ECOLOGICAL ASSESSMENT OF THE MARINE INVASIVE ALIEN SPECIES (IAS) *PERNAVIRIDIS* IN TRINIDAD AND TOBAGO

Prepared by

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FISHERIES & AQUACULTURE RESEARCH PROGRAMME

Submitted

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Photo Credit: Pernaviridis on red mangrove prop roots in Caroni Swamp, John Agard. Circa 1990

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Executive Summary

Invasive Alien Species (IAS) has been recognised as a major threat to the fragile ecosystems of the Caribbean. Based on the complexity of this problem, a regional response, the "Mitigating the Threats IAS in the Insular Caribbean (MTIASIC)" Project has been developed, involving 11 international, seven regional and more than 25 national partners from five island nations in the Caribbean namely the Bahamas, Jamaica, the Dominican Republic, St Lucia and Trinidad and Tobago.

At the national level, apart from participating in regional activities, key components include the development of a National Invasive Species Strategy (NISS) and accompanying critical situation analysis as well as a public awareness and education strategy. Three national pilot projects were undertaken from which experiences and lessons-learnt serve to help the NISS. One of these projects "Management and Control of the marine invasive Pernaviridis in Trinidad and Tobago" was conducted in 2011 by the Institute of Marine Affairs. This report details one of its objectives; an ecological assessment to determine current distribution, community structure and investigation of the occurrence of natural predators of the green mussel, Pernaviridis in Trinidad and Tobago.

The assessment found that P. viridis was still primarily found on the west coast of Trinidad with some incursion on the north coast at Maracas Bay, Tyrico Bay and Las Cuevas Bay. It was absent from the east and south coasts of Trinidad and Tobago. On the north coast, the green mussel is found intertidally among the crevices of rocks as well as on pier pilings. On the west coast of Trinidad, the mussel can be found in both the intertidal and sub-tidal regions. An extensive mussel bed at Cedros heavily exploited by fishers and shellfish gatherers in the 1990s no longer exists as well as a smaller bed at Claxton Bay.

Highest densities were found at the ALCOA Jetty in Chaguaramas and at the Coast Guard Jetty in Cedros. Length frequencies distribution showed that while the green mussels at the ALCOA Jetty were the largest, no young recruits were present, unlike the other regions, where recruits were found. The condition index, which gives an overall indication of the 'health' of the green mussels were highest for San Fernando and lowest at the ALCOA Jetty. The physico-chemical environmental conditions recorded during the survey, and from past studies for Trinidad,

suggested that in the Atlantic region, the tolerance range of the green mussel for both salinity (17-38‰) and temperature (25-31 ^{o}C) have been extended.

The qualitative assessment of the community structure of pier pilings, the most ubiquitous habitat for P. viridis, showed the presence of seventy five (75) macrobenthic species in addition to Pernaviridis. In this ecological assessment, the species recorded comprised eight (8) phyla or major taxonomic groups – Annelida (polychaetes or marine worms), Arthropoda (shrimp,crabs, amphipods, isopods and barnacles), Echinodermata (brittle stars), Mollusca (snails and shells) Sipunculida (peanut worms), Tunicates (Sea squirts), Porifera (Sponges) and teleost fish. The largest phyla were the Mollusca and Arthropoda. As expected, softer-bodied organism such as the annelids and ascidians were found mainly in the subtidal region, while barnacles and bivalves dominated the intertidal region. An examination of the biodiversity of these habitats using a number of biodiversity indices; species richness, species evenness, Shannon Weiner Index and the Simpson Index showed that highest biodiversity was found in Tobago. Further analysis using Multi-Dimensional Scaling (MDS) plot and the associated dendrogram show that the fauna at the Port of Point Lisas and the LABIDCO Port were similar, so too were the north coast and the north-western peninsula while Tobago showed a separate and distinct clustering. No known natural predators were identified and none of the P. viridis samples had sign of boring on their shells indicative of predation.

The pathway for the introduction of P. viridis into the Atlantic Region, and initially to Trinidad, has been suggested as the shipping industry with the vectors being either ballast water or ship hull fouling. Research has shown that the origin of the Atlantic spread of the green mussel was from Trinidad. Most of the sites where P. viridis is still found in Trinidad are predominately at port and port facilities and as such, it remains an opportunity for introduction and further expanding of the introduced range of P. viridis.

There are no systems in place to monitor the introduction of marine IAS in Trinidad and Tobago. The role played by the maritime industry is of paramount importance because over 95% of the country's international trade is moved by maritime sea transport. Since shipping is global industry, addressing the problem of introduction of marine IAS must be global in nature; Trinidad and Tobago has been part of the International Maritime Organisation (IMO)

GloBallast Partnerships (GBP) which assisted developing countries to reduce the risk of ballast water mediated bio invasions and; prepare the countries for implementation of the IMO Ballast Water Management Convention, and compliance with its requirements at all levels.

As part of monitoring the implementation of national strategies to prevent the entry of marine IAS as well as to test the efficacy of any proposed legislation, it is recommended as a priority to implement biological baseline surveys of the port and port facilities in Trinidad and Tobago. This is considered urgent, as the accession by Finland has triggered the entry into force of the Ballast Water Management (BWM) Convention on 8 September 2017 (IMO 2016) which would have major implications for trade via ships around the world including Trinidad and Tobago.

1.0 BACKGROUND

Invasive alien species (IAS) are emerging as one of the major threats to sustainable development, on a par with global warming and the destruction of life-support systems. These aliens come in the form of plants, animals and microbes that have been introduced into an area from other parts of the world, and have been able to displace indigenous (native) species. Marine IAS has been identified as one of the four greatest threats to the world's oceans and to biodiversity globally (Globallast 2014). The other three are land-based sources of marine pollution, overexploitation of living marine resources and physical alteration/destruction of marine habitat. In addition, marine IAS also threaten marine industries (trade, fishing and tourism), coastal infrastructure and human health.

IAS has been recognised as a major threat to the fragile ecosystems of the Caribbean (Lopez *et al*. 2007). With a global focus on food and nutrition security amidst serious concerns about impacts of climate change on water resources and food production, it is important to be aware of, and understand how IAS impact on our lives, livelihoods, economy and environment. Such understanding can go a long way in determining our roles in preventing the introduction of IAS and managing them once they have 'arrived'.

Based on the complexity of this problem, a regional response, the **"Mitigating the Threats of Invasive Alien Species (IAS) in the Insular Caribbean (MTIASIC)"** Project was developed, involving 11 international, seven regional and more than 25 national partners from five island nations in the Caribbean namely the Bahamas, Jamaica, the Dominican Republic, St Lucia and Trinidad and Tobago. The Project aimed to broaden the approach to deal with IAS by both strengthening existing national capacity and measures, and fostering regional cooperation frameworks through which Caribbean-wide strategies can be developed. Implemented regionally and nationally by the Centre for Agriculture and Bioscience International (CAB International) and the Ministry of Food Production respectively, and executed by the United Nations Environment Programme (UNEP), the MTIASIC Project was funded by the Global Environment Facility (GEF) with co-financing from governments and national agencies in participating countries.

At the national level, apart from participating in regional activities, key components included the development of a National Invasive Species Strategy (NISS) and accompanying critical situation analysis as well as a public awareness and education strategy. Additionally, a significant conclusion to the project was the development a framework mechanism to deal with the threat of IAS at a national level.

Three national pilot projects were undertaken from which experiences and lessons learnt serve to help the NISS:

1. Maintenance of the native biodiversity of the ecologically sensitive area- nariva Swamp by the production and transplanting of IAS-free palm seedlings. Species targeted : Red Palm Mite and Coconut Moth;
2. Preventing the entry of Moniliophtorararoreri (Causal agent of Frosty Pod Rot of Cocoa) into Trinidad & Tobago;
3. Management and Control of the marine invasive, *Pernaviridis* in Trinidad and Tobago.

Implementation of the third project "Management and Control of the marine invasive *Pernaviridis* in Trinidad and Tobago" was carried out by the Institute of Marine Affairs.

The specific project objectives were:

1. An ecological assessment to determine current distribution, community structure and investigation of the occurrence of natural predators;
2. An economic assessment to determine the cost of its control as a fouling organism since its introduction;
3. Determination of an effective control of *P. viridis* as a fouling organism.

Objective (3) was amended given the reduced population of *Pernaviridis* during the ecological survey, and based on interactions with varied stakeholders during the project, was replaced with a national workshop on marine IAS.

This report details the findings of the ecological assessment (Objective 1).

2.0 LITERATURE REVIEW

The following literature review with a focus in the Atlantic region, highlights the distribution of the green mussel, *P.viridis*, and its biological characteristics which promotes its ability to colonise and establish itself as an as an invasive species and subsequent impacts on the ecology, economy and human of affected areas.

2.1 Distribution

Perna.viridis (Linnaeus, 1758), the green mussel or green-lipped mussel belongs to the genus *Perna* which has two other species: *P. canaliculus and P. perna* (Siddall 1980). In its natural range, *P. viridis* is widely distributed throughout the Indo-Pacific area, extending to the north as far as Hong Kong, the southern provinces of Guangdong and Fujian in China, Northeast Vietnam and southern Japan (Vakily 1989 and Baker *et al*. 2007). The east west distribution ranges from the Persian Gulf to the Indonesian coast west (Philippines plus Sumatra, Borneo, Bali and Sulawesi but excludes the remainder of Indonesia) of New Guinea (Siddall 1980, Baker *et al.* 2007), and to some of the Pacific islands, where *P. viridis* has been experimentally introduced (Vakily 1989). Additional, localities not reported by Siddall (1980) and Vakily (1989) include the Andaman Islands, Vietnam, Java and southern Sulawesi (Baker *et al.* 2007). Siddall (1980) noted that *P. viridis* has the potential to increase its geographical distribution by "island hopping", a kind of stepwise larval dispersal (Rajagopal 2006).

2.2 Introduction to the Atlantic

The first known Atlantic basin invasion of *P. viridis* occurred in western Trinidad (southern Caribbean) about 1990 at the port of Pt. Lisas (a commercial harbour**)** (Agard *et al.* 1992). By 1992, *P. viridis* had spread along the entire western coast of Trinidad on a variety of substrate (mangrove roots, rocks and pilings) primarily with the prevailing currents (**Figure 1**). Identification was confirmed with electrophoretic analysis of allozymes coding for enzymes (Agard *et al.* 1992). Since its introduction into Trinidad, the green mussel has spread throughout the Atlantic Ocean to Venezuela, Margarita Is., Jamaica, Cuba, and Florida and Georgia states in the United States and Colombia (**Figure 2**).

In 1993 *P. viridis* appeared in Venezuela, across the Gulf of Paria with the prevailing currents (Rylander 1996) and by 1996, it had spread across the northern coastline of the Araya Peninsula where they were patchily distributed along the northern coast at La Esmeralda, Chacopata, Punta Escarceo, La Orqueta and as far west as Punta Araya, and to Isla de Margarita where it has become a vibrant fishery and where it began to displace the non-native *P. perna*. This spread is consistent with larval dispersal via local oceanic currents (Agard *et al.* 1992; Rylander 1996).

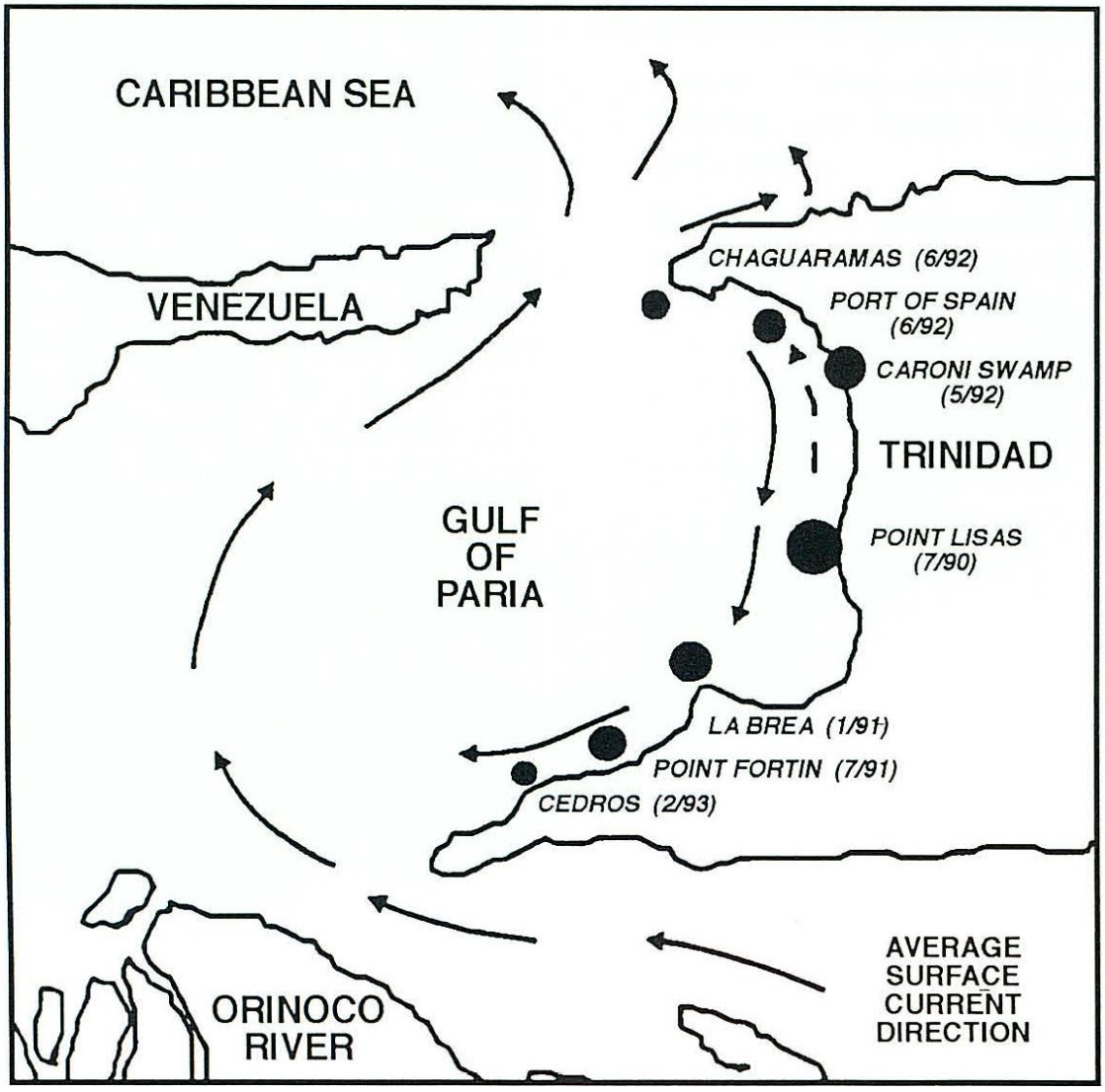

Figure 1: Distribution of *Pernaviridis* in Trinidad with dates of first sightings in parentheses, and solid circles of diameter proportional to their relative abundances at these sites (Agard *et al.* 1992)

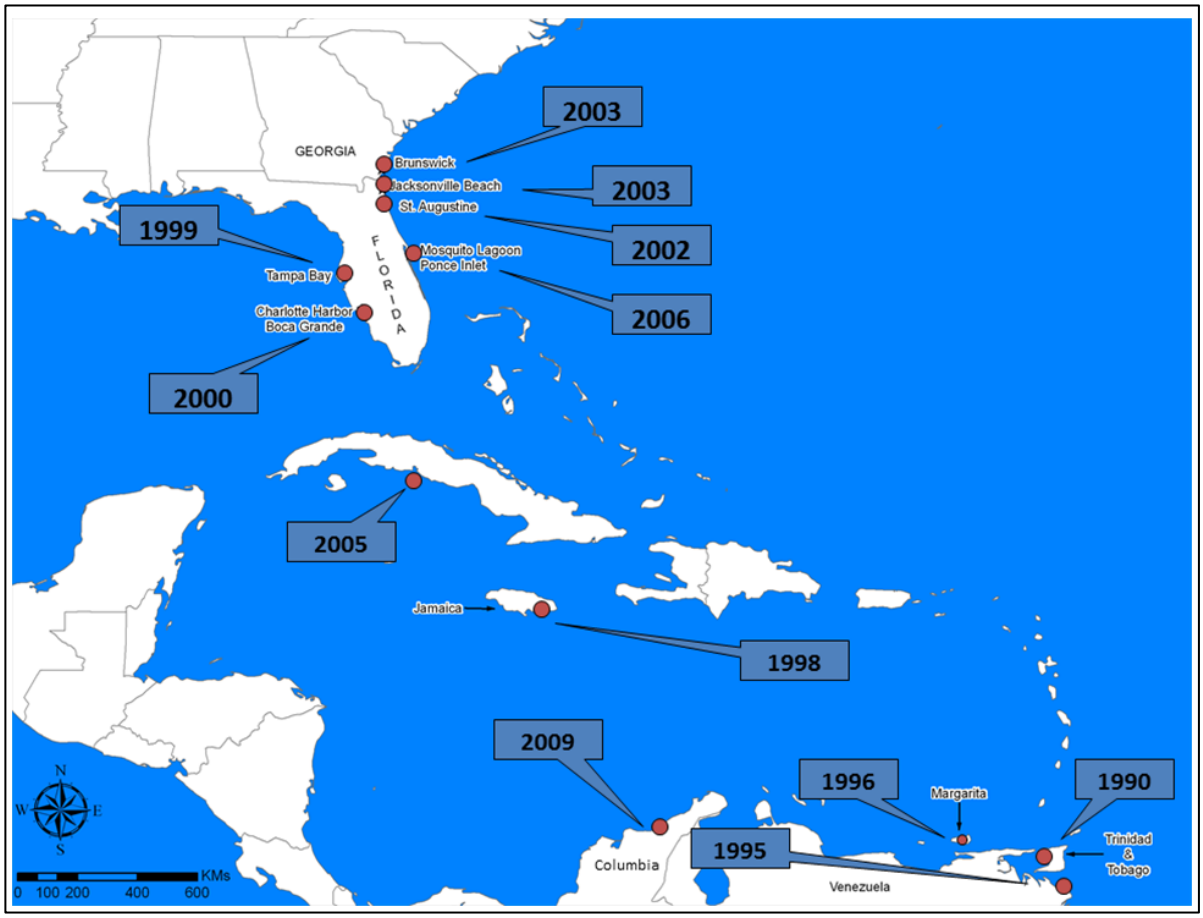

Figure 2: Introduction of *Pernaviridis* into the Atlantic region showing dates of first sightings (taken from Kishore 2013)

In February 1998, *P. viridis* was reported on mangrove roots in Kingston Harbour, Southern Jamaica (Buddo *et al.* 2003). It was most likely introduced via ballast water which contained the larval stages which subsequently settled out of the water column as spat onto various habitats around the harbour (Baker *et al*. 2007 and Buddo *et al.* 2003).

