, yet in poorly studied areas it may be a challenging logistical and financial undertaking that should be considered in the planning phase of such projects.

For example, the equatorial front (surface currents) divides the coast of Ecuador into a northern and southern region, which exposes the coast to distinct seasonal fluctuations, in temperature and exposures to the ENSO (Glynn 2003). Monitoring initiatives on either side of the equatorial front may allow valuable insights into differences in the reaction and adaptation of the respective invertebrate communities.

Adjusting survey methods to local conditions

The use of weighted transect lines rendered much needed "stability" of the transect under the commonly turbulent conditions and is recommended for studies with similar objectives in the region, as its construction requires nominal investments. The same would apply to the use of weighted quadrats. While PVC measuring tapes are quickly deployed, recoiled and commonly used as transect lines, they do not contour the seafloor and are highly instable in turbulent conditions.

Thanks to the ready availability of SCUBA equipment, researchers have been able expand in situ observations beyond the intertidal zone. Nonetheless, actual observation times at depths greater than five meters remain relatively brief and the logistical effort incurred is a significant limiting factor in many regions of the world. Selecting the proper field survey method in order to characterize sublittoral epibenthic marine communities is a pivotal and often challenging feat.

A monitoring protocol for selected invertebrates of continental Ecuador

Balanomorph cirripeds are among the most common encrusting invertebrates found at El Pelado area. In this study, we only recorded the group as a substrate component and haven't examined whether the veneer sheets that they construct are composed of a single or multiple barnacle species. Given their structural significance in the makeup of the Balanomorpha Zone, their documented role as bio-indicators and invasive species (*i.e*. Carlton *et al*. 2011, Dionísio *et al*. 2013), a closer look at this group and their incorporation into monitoring protocols seems pertinent.

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MAPS AND NAUTICAL CHARTS

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IOA 105111, WGS-84 (1:7500) for Bahía de Ayangue

National Geological Map of Ecuador, (1982), Scale 1:1,000,000 2nd Approximation

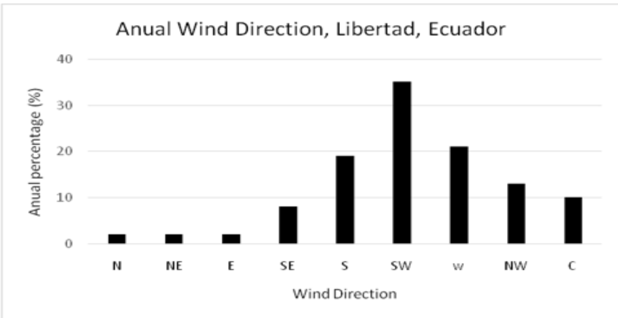
INTERNET RESOURCES

Government of Ecuador

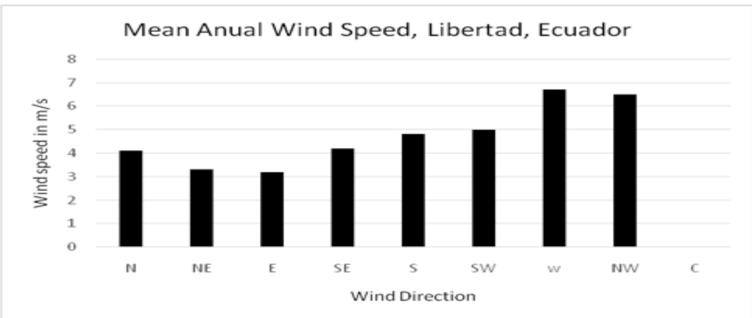
http://areasprotegidas.ambiente.gob.ec/areas-protegidas/reserva-marina-el-pelado), viewed 23 February 2016

APPENDIX

Appendix 1. Annual wind direction, at Libertad, Santa Elena, adapted from Gálves and Regalado's (2007) and their analysis of wind data from 1948 to 2006.



Appendix 2. Annual wind speed at Libertad, Santa Elena, adapted from Gálves and Regalado's (2007) and their analysis of wind data from 1948 to 2006.



Appendix 3. Rocky mounds and isled on the EPP as labeled in Figure 2, with coordinates and common names.

