First report of marine alien species in mainland Ecuador: threats of invasion in rocky shores

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Abstract Invasive species are of significant concern, especially in mega-diverse countries, because they cause negative effects such as loss of native biodiversity, ecological alterations, disease spread, and impacts on economic development and human health. In mainland Ecuador, information on invasive invertebrates in marine ecosystems is scarce. The objective of this study was to describe and locate the invasive species present in the rocky shores of the intertidal and subtidal zones along 10 areas (83 sites) covering most of the Ecuadorian coast during 2015–2016. Benthic macroinvertebrates communities were measured over quadrats located randomly on a 50 m transect positioned parallel to the coast in the intertidal and subtidal zone, covering an area of 1,860 km^2. Six invasive species were recorded: Arthropoda *(Amphibalanus amphitrite),* Cnidaria *(Pennaria disticha, Carijoa riisei*), Bryozoa (*Bugula neritin*a), Rhodophyta (*Asparagopsis taxiformis*) and Chlorophyta (*Caulerpa racemosa*). The areas with highest abundance of invasive species were in Jama (not a protected area), Marine and Costal Wildlife Reserve Puntilla of Santa Elena and Santa Clara Island Wildlife Refuge (protected areas). The most abundant species was *Carijoa riisei* with a relative abundance of up to 80%. It was the most aggressive of the invasive species registered in the subtidal zone, mainly in northern centre of the Ecuadorian coast. *C. riisei* is growing on native coral (*Pocillopora* spp.*)* and on sessile macroinvertebrate communities (*Pinctada mazatlanica*, *Muricea appresa* and *Aplysina* sp.) that are being affected by its invasion. This study must be taken into account by local and regional government authorities to create public policy programmes of monitoring for surveillance and control of invasive species. These programmes should focus on integration of socio-economic and ecological effects. They should be complemented by experimental design and analysis of environmental variables to provide technical information for a baseline of bio-invasion analysis along the Ecuadorian coast and Galápagos, to avoid the expansion of invasive species negatively affecting the marine biodiversity of mega-diverse countries such as Ecuador and other countries of South America.

Keywords: *Carijoa riisei*, continental Ecuador, Galapagos Islands, invasive species, macroinvertebrates, marine ecosystems, marine protected areas

INTRODUCTION

Invasive species are a cause of worldwide concern especially in mega-diverse countries because they can cause loss of native diversity, ecological alterations, increases in pests, diseases (Prenter, et al., 2004), impacts on benthic communities, impacts to the water column (Darrigran & Damborenea, 2011). Additionally, they can affect economic development and human health (Lowe, et al., 2000; Pimentel, et al., 2005). Many species are transported accidentally through anthropogenic means breaking geographic barriers that once restricted their range of expansion (Schüttler & Karez, 2008); they invade new areas, where they can settle, reproduce, spread and compete with native species.

Biological invasions, along with climate change, are key processes that feedback and affect global biodiversity. Climate change facilitates the dispersal and establishment of species which aggravates their impacts and makes their control more difficult, while invasive species can influence the magnitude of the environmental impacts by altering the structure and function of ecosystems (Mendoza, et al., 2014).

At present, there are numerous global and regional initiatives dedicated to optimising information and management of invasive alien species, including the Global Invasive Species Program (GISP), the IUCN-ISSG Invasive Species Global Information Network on Invasive Alien Species (GISIN), the Global Invasive Species programme of The Nature Conservancy (TNC-GISI) and the Inter-American Invasive Species Information Network (IABIN-I3N) (Schüttler & Karez, 2008).

On mainland Ecuador, information on invasive invertebrate species on intertidal rocky shores and subtidal zones is limited, fragmented and scattered. However, research on non-native species conducted in the Galapagos Islands (1,000 km off the coast of mainland Ecuador) has increased in the last decade, both in the terrestrial and marine environments (Campbell, et al., 2015). In 2012, the Charles Darwin Foundation (CDF), in collaboration with the Galapagos National Park Directory (GNPD), the Galapagos Biosecurity Agency (ABG), the Ecuadorian Navy and the Ecuadorian Navy Oceanographic Institute (INOCAR), initiated the Marine Invasive Species Project in the Galapagos Marine Reserve (Keith, et al., 2015).