The first report in the USA was in July 1999 from the seawater cooling system of TECO Gannon electrical power plant facility in Hillsborough Bay portion of Tampa Bay, a large subtropical estuary in peninsular Florida (Baker *et al*. 2007; Baker *et al*. 2011). Based on genetic work, Trinidad was proposed to be the single source for both Jamaica and Florida invasions based on transhipment records (Gilg *et al.* 2012). By August 2000, the species had spread as far south as Charlotte Harbour and north of Tampa Boca Ciega Bay. This spread along the Florida coastline is consistent with larval dispersal in local oceanic currents (Baker *et al.* 2007). In 2002, reports of the green mussel surfaced in St Augustine on the northeast coast of Florida.

During early 2003, currents carried the mussels as far north as Jacksonville Beach Florida (Power *et al.* 2004). Its establishment in Georgia was a result of larval dispersal around the peninsula, and may have represented an entirely new introduction; however it is more likely to have resulted from boats and equipment being transferred between coasts without adequate cleaning of attached organisms (hulls and propellers) and the draining of bilge water (Baker *et al.* 2007).

In 2005, *P. viridis* was first reported in Cuba interfering with the operations of a local thermoelectric plant in the Cienfuegos Bay (Castillo *et al*. 2014). Almost 20 years since its introduction into the Atlantic region, *P. viridis* was found at port facilities in Cartegena, Colombia, (da Costa Fernandes and Coatanroach 2009).

The natural habitats of *P. viridis* are midlittoral and sublittoral zones, where they often constitute dense populations on rocky substrata that are rich in plankton and organic matter and carry low loads of suspended sediments (Vakily 1989; Rajagopal *et al.* 1998). *P. viridis* is able to adapt to a wide range of different environmental conditions, such as brackish estuaries and in open though not very exposed (subjected to wide tidal fluctuation) waters. The mussel attaches itself by means of byssal threads to a large variety of substrata, such as rocks where it plays an important role in the ecology of rocky shore ecosystems (Rajagopal *et al.* 1998), stones, piers, dead shells and even compact mud or sand. Mangrove mudflats represent a major potential habitat for the tropical mussel (Macintosh 1982). *P. viridis* was found on hard substrates, soft substrates, man-made and natural substrates in the Trinidad, Jamaica, Venezuela and Florida (Vakily 1989, Agard *et al.* 1992; Buddo*et al*. 2003).

2.3 Reproduction

The ability of one species of mussel to out- compete another is largely determined by differences in reproductive strategy (Barber *et al*. 2005). This includes both reproductive effort (fecundity) and the timing and frequency of spawning. The species that produces the most larvae will likely be able to out-compete other species for settlement space and energy.

The green mussel is a dioecious broadcast spawner, displaying a wide variation in spawning pattern within its native range, encompassing unimodal, biomodal and asynchronous modes (Benson *et al.* 2001). It is sexually mature at two to three months of age and live for about three years. The larvae remain free-swimming for 2-3 weeks in the plankton, typically attaching to hard structures with byssal threads in densities of up to 35 - 200 individuals /m². In parts of their native range, they exhibit rapid growth rates of between 6 and 10 mm/ month. These biological attributes facilitate opportunistic colonization of new areas (Power 2004).

Maturation and spawning of gametes occurs very rapidly by *P. viridis*, which indicate that *P. viridis* is capable of more rapid response to environmental factors that affect gametogenesis. Continuous production and release of few gametes over an extended period of time is not uncommon, particularly for tropical species like *P. viridis*. Gametogenesis has been reported in specimens as small as 21.3 mm in Thailand and 28.6 mm in Florida, but 50 mm is more typical minimum size for production of mature gametes (i.e. spawning) in Taiwan (Baker *et al.* 2007).

Green mussels in Trinidad were found to reproduce throughout the year with one or two annual peaks in reproductive activity concordant with changes in salinity and two pulses of recruitment (Phillip *et al*. 2001). This was found to be typical in tropical environments, and in its native region, the green mussel had a pattern of year round spawning which was a result of differences in the timing of spawning between the various age groups of green mussels in the same population (Phillip *et al.* 2001). In the Gulf of Paria, spawning appeared to be most closely associated with salinity, and to a lesser extent with temperature (Phillip *et al.* 2001). It was found that maturation and spawning were associated with the seasonality of food availability, and temperature in determining gonad maturation in green mussels (Parulekar *et al*. 1982). Gonadal development and spawning activity of *P. viridis* in India were found to be linked to the water temperature which showed two clear peaks, one in April-June (30.9 ^{o}C) and another in October (31.3 ^{o}C). Obviously, local hydrographical conditions and food availability may act as key factors that influence the timing of spawning by a population (Rajagopal *et al*. 2006).

2.4 Growth Rates

P. viridis is capable of high growth rates, especially under favourable conditions (Vakily 1989). Rajagopal *et al.* (1998) reported that on the southeast coast of India, the mussel could reach 119 mm shell length in the first year and 152 mm in the second year. However, such rates were observed at higher than normal flow conditions i.e., on the screen walls of a power station intake. In Trinidad, growth rates is also high as the largest shell found was 180 mm at Pt. Lisas and was deduced to be approximately 2.5 years old (Phillip *et al.* 2001). Comparison of growth rates from Trinidad and the Indo-Pacific showed that the highest growth rate obtained for wild mussels was at Pt. Lisas and the lowest at Vessigny from Trinidad. Such a high growth rate was attributed to increased food flux rate caused by the high flow (Rajagopal *et al.* 1998).

Growth of mussels is determined by several factors, the most important of which are believed to be food supply and environmental conditions (Parulekar *et al.* 1982; Jamieson 1989). In many studies, the influence of seasonality was highlighted (Chatterji *et al.* 1984) recorded maximum growth rate at Goa (west coast of India) during March–May, coinciding with the phytoplankton maximum. However, polluted waters slows growth. In the Victoria harbour,

Hong Kong, Lee (1986) reported low growth rate of 5 mm per month in its first year while Cheung (1993) reported a growth of 49.7 mm in the first year in another polluted harbour in Hong Kong.

Turbulent coastal waters often contain substantial amounts of suspended particulate matter, which can be an impediment to the growth of filter-feeding organisms such as mussels (Rajagopal *et al*. 2006). However, *P. viridis* was found to be able to tolerate a high level of suspended particulate matter (up to 1200 mg l^{-1}). It was found that in Trinidad, *P. viridis* growth was influenced by the food quality suspended and tidal height (Phillip *et al.* 2001).

2.5 Behavioural Responses

The green mussel tolerates a temperature range of 15-32.5 ^{o}C but can survive at temperature of 39 ^{o}C for about 200 minutes, and thrives well in winter water temperatures as low as 12 ^{o}C (Benson *et al.* 2001). The optimum temperature for *P. viridis* has been found to be between 26 ^{o}C – 31 ^{o}C, with 31 ^{o}C being the best for growth and 29 ^{o}C being the best for settlement (Nair and Appukuttan 2003). The significance of understanding the role of thermal tolerance on marine invasive species like *P. viridis* is important in light of the potential climate changes associated with global warming. Currently, green mussel populations are confined to a portion of the Gulf coast of Florida and the southeastern Atlantic coast as far north as southern Georgia. More extreme air temperatures would be likely to cause periodic mortality events within the intertidal portion of green mussel populations and may cause them to become predominately subtidal along the northern edge of their range. Conservative estimates of a 1.4 ^{o}C water temperature increase over the next 100 years would cause the coastal water temperatures near Savannah, Georgia to reach 12 ^{o}C and could allow the current distribution of *P. viridis* to spread farther north into the coastal waters of Georgia (Urian *et al*. 2003).

Pernaviridis can tolerate fairly large changes in salinity and they are very adaptable to these changes (Rajagopal *et al.* 2006; Vakily 1989). They are able to survive in estuarine habitats which have normal fluctuations in salinity (27-33 ‰). *P. virdis* optimal preference is at 30 ‰ and can tolerate salinities as low as 20 ‰, with a lethal salinity at below 5 ‰ (Rajagopal *et al.* 2006 and Vakily 1989). Segnini de Bravo (2003) found that RNA/DNA ratio during salinity changes indicated that *P. viridis* shows physiological compensation to salinity increases and decreases. The gill tissue was the most affected by salinity changes, however, the protein in the abductor muscle increased in response to physiological stress. In the Atlantic region however, the green mussel appears to have a lower salinity limit as the green mussel has been found in Tampa Bay, Florida at 14 ‰ (Baker *et al.* 2004). In Addition, for Trinidad, Phillip *et al.* (2001) found P. *viridis* in the Gulf of Paria at salinities ranging from 18 to 38 ‰.

P. viridis is asuspension feeder selectively feeding primarily on algae (Benson *et al*.2001) and can assimilate various kinds of suspended food like plankton, dissolved substances, bacteria, heterotrophic nanoflagellates, dinoflagellates, diatoms, blue-green algae and detritus all form part of their diet (Rajagopal *et al.* 2006, Buddo *et al*. 2003). *Perna* spp. thrives in areas such as estuaries and bays which contain a high concentration of organic matter (Buddo*et al.* 2003). Optimum filtration rates of *P. viridis* were observed at a temperature and salinity of 30 °C (Rajagopal *et al.* 2006). Fast growth in *P. viridis* resulted from a higher average clearance rate, a higher average organic enrichment of filtered relative to available matter and a higher organic enrichment of ingested relative to filtered matter than have so far been recorded for any species of filter-feeding bivalve (Rajagopal *et al.* 2006).

The presence of natural hard substrata does not seem to be necessary for large scale colonization of *P. viridis*. There are some conflicting views on the preference of microhabitats of *Pernaviridis*. Spat has been found at greater densities on shells than on man-made structures after 1 to 3 months (Gilg *et al.* 2010), but Baker *et al.* (2011) found that pilings, piers and floating objects are favourable as *P. viridis* habitats while mangroves, flats and seawalls are not and shell and rock aggregations are intermediate. However, the increase in *P. viridis* spat density on man-made substrates that have been in the field for longer periods of time may be explained by initial changes in biofilms of the accumulation of other fouling organisms, such as barnacles, that may influence subsequent *P. viridis* recruitment. Biofilms have been associated with increased larval settlement and various benthic organisms. Baker *et al.* (2011) also observed a single report of high density on red mangrove prop roots and a large aggregation of *P. viridis* in a seagrass meadow along the Florida coastline.

2.6 Predators /Natural Enemies

Predation as a form of management and control has been looked at by researchers in different countries where *P. viridis* has invaded. However, none of their natural predators are able to make a considerable impact on the population at least not fast enough for industrial firms. In Trinidad, *Melongenamelongena* and *Pugilinamoria,* two local molluscs, were found to prey on bivalve shellfish (Ammons 2001). In China, mussels are the major diet of the crab, *Thalamitadanae,* which employ different modes of attack, depending on the overall size and relative shell thickness. Crabs are able to cut byssal threads and shell lips using their claws and dislodge individuals from a mussel bed. Crabs are better able to open the mussels if there is a decrease adductor, shell and byssus performance which most times would occur under stressful environmental conditions. However, stressful environmental conditions may also result in the predators leaving but returning after the hypoxia conditions (Wang *et al.* 2012). The diet of *T. danae* includes a high percentage (53.9 %) of mussel *P. viridis*. Other predators of *P. viridis*

include starfish and humans which would undoubtedly increase the predation risk of the mussel under that situation (Wang *et al.* 2012).

In Florida several predators of shellfish already inhabit the waters currently within potential range of *P. viridis*. These native predators include the blue crab, *Callinectessapidus*, and the Caribbean spiny lobster, *Panulirusargus*, both of which are commercially important crustaceans. Other *Panulirus* spp. are known to prey on *P. viridis*. It was found that blue crabs may provide a natural control mechanism for the green mussel. However, predation is apparently having little impact on green mussel populations as their high reproductive potential and rapid growth rate will probably prevent any significant control by predators (Mitchem *et al*. 2007).

2.7 Impacts

The impact potential of a species is expressed in terms of their actual (or potential) human health, economic and environmental impacts. The potential international target species are prioritised by their location in the invasion potential/impact potential space. *P. viridis* has been categorised as one of ten potentially most damaging species. In Australia, *P. viridis* has been categorised as 'High priority' (Hayes *et al.* 2005).

Within the Atlantic region, wherever they have been introduced, initial high densities were observed (Agard *et al.* 1992, Buddo *et al.* 2003, Lopeztegui-Castillo *et al.* 2014), however, co-existence at a reduced level appears to be the situation with *P. viridis* and is found primarily with associated organisms such as *Crassostrea*spp. and *Brachidontesexustus* in Florida, Trinidad and Jamaica. As for the smaller *B. exustus*, not only did it co-occur at all sites with *P. viridis* but there is no evidence to say that it was affected by *P. viridis* density. The nonindigenous *P. viridis* clearly has the ability to dominate certain habitats within certain natural ecosystems and the potential to adversely impact native species such as *C. virginica* through space competition. Findings suggest that these impacts will not be consistent across habitat types, or necessarily widespread within a habitat type (Baker *et al.* 2011). In Cuba, during a 22-month sampling period, the population of the green mussel declined dramatically. Among the suggested reasons for this decline are mortality resulting from stressful environmental conditions and removal through harvest, both coupled with low recruitment rates.

Colonization by *P. viridis* has been shown to also foster a microhabitat for other fouling organisms such as barnacles which would further reduce the water flow in the pipes which leads to more economic costs to clean since barnacles are much more difficult to remove than mussels (Rajgogal *et al.* 2006). The only evidence to support ecological impacts by *P. viridis*is its displacement of *P. perna*in Venezuela (Rylander 1996).

Pernaviridis was commercially harvested in India Thailand, Malaysia, Philippines, Venezuela, Jamaica and Trinidad when it was abundant. It has great potential to become a sustainable and lucrative resource but proper depuration procedures must be carried out before they can be put on the local and export markets (Buddo *et al.* 2003). Guppy (2001) found that the protein and fat content of the *P. viridis* meat was comparable with those reported for other shellfish showing the mussel to be a viable food source. The favourable nutritional value and relatively low contamination levels made this mussel an excellent candidate for commercial cultivation. However, Lopeztegui-Castillo *et al.* (2014) noted that the drastic decline of the population in the Cienfuegos Bay, Cuba suggested that this species is not a candidate for a commercial fishery and that manual harvesting may be an efficient means of eradication, if desired.

Apart from being a source of food, other benefits of mussels have been their use as bioindicators to study contamination of coastal waters by a variety of organic and inorganic pollutants, including heavy metals, organochlorines and polychlorinated biphenyls (PCBs). *P. viridis* has been used in its native region to monitor heavy metals such as zinc, manganese, copper, chromium, nickel and cadmium. Not only can the meat of the *P. viridis* be analysed for heavy metals, but the byssal threads too have been found to be reliable indicators of heavy metal pollution in the environment (Rajagopal *et al.* 2006).

Guppy (2001) investigated *P. viridis* as a biomonitor for pollution in the Gulf of Paria, Trinidad and found that smaller mussels were generally found to contain higher levels of cadmium, copper, zinc and lead than larger mussels, possibly due to the absence or lower rates of metallothionein production (protein). This study also found that the various soft tissues and foot was the site of least metal deposition while the digestive gland was the site of the highest metal concentration. The levels of heavy metals in the soft tissues of the mussel (minus the digestive gland) in 1995 and 1996 were found to be below maximum permissible levels set by local and international regulatory agencies (Guppy 2001). The complete removal of the digestive gland as an alternative to depuration appears feasible, if the mussel is to be used as a bio-indicator of heavy metal contamination (Rajgopal *et al.* 2006). The shells and byssal threads were also found to contain relatively high levels of heavy metals (Rajgopal *et al.* 2006).

A comparison between the green mussel and the commonly found oyster, *Crassostrearhizophora* (Rojas de Astudillo *et al.* 2005), found that cadmium, mercury and zinc in sediments were significantly correlated with those of the mussels, but not of the oyster, in which copper and zinc at several sites in the Gulf of Paria, Trinidad exceeded local maximum permissible levels (Cu =20 µg g^{-1} wet wt; Zn =50 µg g^{-1}wet wt) for human consumption (World Health Organization MPL of 13.0 µg g^{-1}wet wt).

The presence of saxitoxin, a paralytic shellfish poisoning (PSP) toxin was detected in *P. viridis* along the west coast in Trinidad in 1995, 1996 and 1997, and the mussels at the ALCOA port facilities were of great concern. However, detected levels were lower than the

Eastern European Countries (EEC) standard of 40 micrograms/100 g above which shellfish harvesting is banned (Guppy 2001). The specific PSP toxins persisted in the green mussel in 1999 and 2000 at a single site on the west coast of Trinidad (Chang Yen *et al.* 2004). Given that PSP-producing dinoflagellates have been identified in Trinidad waters (Heileman and Siung Chang 1990; Lee Lum and Guppy 1999), it was recommended by Guppy (2001) and Chang Yen *et al.* (2004) that the Food and Drugs Act (1998) be modified as it currently does not permit the presence of any PSP in samples for human consumption. Nevertheless, there is also the recommendation that this species be used for future monitoring for PSP.

The impact of the green mussel as a fouling organism of sea water cooling systems of industrial plants is well documented (Agard *et al.* (1992), Benson *et al.* (2001), (Rajgopal *et al*. (2006) and Lopeztegui-Castillo *et al.* (2014). This was the case in Trinidad at an industrial estate where its presence was noted for the first time in 1990 as well as an electrical company in Tampa Bay, Florida in 1999. Economic aspects of fouling-induced effects on electric power plants can be very significant, such as reduced flow of cooling water for steam condensation, causing increased condenser backpressure and below-optimum performance. Live mussels or empty shells can cause mechanical damage to pumps and block the flow of water in condenser tubes and reduce the heat transfer efficiency. Mussel shells can also cause accelerated corrosion of the condenser tubes.

An economic assessment of these impacts by fouling organisms including the green mussel on industrial plants in Trinidad was conducted by Chase (2016) who identified that the presence of the green mussel contributed to the clogging of pipelines thus hampering the efficiency of cooling systems and causing overall equipment damage. These firms produce petrochemicals, ammonia, methanol, fertilizer and urea and one is a desalination plant. Generally, the fouling of *Pernaviridis* caused equipment damage; flocculation/sediment tanks, media filters, membranes, pumps, intake screens, and intake pipelines. Moreover, the study revealed that amongst the six (6) firms affected by *Pernaviridis*, the fouling of the species in the peak years of invasion (1990 – 2003) had been responsible for as much as 60-90 % of their cooling water and pipeline maintenance costs. Maintenance costs per firm during that period ranged from USD 40,000.00-USD160, 000.00 annually amongst firms affected. Currently firms spend as much as USD1, 129,267.00 per annum in their control of fouling organisms including the marine invasive *Pernaviridis*.

There have been reports of extensive growth of green mussels in the condenser cooling systems of power stations in India (Rajgopal *et al*. 2006). In Cuba, its presence was recorded for the first time in Cienfuegos Bay, and has negatively affected the operations of a local thermoelectric plant Lopeztegui-Castillo *et al.* (2014). In China, the green mussel is also a major part of the biofouling community where they are notorious for fouling navigation buoys with biomasses as much as 72 kg/m². *Pernaviridis* is also a new member of the biofouling community in Japan.