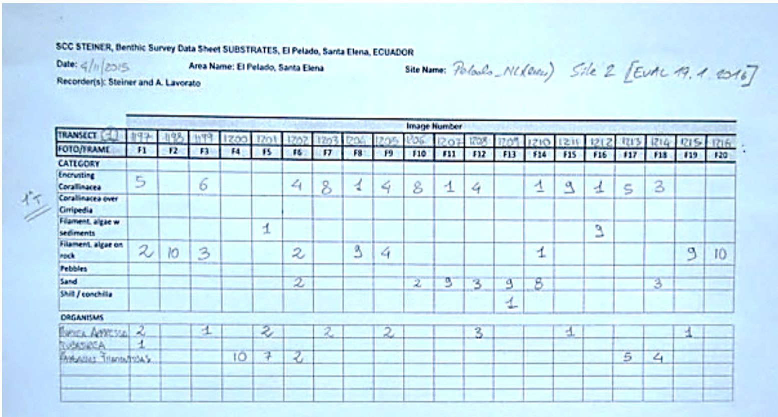
Rocky Mounds on EPP	Survey Site	Coordinates	Common Name
NW	1	1°55’58.25’’S 80°47’32.83’’W	La Pared
NW	2	1°56’01.35’’S 80°47’31.94’’W	El Planchon
NW/E?	3	1°55’58.25’’S 80°47’32.83’’W	La viejita
NW	3A	1°55’56.86’’S 80°47’30.46’’W	None
NW	3B	1°55’58.25’’S 80°47’32.83’’W	None
Islet SW	~~4~~	1°56’07.72’’S 80°47’22.80’’W	El Laberinto
Islet North	5	1°56’00.59’’S 80°47’19.35’’W	Rabo del Viejo
NE	6	1°55’53.10’’S 80°47’14.25’’W	Bajo San Ignacio
Islet East	7	1°56’05.20’’S 80°47’19.63’’W	Zona Protegida
Islet South	8	1°56’10.20’’S 80°47’20.12’’W	El Acuario
SE	9	1°56’19’’S 80°47’13’’W	El Cuarenta
SE	10	1°55’58.25’’S 80°47’32.83’’W	El Cuarenta / La reina
A. P Depteco	11	1°58’57.82’’S 80°45’39.16’’W	Punta Delteco
A. P Depteco	12	1°59’01.84’’S 80°45’37.10’’	Punta Delteco

Appendix 4. Substrate Habitat types on the easterly slopes from the El Pelado Platform and near Ayangue*.

Type	**Depth range (m)**	**Terrain morphology**	**Structural complexity**	**Common encrusting organisms and/or secondary hard substrates**	**Common sessile invertebrates**	**Name**
A	1-10	planar, horizontal or inclined, rocky, without sediment and rubble accumulations	low	Balanomprph Cirriedia, Corallinacaea	*Megabalanus* sp.	Balanus Zone
(A1)	3-10	planar, horizontal or inclined, rocky, with	moderate	Corallinacaea	*Pocillopora*	Pocillopora Zone

		sediment and rubble accumulations				
B	5-12	planar, horizontal or inclined, rocky, with sediment and rubble accumulations	low	Corallinacaea, turf algae	n/a	Turf Zone
C	10-25	convoluted, rocky, angular slabs and rounded boulders with sediment and rubble accumulations	high	Corallinacaea,	*Muricea* spp. *Tubastraea* spp.	Muricea Zone
D	8-25	soft sediments, primality mollusk and echinoderm shell fragments	low	n/a	n/a	Sandy Substrate
E*	3-7	convoluted, along-shore deposits of rocks and boulders without sediment and accumulations	moderate	Zoantharia, Actinaria	Zoantharia, Actinaria	Zoantharia Zone

Appendix 5. Example of Substrate table, bases on 10 random points per frame (F) for each of the 20 images per transect were recorded in the following format.



Appendix 6. Example of compilation of quantification of substrate categories and invertebrate species per transect.

