# SAMPLE COLLECTION METHODS AND PRACTICAL CONSIDERATIONS FOR INTRODUCED SPECIES' SURVEYS AT TROPICAL PORTS

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**FOREWORD** 

The School of Marine Biology and Aquaculture at James Cook University is a partner

in the CRC Reef Research Centre. It is interested in all aspects of the management of

the marine environment as well as maintaining a first class research capability. The

Ports Corporation of Queensland (PCQ) is responsible for the management and

operation of eight major ports on the Queensland coast and a major part of these

responsibilities is to maintain the environmental integrity of ecosystems in and

around these ports. As part of PCQ's EcoPorts program, systematic surveys are

underway to search for exotic marine organisms that have been imported through

international shipping movements.

In 1998, discussions between PCQ and CRC Reef identified there would be mutual

benefits if an arrangement could be put in place whereby CRC Reef could carry out

these surveys. PCQ would benefit by having the surveys carried out at a

commercially competitive rate and CRC Reef would acquire a taxonomic capability

that would enhance its research and teaching program. While PCQ commissioned

CRC Reef to carry out the work on a commercial basis, it recognised and strongly

supported the various research spin-offs and knowledge-base increases that would

accrue as a result of the collection and classification of large numbers of local

organisms.

It became clear that there were some particular considerations to be addressed to

ensure adequate sampling in tropical ports. These included such things as the high

diversity of organisms and sampling regimes, and not getting eaten by crocodiles. As

a result, this report has been written to document the requirements of sampling in

tropical ports so that a consistent approach can be taken that will allow temporal and

spatial continuity of data sets. This will have much wider applications than just the

sampling for exotic organisms, and both the Ports Corporation and the Marine

Biology Department are pleased to have been involved with this work.

**Derek Andrews** 

**Chief Executive Officer** 

**Ports Corporation of Queensland** 

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#### **EXECUTIVE SUMMARY**

There is growing worldwide recognition of the detrimental impact of marine pest species in inshore waters. Most of the documented marine pest species in Australian waters are believed to have been transported through mariculture and shipping activities. In response to this problem, the Australian Association of Ports and Marine Authorities and the CSIRO Centre for Research on Introduced Marine Species (CRIMP) developed terms of reference for a nationwide series of baseline port surveys. CRIMP has written a set of guidelines for sampling methods for these surveys (Hewitt and Martin 1996) to ensure that the results of the port surveys are comparable. The Ports Corporation of Queensland's environmental program (Ecoports) is presently undertaking biological surveys of all of its Queensland ports to determine native biodiversity and detect introduced marine species.

In comparison with temperate waters, little is known about the presence and impact of exotic marine species in tropical Australian waters. Baseline knowledge of both endemic flora and fauna and any introduced species at ports is essential for future monitoring and management of the potential problem of introductions. Queensland coastal waters, which include the Great Barrier Reef, are part of biologically diverse marine zone known as the central Indo-West Pacific province. Monitoring of introduced species at potential points of entry is an important part of managing and protecting this unique coastline.

The first tropical port surveys in Australia were conducted by CRIMP in Hay Point and Mackay in Queensland in 1998. Subsequently, the School of Marine Biology and Aquaculture at James Cook University, on behalf of CRC Reef Research Centre undertook surveys at Mourilyan Harbour, Abbot Point, Lucinda, Weipa and Karumba. For these surveys, we adopted the methods of Hewitt and Martin (1996) but found it was desirable to implement some modifications to allow for the biological and environmental characteristics of tropical ports. We feel that some modifications are required for tropical ports so that sampling methodologies are appropriate for these environments. This report addresses the need for guidelines specific to tropical port surveys. It discusses the characteristics of tropical inshore marine communities and their habitats that are relevant to port survey planning and

the sampling methods appropriate for these environments. Data from two of the surveys (Mourilyan Harbour and Abbot Point) are used as a basis for discussion in this report. The recommendations in this report can serve as a guideline for the ongoing surveys of tropical Queensland ports and will also provide a technical guide for biological/environmental surveys in tropical waters.

A number of environmental and biological characteristics of tropical inshore marine environments were identified as requiring consideration when planning biological collecting or monitoring programs in tropical ports. Warm temperature, the potential presence of dangerous marine animals, turbidity, habitat type and monsoonal seasonality need to be considered in relation to specimen handling, diving operations, and time of year and frequency of sampling. Biological characteristics of relevance included high species diversity and high biological process rates. High species diversity coupled with the lower proportion of described species of tropical Australian marine invertebrates compared with temperate species increase sorting and analysis time. The high rates of biological processes that characterize tropical waters will influence the frequency of ongoing monitoring.

Sampling devices recommended in Hewitt and Martin (1996) were for the most part effective. The methods for sampling hard substrate invertebrates (wharf pile scraping samples), crabs (traps), crab exuviae (beach surveys), fish (seine net), zooplankton (plankton nets), dinoflagellate cysts (sediment samples) and phytoplankton (plankton nets) successfully collected the target organisms. However, significant problems were encountered with aspects of the diver-mediated sampling and surveying, shrimp-trapping and beam trawl sampling and these methods need to be reappraised.

Diver sampling and surveying comprise a significant component of the recommended minimum sampling methodology in Hewitt and Martin (1996) but it is evident that for some tropical ports, alternative methods need to be considered. Soft-bottom surveying and sampling for epibenthic organisms by divers was problematic at both ports but particularly in the estuarine Mourilyan Harbour. Extremely limited visibility reduced the usefulness of diver surveying. Furthermore, safety risks unique to the tropics, such as the presence of estuarine crocodiles and marine stingers,

warrant the reduction of diver sampling where possible in tropical ports, particularly in estuarine habitats.

An Ocklemann sledge was trialed at Mourilyan Harbour and Abbot Point as an alternative method to diver sampling of soft bottom epibenthic organisms. This device effectively collected a wide range of taxa in this habitat. The Smith-McIntyre grab was also trialed as an alternative to diver hand-coring to sample benthic infauna, dinoflagellate cysts and sediment samples. At Mourilyan Harbour and Abbot Point, the Smith-McIntyre grab successfully collected a large number of benthic infauna taxa. It also collected sediment samples for dinoflagellate cyst and sediment analysis. Furthermore, there was little overlap between the taxa collected in the Ocklemann sledge-dredge and the Smith-McIntyre grab.

Shrimp traps as recommended in Hewitt and Martin (1996) did not collect the target organisms at the two ports surveyed. Fortunately, other methods did catch shrimps; 11 and 19 taxa were collected at Mourilyan Harbour and Abbot Point, respectively in wharf pile scraping, benthic sled and Smith-McIntyre grab samples. Shrimp can also be sampled using other methods including light traps, fine mesh seine nets and push nets. Beam trawl catches at both ports were low in abundance and diversity, probably because the device was used in daylight hours. Subsequent night sampling yielded greater catches indicating that this device must be operated at night for optimal catches.

The adequacy of sampling intensity was examined by plotting cumulative species' curves for sampling devices used at the two ports. In some instances, the curves had not reached the asymptote indicating that the number of samples collected at tropical ports may need to be re-assessed for certain sampling gears.