It is anticipated that the cost of maintenance in these structures, as well as on navigational buoys and vessels will increase substantially as the *P. viridis* population continues to increase (Benson *et al.* 2001).

3.0 METHODOLOGY

The main focus of the ecological assessment was to determine the current distribution pattern of *P. viridis*and to characterise the community structure of habitats common to the green mussel including the occurrence of any natural predators.

3.1 Distribution

The physical survey to investigate the distribution pattern of the green mussel was guided by the following approach

- Past distribution pattern of *P. viridis* (Agard *et al*. 1992);
- Substrate type known upon which *P. viridis* colonises (as identified from the literature);
- Key informants (shellfish collectors and fishers, selected industrial plants personnel, research scientists);
- Published reports which included locations for *P. viridis* (Phillip *et al.* 2001, Bullock & Moonesar 2003, Guppy 2001).

The mussel attaches itself via byssal threads to hard substrates both in the intertidal and sub-tidal zones. These hard-substrates include wharfs, pier pilings, rocky outcrops, mangrove prop roots as well as can form mussel reef/banks in the sub-tidal zones. Such areas around Trinidad were sampled for the presence of the green mussel but was restricted to the north coast from west of La Fillette, the north-western peninsula and the west coast. The south coast was not sampled after preliminary discussion with fishers, using the green mussel shell as a visual aid, indicated that the green mussel was not found on the south coast. The east coast was not sampled because of the absence of the green mussel during the Census for Marine Life survey (pers comm. A. Jute, the University of the West Indies, St Augustine) as well as based on interviews with shellfish harvesters on that coast.

In Tobago, the presence of the green mussel was not reported from on-going and ad-hoc surveys of the IMA on coral reefs, mangroves and sea grass beds. As a result, the only habitat investigated during this assessment was the pier pilings at four sites; Scarborough, Roxborough, Charlotteville and Palatuvier which had the largest of these structures.

One hundred and six (106) sites from Trinidad and Tobago were thus sampled. Surveys were both qualitative and quantitative (mainly for community structure) spanning from February to November 2011. Each site was sampled once. Physico-chemical parameters- temperature, salinity, dissolved oxygen and pH were recorded using the YSI meter and GPS coordinates

were collected for all sampling stations. Photos of the sites were taken including underwater shots whenever water clarity permitted.

3.2 Description of Study Area

Sampling areas were grouped into regions based on proximity (**Table 1**).

Table 1: Description of sample sites grouped by regions used for quantitative analysis

Region	**Sample Area**	**Type of Assessment**
Region 1	Maracas Bay, Tyrico Bay, Las Cuevas, Dhein Bay, Blanchisseuse, La Fillette	Qualitative
Region 2	Northwestern peninsula of Trinidad Nelson Island, Pelican Is., Caledonia Is., Lenegan Is., Chacachacaree Is., Huevos Is., Monos Is., Scotland Bay, Tetron Bay, Gasparee Island, Staubles Bay -Carenage (jetties & marinas) Alcoa Jetty to Carenage	(Qualitative) Quantitative
Region 3	Caroni Swamp	Qualitative
Region 4	Pt. Lisas – Jetties at port facilities - PLIPEDCO, NEC, YARA LTD. TCL Mangroves prop roots and man-made 'island'	Quantitative Qualitative
Region 5	San Fernando, Godineau Bridge	Quantitative Qualitative
Region 6	La Brea – Brighton Jetty and LABICO jetty	Quantitative
Region 7	Cedros Jetties	Qualitative
Region8	Tobago – Scarborough port, jetties at Charlotteville, Parlatuvier and Roxborough	Quantitative

Region 1

The north coast is open to the Caribbean Sea and also to the direct influence of the Northeast Trade Winds. The north coast is fairly exposed and as such is a high wave energy environment (IMA 2013). The sample sites of Maracas Bay, Las Cuevas Bay, Blanchieusse Bay, Tyrico Bay and La Fillette Bay are situated along the western region of the north coast of Trinidad and are four of several bays located along the coast. All five aforementioned bays, are popular tourist destinations and with the exception of Tyrico Bay, are the locations of fish landing sites for the artisanal fishery of Trinidad. Much of the shoreline consists of rocky shore alternating with cliffs and beaches. **Plate 1** illustrates habitats examined for the presence of the *P. viridis.*

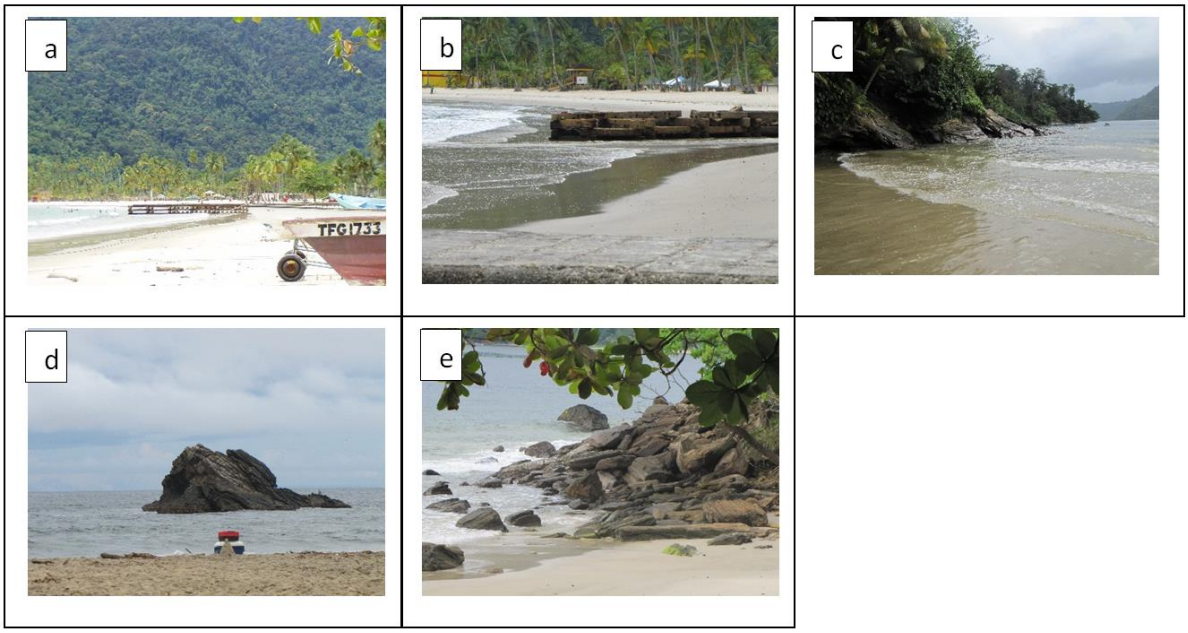

Plate1: Sampling locations at the various bays along the North Coast of Trinidad (a-b) Maracas Bay jetty. (c) Cliff faces at Las Cuevas Bay. (d) Stack at La Fillette (e) rocky intertidal region at Blanchieusse Bay

Region 2

The Chaguaramas Peninsula is situated on the north-western end of Trinidad. The Peninsula forms the western end of the Northern Range, and includes the mainland of Trinidad as well Monos, Huevos and Chacachacare Islands (Cropper Foundation 2008). Offshore islands also exists such as Gaspar Grande, Diego and Five Islands (Nelson, Pelican, Caledonia, Lenegan and Rock Islands). The area is a multiple use one with many economic, recreational and ecological activities. It is home to the Port of Tembladora, one of the eight ports of Trinidad and Tobago and several port facilities including the largest dry-docking facility in the Caribbean CL Marine Ltd (Kishore 2013). There are also services facilities of energy companies such as British Gas and BpTT.

It is a hub for recreational activities including several marinas with yachts, sailing and sea bathing and swimming at the various beaches and nature trails. Recreational activities in the Chaguaramas Peninsula have seen notable increases over the last decade. This has been largely due to Chaguaramas Development Authority's (CDA) efforts to promote tourism in the area, and increase interest of Trinidad as a hotspot for yachters in the Caribbean region (Alkin Koo, 2008).

The Chaguaramas Peninsula supports terrestrial and marine ecosystems; beaches, seagrass beds and coral reefs on different areas of the Peninsula. Coastal ecosystems include the smaller islands adjacent to the peninsula. As with other coastal areas around Trinidad, the marine conditions and communities are influenced by the seasonal freshwater outflow from the mainland and local rivers during the wet season. Shallow water coastal communities include patch reefs dominated by finger coral, *Porites*, small fringe reefs are found around the Five Islands, Monos and Chacachacare Is, and "deep-water" coral communities found at relatively shallow depths (e.g. 25 m). These communities include ivory coral, non-reef building corals and black corals (Alkin Koo, 2008; EMA, 2004). There are also sand and mud substrates with seagrass beds as well as rocky substrates covered with an assortment of hydroids and sponges (Alkin Koo 2008, EMA 2004). The Chaguaramas Peninsula was designated a National Heritage Park in 1975 with the objectives of protecting biodiversity such as the red howler monkey, and conserving ecological, historical and archaeological resources (Alkin Koo, 2008). **Plate 2** identified some of the locations and habitats sampled.

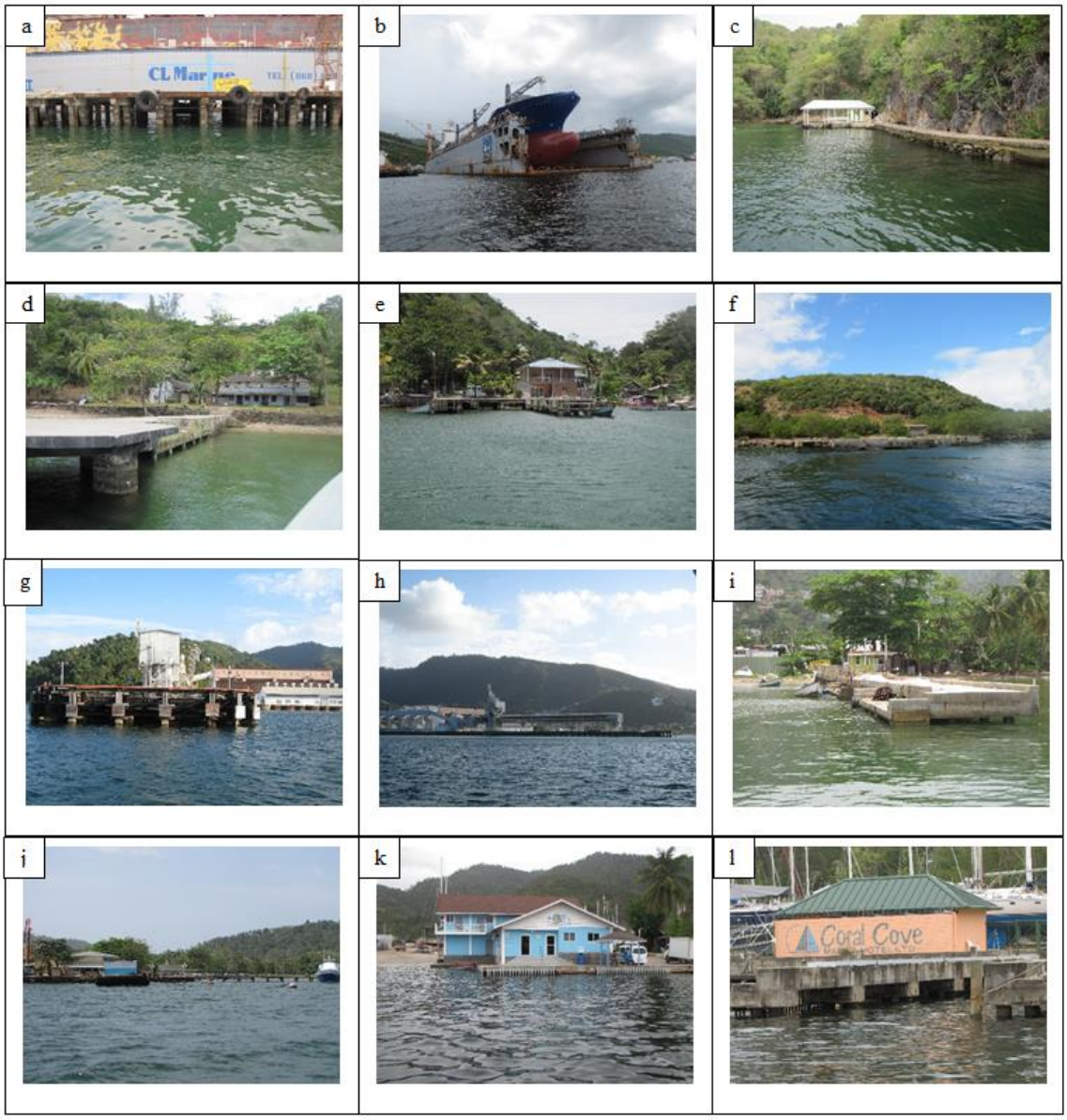

Plate 2: Sampling locations on the north western peninsula of Trinidad (a-b) Jetty and ship drydocks at CL Marine (c) Retaining wall at Staubles Bay (d-f) Jetties and rock walls along "Bocas Islands (g-h) Jetty and pier pilings at the non-operational ALCOA bauxite transshipment terminal (i) Jetty at Carenage (j) Jetty and pier pilings at Pier 1 marina (k) waterfront decking at Powerboats marina (l) Jetty at Coral Cove marina

Region 3

The Caroni Swamp is located on the west coast of Trinidad, 3.5 km south west of the capital, Port of Spain. It is the second largest wetland in Trinidad and Tobago, estuarine in nature with 8,340 ha of mangrove and herbaceous marsh interrupted by channels and brackish and saline lagoons (Juman 2010). It was designated a wetland of international importance under the Ramsar Convention in 2005. The swamp is important economically for oysters and fish harvesting, for hunting and eco-tourism. Some 160 species have been reported and is a major roosting site for one of the national birds, the Scarlet Ibis. It is affected by river-born pollution from land-based sources, encroachment and pollution (Juman 2010). Qualitative surveys for *P. viridis* were undertaken in numerous channels with the Swamp, (**Plate 3**).

Plate 3: Sampling locations in Caroni Swamp, west coast of Trinidad (a) Channel typically sampled within the Caroni Swamp. (b-c) Flora and fauna found on the red mangrove, *Rhizophora mangle* prop root

Region 4

This region represent a heavily industrialised area centrally located on the west coast of Trinidad (**Plate 4**). It comprised the Port of Point Lisas and port facilities at YARA, Savonetta Piers, and Trinidad and Tobago Cement Limited (TCL). The Port of Point Lisas (World Port Index No. 11750) is a multipurpose port as the nature of its business is the receipt, storage and delivery of cargo including containers, breakbulk, dry bulk and roll on – roll-off. It is 19.2 ha with 5 berths ranging in size from 35 to 200 m and with an associated draft of 7.5 – 11.76 m. Associated with the Port is the PLIDECO estate, which is the heart of Trinidad and Tobago's petrochemical sector covers 860 ha and represents an investment of over US $2 billion. The Estate is home to 94 companies involved in a range of activities. The petrochemical sector is dominant, however, with many multi-national production plants operating on the Estate. Ease of access to vast resources of natural gas has been a strong incentive for some of the world's leading manufacturers of ammonia, urea, methanol, other petrochemicals and steel (The Shipping Association of Trinidad and Tobago 2012).

The liquid and dry bulk port facility, Savonetta Piers, is owned by the National Energy Corporation of Trinidad and Tobago and the nature of business is cargo such as ammonia, methanol, urea, direct reduced iron, hot briquetted Iron, and import of bulk base oils, iron ore fines, grains and aggregates. There are six active terminals in addition to ISCOTT Dock. The terminals ranged in length from 115-245 m. YARA (including Tringen 2 Deepwater Port Terminal) is owned by Yara Trinidad Ltd and Trinidad Nitrogen Ltd. There are three terminals 170 - 219 m in length used to move the cargo, anhydrous ammonia, propane, butane, sulfuric acid and caustic soda. The Claxton Bay Marine Terminal is owned by Trinidad Cement Ltd. It is a sufferance wharf handling dry bulk; discharge and import of gypsum and clinker and export of bagged, bulk, jumbo cement (The Shipping Association of Trinidad and Tobago 2012).

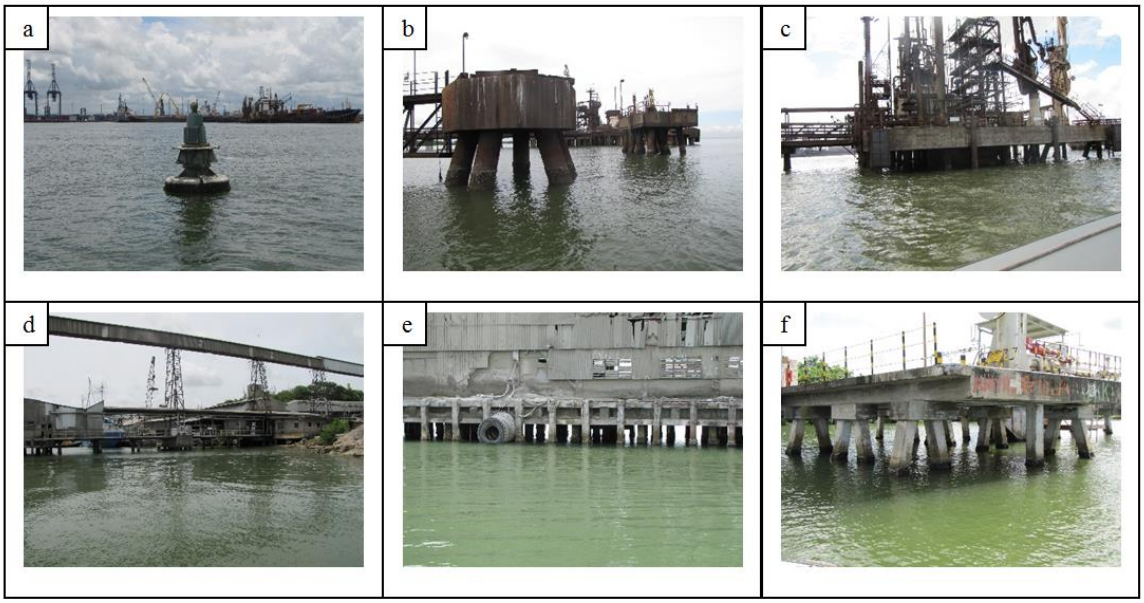

Plate 4 Sample locations for the central west coast of Trinidad (a) Marker buoy for the Savonetta channel. (b) Savonetta Piers pilings (c) Jetty at the Port of point Lisas (d-e) pier pilings and jetty at Claxton Bay Marine Terminal (f) Pier pilings one of the YARA terminal

Region 5

The Godineau Swamp also known as the South Oropouche Swamp is located in southwest Trinidad. It is an estuarine wetland, the third largest in the country and the second largest on the Gulf of Paria coast. It is 3,171 ha but receives drainage from a large catchment basin of 42, 473 ha. The surrounding urban communities include Fyzabad, Siparia, Penal and Debe. Traditionally, the wetland has supported a fish and shellfish industry and also meet the needs of rural communities by providing land for farming and housing (Juman 2010). Oil exploration has taken place in the Godineau Swamp since the early part of the twentieth century and the Swamp continues to provide several opportunities for ecotourism (Clarke-Marshall and Ghouralal 2006).