The study of non-native species in Ecuador has mainly been done in the Galápagos Islands, due to the importance of this unique ecosystem in the world and the relative lack of scientific funding on the mainland. In the Galápagos Marine Reserve (GMR), an initial baseline study produced a list of seven non-native species in the GMR (Keith, et al., 2016). The marine invasive species team of the CDF have continued the research and applied different methodologies to learn more about non-native species in the GMR and

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the Eastern Tropical Pacific (ETP) region (I. Keith, pers. comm.). The objective of this study was to identify invasive species located in rocky shore habitats of the intertidal and subtidal zones covering 1,860 km^2 of the Ecuadorian coast during 2015–2016, that could be considered as threats for Ecuadorian mainland as well as Galapagos vulnerable ecosystems.

MATERIALS AND METHODS

Study area

Fieldwork was carried out in 10 areas along the Ecuadorian coast in six protected coastal marine areas (acronym in Spanish: AMCP) and four non-protected areas. The study areas ranged from Playa Escondida (0°49'8.05" N, 80° 0'22.66" W) in the north of Ecuador in Esmeralda province to Santa Clara Island in the south of Ecuador (3°11'21.11" S, 80°27'10.21" W) in El Oro province, covering 1,478 km^2 of protected areas and 382 km^2 of additional areas on the mainland coast. This survey included the protected areas (Fig. 1) from the north of Ecuador in the Galeras San Francisco Marine Reserve (acronym in Spanish: RMGSF) (Esmeralda province); Wildlife Refuge and Marine Coastal Pacoche (Pacoche) and Machalilla National Park (acronym in Spanish: PNM) (Manabí province); El Pelado Marine Reserve (acronym in Spanish: REMAPE); and Wildlife Coastal Marine Reserve Puntilla of Santa Elena (acronym in Spanish: REMACOPSE) (Santa Elena province) to Santa Clara Island Wildlife Refuge (Santa Clara) (El Oro province). The non-protected areas (Fig. 1) were: Jama, Canoa (Manabí province), Ayampe-La Entrada (between Manabí and Santa Elena provinces) and Copé (Santa Elena province). The Ecuadorian coast has an extension of 2,900 km corresponding to 45% of open coastal and 55% of inner coastal waters (Ayón, 1988). There is a wide range of geological features along the coast, including bluffs, barriers and sandplains, estuaries and lagoons, and engineered shoreline structures (Boothroyd, et al., 1994).

The climate on the coast varies seasonally from dry season (May to November) to the rainy season (December to April). The average annual temperature is above 22°C, with maxima fluctuating between 32–38°C and minima fluctuating around 15°C (Sonnenholzner, et al., 2013). Ecuador belongs to the Tropical East Pacific (TEP) region, with two sub-regions known as Panama Bight Ecoregion and Guayaquil Ecoregion (Sullivan & Bustamante, 1999; Miloslavich, et al., 2011). The northern half of the Ecuadorian mainland coast corresponding to the Panama Bight Ecoregion extends from Azuero Peninsula of Panamá to Caráquez Bay. It is characterised as a tropical zone, covered mostly by mangroves and dense rainforest vegetation (Miloslavich, et al., 2011), with >2,000 mm/yr of rainfall and without ecologically dry months through the year (Sonnenholzner, et al., 2013). The southern Ecuadorian coast, falling within the Guayaquil Ecoregion, extends from Caráquez Bay to Illescas Peninsula in the north of Perú and is characterised by a drier climate with <100 mm/yr of rainfall (Miloslavich, et al., 2011).

Survey

A total of 83 sites were sampled from February 2015 to February 2016 along the four coastal provinces of Ecuador (Esmeraldas, Manabí, Santa Elena and El Oro). These sites were established considering aspects such as representativeness of ecosystems; areas with greater and lesser anthropogenic intervention, biological processes (reproduction hotspots, feeding areas, seabirds and sea turtle nesting sites); and sensitive habitats or areas of great ecological importance according to the requirements established in the terms of reference of the Environmental Ministry (Ministry of Environment, 2014). The composition and abundance of the macroinvertebrates present in the rocky shore in the intertidal and subtidal zones were quantified using a band-transect system parallel to the coastline.