A preliminary analysis of differences in taxonomic diversity indicates that tropical ports are more diverse than temperate ports. At Mourilyan Harbour (MH) and Abbot Point (AP), 401 and 593 taxa respectively were collected. The major component of time in processing samples was devoted to identification of specimens. The higher diversity in tropical ports increases the time needed to complete a biological survey.

We noted marked differences in community composition at the two ports surveyed. The biological assemblages at the two ports differed as a consequence of contrasting habitat types. Within two of the more diverse groups, the bivalve molluscs and polychaete worms (38 and 44 species of bivalve, and 125 and 166 species of polychaete were collected at MH and AP respectively), between 62% and 71% of the species in each group were found only at one port.

It can be difficult to identify tropical inshore marine invertebrates. The status of taxonomic knowledge for tropical Australian invertebrate groups is typically less than for their counterparts in temperate waters. This has two important ramifications. Firstly, a higher proportion of taxa cannot be identified to species level by taxonomic experts compared with temperate ports. Secondly, it is likely that a greater proportion of specimens in a tropical collection will require the assistance of taxonomic specialists for identification. Tropical port surveys conducted in collaboration with taxonomic specialists represent a unique opportunity to increase the existing knowledge of tropical marine invertebrate taxonomy.

A feature of tropical systems that should be considered when planning the timing of surveys for monitoring introduced species are the monsoonal seasonal changes in environmental conditions and consequently, community structure. Given the high rate of biological processes in the tropics which can translate into rapid colonisation by introduced species, it is recommended to continue regular cost-effective monitoring of ports.

#### 1. INTRODUCTION

#### 1.1 Background

There are over 200 exotic marine species recorded from Australian waters (Thresher, 1999). Most of the introduced species are believed to have been unintentional introductions associated with mariculture and shipping activities. Hull fouling and ballast water discharge by shipping has been implicated as important vectors for the transport of exotic species to Australian waters. A number of species including the European fanworm (*Sabella spallanzanii*) and the northern Pacific sea star (*Asterias amurensis*) in temperate Australian ports and the black striped mussel (*Mytilopsis sallei*) in the tropics, have flourished at points of entry causing obvious disruption to the inshore ecosystems.

A prerequisite to control the spread of introduced marine pest species is knowledge of the present distribution and abundance of exotic species in Australian ports. A sampling protocol for surveys of Australian ports to detect exotic species has been devised by the Centre for Research into Introduced Marine Pests (CRIMP) in association with the Australian Association of Port and Marine Authorities (AAPMA) (Hewitt and Martin 1996). The aim of this protocol was to ensure that different agencies and research organisations follow a similar approach to port surveys and employ standardised survey methods. In Queensland, the Ports Corporation of Queensland's *Ports Corporation Environmental Program (EcoPorts)* is conducting biological surveys in all of its Queensland ports. The first tropical ports in Australia were conducted by CRIMP at Hay Point (a Ports Corporation of Queensland port) and Mackay in 1998.

In June and July 1998, the Department of Marine Biology and Aquaculture of James Cook University on behalf of CRC Reef Research Centre undertook surveys of Mourilyan Harbour and Abbot Point to monitor the endemic coastal biota and detect invading species. The aim of these surveys was to make a comprehensive taxonomic description of the existing marine communities and recognise species which may have been introduced. The specific environmental conditions of these tropical ports necessitated the implementation of some modifications to the existing sampling protocols of Hewitt and Martin (1996). The results from these surveys

further highlighted the unique environmental and biological characteristics of tropical inshore marine communities (cf. temperate communities) and implications for planning sampling and analysis. Subsequent implementation of modifications proved successful for surveys of the ports of Lucinda, Weipa and Karumba. This report discusses the relevant environmental and biological features of tropical inshore marine systems and the appropriate sampling methodologies for biological monitoring of tropical ports using the results of the Mourilyan Harbour and Abbot Point surveys as a basis for discussion.

#### 1.2 Objectives

The aim of this report is to discuss the implementation of the national sampling protocols for introduced marine pest surveys (Hewitt and Martin 1996) in tropical ports and suggest some areas of flexibility in approach. We hope to contribute to the existing sampling methods and guidelines for Australian port surveys, to improve the effectiveness of future tropical port surveys. The specific aims of this report are:

- ?? To determine the performance of sampling devices recommended in Hewitt and Martin (1996) at catching target organism groups at the tropical ports of Mourilyan Harbour and Abbot Point.
- ?? Where a sampling method or device did not collect target organisms, suggest improvements in sampling methodology and/or alternative sampling devices.
- ?? To report the performance of, and assess sampling overlap, by comparing catches between the two additional sampling devices trialed at these ports: the Smith-McIntyre grab and the modified Ocklemann sledge.
- ?? To provide useful guidelines for the operation of sampling equipment.
- ?? To examine adequacy of survey intensity for selected devices using cumulative species curves.
- ?? To quantitatively compare temperate and tropical diversity of marine fauna in Australian ports and discuss the implications for survey planning.
- ?? To discuss the relevance of differences in the status of the taxonomic knowledge of inshore invertebrates between temperate and tropical zones to port surveys.

#### 2. METHODS

The sampling methods used during the surveys at Mourilyan Harbour and Abbot Point were based on the recommended sampling protocols outlined in Hewitt and Martin (1996). However, some modifications were made to adapt to the specific environments of these ports (Table 1). In the Results section, "Recommended sampling methods" refer to the sampling method suggested in Hewitt and Martin (1996) and "alternative or additional sampling methods" refers to sampling methods additional to those in Hewitt and Martin (1996).

Table 1. Sampling methods recommended in Hewitt and Martin (1996) and those used at Mourilyan Harbour (MH) and Abbot Point (AP). Methods which are modifications or additional to those recommended are highlighted.

TARGET ORGANISMS /	RECOMMENDED METHOD	METHOD USED AT MH
SAMPLE	(HEWITT & MARTIN 1996)	AND AP
Shrimps	Shrimp traps	Shrimp traps
Crabs	Crab traps, beach drift surveys	Same and surveys of
	for exuviae	intertidal rocky
		substrates
Benthic infauna	Diver hand coring	Smith-McIntyre grab
Dinoflagellate cysts	Diver hand coring	Smith-McIntyre grab
Sediment	Diver hand-coring	Smith-McIntyre grab
Soft substrate invertebrates-	Diver surveys	Diver surveys,
epibenthos		Ocklemann sledge
Soft substrate invertebrates-	Beam trawl	Beam trawl
mobile epibenthos		
Hard substrate invertebrates-	Diver scrape samples	Diver scrape samples
wharf piles		
Phytoplankton	20 micron plankton net and	Same but <b>fixed in 2%</b>
	live culture	glutaraldehyde
Zooplankton	100 micron plankton net	100 micron plankton net
Fish	Seine net, rotenone	Seine net
Intertidal fouling organisms	No recommendation	Intertidal rocky substrate
		survey at low water

The abundance and diversity data for Mourilyan Harbour and Abbot Point used in this report are based on sorting and identifications by a team of workers with broad experienced in invertebrate taxonomy. Following primary sorting, taxa from selected groups were identified by group-specific specialists. Specialists were consulted for dinoflagellate cysts, phytoplankton, algae, sponges, hard corals, hydrozoans, gorgonians, soft corals, bryozoans, polychaetes, crabs, shrimps, amphipods, isopods and molluscs.