The San Fernando Wharf is use by small coasters (The Shipping Association of Trinidad and Tobago 2012), predominantly fishing vessels. The Wharf, 122 m long, extends from the shore abreast the town and has a depth of 1.8 m alongside. It was reported in 1986 that the channel was disused and the buoys removed and in 2008 the channel repositioned. Quantitative surveys were undertaken in this region (**Plate 5**).

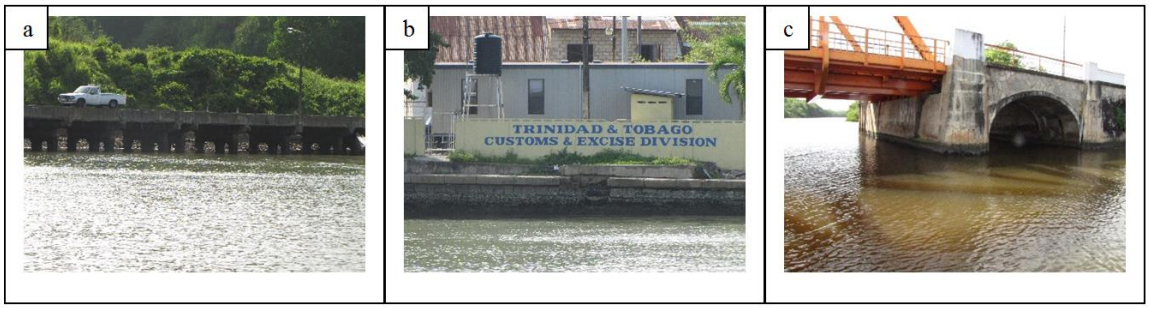

Plate5: Sample locations on the west coast, Trinidad (a-b) San Fernando jetty and wharf. (c) Bridge at the mouth of the Goudineau Swamp

Region 6

The Port of Brighton (World Port Index No. 11760) is a breakbulk facility operated by La Brea Industrial Development Company (LABIDCO), a subsidiary of National Gas Company of Trinidad and Tobago (**Plate 6**). LABIDCO Port Services Ltd and associate companies are engaged in providing facilities and services for the Oil & Gas Service Industry in Trinidad & Tobago, the Southern Caribbean and parts of Latin America (Labidco Port Services Ltd. 2012). T he old Brighton jetty is no longer active, but is used by fishers from nearby fishing villages and for recreational swimming and fishing.

Plate 6: Sampling locations at la Brea, west coast of Trinidad (a-b) Port facilities and jetty at LABIDCO (c) Old jetty at Brighton.

Region 7

Located at the tip of the southwestern peninsula, Cedros lies mere miles off the coast of Venezuela. It has historically been a fishing village and coconut grove, producing much of the coconuts for harvest. There are Coast Guard/Customs port facilities, however only small vessels such as fishing pirogues can be accommodated because of siltation in the bay. Qualitative surveys were conducted at the Coast Guard Jetty and derelict jetty (**Plate 7**).

Plate 7: Sampling locations at Cedros, west coast of Trinidad (a) Derelict jetty on Cedros beach. (b) Cedros beach with fishing vessels (c) Coast Guard jetty.

Region 8

Quantitative surveys were done at the Port of Scarborough and the jetties at Charlotteville, Parlatuvier and Roxborough. The Port of Scarborough (World Port Index No. 11700) is located on the windward side of Tobago, in Scarborough the capital of Tobago. The nature of its business is passenger, general cargo, fishing, yachting, and cruise ships (The Shipping Association of Trinidad and Tobago 2012). The other three jetties are associated with fish landing sites on the windward side of the island (**Plate 8**).

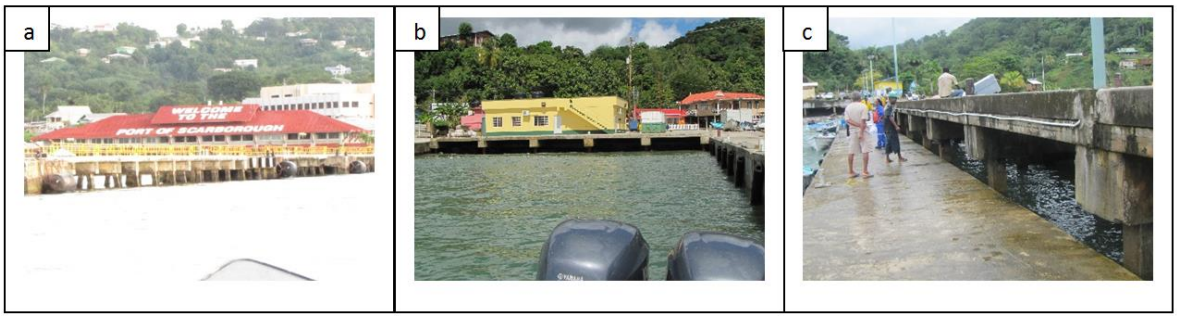

Plate 8: Sampling locations in Tobago (a) Wharf and Jetties at the Port of Scarborough (b) Jetty and pier pilings at Roxborough (c) Jetty and pier pilings at Charlotteville

3.3 Community Structure

Four (4) main habitat types were initially selectedfor comparison based on Agard *et al*. (1992). They were:

1. a large mussel bed in the intertidal region at Cedros on the southwest coast of Trinidad;
2. pier pilings at the Alcoa bauxite transhipment port facility on the north western peninsula of Trinidad;
3. mangrove prop roots at Caroni Swamp;
4. concrete intake water channel at Yara Ltd from the Point Lisas industrial estate on the central west coast of Trinidad.

The absence of the large mussel bed at Cedros and a smaller one at Claxton Bay initially located in 2000 (perscomm. R. Juman, Institute of Marine Affairs), the limited presence of the green mussel on mangrove prop roots (only one individual was found in the Caroni Swamp during the survey) and the inability to get final permission from the management of Yara Ltd. to sample the man-made channel, resulted in a re-focus of the community structure of pier pilings across its natural range in Trinidad.

3.3.1 Sampling strategy

The sampling strategy for a quantitative assessment of the community of pier pilings was adapted from the Centre for Research on Invasive Marine Pests (CRIMP) Protocol (Hewitt and Martin 2001). Three 0.1 m^2 (0.33 m x 0.33 m) quadrat were originally designed at 0.5 m. 3.0 m and 7.0 m depth (from the high water mark) (**Figure 3**). After preliminary testing of the apparatus at the ALCOA jetty, due to the narrow pilings, the quadrat was reduced to half its width. Four to nine quadrats from 0.5 m, 3.0 m and 7.0 m depth zones were sampled depending on the length of the pilings and number of adjacent jetties. With the exception of ALCOA, all other samplings were conducted at the 0.5 m and 3.0 m depth because of the restricted visibility of the seawater. Scrapings of the encrusting fauna were taken from the exterior wall of the piling and collected in the attached bag and later brought to the surface (**Plate 9**). Samples were placed in plastic buckets on ice and brought back to the lab, placed in a chiller (4 ^oC) overnight for processing the next day.

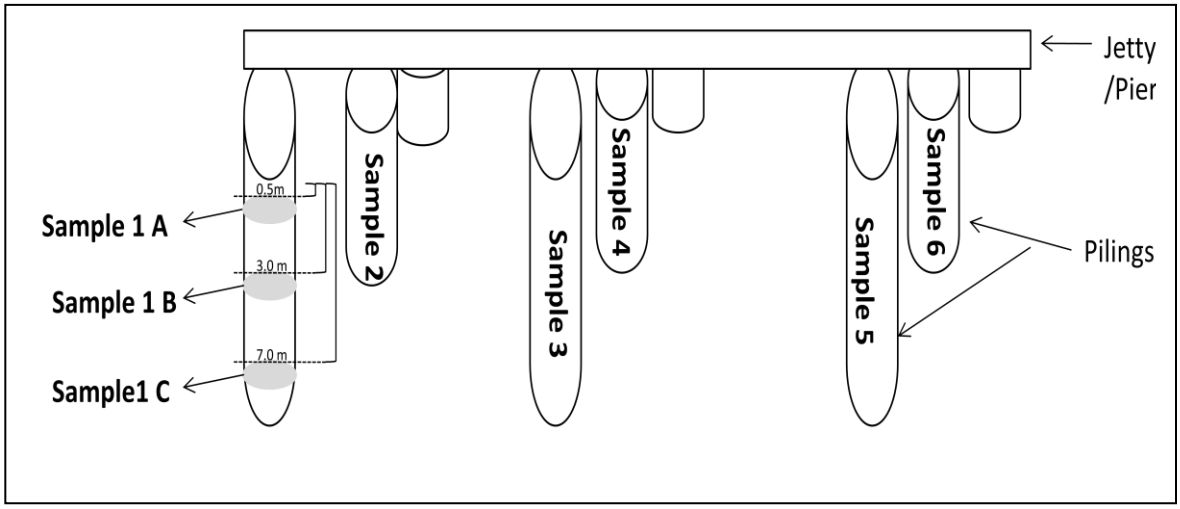

Figure 3: Sampling strategy for pier pilings adapted from the CRIMP Protocol by Hewitt and Martin (2001)

Plate 9: Sample collection for community structure of pier pilings from Trinidad and Tobago. (a) Diving team (b-f) Quadrat sampling at designated depths on pier pilings. (g-h) Scraping encrusting fauna at 0.5 m and 5.0 m depths (i) *Pernaviridis* and associated fauna in situ on pier pilings (j-l) Collecting *P. viridis* and associated fauna of pier pilings

Mangrove Prop Roots

Seventeen (17) areas within the Caroni Swamp bordered by the Blue River and the Madame Española River as well as from the sea inwards to the more riverine conditions of the North/South Drainage were sampled. Prop roots at all of these locations were examined focusing on the larger roots which had a greater extent of encrusting organisms Destructing sampling was carried out where the root was cut just above the high water mark and brought to the lab for processing.

Rocky outcrops/rockwall facings

These habitats were samples on the north coast, the northwestern peninsula, San Fernando, Vessigny and Cedros. Qualitative visual surveys on the west coast were restricted at the 0.5 m depth because of the turbidity of the water. Quantitative surveys were done at the 0.5 m depth and occasionally, when the clarity of the water permitted, at the 1.0 m depth.

Sample Processing

Samples were roughly sorted separating hard- and soft-bodied organisms. Hard-bodied organisms were fixed in 4 % formalin and soft-bodied in 70 % alcohol for later processing. Those samples stored in 4 % formalin (less than one week) were rinsed free of formalin over 24 hrs and stored in 70 % alcohol. Samples were later sort into broad taxonomic groups in most cases by families and stored in glass vials and containers (**Plate 10**).

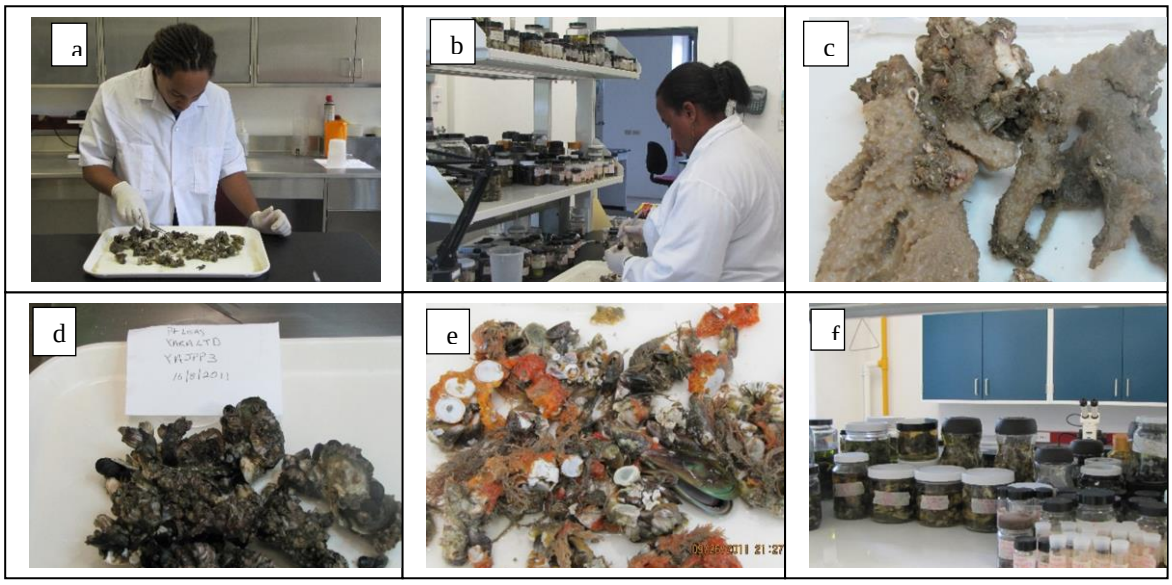

Plate 10: Sorting and identifying fauna collected from intertidal and sub-tidal zones on the north and west coasts of Trinidad. (a-b) sorting and identify samples (c) samples dominated by sponges (d) sample mainly of oysters and barnacles (f) sample containing the green mussel (f) sorted samples into major taxonomic groupings

Taxonomic Identification

Taxonomic identification was done with the use of published and online keys as well as local taxonomic experts. For the polychaetes, Dr. Judith Gobin, Senior Lecturer, The University of West Indies, St Augustine provided her expertise as well as the use of taxonomic keys by Uebelacker and Johnson (1984) and Hussain Ahmed (2011). Molluscs were identified using Sutty (1990), Abbott and Morris (1995), Kaplan (1988), Jones and Jones (2005) and Tunnel *et al.* (2010). Arthropods (barnacles and amphipods), using (Bacon *et. al.* 1984) Farrapeira (2008), Agard, and Barnard (1986), Phillips (2006) and Thomas (1993). Sponges, ascidians and echinoderms were identified from Hubbard (1982) Rocha *et al.* 2(012), Cole (2012) and Borrero-Perez *et al.*(2008). All samples were identified down to the lowest taxonomic level as far as possible.

3.4 Analysis of data

For all sample stations at each site, counts of individuals were made for each taxonomic group. The data was then pooled for each depth per site and raised to express the counts as density per m^2. Additionally, adjacent sites were further grouped into regions as described in **Table 1**. Pier pilings at the four sampling locations in Tobago were grouped into one region, due to insufficient replicates at Roxborough, Palatuvier and Charlotteville as a result of rough sea conditions making visibility poor, and sampling difficult.

Comparison of the community structure were made using the biological indices of Species Richness, Simpson Index, Shannon Weiner and Evenness.

The community structure of the various regions were further analysed by comparing the taxonomic groups using the Bray Curtis Similarity Index, Cluster Analysis and resulting Multi-Dimensional Scaling (MDS) Plots using Primer-E Version 6.1.16.

Condition Index of Pernaviridis

The wet or dry weight of the mussel meat reflect the mussel general 'condition (Vakily 1989) and Yildiz *et al.* (2006) noted that is useful to both researchers and mussel growers for predicting good environmental condition for growth. The condition of mussel depend on factors such as food availability, temperature, salinity and metabolic activities suchas growth and reproduction. A condition index (C.I.) for all the green mussels collected from the survey (both qualitative and quantitative samples) was thus developed.

They first separated from the rest of the sample, placed on ice in the field, brought back to the laboratory and stored in a chiller overnight at 4 ^{o}C. Each mussel was then scraped clean of encrusting organisms, wet weight (g) taken of the whole mussel and the length, width and height of the shell was recorded using a pair of vernier callipers (±0.05 mm) (**Figure 4**). The mussel was sexed based on macroscopic examination; white gonad for male (within the gill and body

tissue) and orange for female as similarly described by (Yap *et al.* 1979). None of the mussels with developed gonads shown any sign of spawning which would have affect the overall C.I.

To obtain the tissue dry weight, the wet tissue from each mussel was placed in pre-weighed containers and placed in a convection oven for 48 hrs at 60 ^{o}C. The containers with the dried tissues were then cooled in a dessicator to a constant weight and then weighed using a Sartorius Balance (±0.001 g). To calculate the shell cavity volume of the mussel the method by Kishore (1993) was used. Cling wrap was used to cover one half of each shell, two holes were made at the anterior end of the valve close to the umbo, water was introduced into the shell using 10 ml and 1.0 ml syringes, the second hole facilitated the displacement of air as the cavity was filled with water. Since the mussel shells are equivalve, the total shell cavity volume was thus twice that of each valve.

The Condition Index (CI) was then calculated using the following equation based on Walne (1970):

$$\text{Dry tissue(g)} \div \text{Shell cavity volume (ml)} \times 1000$$

Tukey Honestly Significant Different (HSD) multiple comparison test (MINITAB 16) was used to compare the condition indices of the green mussel from the different regions.

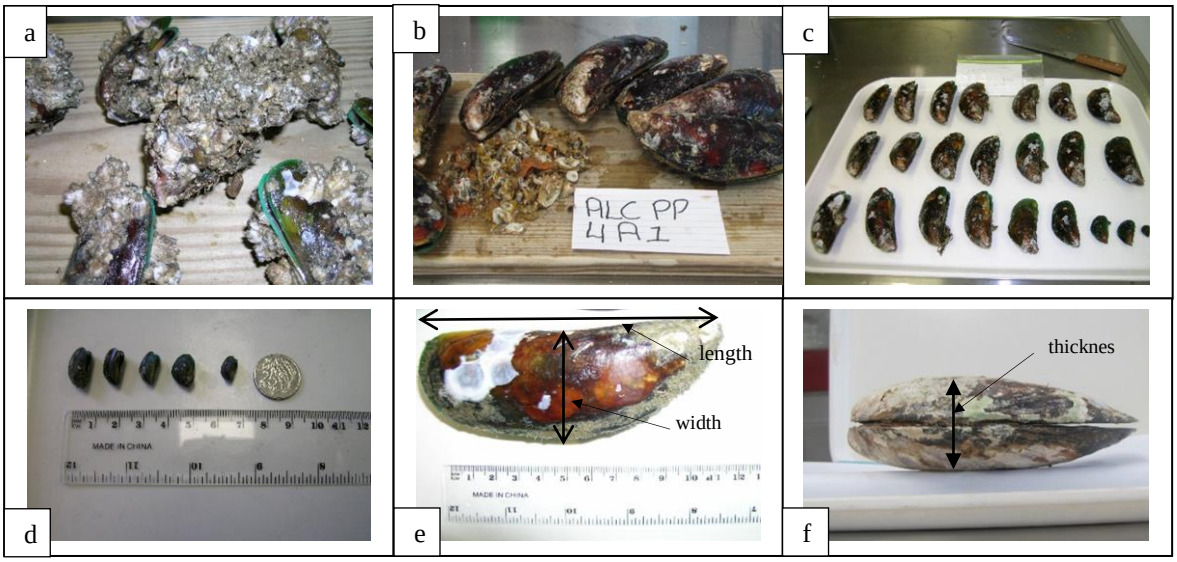

Figure 4: (a-f) Removal of encrusting organisms from the green mussel, *Pernaviridis* in preparation for morphometric measurements of the shell collected from jetty pilings on the northwestern peninsula of Trinidad

4.0 Results

4.1 Physico-chemical parameters

These physico-chemical results are summarised in **Table 2**. Sample stations were grouped by regions as described in **Table 1**.