Data were collected in the intertidal zone following the standardised protocol from the South American Research Group on Coastal Ecosystems (SARCE) (SARCE, 2012). At each site, 10 quadrats (50 × 50 cm each) were randomly placed and sampled along a 50 m transect positioned parallel to the waterline in the mid, low and high intertidal level. A total of thirty replicates was sampled for each site. The abundance of colonial organisms was estimated by percent cover and all mobile individuals (>2 cm long) were counted. Most identification of biota was done in the field, although occasional problematic specimens were collected for reference and sent to specialists for identification. For the subtidal zone, at each site the organisms were separately estimated in two transect blocks by a diver, one on each side of 50 m transect line set along a shallow depth (normally 6–8 m). Every transect block encompassed a total reef area of 50 m × 5 m. The next diver scanned the nearby transect block by swimming back parallel to the initial transect at a distance of 5 m from the transect line (Edgard, et al., 2011). This up and back procedure for two adjacent blocks was

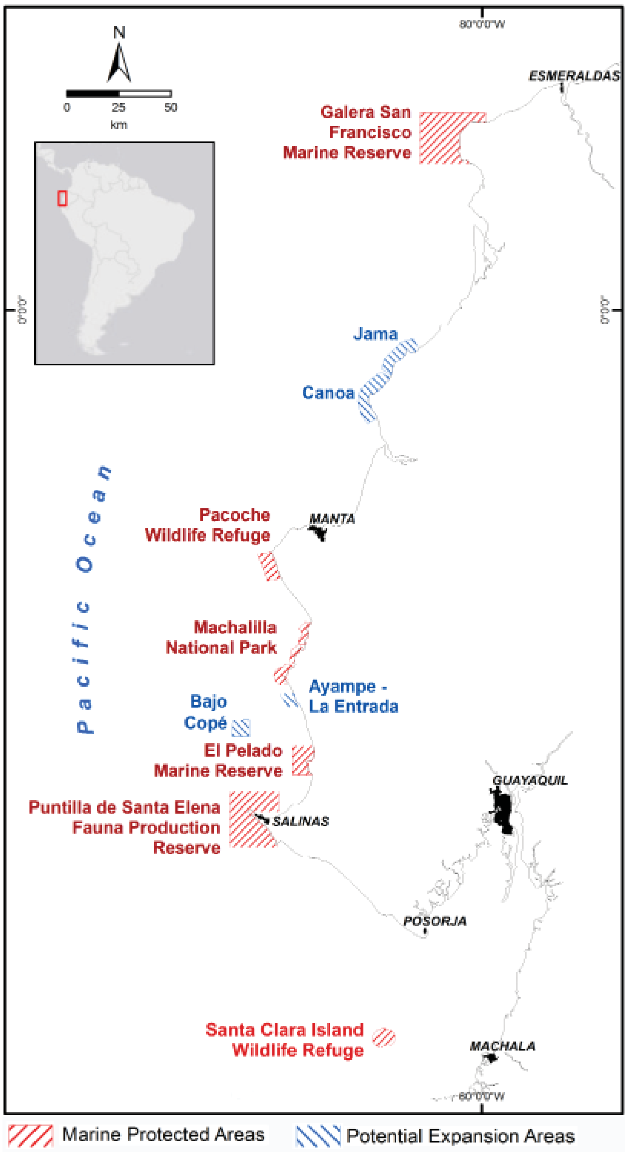

Fig. 1 Study area and location of the sampling sites on the Ecuadorian coast.

repeated along the shallower depth contour, generating a duplicate transect block data at each site. Sessile organisms were estimated by percent coverage of different taxa and grouped in substratum classes (crustaceans, cnidarians, sponges, ascidians, bryozoans, hydroids) within the transect lines. The cover was generally recorded by divers within 10 quadrats (0.5 × 0.5 m), placed sequentially every 5 m along the 50 m transect, and mobile organisms were counted along each quadrat. Digital photo quadrats were taken during the field work. We summed counts across all quadrats to create site totals.