In this report, the "performance" of each sampling method is discussed based on catches at MH and AP. In the absence of the data to quantitatively assess sampling effectiveness, we make qualitative judgements based on catch data.

#### 3. RESULTS AND DISCUSSION

#### 3.1 Sampling methods for specific invertebrate groups or communities

#### 3.1.1 Shrimps

#### **Recommended sampling method**

Commercially available baited shrimp traps with fine mesh (traps of dimensions  $40 ext{ x}$   $25 ext{ x}$   $25 ext{ cm}$  with 2 mm mesh were used), baited and set in late afternoon and checked the following morning. Traps are tethered to crab traps adjacent to wharf piles.

#### Sampling performance and suggestions for improvement of method

No shrimps were caught in pilchard-baited shrimp traps deployed on successive nights at MH and AP. Subsequent trials with various baits and site locations at other ports also, usually, did not catch shrimps.

#### Alternative devices for collecting shrimp

Shrimps were common (11 and 19 taxa identified from MH and AP respectively) in wharf pile scraping samples, beam trawl, modified Ockelmann sled (referred to as benthic sled) and Smith-McIntyre (SM) grab samples indicating that these are useful collecting methods. Modifications to beam trawl operational procedures (night sampling as suggested in section 3.4.2) were implemented in subsequent surveys and significantly increased catches of shrimps and other mobile epibenthic organisms. Some additional sampling methods which could potentially catch shrimp include:

- ?? Light traps (Doherty 1987), although designed to collect young fish, also collect small crustaceans.
- ?? Intertidal collections, shrimps are common in the intertidal zone. At low tide, shrimps can be collected by hand from under small rocks.
- ?? Nearshore sampling; a fine mesh seine net or push net (Schlieper 1972) may also catch shrimps and prawns; night and day samples are recommended.

#### 3.1.2.1 Traps

#### **Recommended sampling method**

Commercially sold, collapsable, baited crab traps. These are set at the base of wharf piles overnight.

#### Sampling performance and suggestions for improvement of sampling method

Standard commercial crab traps (dimensions  $80 \times 55 \times 25$  cm with 4 cm mesh) in addition to a smaller variety ( $60 \times 45 \times 20$  cm with 12mm mesh) were used at MH and AP (six traps set per night over three nights). We observed that the two trap sizes used each caught different-sized crabs and different species of crab. We therefore recommend the use of two-sizes of crab trap. Further these traps regularly collected fish such as cod *Epinephelus* sp.

#### 3.1.2.2 Beach drift surveys for crab exuviae

#### **Recommended sampling method**

Surveys along beaches searching beach drift for crab exuviae.

#### Sampling performance and suggestions for improvement

Crab exuviae (principally carapaces) were found during beach drift surveys at MH and AP. To increase the effectiveness of these searches, a few points should be noted. Drift material may accumulate in specific areas of the beach due to beach morphology and wind direction (eg. the windward side of sand spits). Therefore, different beach zones in the area should be surveyed. Intertidal areas in mangroves and rocky shores should also be surveyed. The abundance of crab exuviae in drift material on beaches may exhibit temporal changes due to seasonality of prevailing wind direction and intensity and this could be considered. During these surveys, shells from dead molluscs can also be collected to provide valuable information on the mollusc community in the locality.

#### 3.1.3 Benthic infauna

#### **Recommended sampling method**

Benthic infauna samples are collected by divers using hand-held coring devices  $(0.025 \text{ m}^2, 200\text{-}250 \text{ mm deep})$ .

#### Alternative method: the Smith-McIntyre grab

Diver hand-coring was not used during the MH and AP surveys. A Smith-McIntyre (SM) grab (Smith and McIntyre 1954) was used instead (Figure 1a). The SM grab has some advantages over diver hand-coring in tropical locations. Firstly, diver time and associated costs, safety risks and complex organisational procedure, are avoided. Two persons can easily deploy the device from a small boat. Successive samples can be collected from a wide area within a relatively short time. Furthermore, the surface area of the SM grab bite is greater than that of the recommended hand-corers so that a larger area is sampled and more infaunal organisms are collected. The SM grab collects a sediment block of approximate 25 x 25 x 13 cm. In Halifax and Cleveland Bays north Queensland, the majority of benthic infaunal organisms are found in the top 5-10 cm of sediment (P. Arnold, personal communication). The presence of underlying clay in this region may contribute to this finding. Schlieper (1972) also states that the majority of organisms are found in shallow, benthic sediments. Parry et al. (1997) collected samples at Portland Harbour using both hand-corers and an SM grab and found that the SM grab collected three species of exotic species not found in core samples.

#### **Sampling performance**

A wide variety of benthic infaunal species were collected by the SM grab. A total of 76 and 197 taxa were collected at MH and AP respectively (Tables 2 and 3) indicating that this is a suitable device for sampling tropical benthic infauna.

#### 3.1.4 Soft substrate invertebrates

#### 3.1.4.1 Epibenthos

#### **Recommended sampling method**

Visual searches by SCUBA divers conducted on the benthos along transects adjacent to sampled wharf piles.

#### **Sampling performance**

Diver searches were problematic at MH and AP and yielded limited information due to suboptimal conditions. At MH, very poor visibility (due to the presence of fine suspended silt on the bottom) prevented diver visual searching to observe and collect soft substrate invertebrates. Visual diver surveys were conducted along two transects at AP. Again, environmental conditions (strong currents and limited visibility) compromised the effectiveness of visual searching.

Table 2. Numbers of taxa in Smith-McIntyre grab samples from Abbot Point.

PHYLUM	GROUP	NUMBER OF TAXA
Phycophyta		5
Foraminifera		4
Porifera		5
Cnidaria	Alcyonacea	1
	Gorgonacea	1
	Zoanthidea	5
Mollusca	Bivalvia	13
	Gastropoda	3
Annelida	Polychaeta	95
	Hirudinea	1
Crustacea	Amphipoda	8
	Anomura	8
	Brachyura	6
	Caridea	5
	Isopoda	3
	Ostracoda	3
	Tanaidacea	1
	Pycnogonida	1
Echinodermata	Echinoidea	2
	Asteroidea	1
	Ophiuroidea	5
Bryozoa		5
Sipuncula		2
Urochordata	Ascidiacea	4
Cephalochordata		1
Chordata	Pisces	1
Unidentified		8
TOTAL TAXA		197

Table 3. Numbers of taxa in Smith-McIntyre grab samples from Mourilyan Harbour.

PHYLUM	GROUP	NUMBER OF TAXA
Phycophyta		2
Cnidaria	Zoanthidea	1
Mollusca	Bivalvia	1
	Gastropoda	1
Annelida	Polychaeta	45
Crustacea	Amphipoda	5
	Anomura	2
	Caridea	4
	Isopoda	3
	Ostracoda	1
	Penaeiodea	1
	Tanaidacea	2
Echinodermata	Ophiuroidea	1
Sipuncula		1
Urochordata	Ascidiacea	1
Cephalochordata		1
Unidentified		4
TOTAL TAXA		76

#### Additional sampling method-modified Ockelmann sledge

A modified Ocklemann sledge (Figure 1b) or benthic sled was trialed at MH and AP as an alternative to diver bottom surveys. The Ocklemann sledge was chosen to compensate for the reductions or absence of benthic diver surveys. Diver observations at AP indicated that large epibenthic organisms were rare on the soft bottom substrate; small invertebrates were numerically dominant. The device samples over long distance and has the advantage of collecting smaller epibenthic organisms that are overlooked by divers. It is designed to minimise sinking into the sediment, thus taking a wider range of the sparsely distributed epifaunal organisms by skimming along the sea floor (English *et al* 1994).