Temperature ranged from 27.33 to 30.68 ^{o}C**,** with the highest mean temperature on the south coast at Cedros (Region 7) and the lowest 27.33 ^{o}C, on the north western peninsula (Region 2). The pH values showed little variation, ranging from 7.15-8.3.

Salinity range showed the highest degree of variation, ranging from 7.04 to 34.26 ppt. Most of the stations were sampled in the rainy season, with a salinity range of 7.04-34.26 ppt, whilst for the dry season (Regions 2 and 7), the range was 27.81-34.02. This variation can be accounted for largely due to the degree of riverine influence especially during the rainy season. Areas not under such an influence to any large extent such as the north western peninsula (Region 2), north Coast (Region 1) and Tobago (Region 8) reflect condition closer to oceanic waters. Within the Caroni Swamp, the salinity range 7.04-27.77 ppt illustrate almost freshwater conditions landward side further away from the Gulf of Paria. Low salinity; 10.10 ppt at the Godineau Swamp (Region 5) and in the nearshore waters of the Port of Point Lisas (16.04-16.12ppt) reflect the contribution of the Godineau River and Couva River respectively.

Dissolved oxygen (DO) ranged from 0.7 to 6.88 mg l^{-1}. Lower mean DO values were found in the Gulf of Paria from the north western peninsula to the south western peninsula when compared to the north coast of Trinidad (Region 1) and Tobago (Region 8). The Caroni Swamp (Region 3) accounted for the lowest mean values obtained 2.88 ± 0.30 mg l^{-1} and is indicative of general poor water quality. Within the Swamp, lower values 0.7-2.93 mg l^{-1} were recorded for areas more landward such as the North-South Drain, feeding basin, feeding tower and the mouths of No. 2 and No. 3 Drains. Dissolved oxygen on the seaward side of the Swamp ranged from 3.47- 4.92 mg l^{-1}.

Table 2: Summary of physiochemical data for sample sites grouped by regions. (parameters ranges are in parentheses). *physiochemical data shown for the 0.5 depth

Region	**Sample Area**	**Mean Temperature (^oC) ± s.e**	**Mean Dissolved Oxygen (mg l^{-1}) ± s.e**	**Mean Salinity ppt (‰) ± s.e**	**Mean pH ± s.e**	**Sample Size (n)**
Region 1	North Coast Trinidad	28.28 ± 0.91 (26.1-35.20)	5.79 ± 0.17 (5.08-6.70)	28.43 ± 1.64 (18.48-34.26)	7.52 ± 0.10 (6.98-7.88)	9
Region 2	Northwestern peninsula of Trinidad	27.33 ± 0.11 (26.10-28.40)	4.50 ± 0.05 (3.95-5.28)	32.18 ± 0.15 (27.81- 33.01)	8.38 ± 0.0 (8.17-8.53)	7
Region 3	Caroni Swamp	29.53 ± 0.25 (27.60-30.70)	2.88 ± 0.30 (0.70-4.92)	18.30 ± 1.86 (7.04-27.77)	7.40 ± 0.07 (6.97-7.88)	17
Region 4	Pt. Lisas – Jetties at port facilities	29.49 ± 0.32 (28.50-31.40)	4.82 ± 0.11 (4.41-5.60)	20.65 ± 1.62 (16.04-29.56)	7.50 ± 0.06 (7.30-7.89)	10
Region 5	San Fernando Wharf, Godineau Bridge	28.88 ± 0.14 (28.60-29.40)	3.59 ± 0.46 (2.60-5.69)	25.49 ± 3.85 (10.10-29.84)	7.15± 0.11 (6.83-7.47)	5
Region 6	La Brea, Vessigny*	29.55 ± 0.25 (29.30-29.80)	4.40 ± 0.31 (4.09-4.70)	18.22 ± 0.18 (18.04-18.40)	7.46 ± 0.02 (7.44-7.48)	2
Region 7	Cedros	30.68 ± 0.23 (30.40-31.60)	4.71 ± 0.06 (4.55-4.86)	33.78 ± 0.06 (33.70-34.02)	7.37 ± 0.03 (7.26-7.44)	3
Region 8	Tobago	28.64 ± 0.14 (28.10-29.00)	6.41 ± 0.10 (6.18-6.88)	31.17 ± 0.28 (29.70-31.86)	8.11 ± 0.01 (8.07-8.14)	9

*Vessigny sampled in the dry season and physio-chemical parameters; temperature (31.6^{o}C), salinity (34.02ppt), dissolved oxygen (4.55 mg L^{-1}) and pH (7.26) not included in mean values for Region 6

4.2 Distribution of *Pernaviridis*

The green mussel, *Pernaviridis,* is still primarily found on the west coast of Trinidad with some incursion on the north coast at Maracas Bay, Tyrico Bay and Las Cuevas Bay (**Figure 5**).

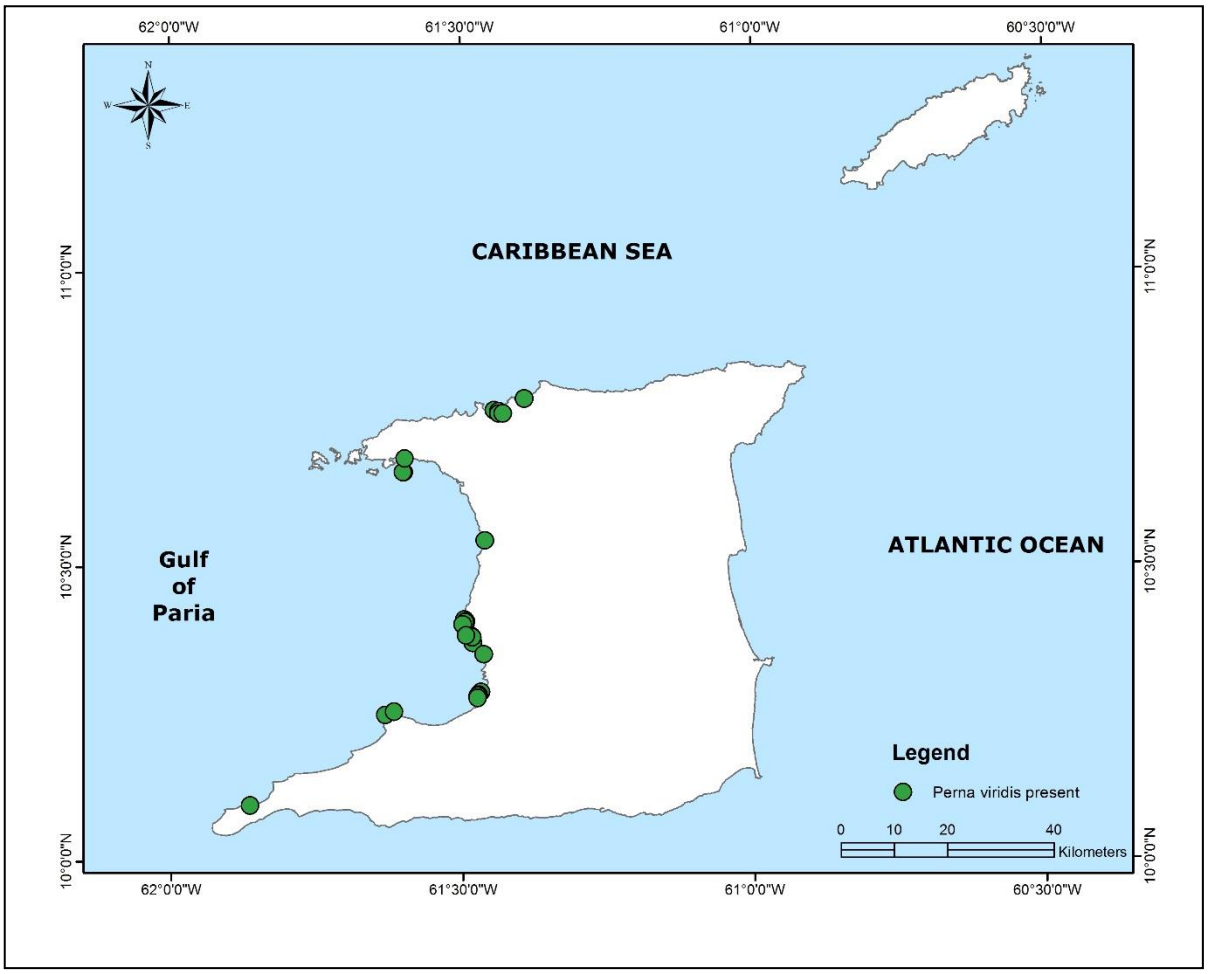

Figure 5: Distribution of the green mussel, *Pernaviridis* in Trinidad and Tobago in 2011

The mussel is vertically distributed both in the inter-tidal and sub-tidal regions. On the north coast, the green mussel is found intertidally among the crevices of rocks at Maracas Bay, Tyrico Bay and Las Cuevas Bay as well as on pier pilings of the jetty at Maracas Bay.

Along the west coast of Trinidad, from Chacachacare Island in the north to Cedros in the south, the mussel can be found in both the intertidal and sub-tidal regions. During the survey, no mussel beds were found and the population of the green mussels attached to red mangrove prop roots were so low that only one specimen was collected after an eight (8) hr search in the Caroni Swamp.

4.3 Morphometric Measurements of *Pernaviridis*

4.3.1 Length Frequency Distribution

For comparative purpose, the length frequencies of the green mussels collected were grouped into regions (**Table 1**). The length frequency distribution showed that Region 2 comprised only very large individuals > 89 mm, with no juveniles, indicating the absent of recruits. Additional samples (n=54) collected from another IMA research project in November 2012 was added to Region 2 data set and reflected in **Figure 6** as Region 2 (added samples). While this sampling obtained smaller green mussels and extended the size range for Region 2 to 52 – 194 mm from 89 – 194 mm, there still were no young recruits for the northwestern peninsula.

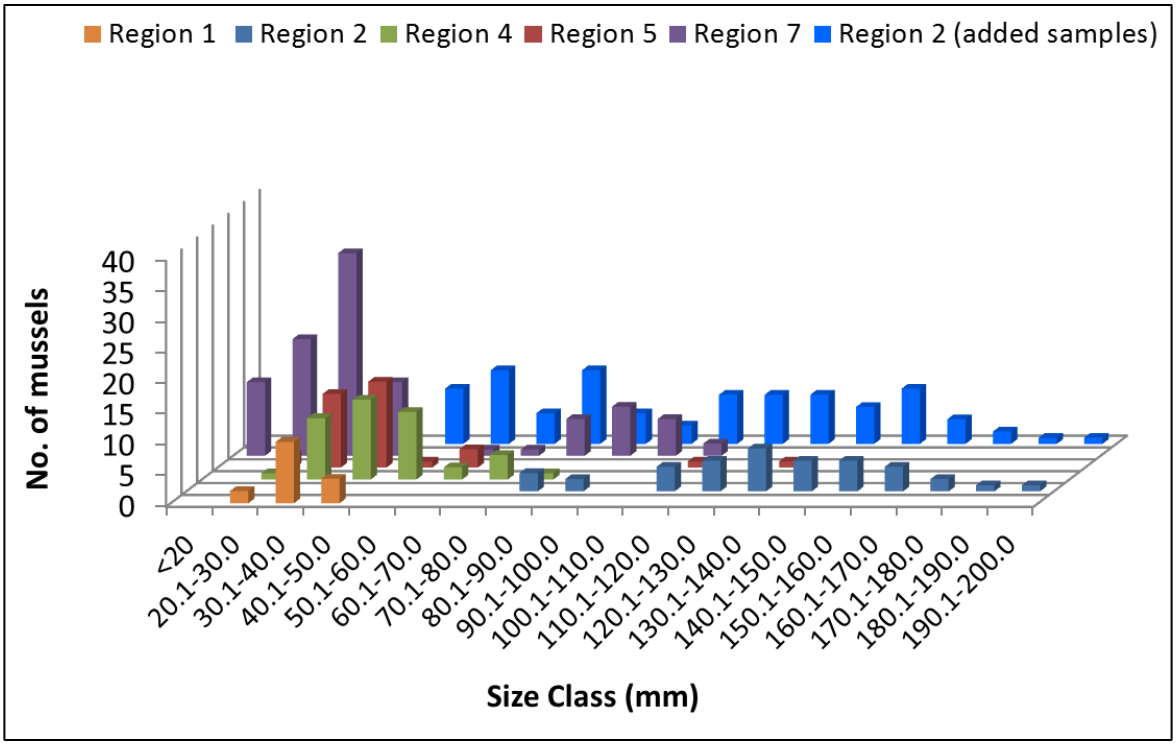

Figure 6: Length frequency distribution of *Pernaviridis* from pier pilings along the west coast of Trinidad

For the north coast, Region 1, based on the length frequency (small and therefore young individuals), the presence of the green mussel appear to be a recent establishment. For all other regions, the size range was wider with a high proportion of smaller individuals, indicative of active recruitment (**Figure 6**).

4.4 Sex Ratio

Table 3 shows the sex composition of the mussels. Macroscopic differences between the sexes were distinguishable up to 11.0 mm (at Cedros).

Table 3: Sex Ratio of the green mussel, *Pernaviridis* on the north and west coasts of Trinidad.

Region	**n (males)**	**n (females)**	**Indeterminate Sex**	**Sex ratio (M:F)**
Region 1	2	11	3	
Region 2	19	20		1:1.05
Region 3*		1		N/A
Region 4	27	15		1:0.56
Region 5	18	16		1:1.13
Region 6	2			
Region 7	51	38	11	1:1.34

Region 3 was not included in any analysis

Chi-square test of the regions 2, 4, 5 and 7 showed no bias (χ^2=0.026, 0.02, 3.43, 1.90, df = 2, p = 0.05), indicating that the sex ratio was not significantly different from 1:1, while for Region 1, the number of females outnumbered males, however, the sample size was much smaller(n = 13). Collectively, male:female sex ratio, 1:0.85, was not significant ((χ^2 = 1.33, df = 1, p=0.05).

4.5 Condition Index of *Pernaviridis*

Mean Condition Index (CI) comparison of the green mussel from region 2, 4, 5 and 7 (**Table 4**), showed that they were all significantly different from each other with Region 5 having the highest CI followed by Region 7, Region 4 then Region 2 (ANOVA, F=49.48, p < 0.001; Tukey test, p <0.05).

Table 4: Mean Condition Index for the green mussel on the northwestern peninsular and the west coast of Trinidad

Region	**Mean C.I ± S.E.**	**n**
Region 2	57.46 ± 6.61	39
Region 3		
Region 4	112.55 ± 1.29	29
Region 5	201.98 ± 14.73	39
Region 6		
Region 7	149.30 ± 5.46	81

4.6 Community Structure

Community structure of habitats associated with the *P. viridis*during both the quantitative and qualitative surveys were examined. Appendix 1 details the species found at the various sampling stations. Seventy-five (75) macrobenthic faunal species were collected in addition to *Pernaviridis*. In this ecological assessment, the species recorded comprised eight (8) phyla or major taxonomic groups – Annelida (polychaetes or marine worms), Arthropoda (shrimp, crabs, amphipods, isopods and barnacles), Echinodermata (brittle stars), Mollusca (snails and shells) Sipunculida (peanut worms), Tunicates (Sea squirts), Porifera (Sponges) and teleost fish. The dominant (in terms of number of individuals) phyla are the Mollusca (26%) and Arthropoda (61%) with bivalves within the Mollusca being very dominant at all sites sampled. Though *Pernaviridis* was the main focus of the study, one species of bivalve (*Brachidontesexustus*) was very dominant at most of the sites and at the varying depths sampled. Cirrepedia (barnacles) within the large Arthropoda group was very dominant at most of the sites sampled. Some of the other large phyla were Annelida with a variety of polychaetes being found at some of the sites. There were some phyla found only at specific sites such as sponges (Porifera) and corals (Cnidarians), which were only found in Chaguaramas and Tobago. No new or first record of any species was collected in this assessment.

A closer examination of the fauna associated with *P. viridis* on jetty pilings at the Alcoa jetty, within Region 2, historically a known region for the green mussels, showed how species varied at the three depths sampled (**Figure 7**). The biodiversity was greater at depths 0.5 m and 3.0 m, with 27 spp. and 24 spp. respectively and less at 7.0 m. with 18 spp. present. The density of *P. viridis* at 0.5 m and 3.0 m depth were the same, 36 individuals per m^{-2} and less, 24 m^{-2} at 7.0 m depth. One species of barnacles, *Amphibalanusreticulatus*, dominated the intertidal zone

(0.5 m depth), but was absent at deeper depths. Another dominant group for this region was the brittle stars. The amphipod was ubiquitous among the three sampled depths, but more so in the intertidal region. Apart from the amphipods, another soft-bodied group, the annelids, were more common above the 7 m depth (**Figure 7**).

On pier pilings of jetties at industrial plants on the central west coast of Trinidad (Region 4) only 0.5 m and 3.0 m depths were examined because of poor visibility. Similar to Region 2, the intertidal zone containing the 0.5 m depth had a higher number of species present, 34 when compared to 23 at 3.0 m (**Figure 8**). Dominant species again were the barnacles, bivalves and polychaete annelids. The density of *P. viridis* was 28 individuals per m^{-2} at the 0.5 m depth when compared to only one individual per m^{-2} at the 3.0 m depth.

The barnacles, *Amphibalanusreticulatus* was again the dominant species, but in Region 4, four other species of barnacle were present at both depths samples, *Amphibalanuseburneus*, *Chthamalusproteus*, *Amphibalanusamphitrite* and *Megabalanus* sp. Other bivalves present along with the green mussel were *Crassostrearhizophorae*, *lsognomonbicolor*, *Ostreolaequestris*, *Brachidontesexustus*, *Mytellafalcata*, *Pernaperna*, *Brachidontesdomingensis* and an unidentified species.