Data analysis

To explain the biological assemblage, an X sites by Y species matrix of abundances was built to perform a non-metric multidimensional scaling (n-MDS) and cluster analysis to visualise the similarity of studied areas. For both analyses a similarity matrix was generated using the Bray-Curtis index on the fourth root transformed data to remove the weight of the dominant species (Clarke & Gorley, 2006). Further bubbled MDS analyses were performed to visually establish the differences among the abundances of *Carijoa riisei* between zones, using the statistical package Plymouth Routines in Multivariate Ecological Research (Primer). In order to determine the difference of organism abundance by province, a nonparametric ANOVA was performed, after the assumptions were not fulfilled, using the Kruskal Wallis test. In addition, to determine the difference in abundances between protected and unprotected zones, we applied the ANOSIM test using PRIMER V6 (Clarke & Gorley, 2006).

Distribution maps of species were prepared using the information collected from the fieldwork. These maps represent the relative abundance with a percent of coverage/m^2 on every site for inter-tidal and subtidal zones, with scales ranging between 0.1–50%, indicating a spatial approximation of alien invasive species location and coverage. Besides the status, invasiveness of each species was established using international databases such as: the IUCN list of 100 most harmful invasive alien species in the world (Lowe, et al., 2000); Global Invasive Species Database, ISSG (IUCN/SSC, 2014); and Invasive Species Compendium (<www.cabi.org/isc>).

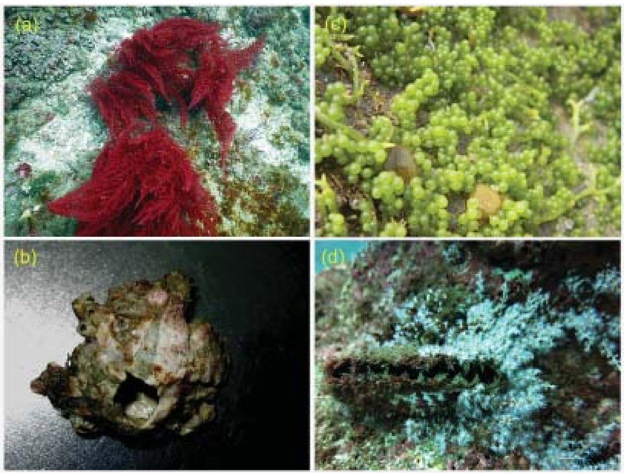

Fig. 2 Alien species found in the survey: a) *Asparagopsis taxiformis,* b) *Amphibalanus amphitrite* c) *Caulerpa racemosa*, and d) *Carijoa riisei* growing on the bivalve *Pinctada mazatlanica*.

RESULTS

A total of six alien invasive species from five phyla were recorded: Cnidaria (*Pennaria disticha*, *Carijoa riisei*), Bryozoa (*Bugula neritina)*, Arthropoda (*Amphibalanus amphitrite),* Rhodophyta (*Asparagopsis taxiformis*) and Chlorophyta (*Caulerpa racemosa*). Assemblages were numerically dominated by cnidarians. The most abundant species was *Carijoa riisei* (Table 1; Fig. 2). Invasive species were recorded at 24 sites (14 sites in the subtidal zone and ten in the intertidal zone). In the subtidal zone, the area with the highest presence of invasive species was the RMGSF in the north of Ecuador (Esmeralda province) while in the intertidal zone it was Punta Carnero site in the REMACOPSE, south-central part of the coast in Santa Elena province· (Table 1; Fig. 3).

The n-MDS of invertebrate invasive species abundance showed four groups with major similarity (60%), one group formed by Jama, REMAPE and RMGSF, the second group clustered the sites of REMAPE (south-central coast); the third group formed REMACOPSE, Ayampe, Copé and Santa Clara (central and south-central coast) and the last one grouped by REMACOPSE, Pacoche, Santa Clara, Ayampe and Canoa (Fig. 4).