The Ocklemann sledge used during the surveys was constructed of aluminium. The dimensions of the device are  $50 \times 14$  cm (mouth) with 6 mm aluminium mesh. The sled has an odometer to record distance travelled, allowing calculation of area sampled. The mouth of the benthic sled partially digs into the sediment and collects organisms within the surface layer of sediment to a depth of a few centimetres; the runners prevent it from sinking completely into the sediment and enabling the exposed mouth area to collect epibenthic fauna.

The mesh size of this device is appropriate for sampling fine inshore sediments. However, if the benthic sediments have less silt and clay and more biogenic rubble or dense seagrass, the fine mesh clogs quickly. In such instances, a larger sledge with courser mesh is recommended.

Notes on the operation of the Ocklemann sledge are given in Appendix 1.

#### Sampling performance of Ocklemann sledge

High numbers of taxa were collected in the benthic sled (70 and 64 taxa were collected at MH and AP respectively) indicating that the device successfully sampled the benthic community (Tables 4 and 5). The device collected both infauna and epifauna because bivalves from within the sediment and mobile surface dwelling fauna such as echinoids and crabs were collected in the samples (Tables 4 and 5).

Table 4. Numbers of taxa in benthic sled samples from Abbot Point.

PHYLUM	GROUP	NUMBER OF TAXA
Phycophyta		9
Porifera		1
Cnidaria	Scleractinia	3
	Hydroida	4
Mollusca	Bivalvia	8
	Gastropoda	8
Annelida	Polychaeta	7
Crustacea	Anomura	1
	Brachyura	5
	Caridea	3
	Penaeidea	1
Echinodermata	Asteroidea	1
	Echinoidea	1
	Ophiuroidea	2
Bryozoa		
Chordata	Pisces	1
Unidentified		2
Total		64

#### 3.1.4.2 Mobile epibenthos

#### **Recommended sampling method**

A lightweight beam trawl towed at 1, 2, 5 and 10 m depths is recommended for sampling mobile epibenthic organisms.

#### Sampling performance and suggestions for improvement of method

A beam trawl towed during daylight hours at MH and AP yielded low catches of mobile epibenthic organisms. Small numbers of shrimp, prawns, crabs, polychaetes, cephalopods and fish were caught. The low catches suggested that the device was not effectively sampling the mobile epibenthic community.

Small beam trawls have successfully sampled mobile epibenthic organisms in tropical seagrass meadows (L. McKenzie, personal communication). The method and time of operation are important considerations for this device. Small beam trawls are best deployed at night. This increases the probability of collecting epibenthic organisms that burrow during the day. It also reduces visual net avoidance. Moon phase can influence catch success; catches of mobile epibenthos over seagrass meadows are higher during the new moon phase (L. McKenzie, personal communication). Subsequent to the surveys at MH and AP, night sampling trials

were undertaken at the Port of Lucinda. Weipa and Karumba which all yielded catches with significantly higher abundance and diversity than diurnal samples at AP and MH. It is strongly recommended that beam trawl samples be collected after dusk. We recommend that at least 10 samples be collected to represent the community.

Notes on the operation of beam trawls are given in Appendix 1.

#### 3.1.5 Hard substrate invertebrates

#### 3.1.5.1 Wharf piles and dolphins

#### **Recommended sampling method**

Fouling organisms on wharf and dolphin pile surfaces are scraped by divers from within 0.1 m<sup>2</sup> quadrats at three depths. Video and photographic records of surface fouling are made.

#### Sampling performance

Wharf piles were sampled followed the procedure outlined in Hewitt and Martin (1996). The high abundance and diversity of the fauna collected from wharf pile scrape samples at MH and AP indicated that the method was effective for collecting fouling organisms (Tables 6 and 7). Up to 80 taxa per quadrat were collected at AP and 181 and 299 taxa were recorded from wharf piles at MH and AP respectively (Tables 6 and 7). Notes on sampling equipment design and use are given in Appendix 1.

#### Suggestions for improvement of sampling method

The design of equipment used for collecting wharf pile scraping samples is important. The quadrat used at MH and AP was effective but could be further improved. The quadrat frame was 3 cm thick and it was sometimes difficult to remove all fouling biota from corners and directly adjacent to the quadrat frame. Incorporating a thin, flexible frame would minimise this problem.

The use of fixed quadrates to sample wharf piles should be supplemented by qualitative collection of larger colonial organisms such as corals, sponges and ascidians as these may occur in low numbers or be damaged or only partially sampled when using quadrats.

#### 3.1.5.2 Breakwaters, groynes, rock walls, natural rocky reefs, wrecks

#### **Recommended sampling method**

These are targeted for visual surveys. For shallow (<5 m) rocky areas, a 50 m transect line is surveyed and paired 0.1 m² quadrats (-0.5 m and bottom) are sampled at five randomly selected locations along the transect. In areas where rocky areas extend to more than 7 m depth, quadrat scraping procedure is similar to wharf pile sampling.

Table 5. Numbers of taxa in benthic sled samples from Mourilyan Harbour.

PHYLUM	GROUP	NUMBER OF TAXA
Phycophyta		10
Porifera		1
Cnidaria	Hydrozoa	11
	Gorgonacea	6
	Alcyonacea	3
Mollusca	Bivalvia	4
	Gastropoda	3
Annelida	Polychaeta	12
Crustacea	Brachyura	5
	Caridea	3
	Penaeiodea	1
Pycnogonida		1
Echinodermata	Ophiuroidea	1
Bryozoa		8
Unidentified		1
Total		70

#### **Sampling performance**

A visual survey and sample collection over subtidal rocky reef was undertaken by snorkel divers in shallow water at AP. The method found four algae and four invertebrate taxa that were not sampled by other methods. This sampling technique is recommended in waters with visibility of at least 3 m.

# 3.1.5.3 Additional sampling methods - surveys of intertidal rocky substrate, boat hulls and floating pontoons

Algal and invertebrate taxa can be collected from natural and man-made hard substrates at low tide. At MH and AP, 43 and 29 taxa (algae and invertebrates)

respectively, were collected during intertidal surveys of rocky substrates. Samples scraped from boat hulls and floating pontoons are also expected to provide useful samples of fouling species.

#### 3.1.6 Phytoplankton

#### **Recommended sampling method**

Phytoplankton are collected using a small hand-hauled plankton net (20 micron mesh). Phytoplankton samples are maintained live and cultured. Suspected toxic species are tested for toxin production.

Table 6. Numbers of taxa in pile scrape samples from Abbot Point.