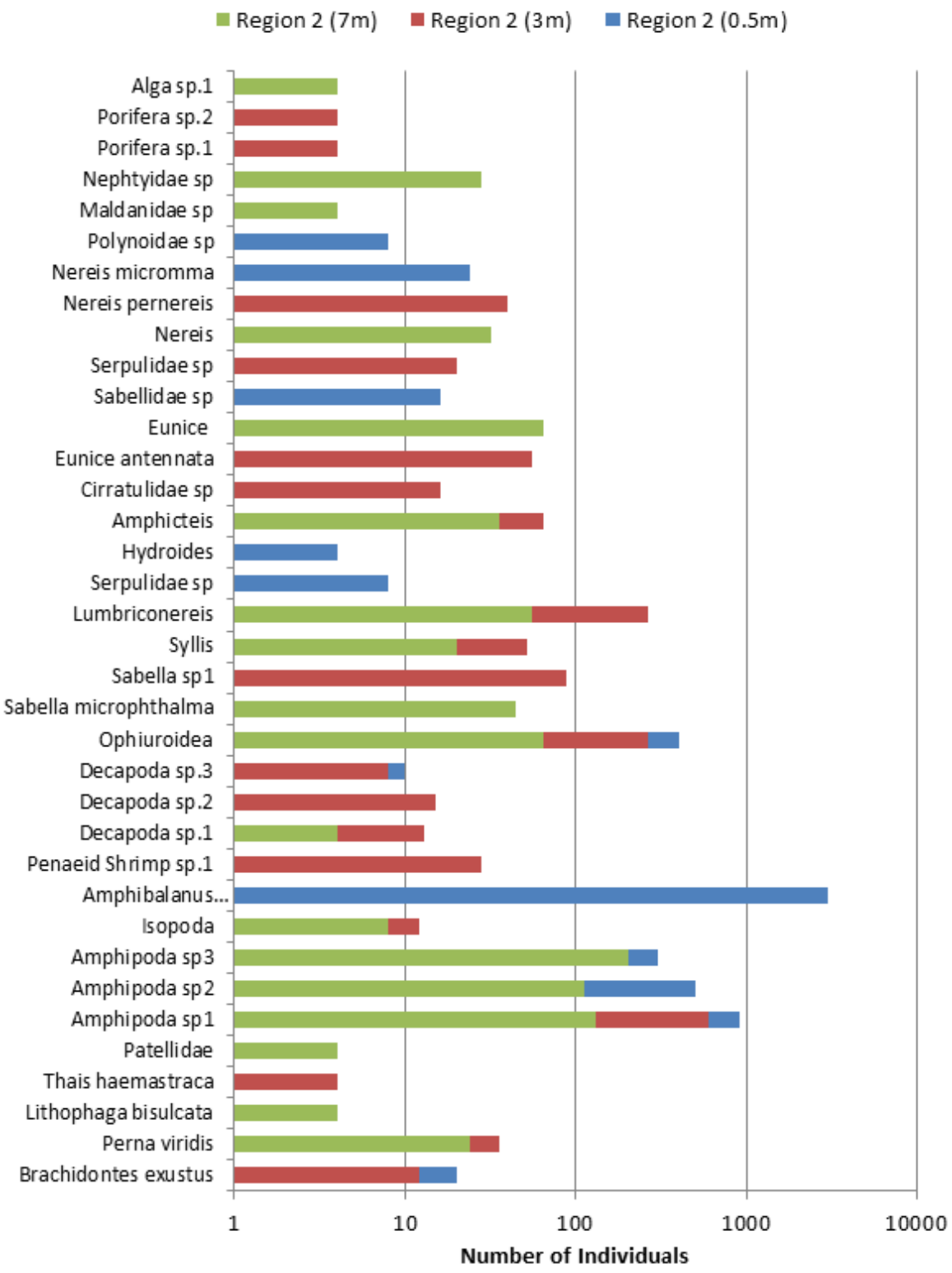

Figure 7: Community structure of the pier pilings associated with *Pernaviridis* at the Alcoa Jetty, Region 2) Chaguaramas, Trinidad

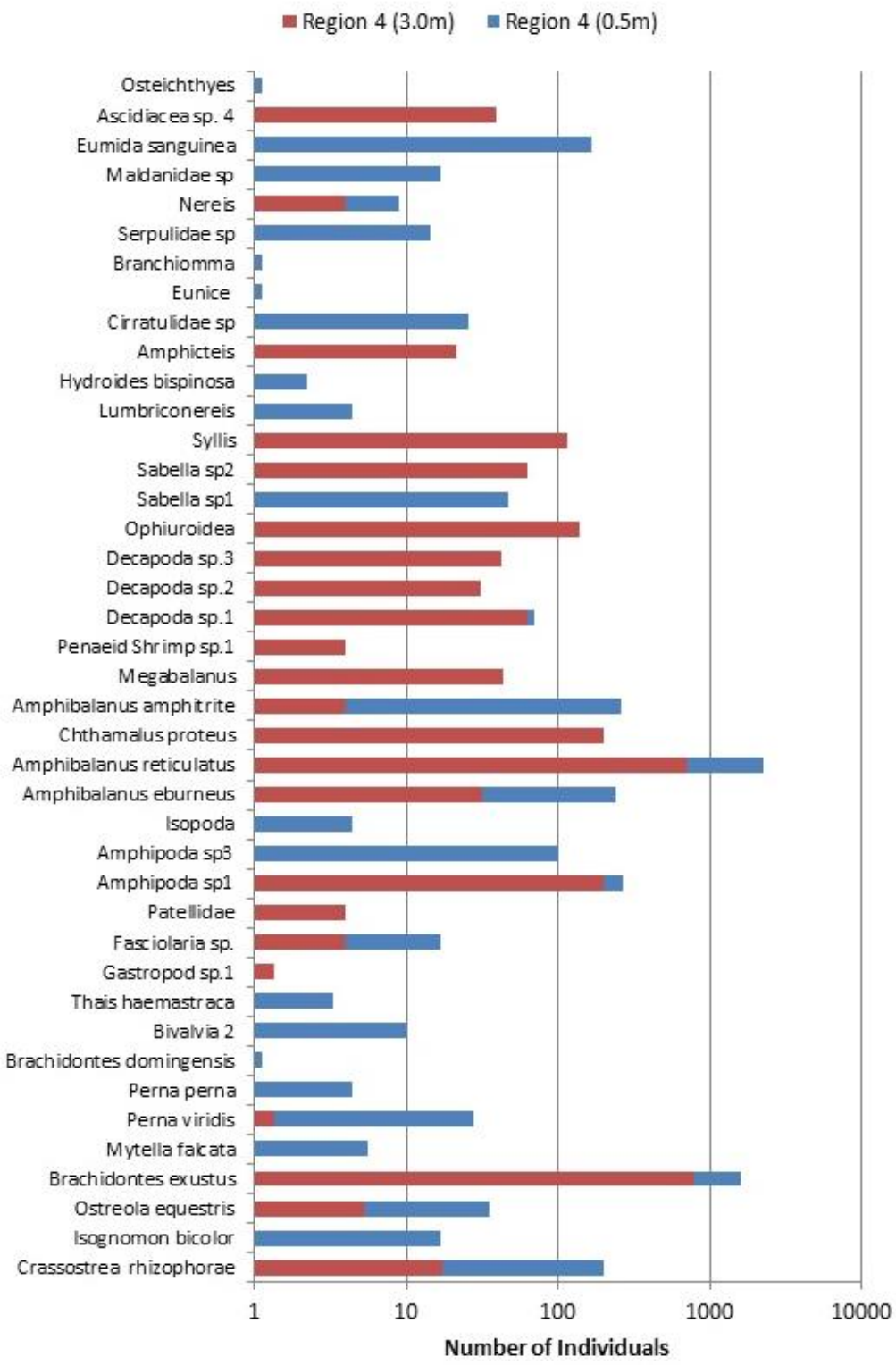

Figure 8: Community structure of the pier pilings associated with *Pernaviridis* at the Alcoa Jetty, Region 2) Chaguaramas, Trinidad

At San Fernando (Region 5), the number of species were less than half that found in Region 2 and Region 4, with more soft-bodied organisms such as annelids and ascidians found sub-tidally (3.0 m depth). The density of *P. viridis* was much greater at 170 individuals per m^{-2} when compared to Regions 2 and 4 (**Figure 9**).

The lowest densities of *P. viridis*, 3 individuals per m^{-2} at the 0.5 m depth with none present at the lower depth (**Figure 10**) were found at Region 6 –La Brea. Numerous large shells of the green mussel, however, were found on the bottom substrate of the pier pilings (**Plate 11**). The number of species and composition were similar to Region 5 but in this case, bivalves; *Crassostrearhizophorae* and *Brachidontesexustus*, were more dominant. The barnacle, *Amphibalanusreticulatus*, was also numerous, but unlike at the other regions, at Region 6, the density was higher subtidally than intertidally (**Figure 10**).

All pier pilings data from the Port of Scarborough and those associated with fish landing sites at Parlatuvier, Charlotteville and Roxborough in Tobago were combined and presented in **Figure 11**. In general, the number of species (50) found on pier pilings around Tobago (Region 8) was much higher than those from Trinidad with the barnacles; *Amphibalanusreticulatus* and *Tetraclitastalactifera*and the bivalves, the mangrove oyster, *Crassostrearhizophorae*, found exclusively at the Port of Scarborough) and the flat-tree oyster *lsognomonbicolor* being the dominant species.

To better examine the biodiversity across all regions, **Figure 12** and **13** detailed the total number of species, individuals and the resulting diversity indices. Region 2 and 4 have the highest number of individuals (**Figure 12**), but Region 8 had highest species richness (**Figure 13**). Better indices such as the Shannon Weiner Index and the Simpson Index which takes into consideration both the species richness and the species evenness (how close in numbers each species in this case each habitat is) shown in **Figure 13**, affirmed that Region 8 has the highest species richness and diversity indices.

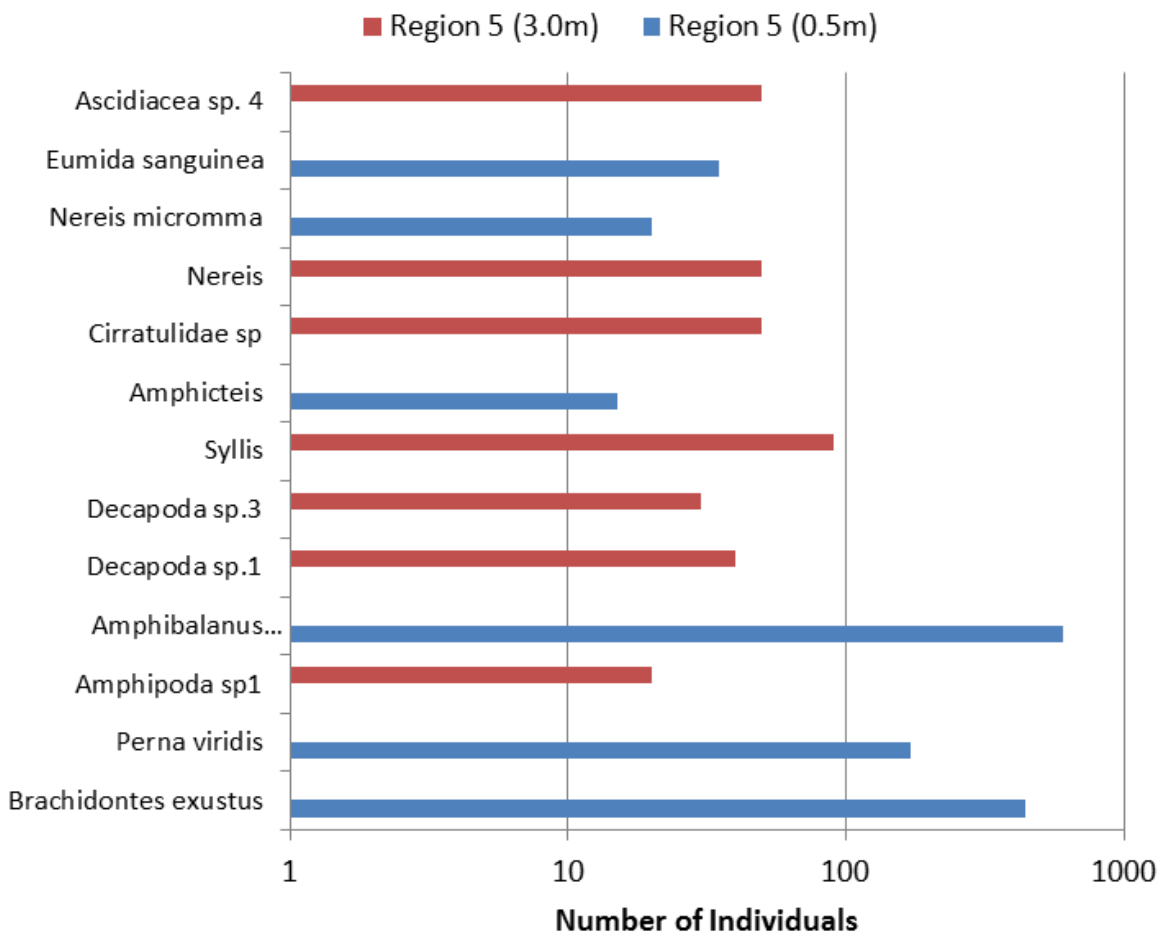

Figure 9: Community structure of the pier pilings associated with *Pernaviridis* at San Fernando (Region 5) on the west coast of Trinidad

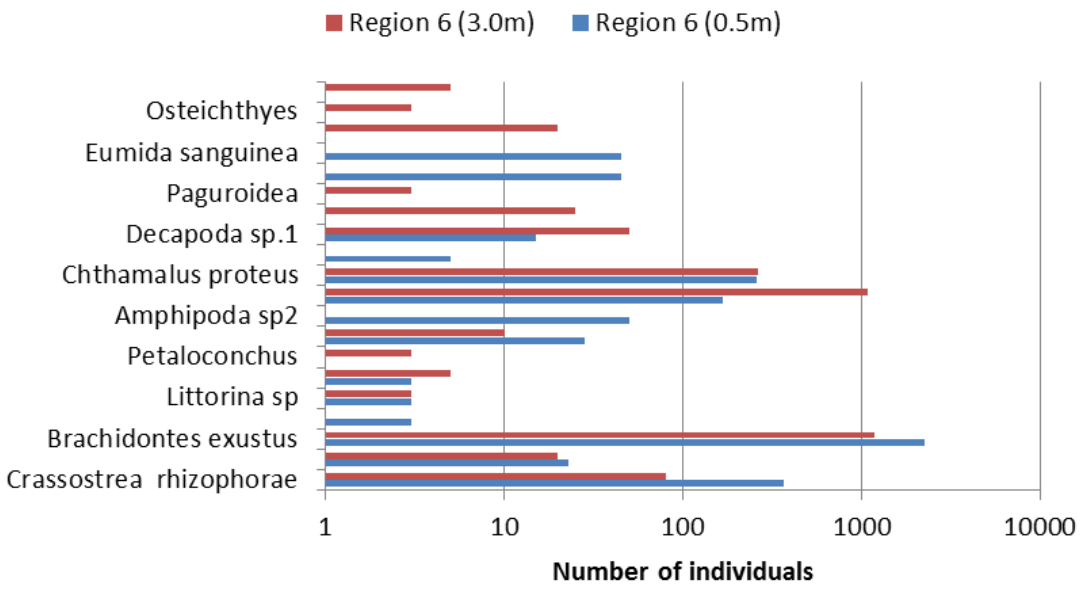

Figure 10: Community structure of the pier pilings associated with *Pernaviridis* at La Brea (Region 6) on the west coast of Trinidad

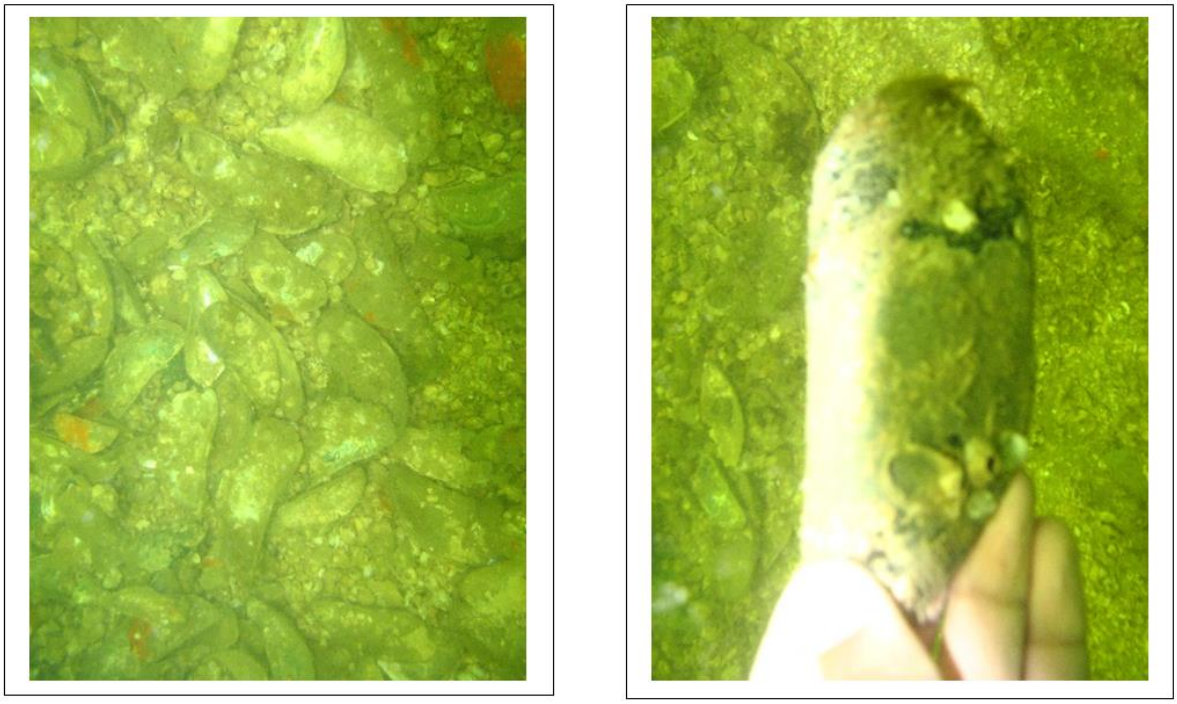

Plate 11: Shells of dead *Pernaviridis* on the bottom substrate of pier pilings at La Brea (Region 6) on the west coast of Trinidad