Amphibalanus amphitrite, Pennaria distincha and *Carijoa riisei* were the invasive species with greatest

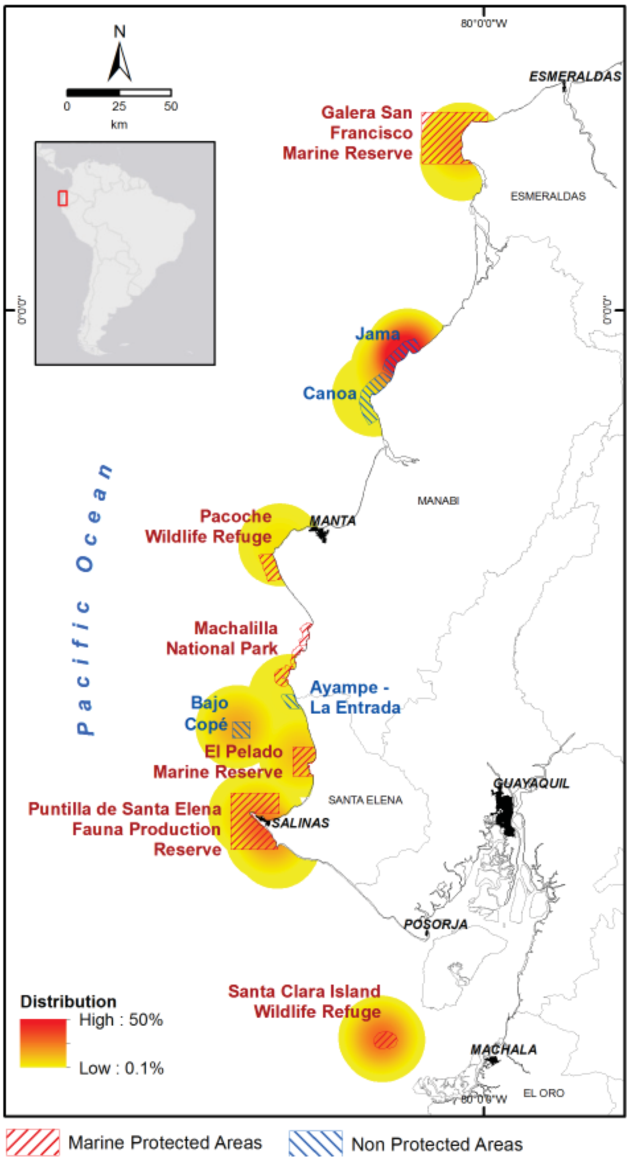

Fig. 3 Relative abundance and distribution of invasive species along the Ecuadorian coast during 2015–2016.

occurrence. Of these three, C. *riisei* was most abundant (Fig. 5) in the non-protected area located in the central coast of Ecuador (Jama) (Table 1). However, it was also recorded in the north zone of the Galera San Francisco Marine Reserve (Punta Alta, Piedra de Quingue) and in the south-central coast at El Pelado Marine Reserve (La Pared).

Statistically, no significant differences were found between the abundance of invasive species by provinces (global R=0.08, p>0.001) or by protected and unprotected zones (global R=-0.06, p>0.001).

The MAP's that presented the greatest number of invasive species were REMACOPSE (four species) and Ayampe (three species), followed by Jama and REMAPE (less than three species. Galeras San Francisco, Canoa, Pacoche and Copé recorded low benthic numbers of invasive species (Table 1).

DISCUSSION

This is the first report investigating the presence of invasive species along the Ecuadorian coast, including marine protected areas and unprotected areas, covering the coast from north to south of the country and two ecoregions in four distinct provinces. There are four species classified as macroinvertebrate invasive species worldwide, of which the majority are the cnidarians, mainly the Anthozoa class. Although the invasive species recorded are not listed in the 100 world's worst invasive alien species according to IUCN (Lowe, et al., 2000), two species (*Carijoa riisei* and *Bugula neritina,*) are listed in the Global Invasive Species Database (ISSG) and four species (*Carijoa riisei*, *Bugula neritina, Pennaria disticha* and *Amphibalanus amphitrite)* are registered by the Global Register of Introduced and Invasive Species (GRIIS).

Table 1 Invasive species recorded by provinces, areas and sites on the Ecuadorian coast, including abundance (coverage percentage) in the subtidal and intertidal zones.