GROUP	SUBGROUP	NUMBER OF TAXA
Plantae		24
Porifera		21
Coelenterata	Hydrozoa	18
	Zoanthidea	5
	Actinaria	5
	Scleractinia	4
	Alcyonacea	2
Platyhelminthes		1
Mollusca	Bivalvia	28
	Gastropoda	8
	Cephalopoda	1
Annelida	Polychaeta	63
	Anomura	4
	Amphipoda	21
Crustacea	Brachyura	14
	Caridea	15
	Cirripedia	6
	Mysidacea	1
	Penaeiodea	1
	Tanaidacea	1
Bryozoa		20
Echinodermata	Ophiuroidea	12
	Echinoidea	1
	Holothurioidea	1
Urochordata	Ascidiacea	19
Sipuncula		1
Chordata	Pisces	3
Other		9
TOTAL		299

#### Sampling performance and alternative treatment of samples

Due to the absence of nearby live-culture facilities, samples were fixed in 12% glutaraldehyde for long-term storage and subsequent identification of taxa (G. Hallegraeff, personal communication). The method successfully collected target

organisms, 8 and 11 species of phytoplankton were collected at MH and AP, respectively.

#### 3.1.7 Zooplankton

#### **Recommended sampling method**

Zooplankton are collected using 100 micron mesh plankton net.

#### Sampling performance

The device successfully collected this target group, 29 and 21 zooplankton taxa were collected at MH and AP, respectively.

#### 3.1.8 Dinoflagellate cysts

#### **Recommended sampling method**

Sediment samples are collected from selected areas by divers using hand-corers. Sample sites are chosen on the basis of bathymetry and hydrography. Areas where sediments are relatively undisturbed (usually deeper holes are best) are suitable. Samples are sent fresh to a laboratory for cyst culture and identification. It is recommended that at least eight samples be collected.

#### Alternative method-formalin preservation

Sediment samples for dinoflagellate cyst detection were successfully collected using the SM grab. At MH and AP, samples were preserved in 7% formalin seawater and cysts were identified. Live-culture of cysts is more desirable but requires overnight transport of fresh samples to a specialised dinoflagellate cyst culture laboratory. The latter method has been successfully adopted in subsequent port surveys and is highly recommended.

#### Sampling performance

The sampling/preservation method was moderately successful at AP where four dinoflagellate cyst taxa were identified in samples. No cysts were detected from samples at MH. At Lucinda, live cyst-culture methods yielded 10 dinoflagellate taxa.

#### 3.1.9 Fish

#### **Recommended sampling method**

A beach seine (25 m, 15 mm mesh) is recommended for collecting fish.

#### **Sampling performance**

The beach seine was successful for collecting fish at MH and AP; 17 and 9 taxa were collected at MH and AP, respectively. Night sampling and the use of a range of net mesh sizes should yield a greater diversity of catch.

Table 7. Numbers of taxa in pile scrape samples from Mourilyan Harbour.

GROUP	GROUP	NUMBER OF TAXA
Porifera		16
Cnidaria	Actinaria	1
	Hydrozoa	13
	Zoanthidea	5
Platyhelminthes		1
Nematoda		1
Annelida	Polychaeta	70
Mollusca	Bivalvia	30
	Gastropoda	1
Crustacea	Amphipoda	1
	Anomura	1
	Brachyura	3
	Caridea	5
	Cirripedia	3
Echinodermata	Ophiuroidea	3
	Holothurioidea	1
Brachiopoda		1
Bryozoa		19
Urochordata	Ascidiacea	3
Chordata	Pisces	1
Unidentified		2
TOTAL		181

#### 3.2 Comparison between catches of the Smith-McIntyre grab and benthic sled

Catches made by the SM grab at AP were compared with those of the benthic sled. Of the 197 and 62 taxa collected in the grab and sled respectively, only 21 were common to both (Table 8). This indicates that the two devices sample different components of the benthic species assemblage.

#### 3.3 Adequacy of sampling intensity

For biological surveys, increasing sample number will increase the number of taxa collected. However, the number of additional taxa per sample will decrease as more samples are collected until all species in the area have been collected. When surveying for introduced marine species it is important to collect adequate samples to be representative of species present. Cumulative species curves are a means of qualitative assessment of the likelihood of collecting more taxa using a given sampling device in a given habitat. Flattening of the curve as it approaches the asymptote indicates that the number of new taxa in samples is rapidly diminishing.

Cumulative species curves were plotted for the SM grab, benthic sled and wharf pile scraping samples at the MH and AP (Figs 3, 4 and 5). The cumulative species curve for the SM grab at MH (eight samples) appeared to have entered an asymptotic phase indicating that a representative component of the fauna in that habitat were collected (Fig. 3a). The cumulative species curve for the SM grab at AP (12 samples) did not exhibit a clear asymptotic phase (Fig. 3b). Hewitt and Martin (1996) recommend a minimum of two grab samples at each scrape-sampled wharf pile (at least six samples per berth). Both MH and AP have only one berth. Based on the cumulative species curves, the recommended six samples would have been insufficient at AP. The cumulative species curves show that taxonomic coverage was considerably enhanced by taking additional samples at this port.

It is generally recognised that different sediment types (clean sand, high silt clay) have characteristic taxa of benthic infauna. Heterogeneity of sediment types at a port will warrant the collection of a greater number of samples.

The cumulative species curves for Ocklemann sledge samples (five at each port) show that the rate of increase in additional species had not yet reached the asymptotic phase (Figs. 4a and b). This analysis suggests that more than five sledge samples are needed for adequate taxonomic coverage in tropical ports. Subsequently we have collected 10 samples (paired samples at 1, 2, 5, 10 and 15 m).

Cumulative species curves were plotted for wharf pile scraping samples at MH (14 samples) and AP (16 samples) (Fig. 5a and b). New species continued to appear in samples but at a decreasing rate for both locations; the curve for AP appeared to be entering the asymptotic phase (Fig. 5a). Based on these results, we recommend a minimum of between 10 and 15 scrape samples.

Comparing our results with those from port surveys from the temperate waters of Portland Harbour, Victoria, suggests that tropical ports need more samples to adequately sample communities. At Portland Harbour, Parry *et al.* (1997) found that 10 grab and five wharf-pile scraping samples were adequate to sample these habitats. However, Currie *et al.* (1998) suggested that most species on wharf piles at the Port of Geelong were collected after 11 pile scrapes.

#### 3.4 Regional variations in taxonomic diversity

#### Differences in species assemblages and diversity between tropical ports

Taxonomic diversities recorded at MH and AP differed (Table 9) demonstrating that diversity can vary between tropical ports. The AP collection (including plankton) was more diverse (593 taxa) than MH (401 taxa). A significant component of the species were present only at one of the two ports indicating that the ports harboured different communities. Of the bivalve molluscs, 25 out of 38 species (65%) at MH and 27 out of 42 species (64%) collected at AP were found at only one port. For the polychaete worms, 78 out of 125 (62%) at Mourilyan Harbour and 118 out of 166 (71%) species at Abbot Point were found at only one port. The species composition of most groups differed between ports. The difference in taxonomic composition between ports may lie in the habitat differences at each location. AP was located on a rocky headland (open coastline) over a sandy bottom. In contrast, MH was located in a semi-enclosed estuarine system. The difference in composition of taxa between ports means that, when sampling successive ports, similar resources need to be allocated to taxonomic identification due to the presence of new suites of species.

Table 8. Comparison of numbers of taxa collected in SM grab and Ocklemann sledge samples from Abbot Point.