	A	B	C	D	E	F	G	H	I	J	K	L	M
7	SITE AND TRANSECT	1T1	1T2	1T3	1T4	2T1	2T2	2T3	2T4	3AT1	3AT2	3AT3	3AT4
8													
9	DEPTH in m	12,8	12,8	14,6	14,6	15,2	15,2	13,4	13,4	10,0	10,0	12,1	12,1
10													
11													
12	SUBSTRATE CATEGORY /200pts												
13	CCA Crustose Coraline Algae	27	79	57	50	60	36	74	17	45	19	94	64
14	CCA over balanom. Cirripedia	0	12	0	107	0	2	105	19	23	0	0	2
15	FTA-S Fil Turf Algae on sed	23	33	8	0	10	31	0	16	45	84	12	22
16	TTA-R Fil Turf Algae on rock	81	55	68	10	50	91	11	66	52	59	51	63
17	Pebbles	14	2	6	1	0	0	0	0	0	3	0	0
18	Sediment	30	0	27	10	36	28	2	43	17	5	8	22
19	Shill	11	0	0	2	1	0	0	25	3	23	14	8
20	Org / Invert	14	19	34	20	43	12	8	14	15	7	21	19
21	ECHINODERMATA												
22	Eucidaris_thouarsii	38	12	31	8	31	32	15	18	5	4	18	0
23	Centrostephanus_(coronatus)	6	2	3	1	2	0	0	3	2	0	1	0
24	Diadema_mexicanum	0	19	10	22	3	14	274	21	2	6	0	3
25	Astropyga_pulvinata		0	0	0	0	0	0	0	0	0	0	0
26	Echinometra_vanbrunti	190	140	260	245	163	155	135	150	217	227	70	107
27	Toxopneustes roseus	1	0	0	0	0	0	0	0	0	0	0	0
28	Pentaceraster_cumingi	0	0	0	0	0	0	0	0	0	0	0	0
29	Pharia_pyramidata	1	5	3	2	3	5	8	3	1	8	5	3
30	Phataria_unifsciales	24	3	39	23	23	9	7	15	32	39	15	6
31	Cucumaria_flamma	8	6	12	7	2	2	2	3	7	4	2	1
32	Isostichopus_fuscus	0	0	1	0	2	1	1	1	1	0	1	1
33													

Appendix 7. Organisms observed during the study.

PHYLUM CNIDARIA

Class Anthozoa
Order Actinaria
Family Actiniaria
Bunodosoma grande (Verrill, 1869)
Order Alcyonacea
Family Clavulariidae
Carijoa riisei (Duchassaing & Michelotti, 1860)
Family Gorgoniidae
Pacifigorgia firma Breedy & Guzman, 2003
Pacifigorgia irene Bayer, 1951
Pacifigorgia rubicunda Breedy & Guzman, 2003
Leptogorgia alba (Duchassaing & Michelotti, 1860)
Leptogorgia pumila (Verril, 1868)
Leptogorgia taboguilla (Hickson, 1928)
Family Plexauridae
Heterogorgia hickmani Breedy & Guzman, 2005
Muricea austera Verrill, 1869
Muricea fruticosa Verrill, 1869
Muricea plantaginea (Valenciennes, 1846)
Muricea purpurea Verrill, 1864
Muricea squarrosa Verrill, 1869
Psammogorgia sp
Order Antipatharia
Family Antipathidae
Antipathes galapagensis Deichmann, 1941
Family Myriopathidae
Myriopathes panamensis (Verrill, 1869)
Order Scleractinia
Family Dendrophylliidae
Cladopsammia eguchii (Wells, 1982)
Cladopsammia gracilis (Milne Edwards & Haime, 1848)
Tubastraea coccinea Lesson, 1829
Family Rhizangiidae
Astrangia browni Palmer, 1928
Oulangia bradleyi (Verrill, 1866)

Family Caryophylliidae

Phyllangia consagensis (Durham & Barnard, 1952)

Family Pocilloporidae

Pocillopora capitata Verrill, 1864

Pocillopora damicornis (Linnaeus, 1758)

Pocillopora eydouxi Milne Edwards, 1860

Pocillopora verrucosa (Ellis & Solander, 1786)

PHYLUM MOLLUSCA

Class Bivalvia

Order Arcida

Family Arcidae

Arca cf pacifica (G.B. Sowerby I, 1833)

Barbatia reeveana (D'Orbigny, 1846)