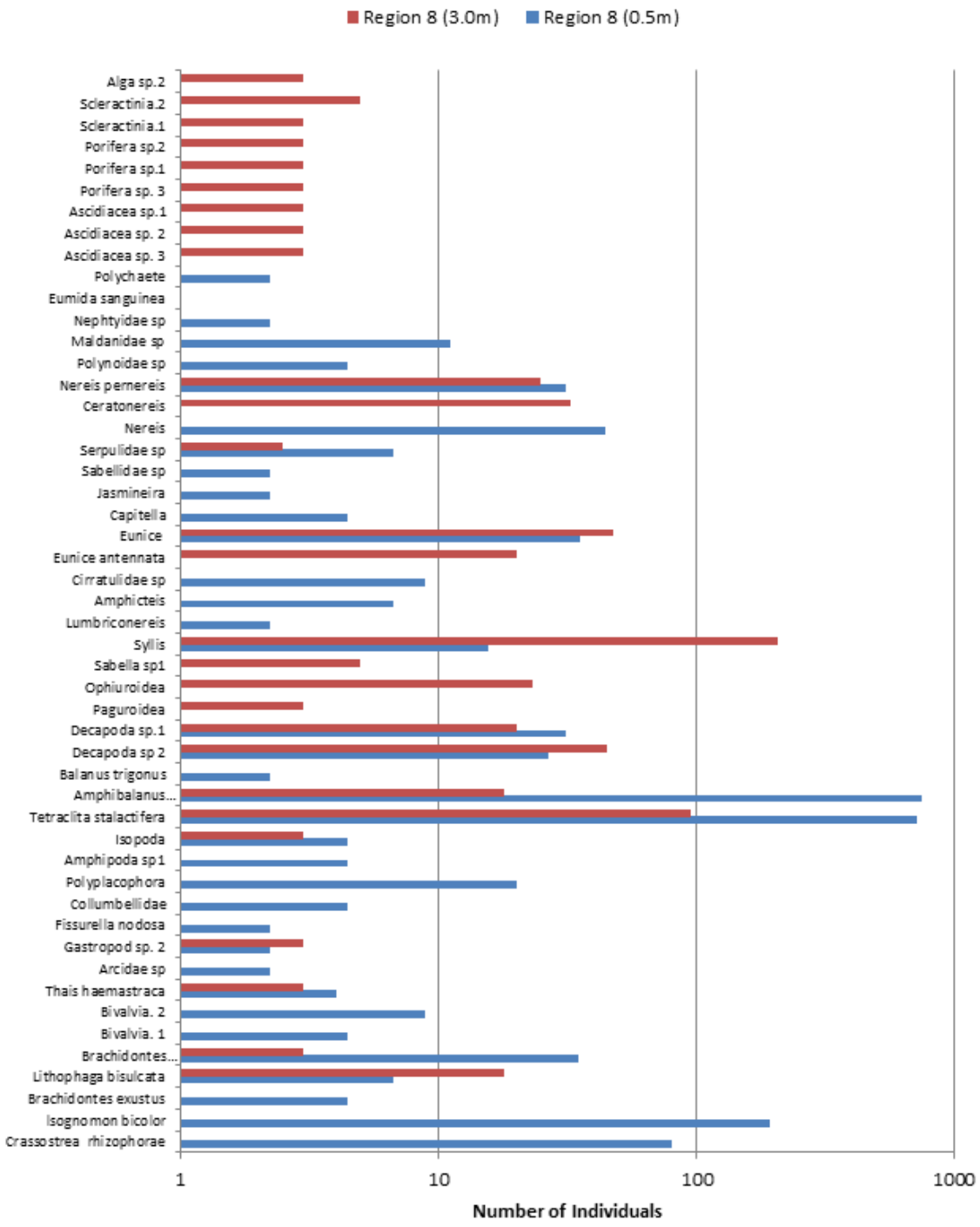

Figure 11: Community structure of the pier pilings in Tobago (Region 8) in Tobago

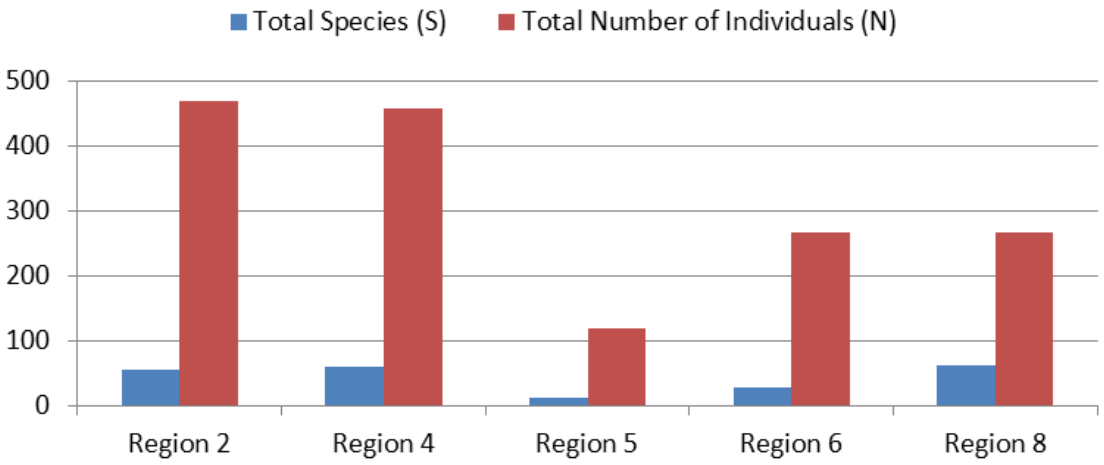

Figure 12: Total species and abundance for quantitative sampling on pier pilings in Trinidad and Tobago

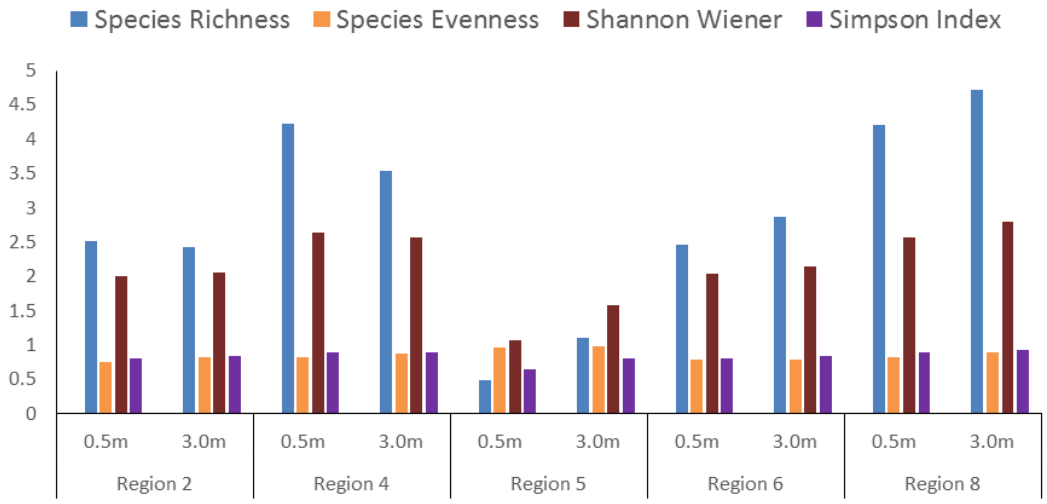

Figure 13: Biological diversity indices of communities on pier pilings in Trinidad and Tobago.

Further analysis using Multi-Dimensional Scaling (MDS) plot and the associated dendrogram showed the clustering or natural grouping of the sample sites from this baseline survey (**Figure 14** and **Figure 15**). Region 4 and Region 6 shows an expected close affinity given their similarity in environmental conditions at both 0.5 m and 3.0 m depths (located on the west coast close to riverine discharges) while Region 8 show its difference by separate and distinct clustering but with some association with Region 2 as the environmental conditions on the north coast of Trinidad and Tobago are more similar.

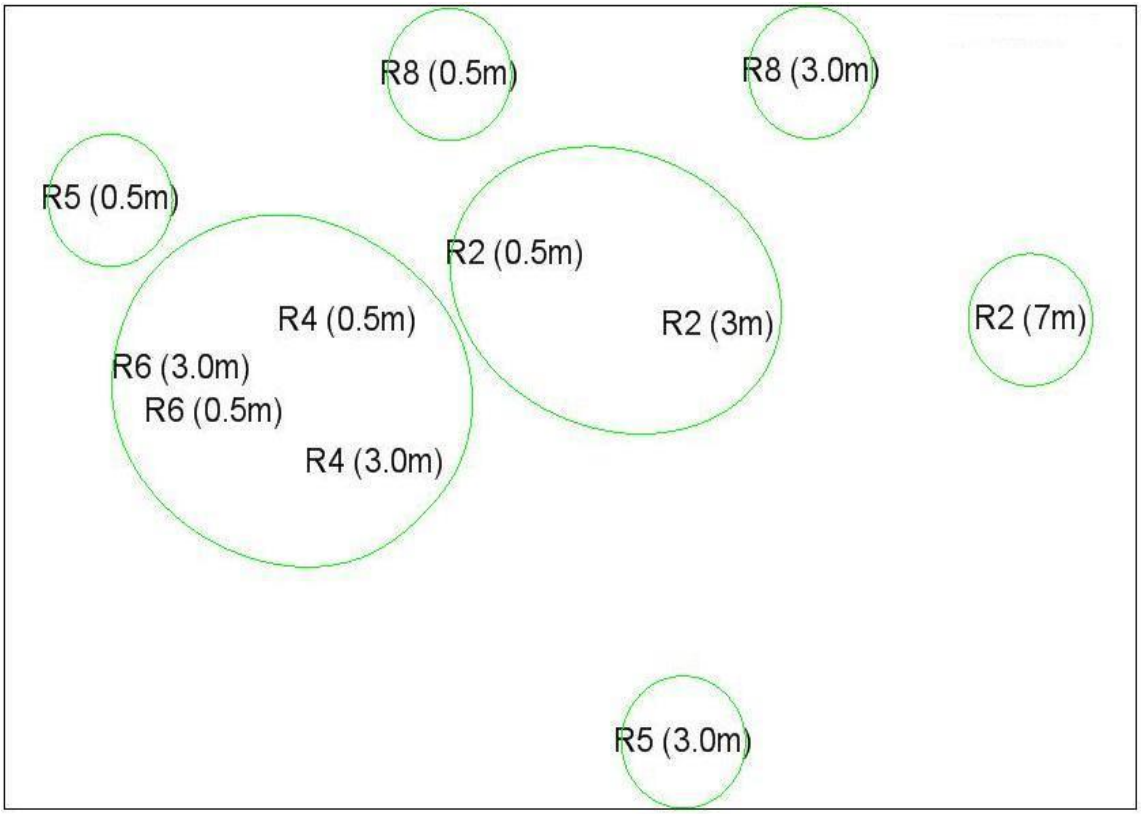

Figure 14: Multi-dimensional scaling (MDS) plot of faunal communities on pier pilings in Trinidad and Tobago

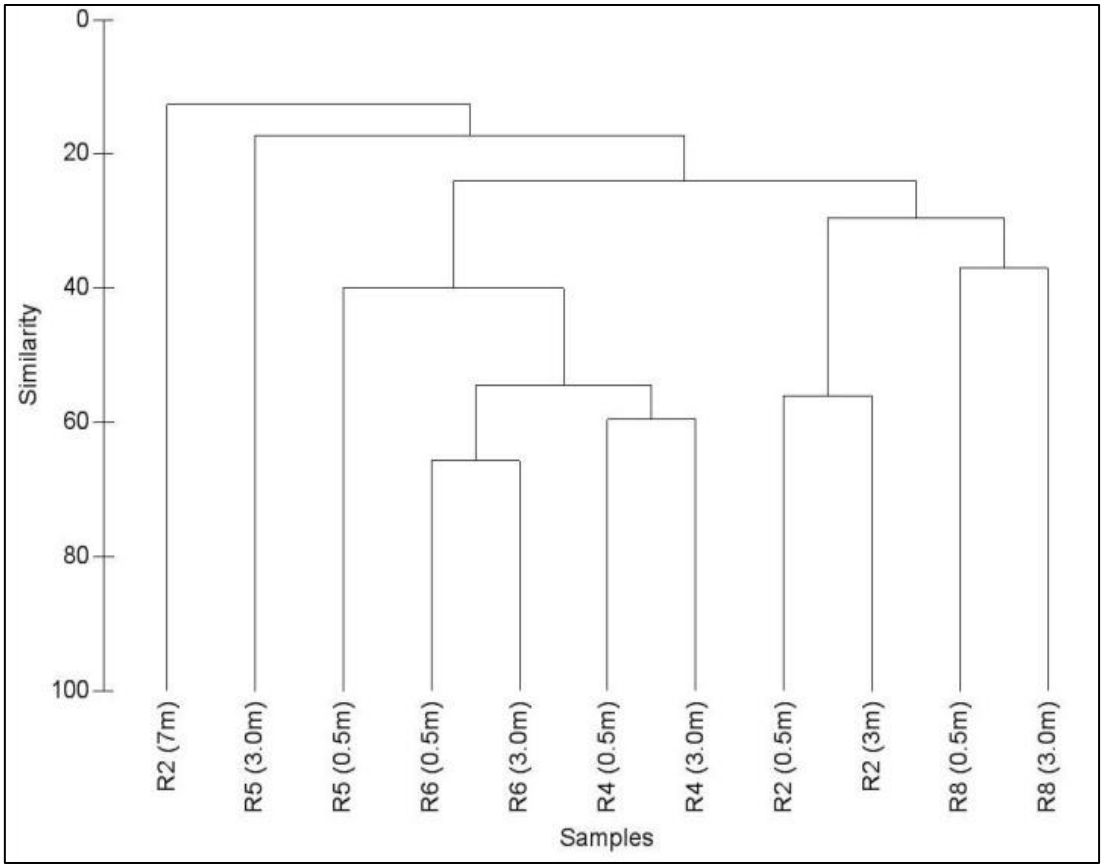

Figure 15: Dendrogram from MSD plot showing hierarchical clustering of sample regions for pier pilings fauna in Trinidad and Tobago

Areas for qualitative sampling were done at Carenage, Godineau, Swamp, Cedros and on the North Coast of Trinidad from Maracas Bay to La Fillette Bay. Nineteen species including *P. viridis* were found for a range of habitats, pier pilings, mangrove prop roots and rock wall facings. None, however, were different from those found during the quantitative sampling. *P. viridis* was more common at Cedros than all of the sites sampled (**Figure 6**). No known natural predators were identified from the organisms identified and none of the *P. viridis* samples had sign of boring on their shells indicative of predation.

5.0 DISCUSSION

The current distribution pattern of *Pernaviridis* in Trinidad and Tobago (**Figure 15**) remains relatively unchanged when compared to Agard *et al.* (1992), and twenty seven (27) years since its introduction in the vicinity of the Port of Point Lisas, on the west coast of Trinidad One exception is its incursion of the north coast, and given the size of the individuals and numbers (**Figure 16**), it seems to suggest that it is a recent introduction. The green mussel is still absent on the east and south coasts of Trinidad and in Tobago.

No prior quantitative assessment exist which can be used to compare past and present population size or densities. However, comparison with qualitative assessment by Agard *et al.* (1992), anecdotal information from biologists, fishers and shellfish harvesters with that of this study, indicate that the population of the green mussel has declined significantly. In the 1990s when large numbers were present in Trinidad, they were collected by traditional oyster collectors and sold to households and commercially to restaurants (Kishore 2013). **Plate 12** illustrates the density of the green mussels on red mangrove prop roots in the seaward drainages of the Caroni Swamp on the west coast of Trinidad in 1990. It was noted by Guppy (2001) that in 2000, consumption of the green mussel was still increasing. In the 1990s, main sites for collection were from jetties at port facilities located at ALCOA, La Brea (now defunct), Caroni Swamp and mangroves at Claxton Bay and Yara Ltd. (then Hydro-Agri Ltd.) (Kishore 2013). Phillip *et al.* (2001) observed that the reproductive potential of the green mussel had declined, coincidence with a 'dieback' of the green mussel in the Gulf of Paria.

Plate 12: Green mussels on mangrove prop roots in Caroni Swamp. Photo taken by John Agard circa 1990

This rapid decline of the invasive green mussel has also been noted in Cuba where it was first observed in Cienfuegos Bay in 2005. During a 22-month sampling period from 2011, the population of the green mussel declined dramatically. Among the suggested reasons for this decline were mortality resulting from stressful environmental conditions and removal through harvest, both coupled with low recruitment rates (Lopeztegui-Castillo *et al*. 2014). A similar decline was also observed in Tampa Bay, Florida in 2002-2003 where the winter of 2002-2003 killed most of the intertidal green mussels. Subtidal winter mortality of mussels was observed in northeast Florida and Georgia (Baker *et al*. 2004). In this study, lower thermal limit under laboratory conditions for the green mussel was 10-12 ^{o}C.

*Pernaviridis*are able to survive in estuarine habitats which have normal fluctuations in salinity (27-33 ‰) with an optimal preference of 30 ‰ and can tolerate salinities as low as 20 ‰, with a lethal salinity at below 5 ‰ (Rajagopal *et al.* 2006 and Vakily 1989). In the Atlantic, lower salinity limits are observed with the green mussel able to survive at 14 ‰ in Tampa Bay, Florida (Baker *et al*. 2004). This also seems to be the case for Trinidad with *P. viridis* found in regions of the Caroni Swamp with a salinity of 17 ‰ on the west coast of Trinidad. Additionally, higher salinity tolerance of up to 38 ‰ in the Gulf of Paria, Trinidad has been found by Phillip *et al.* (2001). Therefore, for Trinidad, the salinity tolerance range of *P. viridis* appear to be extended.

The water temperature range around Trinidad and Tobago in this study was 27.33 to 30.68 ^{o}C as compared to 24-27 ^{o}C (Kenny and Bacon 1983), and 27.6-29.7 ^{o}C (Nelson, 2014) and for the Gulf of Paria of 25-31 ^{o}C (Phillip *et al.* 2001). Barring differences due to seasonality, this temperature range fell well within the optimum temperature range of 15 –32.5 ^{o}C (Rajagopal *et al.* 2006) for *P. viridis*. Nelson (2014) noted that the temperature recorded off the north coast was lower than the other coasts, and suggested that the difference may be attributed in part to the land-based runoff/ discharge from the Orinoco River in South America that affect the southern and western parts of the island. This study also noted lower water temperatures for the north coast of Trinidad similarly conducted during the wet season by Nelson (2014), and as all sampled areas were nearshore, this may be more due to larger riverine discharges from local rivers that flow into the west coast of Trinidad where *P. viridis* was found. The higher condition indices for *P. viridis* at Region 5 (San Fernando), Region 7 (Cedros) and Region 4 (Pt. Lisas and Claxton Bay) seems to supports this, in addition to the nearby presence of mangrove systems which provide large supplies of detritus.

The dissolved oxygen (DO) range of 0.7 to 6.88 mg l^{-1} and in particular on the west coast in this study, was even lower (even if the landward side of the Caroni Swamp is excluded) ranging from 3.71-5.28 mg l^{-1} when compared to 5.10- 6.66 mg l^{-1} as was found by Nelson (2014). This may be as also suggested by Nelson (2014), that the west coast is subjected to greater quantity of organic pollutants from sewage, agricultural runoff and food waste; which would

utilize oxygen in the water during decomposition. The Gulf of Paria is also semi-enclosed and more sheltered when compared with the other sampling areas resulting in less aeration of this water body.

Nevertheless, the physico-chemical environmental conditions recorded during the survey are within the tolerance range of *P. viridis* with respect to salinity, temperature and dissolved oxygen as detailed by Vakily (1989), Rajagopal *et al.* (2006), Benson *et al.* (2001) and (Baker *et al.* 2004) and so demonstrate no limitations on its distribution. Even more so, based on Phillip *et al.* (2001) *P. viridis* in the Gulf of Paria, Trinidad has been able to survive in salinity of 38 ‰, well outside of its optimal preference of 30 ‰ (Rajagopal *et al.* (2006).