PHYLUM	GROUP	GRABT	SLED TAXA	COMMON TAXA
Phycophyta		5	9	3
Foraminifera		4	-	
Porifera		5	1	0
	Alcyonacea	1		
Cnidaria	Hydrozoa	-	2	
	Gorgonacea	1	-	-
	Scleractinea	2	3	2
	Zoanthidea	5	-	-
Mollusca	Bivalvia	13	8	3
	Gastropoda	3	8	1
Annelida	Hirudinea	1	-	-
	Polychaeta	95	7	5
	Amphipoda	18	-	-
Crustacea	Anomura	8	1	-
	Brachyura	6	5	0
	Caridea	5	3	2
	Isopoda	3	-	-
	Ostracoda	3	-	-
	Tanaidacea	1	-	-
Pycnogonida				
	Asteroidea	1	1	1
Echinodermat	Echinoidea	2	1	1
	Holothuriiodea	-	2	-
	Ophiuroidea	5	2	1
Bryozoa		5	7	2
Sipuncula		2	-	-
Urochordata	Ascidiacea	4	-	-
Cephalochord		1	-	-
Chordata	Pisces	1	1	
Unidentified		8	2	
TOTAL		197	62	21

The taxonomic diversity between ports impacted sorting and analysis times for each port. Wharf pile scraping samples from AP exhibited high levels of abundance and diversity compared with those from MH. It took experienced sorters an average of 21 hours to fully sort of a pile-scraping sample from AP (includes primary identification of taxa) compared with 12 hours for a MH sample. The time component of work for AP was considerably greater han for MH as a result of the different taxonomic diversities in the two ports.

#### Comparisons of taxa diversity between tropical and temperate ports

The identification of taxa, both during primary sorting and by taxonomists after sorting, is a major time component and cost of port flora and fauna surveys. Therefore, regional patterns in taxonomic diversity should be taken into account when planning port surveys.

Tropical marine benthic communities can be extremely diverse (Hammond and Synnot 1994). There is a trend to increasing diversity in marine shelf invertebrate communities from high to low latitudes ie. from temperate to tropical waters (Began *et al.* 1995, Levinton 1995). Tropical Queensland's coastal biota exist in a highly diverse marine zone known as the central Indo-West Pacific province (Hilliard *et al.* 1997). However, high diversity is not a characteristic of tropical habitats alone. For example, some temperate benthic communities in Australia exhibit high diversity of invertebrate groups (Hammond and Synnot 1994, Coleman *et al.* 1997).

To make a preliminary comparison of taxonomic diversity in temperate and tropical locations, we compared data from six port surveys (Table 9). Five were undertaken in Australian waters using similar sampling methodologies. Data from a survey in Hawaii using comparable sampling methods were also compared. The number of taxa within 10 of the more diverse phyla were compared.

Based on the data in Table 9, it is evident that the average number of taxa collected is higher for tropical ports (447 taxa) compared with temperate ports (289 taxa). However, the species count for a port survey depends on the use of effective sampling methods, the number of samples collected, and careful sorting and identification of specimens. The two groups with greatest difference were chidarians and polychaetes. Both of these groups are exceptionally time-consuming to identify. These preliminary data indicates that tropical port surveys may require more time for analysis than temperate ports. More data are needed to quantify differences in diversity between tropical and temperate ports.

Table 9. Comparison of taxa diversity between four tropical and three temperate ports.

	TROPICAL PORTS				TEMPERATE PORTS		
	MH* AP HP Haw				Н	G	P
Phycophyta	11	37	4	36	6	14	18
Porifera	16	28	**	33	24	12	9
Cnidaria	31	45	30	10	4	4	3
Mollusca	74	71	109	85	65	44	28
Crustacea	51	110	129	110	126	72	83
Echinodermata	8	26	15	7	11	17	4
Annelida	119	158	100	54	55	68	49
Bryozoa	25	26	14	13	10	11	10
Urochordata	3	22	21	19	20	16	7
Chordata (fish)	21	18	21	59	26	32	16
Total taxa/port	359	541	443	426	347	290	229
Average/region		44	7			289	

<sup>\*</sup>Where AP = Abbot Point; MH = Mourilyan Harbour; HP = Hay Point; H = Port of Hastings;

#### 3.5 Taxonomic status of tropical marine invertebrates

The status of taxonomic knowledge in a region should be considered when undertaking tropical port flora and fauna surveys. It is difficult to detect introduced species in areas where the endemic biota is poorly documented. The ease with which introduced species can be detected is inversely related to the amount of taxonomic knowledge of its group (Hilliard *et al* 1997). Within Australia, a smaller proportion of taxa within certain invertebrate groups in the tropics have been described compared with their counterparts in temperate regions. Accordingly, there is a greater dependence on taxonomic specialists when dealing with tropical collections.

Tropical sponges are probably one of the more problematic groups in terms of taxonomic knowledge and they illustrate the taxonomic problems for tropical invertebrates. Only about 350 species of sponges are described for Queensland waters, after about 150 years of exploring this region (including the GBR). However, Queensland Museum staff have collected about 2000 taxa from Queensland alone, most of them new to science (J. Hooper personal communication). This is also the case for a number of invertebrate groups. Consequently, many taxa can only be identified to family or genus level, even by experts. Therefore, tropical port surveys conducted in conjunction with taxonomic experts provide an opportunity to make

G = Port of Geelong; P = Portland Harbour; Haw = Hawaii

<sup>\*\*</sup>Porifera not yet identified.

comprehensive systematic collections of tropical inshore marine invertebrates and improve the taxonomic knowledge of tropical marine invertebrates.

Another factor that impacts the ease of identification of introduced marine pests is the location of tropical Australian ports within the wider Indo-Pacific zoogeographic region. It is more difficult to establish whether species which have not previously been recorded in Australia are invaders or naturally occurring. There may also be a complex of closely related species that can only be recognised with the help of a specialist. This is less of a problem in southern temperate ports which have a higher proportion of endemic species.

#### 3.6 Other considerations for tropical port surveys

#### **Diver safety**

Some tropical estuarine ports are inhabited by estuarine crocodiles. Therefore, it is desirable to minimise dive time and seek alternative methods where possible (see section 3.4). In some far northern ports, it may be necessary for divers to work inside a protective cage. Divers must also wear adequate protective clothing to avoid injury from marine stingers. Care must be taken by divers to avoid dehydration while working both in and out of water.

#### **Treatment of samples**

In warm tropical conditions, it is essential to preserve samples as quickly as possible. Soft-bodied organisms deteriorate if left untreated for several hours which can make later identification difficult. If immediate preservation is not possible, samples should be kept on ice.

#### Seasonality

In tropical ports, it is important to consider the impact of seasonal changes on sampling. The timing and intensity of monsoons at higher tropical latitudes influence the abundance of intertidal and shallow subtidal benthic fauna (Hilliard *et al.* 1997). Where a prolonged monsoon season occurs, major runoff and flushing can cause significant mortality in these habitats. Therefore, it is desirable to sample in the dry season. In areas where the monsoon is less intense, faunal densities in estuarine waters can be highest during the summer wet season (Alongi 1989). Taxa

assemblages in a port may differ between the wet and dry season and ideally, surveys should be done in each season.

#### **Speed of biological processes**

Tropical inshore communities can exhibit rapid turnover rates and consequently communities can change over short periods (R. Willan personal communication). In tropical conditions, an introduced pest species may multiply rapidly and become established within a short time. This is exemplified by the rapid colonisation by the introduced black striped mussel (Mytilopsis sallei) in Darwin (Willan et al 2000). Following initial baseline surveys of ports, regular monitoring using a simplified protocol would be appropriate.