PROVINCE / Area	Sites	*Amphibalanus amphitrite*	*Caulerpa racemosa*	*Aspargopsis taxiformis*	*Pennaria distincha*	*Carijoa riisei*	*Bugula neritina*
ESMERALDAS							
Galeras San Francisco Marine Reserve	Punta Alta	-	-	-	-	1.37	-
	Piedra de Quingue	-	-	-	-	11.5	-
MANABÍ							
Jama	Vaca Brava 1	-	-	-	-	20.25	-
	Punta Ballena*	0.11	-	-	-	-	-
	Bajo Londres	-	-	-	-	44.57	-
Canoa	Cabo Pasado*	-	0.96	-	-	-	-
Wildlife Refuge and Marine Coastal Pacoche	Liguiqui*	3.28	-	-	-	-	-
Ayampe – La Entrada	Los Ahorcados 1	-	-	-	0.49	-	0.12
	La Entrada*	0.03	-	-	-	-	-
Bajo Copé	Seco Manta	-	-	-	4.68	-	-
	Bajo Fer 3	-	-	-	8.86	-	-
SANTA ELENA							
El Pelado Marine Reserve (REMAPE)	La Pared	-	-	-	-	4.44	-
	Bajo 40	-	-	0.37	-	-	-
	Corales	-	-	4.82	-	-	-
Puntilla de Santa Elena Marine and Coastal Wildlife Reserve (REMACOPSE)	Guarro	-	-	-	1.12	-	-
	Bajo Ballena	-	-	-	0.25	-	5.31
	Chocolatera*	0.11	0.03	-	-	-	0.17
	Loberia*	7.8	0.22	-	-	-	-
	Punta Carnero*	16.01	-	-	-	-	-
	Anconcito*	0.5	-	-	-	-	-
EL ORO							
Santa Clara Island Wildlife Refuge	Sur*	-	0.6	-	-	-	-
	Norte*	-	1.68	-	-	-	-
	Sitio 2	-	-	-	18.64	-	-
	Sitio 3	-	-	-	3.67	-	-

* Sites with results of intertidal zones.

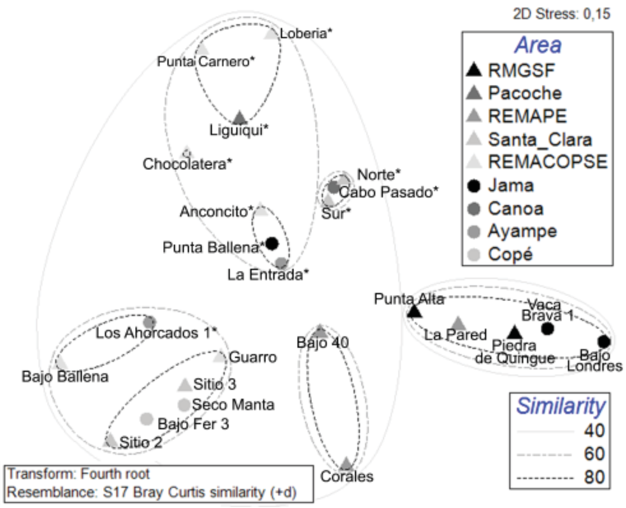

Fig. 4 Non-metric multidimensional scaling ordination, showing relative abundance of marine invasive species registered along the Ecuadorian coast during the period 2015–2016.

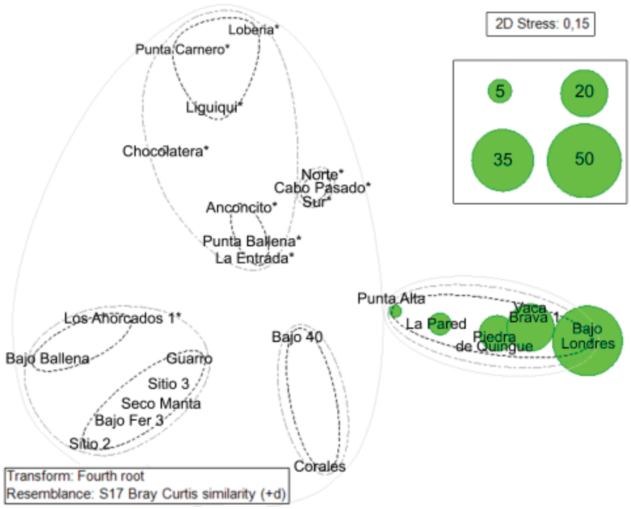

Fig. 5 2D bubble MDS configuration showing relative abundance of *Carijoa riisei*.