Order Pectinida

Family Anomiidae

Pododesmus foliatus (Broderip, 1834)

Family Spondylidae

Spondylus limbatus G. B. Sowerby II, 1847

Superorder Imparidentia

Family Chamidae

Chama buddiana C.B. Adams, 1852

Order Mytilida

Family Mytilidae

Modiolus capax (Conrad, 1837)

Order Ostreida

Family Gryphaeidae

Hyotissa hyotis (Linnaeus, 1758)

Family Pteriidae

Pinctada mazatlanica (Hanley, 1856)

Pteria sterna (Gould, 1851)

Order Venerida

Family Veneridae

Periglypta multicostata (G.B.Sowerby I, 1835)

Class Gastropoda

Order Neogastropoda

Family Columbellidae

Columbella major G.B. Sowerby I, 1832

Family Conidae

Conus brunneus W. Wood, 1828

Family Fasciolariidae

Pustulatirus sanguineus (Wood, 1828)

Latirus sp.

Opeatostoma pseudodon (Burrow, 1815)

Family Muricidae

Hexaplex regius (Swainson, 1821)

Hexaplex radix (Gmelin, 1791)

Hexaplex princeps (Broderip, 1833)

Vasula melones (Duclos, 1832)

Vasula speciosa (Valenciennes, 1832)

Family Turbinellidae

Vasum caestus (Broderip, 1833)

PHYLUM ECHINODERMATA

Class Asteroidea

Order Forcipulatida

Family Heliasteridae

Heliaster cumingi (Gray, 1840)

Order Valvatida

Family Ophidiasteridae

Pharia pyramidata (Gray, 1840)

Phataria unifascialis (Gray, 1840)

Family Oreasteridae

Nidorellia armata (Gray, 1840)

Pentaceraster cumingi (Gray, 1840)

Family Mithrodiidae

Mithrodia cf. *bradleyi* Verril, 1867

Class Echinoidea

Order Cidaroida

Family Cidaridae

Eucidaris thouarsii (L. Agassiz & Desor, 1846)

Order Diadematoida

Family Diademidae

Astropyga pulvinata (Lamarck, 1816)

Centrostephanus coronatus (Verril, 1867)

Diadema mexicanum A. Agassiz, 1863

Order Camarodonta

Family Echinometridae

Echinometra vanbrunti A. Agassiz, 1863

Family Toxopneustidae

Lytechinus semituberculatus (Valenciennes in L. Agassiz, 1846)

Toxopneustes roseus (A. Agassiz, 1863)

Tripneustes depressus (Agassiz 1863)

Class Holothuridea

Order Dendrochirotida

Family Cucumariidae

Cucumaria flamma (Solis-Marin & Laguarda-Figueras, 1999)

Family Psolidae

Lissothuria sp

Order Aspidochirotida

Family Holothuriidae

Holothuria (Thymiosycia) arenicola Semper, 1868

Holothuria (Stauropora) fuscocinerea Jaeger, 1833

Holothuria (Selenkothuria) theeli Deichmann, 1938

Order Synallactida

Family Stichopodidae

Isostichopus fuscus (Ludwig, 1875)

Class Ophiuroidea

Order Ophiacanthida

Family Ophiocomidae

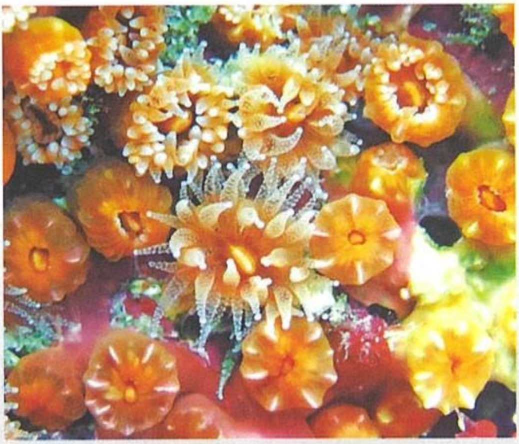
Ophiocoma aethiops Lutken, 1859

Ophiocomella alexandri (Lyman, 1860)

Order Amphilepidida

Family Ophiactidae

Ophiactis savignyi (Müller & Troschel, 1842)



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