#### Small size of tropical benthos

Fauchald (1968) found that tropical benthic infauna (in particular, polychaetes) are relatively small compared with their temperate counterparts. This will have implications for the determination of appropriate mesh sizes and sorting strategies.

#### 4. **CONCLUSIONS**

A number of environmental and biological characteristics of tropical inshore marine environments were identified as requiring special consideration when undertaking collecting or monitoring programs in tropical ports. Warm temperature, soft, muddy bottoms and associated turbidity, presence of dangerous marine animals, seasonal environmental variation factors need to be considered in relation to specimen handling and preservation, use of divers, sorting time allocation and time of year, and frequency of sampling. Relevant biological characteristics include high species diversity and high rates of biological processes characteristic of tropical systems. High species diversity impacts sorting and analysis complexity and time. The high rates of biological processes necessitates ongoing monitoring programs.

Some of the sampling methods listed in Hewitt and Martin (1996) successfully collected the target organisms: wharf pile scraping for hard substrate invertebrates; traps for crabs; seine netting for fish; beach drift surveys for crab exuviae; plankton nets for zooplankton and phytoplankton; and sediment sampling for dinoflagellate cysts. Problems were encountered when undertaking diver surveys of the benthos due to silted mud bottoms with associated high turbidity and safety risks. We recommend that if very poor diving conditions are encountered (including presence of estuarine crocodiles), diver sampling and surveying should be reduced and alternate methods implemented where possible during tropical port surveys. A suitable alternative to diver hand-coring (for benthic infauna, dinoflagellate cyst and sediment sampling) is the use of the Smith-McIntyre grab which can be easily deployed from a small boat. Diver surveys of soft substrate habitats for epibenthic organisms are recommended for ports with good underwater conditions but in estuarine ports with poor diving conditions, alternative sampling devices are suggested.

Shrimp traps have repeatedly been found to exhibit very low catches at the ports surveyed. However, other methods (wharf pile scraping, benthic sled and Smith-McIntyre grab) successfully collected shrimps. Alternative sampling devices including light traps, fine mesh seine nets and push nets may also be useful. Beam trawl sampling must be conducted after dusk to optimise catches.

The results of analysis of sampling adequacy for selected devices at MH and AP indicated that, in some instances, more sampling effort is needed than recommended as the minimum by Hewitt and Martin (1996) to provide adequate taxonomic coverage. Based on our findings we recommend that *a minimum* of 12 grab (benthic infauna) and 10-15 wharf pile scraping samples be collected at a port.

A preliminary analysis of regional differences in taxonomic diversity between ports indicates that, as predicted, tropical ports are more diverse than temperate ports. Taxa identification comprises a major component of time to sort and analyse samples. Increases in the number of taxa collected will consequently increase the time needed to complete a biological survey. Further, the status of taxonomic knowledge for some tropical invertebrate groups is less than for their counterparts in temperate waters. Therefore, it is expected that there will be a more unidentified species from tropical ports. In light of these difficulties, collaboration with taxonomic experts is considered essential. The environments and subsequently the biological assemblages at MH and AP differed. This highlights that tropical ports in the same region may harbour dissimilar species assemblages and the knowledge of the taxa from one tropical port may be of limited use for other ports.

In light of the wet/dry seasonal changes in community structure, the high rate of biological processes in the tropics and the constant threat of introduction of marine pests, it is recommended that, following initial surveys to obtain baseline data, regular cost-effective monitoring of ports is continued.

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#### APPENDIX 1. Notes on operation of sampling equipment.

#### **Smith-McIntyre grab**

The Smith-McIntyre (SM) grab must be lowered to within a few metres of the sea floor before letting the device fall freely to the bottom. If released too far above the bottom, the device may tilt or land on its edge and not take a full bite of sediment. The sample is sieved through 1 mm mesh and washed to remove excess sediment. The sample is placed in a container and preservative and a small volume of 1% erythrocin stain (to assist visual sorting of small organisms) is added. Erythrocin stains living matter pink making it easier to detect organisms in the laboratory.

Where possible, delicate organisms such as nudibranchs, flatworms and shrimps should be removed from grab samples before washing in a sieve and placed in a separate vial of preservative within the sample container. This avoids damage resulting from the washing process. In particular, Crustacea need to be preserved in good condition, because the delicate appendages are needed to identify the specimens.

The mesh size of the sieve used to wash samples is a critical factor for benthic infaunal sampling. Slight increases in mesh size can significantly reduce the numbers of organisms caught. Commonly used sieve mesh sizes for benthic infauna sampling are 0.5-1.0 mm. A 1 mm mesh size was selected over 0.5 mm because of the difficulty of identifying the very small epibenthic organisms. This mesh size yielded a diverse sample.

Benthic infauna samples should not be collected close to wharf piles to avoid the accumulations of calcareous matter from dead fouling organisms that typically occur at the base of piles.

In the laboratory, samples are washed through a 1 mm sieve with fresh water and placed in a tray of water. The sample is lightly shaken from side to side and the water with suspended organisms poured into a 1 mm sieve. This removes most organisms from the sediment. The remainder of the sample is examined under low power microscopy and organisms removed with forceps.

#### **Modified Ocklemann sledge**

The aluminium frame of the benthic sled is lightweight and easily deployed and retrieved by two people from a small boat. When using the benthic sled, care must be taken not to tow too quickly because this makes the sled lift off the bottom. When sampling effectively, the sled digs into the surface sediment layer and increases the tow-rope tension which can be monitored by the boat driver. Sudden loss of tow-rope tension (coupled with increased boat speed) indicates the sled has lifted; slowing down will allow the sled to "dig in" again. The device is most effective over sandy substrates; heavy clay sediments may cause some degree of clogging and reduce the tow distance. Tows of 5-10 minutes are recommended.

A rope length of approximately 50 m was suitable for towing at depths up to 15 m. Vessel speed was maintained at 1-2 m/s for towing.

#### Beam trawl

Correct operation of the trawl is essential. Care must be taken to avoid towing the trawl too quickly, because lifting will occur. The presence of algae and other marine debris in the net indicates that the net has been fishing close to the bottom. A tow of 100 m should take 2.5-5 min or 0.3-0.7 m/s (L. McKenzie personal communication). Beam trawl dimensions should be around 1.5 m x 0.5 m mouth and 2-4 mm mesh.

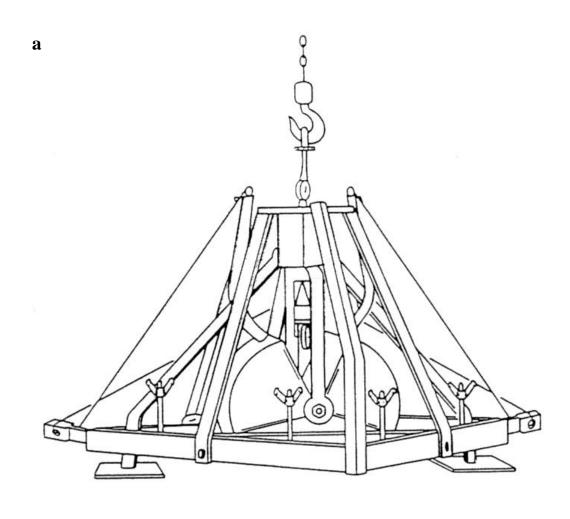
#### Wharf pile scrape samples

The most complex procedure undertaken in the surveys was collecting scrapings from wharf piles. The sampling team comprises two divers, a boatman and one person on the wharf to sort and preserve samples as they are retrieved. A well-planned sampling procedure and good quadrat design are important for efficiency and maximising sample retention.