Little is known about the biodiversity associated with *P. viridis* and most observations are based on its ability to out-compete or crowd out organisms which occupy similar space such as oysters and other native mussels (Firth *et al.* 2011). Such competition with oysters have been noted by Barker *et al.* (2007) in Tampa Bay, Florida and with *Pernaperna* in in Venezuela (Rylander 1996). It is well documented that mussels provide refuge and habitat for a wide variety of associated organisms (Seed 1996) and that this function can vary with size of the mussels (O'Connor and Crowe 2007). Due to differences in size of individuals and patch complexity between oysters and mussels, it is likely that expansion of the green mussel will have potentially long term effects on diversity of epibiota and mobile fauna (Firth *et al.* 2011).

There is no prior assessment of community structure of the pier pilings to compare with this study and as such the ecological survey represents a baseline assessment of fauna associated with the green mussel for Trinidad and Tobago. It does provide the basis for comparison for any future research attempting to look at community structure changes if the population of the *P. viridis* increases again, or if any new marine alien invasive species is introduced.

Initial colonization by *P. viridis* in Trinidad was thought to have occurred with the introduction of a small number of individuals. The initial introduction of low numbers and small pool of genetic material could have given rise to a population bottleneck which may have resulted in a new population with a lower genetic diversity, when compared to the original population (Gobin *et al.* 2013). However, genetic analysis of tissue samples collected in 2009 compared with those collected in 1994 when it was first observed, indicated that there is no genetic variation in the population of *P. viridis* even more than 20 years after initial colonization. Additionally, Benson *et al.* (2001) showed that genetic analysis of tissue samples from Florida and Trinidad revealed that *P. viridis* from Florida were closely related to those from Trinidad, further suggesting that there was no evidence of a genetic bottleneck. This is further supported by Gilg e*t al.* (2012) who noted that genetic data supported the stepping-stone pattern of introduction of *P. viridis* from the native range to Trinidad, and from Trinidad to other locations in the Caribbean and United States. In addition, the lack of genetic variation in

introduced populations, no admixture (inbreeding) suggests a single original source population (Gilg e*t al.* 2012).

The pathway for such an introduction of *P. viridis* has been suggested as the shipping industry with the vectors being either ballast water or ship hull fouling (Agard *et al.* 1992, Benson *et al.* 2001, Buddo *et al.* 2003, Gilg e*t al.* 2012). Most of the sites where *P. viridis* is still found in Trinidad are predominately at port and port facilities, and as such it remains an opportunity for introduction and further expansion of the introduced range of *P. viridis.* Elaborating on this, there are eight (8) major ports included in the thirty (30) International Ship and Port Facility Security (ISPS) compliant port facilities in Trinidad and Tobago. With the exception of two port facilities in Tobago, 26 of the remaining 28 are located along the west coast of Trinidad, largely because of the sheltered nature of the Gulf of Paria, the degree of associated industrial development and available amenities which service these ports. (The Shipping Association of Trinidad and Tobago 2012).

The role the maritime industry play is of paramount importance because over 95% of the country's international trade is moved by maritime sea transport (Maritime Services Division 2010). In terms of vessel traffic, the main ports are at Point Lisas, Chaguaramas, Port of Spain, Point a Pierre and Point Fortin. As expected, the larger vessels are those exporting crude oil and Liquified Natural Gas (LNG) (Kishore 2013).

Based on the Electronic Pre-Arrival Notification (EPAN) database from the Ministry of National Security for 2011, 64 countries traded with Trinidad and Tobago, with 16 of these countries making $\geq$ 20 port of calls per year. These countries share similar ecological characteristics in that the trading countries either share the Atlantic Ocean or the Caribbean Sea making Trinidad and to a lesser extent Tobago, much more susceptible to introductions of marine IAS from these countries. More in common with Trinidad, are the trading partners of Brazil and Venezuela, whose marine environments are also influenced by the influx of the Orinoco and Amazon River systems (Kishore 2013).

There are no systems in place to monitor the introduction of marine IAS in Trinidad and Tobago. Detection of IAS in the marine environment, particularly at low densities, is difficult. Nationally, there are 15 pieces of legislation which provide for environmental protection however, none specifically addresses invasive alien species apart from where it can be labelled as a notifiable pest usually associated with agriculture (Kishore 2013). The proposed Shipping (Marine Pollution) Bill which will give effect to ballast water management will facilitate the implementation of the International Convention for the Control and Management of Ships' Ballast Water and Sediments (2004). Trinidad and Tobago acceded to the Ballast Water Management Convention (2004) in January 2012 (Kishore 2013).

Since shipping is a global industry, addressing the problem of introduction of marine IAS must be global in nature. Trinidad and Tobago has been part of the International Maritime Organisation (IMO) GloBallast Partnerships (GBP) which assisted developing countries to reduce the risk of ballast water mediated bio invasions and; prepare the countries for implementation of the IMO Ballast Water Management Convention, and compliance with its requirements at all levels.

As part of monitoring the implementation of national strategies to prevent the entry of marine IAS as well as to test the efficacy of any proposed legislation, it is recommended as a priority to implement biological baseline surveys of the port and port facilities of Trinidad and Tobago. This is considered urgent as the accession by Finland has triggered the entry into force of the Ballast Water Management (BWM) Convention on 8 September 2017 (IMO 2016) which would have major implications for trade via ships around the world including Trinidad and Tobago.

Although this present study has continued to note the drastic decline of the green mussel since its introduction, the economic assessment by Chase (2016) has reported that due to the substantial impact of the green mussel as a fouling organism at the height of its invasion, affected industrial firms had used *Pernaviridis* as a catalyst to put control mechanisms in place to help avoid heavy infestation of fouling organisms including any new marine IAS. The most effective management strategy used by firms in dealing with the marine invasive *Pernaviridis* along with other encrusting organisms such as barnacles and other smaller mussels, *Brachidontes* spp. was through the use of chemicals.

It is therefore suggested along with biological baseline monitoring of port and port facilities, the public awareness and education as well as stakeholders' specific strategies developed by this project such as the marine IAS DVD, posters and brochures, together with similar materials from the national public awareness campaign of the larger MTIASIC project be continued. Effective means of communication can be disseminated through the IMA's website and related ministries websites. Relationships with the industrial base of Trinidad and Tobago (i.e. ports and port facilities and industrial firms) built during this project should be maintained and through internal programs at these facilities, to continue to promote the awareness of the introduction of marine IAS and their impacts from the shipping industry as well as through other pathways.

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7.0 APPENDICES

Appendix 1: List of species collected at all sample sites during the survey for the green mussel, *Pernaviridis* in Trinidad and Tobago.

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		TRINIDAD																									TOBAGO						
Regions		R1					R2		R3		R4								R5		R6				R7		R8						
Common Name	***Species***	LAF 0.5m	BLA 0.5m	TRI 0.5m	LAC 0.5m	MAR 0.5m	AL 0.5m	AL 3m	CAR 0.5m	CAR 3m	CB 0.5m	PL 0.5m	PL 3m	SV 0.5m	SV 3m	YA 0.5m	YA 3m	GD 0.5m	SF 0.5m	SF 3m	LB 0.5m	LB 3m	BR 0.5m	BR 3m	CED 3m	CED 0.5m	SB 0.5m	SB 3m	CH 0.5m	CH 3m	RB 0.5m	PAL 0.5m	PAL 3m
Mangrove oyster	*Crassostrearhizophorae*	O	O	O	O	O	O	O	X	X	X	X	X	X	O	X	O	X	O	O	X	X	O	O	O	O	X	O	O	O	O	O	O
purse oyster	*Isognomonbicolor*	O	O	O	O	X	O	O		O	X	X	O	O	O	X	O	O	O	O	X	X	X	X	O	O	X	O	X	O	O	O	O
scorched mussels	*Brachidontesexustus*	O	O	O	O	O	X	X	X	O	X	X	O	X	X	X	X	O	X	O	X	X	X	X	O	X	O	O	X	O	O	O	O
Mok	*Mytellafalcata*	O	O	O	O	O	O	O	X	O	X	X	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O
Green mussel	*Pernaviridis*	O	O	O	O	O	X	X	O	X	X	X	O	O	O	X	X	O	X	O	X	O	O	O	X	X	O	O	O	O	O	O	O
mahagony date mussel	*Lithophagabisulcata*	O	O	O	O	O	X	X	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	X	X	O	O	O	O	O
false mussel	*Mytilopsisdomingensis**	O	X	O	O	X	O	O	X	X	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O
Hooked Mussel	*Ishediumrecrvum*	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	X	O	O	O	O	O	O	O
Mussel	*Bivalvia sp.1*	O	O	O	O	O	X	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	X	O	O	O	O	O	O
brown mussel	*Pernaperna*	X	O	X	X	X	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O
crested oyster	*Ostreaegyestris*	O	O	O	O	O	O	O	O	O	O	X	O	O	X	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O
Mussel	*Bivalvia sp.2*	O	O	O	O	O	O	X	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	X	X	O	O	O	O	O
juvenile mussel	*Bivalvia sp.3*	O	O	O	O	O	O	0	O	O	O	O	O	O	O	X	O	O	O	O	O	O	O	O	O	O	X	O	O	O	X	O	O
juvenile oyster	*Bivalvia sp.4*	O	O	O	O	O	O	0	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	X	O	O
Oysters	*Bivalvia sp.5*	O	O	O	O	O	O	0	O	O	O	X	O	O	O	O	O	O	O	O	O	O	O	O	O	O	X	O	O	O	O	O	O
Clam	*Bivalvia sp.6*	O	O	O	O	O	O	0	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	X	O	O	O	O
juvenile clam	*Bivalvia sp.7*	O	O	O	O	O	O	0	O	O	O	O	O	O	O	X	O	O	O	O	O	O	O	O	O	O	X	O	X	O	O	O	O
Triton shell 1	*Thais haemastraca*	O	O	O	O	X	O	X	O	O	O	X	O	O	O	O	O	O	O	O	O	O	O	O	O	X	O	X	O	O	O	O	O
Triton shell 2	Muricidae	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	X	O	O	X	O
Ark shell	Arcidae	O	O	O	O	X	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	X	O	O	O	O
Perwinkle	*Littorina*	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	X	X	O	O	O	O	O	O	O	O	O	O	O
Snail	*Gastropoda sp.1*	O	O	O	O	O	O	O	O	O	O	O	O	O	X	O	O	O	O	O	X	O	O	O	O	O	O	O	O	O	O	O	O
Tulip	*Gastropoda sp.2*	O	O	O	O	O	X	O	O	O	O	O	O	O	O	X	X	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O
Shell	*Gastropoda sp.3*	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	X	O	O	O	O	X
Limpet	Fissurellidae sp.2	X	O	O	O	O	O	O	O	O	O	O	X	O	X	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O
knobby limpet	*Fissurellanodosa*	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	X	O	O	O
Dovesnail	Collumbellidae	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	X	O	O	O	O	O
juvenile chiton	Polypacophora	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	X	O	X	O	O	O	O	O
Amphipods	*Amphipoda sp.1*	X	O	O	O	O	X	X	O	O	O	O	X	X	X	X	X	O	O	X	O	O	X	X	O	O	X	O	O	O	O	O	O
Isopod	***Isopoda sp.1***	O	O	O	O	O	X	X	O	O	O	O	O	O	O	X	O	O	O	O	O	O	O	O	O	O	X	O	O	X	O	O	O
water insect	Arthropoda sp.1	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O	O

		TRINIDAD																									TOBAGO						
Regions		R1					R2		R3		R4								R5		R6				R7		R8						
Common Name	***Species***	LAF 0.5m	BLA 0.5m	TRI 0.5m	LAC 0.5m	MAR 0.5m	AL 0.5m	AL 3m	CAR 0.5m	CAR 3m	CB 0.5m	PL 0.5m	PL 3m	SV 0.5m	SV 3m	YA 0.5m	YA 3m	GD 0.5m	SF 0.5m	SF 3m	LB 0.5m	LB 3m	BR 0.5m	BR 3m	CED 3m	CED 0.5m	SB 0.5m	SB 3m	CH 0.5m	CH 3m	RB 0.5m	PAL 0.5m	PAL 3m
Barnacles 1	Sessilia sp.1	o	o	o	o	o	x	o	o	o	x	x	o	x	x	x	x	o	x	o	x	x	x	x	o	o	x	x	x	o	x	o	o
Barnacles 2	Sessilia sp.2	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	x	o	o	o	x	o	x	x	o	o	o	o	o	o	o	o	o
Barnacles 3	Sessilia sp.3	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o
ribbed barnacles	*Tetraclitastalactifera*	o	o	o	o	o	o	o	o	o	o	o	o	o	0	o	o	o	o	o	o	o	o	o	o	o	x	o	x	o	x	x	x
Barnacles 5	*Chthamalusproteus*	o	o	o	o	o	o	o	o	o	o	x	o	x	x	x	o	x	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o
Barnacles 6	*Amphibalanusamphitrite*	o	o	o	o	o	o	o	x	x	o	o	x	o	o	o	o	x	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o
Ivory barnacle	*Amphibalanuseburneus*	o	o	o	o	o	o	o	o	o	o	x	o	o	x	x	x	o	o	o	o	o	o	o	o	x	o	o	o	o	o	o	o
barnacle 8	*Megabalanus sp.1*	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	x	o	o	o	o	o	o	o
barnacle 9	*Balanustrigonus*	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	x	o	o	o	o	o	o
Crabs	Decapod sp.1	o	o	o	o	o	x	x	x	x	o	x	x	x	x	x	x	x	o	x	x	x	o	o	o	o	x	x	o	o	o	o	o
Shrimp	*Penaeus sp.1*	o	o	o	o	o	x	x	o	o	o	o	o	o	o	o	x	o	o	o	x	o	o	o	o	o	x	x	x	x	o	o	x
hermit crab	*Decapod sp.2*	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	x
Brittle stars	Ophiuroidea sp.1	o	o	o	o	o	x	x	o	o	o	o	x	o	x	x	x	o	o	o	o	o	o	o	o	o	o	x	o	o	o	o	o
Seaworms	*Sabella microphthalma*	o	o	o	o	o	x	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o
Seaworms	*Sabella sp1*	o	o	o	o	o	x	x	o	o	x	x	o	x	x	x	x	o	o	o	o	o	o	o	o	o	o	x	o	o	o	o	o
Seaworms	*Sabella sp2*	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	x
Seaworms	*Syllis sp.1*	o	o	o	o	o	x	x	o	o	x	x	o	x	x	x	x	o	o	x	x	o	x	x	o	o	x	x	x	o	o	o	x
Seaworms	*Lumbriconereis sp.1*	o	o	o	o	o	x	x	o	o	x	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	x	o	o	o	o	o	o
Seaworms	*Janua sp.1*	o	o	o	o	o	o	o	o	o	o	x	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o
Seaworms	Serpulidae sp.1	o	o	o	o	o	x	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o
Seaworms	*Hydroidesbispinosa*	o	o	o	o	o	o	o	o	o	o	x	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o
Seaworms	*Hydroides sp.2*	o	o	o	o	o	x	x	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o
Seaworms	*Amphicteis sp.1*	o	o	o	o	o	x	x	o	o	o	o	o	o	o	x	x	o	x	o	o	o	o	o	o	o	x	o	o	o	o	x	o
Seaworms	Cirratulidae sp.1	o	o	o	o	o	x	x	o	o	x	x	o	x	o	o	o	o	o	x	o	o	o	o	o	o	x	o	o	o	o	x	o
Seaworms	*Eunice antennanta*	o	o	o	o	o	o	x	o	o	o	x	o	o	o	o	o	o	o	o	o	o	o	o	o	o	x	x	o	o	o	x	o
Seaworms	*Eunice sp.2*	o	o	o	o	o	x	x	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	x	x	o	o	o	o
Seaworms	Capitella sp.1	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	x	o
Seaworms	*Jasmineira sp.1*	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	x	o
Seaworms	Sabellidae sp.1	o	o	o	o	o	x	x	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	x	o	o	o	o
Seaworms	*Branchiomma sp.1*	o	o	o	o	o	o	o	o	o	o	x	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o
Seaworms	Serpulidae sp.1	o	o	o	o	o	o	x	o	o	o	x	o	o	o	o	o	o	o	o	o	o	o	o	o	o	x	x	o	o	o	o	o
Seaworms	*Nereis sp.1*	o	o	o	o	o	x	x	o	o	x	x	x	x	o	x	x	o	o	x	o	o	o	o	o	o	x	o	o	o	o	x	o
Seaworms	*Ceratonereis sp.1*	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	x	o	o	o	o	o

		TRINIDAD																									TOBAGO						
Regions		R1					R2		R3		R4								R5		R6				R7		R8						
Common Name	***Species***	LAF 0.5m	BLA 0.5m	TRI 0.5m	LAC 0.5m	MAR 0.5m	AL 0.5m	AL 3m	CAR 0.5m	CAR 3m	CB 0.5m	PL 0.5m	PL 3m	SV 0.5m	SV 3m	YA 0.5m	YA 3m	GD 0.5m	SF 0.5m	SF 3m	LB 0.5m	LB 3m	BR 0.5m	BR 3m	CED 3m	CED 0.5m	SB 0.5m	SB 3m	CH 0.5m	CH 3m	RB 0.5m	PAL 0.5m	PAL 3m
Seaworms	*Nereispernereis*	o	o	o	o	o	o	x	o	o	o	o	x	o	o	o	o	o	o	o	o	o	o	o	o	o	x	x	o	o	o	o	o
Seaworms	*Nereismicromma*	o	o	o	o	o	x	o	o	o	o	o	x	o	o	o	o	o	x	o	o	o	o	o	o	o	o	o	o	o	o	o	o
Seaworms	Polynoidae sp.1	o	o	o	o	o	x	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	x	o	o	o	o	o	o
Seaworms	Maldanidae sp.1	o	o	o	o	o	x	o	o	o	o	x	o	x	o	o	o	o	o	o	o	o	o	o	o	o	x	o	o	o	o	o	o
Seaworms	Nephtyidae sp.1	o	o	o	o	o	x	x	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	x	x	o	o	o	o
Seaworms	*Eumidasanguinea*	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	x	o	o	o	o
Seaworms	*Polychaete sp.1*	o	o	o	o	o	x	x	o	o	x	o	o	o	o	o	o	x	o	o	o	o	o	o	o	o	o	o	x	o	o	o	o
worm snail	*Petaloconchus sp.1*	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	x	o	o	o	o	o	o	o	o	o	o	o	o
Peanutworm	Sipuncula sp.1	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	o	x	x	o	o	o	o	o
Sea squirt	Ascidiacea	o	o	o	o	o	o	o	o	o	o	o	x	o	x	o	x	o	o	x	o	x	o	o	o	o	o	o	o	o	o	o	o
juvenile fish	Osteichthyes sp.1	o	o	o	o	o	o	o	o	o	o	x	x	o	o	x	o	o	o	o	o	x	o	o	o	o	o	o	o	o	o	o	o

LAF – la Fillette
BLA - Blanchisseuse
TRI - Tyrico
LAC – Las Cuevas
MAR - Maracas
AL - ALCOA
CAR - Carenage
CB – Claxton Bay
PI – Port of Point Lisas
SV – Savonetta
YA - YARA
GD - Godineau
SF - San Fernando
LB – La Brea
BR - Brighton
CED - Cedros
SB - Scarborough
CH - Charlottelville
RB - Roxborough
PL - Palatuvier

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