Carijoa riisei showed a greater abundance in the central zone of the Ecuadorian coast, mainly in Jama. This species has increased its colonisation in some areas of the El Pelado Marine Reserve in two years (2013–2015) (Cárdenas-Calle & Triviño, 2014). The invasion of *C. riisei* to new sites is probably caused by marine currents and maritime traffic. The invasive growth of *C. riisei* was noted among colonies of Pocilloporidae corals, *Pinctada mazatlanica, Muricea appresa* and *Aplysina* sp., confirming the imminent threat of this species to the sessile biota of the marine protected areas (Martínez, 2013). This species has an extensive geographic distribution in the Pacific from the Philippines, Indonesia, Australia, and Thailand, South Atlantic (Silva, et al., 2011) and Caribbean region (Kahng & Grigg, 2005; Kahng, et al., 2008;) with a variety of reproductive strategies (Barbosa, et al., 2014) including sexual and asexual reproduction, growing in different habitats, but preferring shallow areas.

Carijoa riisei has caused great impacts and damage to coral areas in Hawaii (Barbosa, et al., 2014) where it is currently considered a pest and has affected over 70% of the colonies of black corals *Antipathes dichotoma* and *A. grandis* (Global Invasive Species Database, 2017). It is considered a common invasive species from Florida (USA) to Santa Catarina (Brazil), displacing native species. It is now known to monopolise benthic surfaces under optimal conditions for its growth, from the intertidal zone to depths of >100 m (Venkataraman, et al., 2016). *C. riisei* competes successfully over black coral and invertebrates (Kahng & Grigg, 2005) and is dispersed through marine vectors (Grigg, 2003), and it is reported as a major biofouler in the Atlantic region (Concepcion, et al., 2010).

The rapid growth of the *C. riisei* colonies and their widespread dispersion in coral ecosystems has begun to generate great concern worldwide for being considered a threat to the diversity of sessile corals and invertebrates. For this reason, it is listed in the database of invasive species of IUCN (Global Invasive Species Database) and there is evidence of ecological impacts of this species in some countries of the South Pacific, as in Colombia, where high mortality of corals and octocoral coating has been reported on the island of Malpelo (Sánchez, et al., 2011).

Orensanz, et al. (2002) detected more than 40 invasive species in the Southern Atlantic Ocean, where poor knowledge of the regional biota makes it difficult to track invasions. For these reasons it is necessary to begin an alliance between national and international academics and environmental authorities (Ministry of Environment) in Ecuador to develop a strategy for surveillance and research on the ecological effects of invasive species in the coastal zones. With *Carijoa riisei* it is necessary to quantify mortality and replacement of existing coral communities in Ecuador, because this information is currently unknown, as is the habitat and biota preferences for colonisation. It is important to know its distribution, its ecological effects on native fauna, and its preferences (habitat, substrates, depths and environmental variables) to allow the establishment of substantial management actions to avoid its dispersion to other sensitive areas, such as the Galapagos Islands where it is still absent.

We found that the greatest abundance of invasive species was in the Ecuadorian central coast (Manabí), belonging mainly to the cnidarians. However, the largest diversity of species was in the south-central coast (Santa Elena). The presence of these invasive species is possibly due to the currents, ballast water and encrustations of invaders on ships. We can speculate that factors such as marine currents, rise of temperature, increase of maritime traffic, global warming and invasive breeding strategies will accelerate the augmentation of invasive alien species and the loss of diversity of corals, octocorals, sponges and other marine sessile invertebrates on the Ecuadorian coast. Four of the six non-native species found on the mainland of Ecuador (*Pennaria disticha, Bugula neritina, Asparagopsis taxiformis* and *Caulerpa racemosa*) from Table 1, are already present in the Galapagos Islands (Danulat & Edgard, 2002; Keith et al, 2016).

This study must be taken into consideration by local and regional government authorities to create public policies and programmes to monitor for surveillance and control of invasive species. These programmes have to be integrated with socio-economic and ecological effects and complemented by experimental design and analysis of environmental variables to provide technical information and a baseline of bio-invasions along the Ecuadorian coast and Galápagos. It is important to avoid or limit the expansion of invasive species that negatively affect the marine biodiversity of mega-diverse countries such as Ecuador and other countries of South America.

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