A weighted rope with depths marked on it was lowered alongside the pile. Three quadrats were lowered from the wharf to the required depth (depth marked on rope). As the divers descend, the pile surface was recorded using video (using the depth-marked rope as a guide). Each quadrat was attached and a still photograph of each taken. The first scraping was taken from the deepest (7 m) quadrat followed by the 3 m and 0.5 m as divers ascended. Fouling organisms are often small, therefore, care must be taken to prevent loss of organisms by drift as they are scraped into a

collecting bag. For this reason, scraping is best done at still water (slack tide). Following sample collection at each depth, the diver sealed the detachable cotton bag liner with clips (see quadrat design below), detached the quadrat from the pile and tugged the rope to the surface to alert the person on the wharf to retrieve the quadrat with the sample. In tropical conditions, it is important that samples are sorted and preserved immediately to optimise the preserved condition of organisms.

The sampling quadrat used in the MH and AP surveys was designed specifically for scraping wharf piles (Fig 2). To easily attach and detach, one side of it was fitted with two sets of rope ties with loops along its length and two elastic (bungee) cord with clips on the other. These could be tied around wharf piles of different sizes to hold the quadrat firmly in place (Fig. 2). To minimise sample losses, an open cotton collection bag with detachable liner was attached by clips to the frame at the quadrat base to collect the sinking sample as the diver scraped. The scraping tools, a hammer and a heavy-duty scraper, were attached to the quadrat (Fig. 2). This design ensured that the diver's hands were free to guide the samples into the bag.



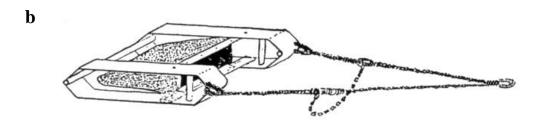


Figure 1. Diagrams of a) Smith-McIntyre grab and b) an Ocklemann sledge-dredge.

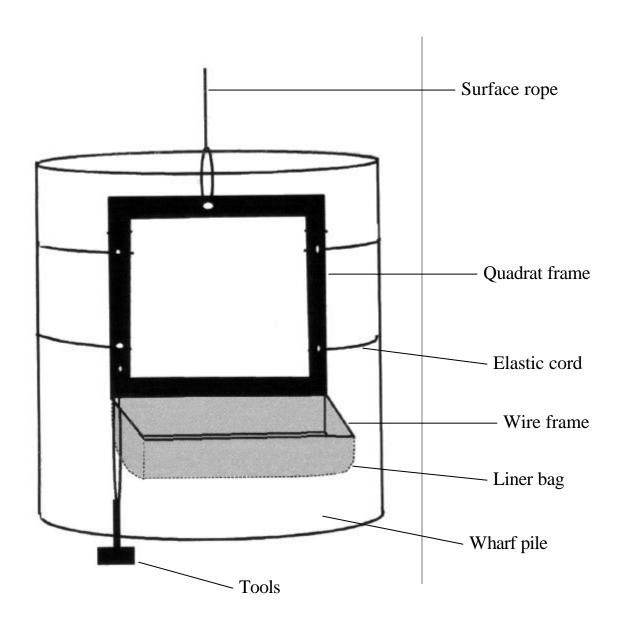
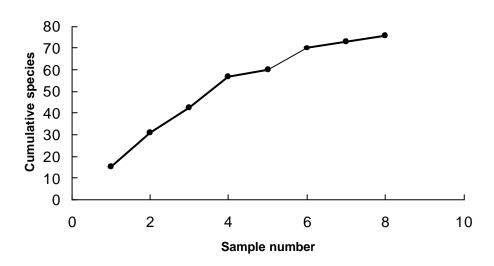


Figure 2. Diagram showing the pile scrape sampling quadrat attached to a wharf pile.

#### Mourilyan Harbour (SM grab)



#### **Abbot Point (SM grab)**

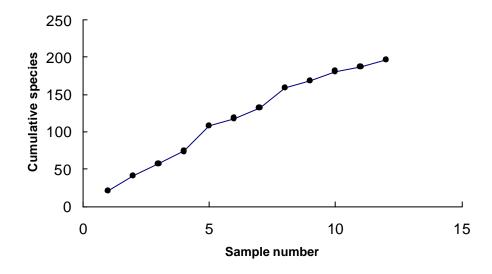
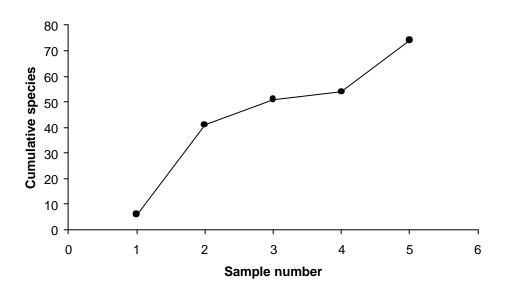


Figure 3. Cumulative species curves for Smith-McIntyre grab samples.

## Mourilyan Harbour (Sled)



## **Abbot Point (sled)**

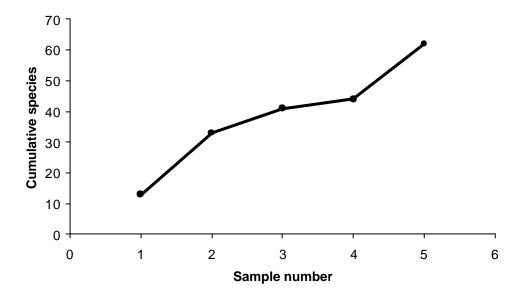
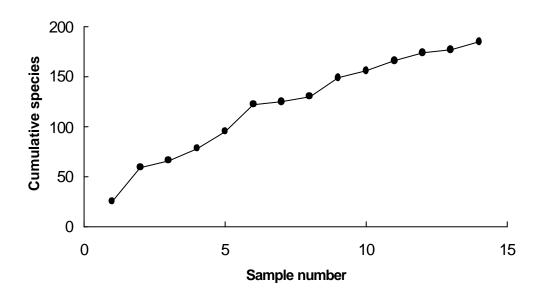


Figure 4. Cumulative species curves for benthic sled samples

# Mourilyan Harbour (scrape)





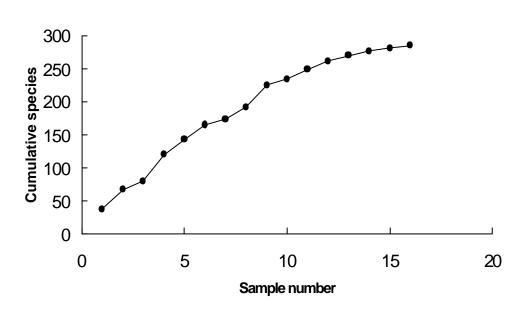


Figure 5. Cumulative species curves for wharf pile-scraping samples.