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Executive Summary

This report describes the results of the continuation of a targeted surveillance programme for marine pests that are currently listed on the New Zealand register of Unwanted Organisms under the Biosecurity Act 1993. Surveillance began in summer 2002/2003 and continued at six-monthly intervals until winter 2004. The results of these first four surveys were reported previously (Inglis et al. 2005a). This report presents the results of the fifth survey conducted in summer 2005-2006. The first four surveys targeted seven species, namely the Asian kelp *Undaria pinnatifida*, the Mediterranean aquarium weed *Caulerpa taxifolia*, the northern Pacific seastar *Asterias amurensis*, the Mediterranean fanworm *Sabella spallanzanii*, the European green crab *Carcinus maenas*, the Chinese mitten crab *Eriocheir sinensis*, and the Asian clam *Potamocorbula amurensis*. The Asian kelp, *U. pinnatifida* is already present in New Zealand. Following the discovery of the introduced clubbed ascidian *Styela clava* in the Waitemata Harbour in August 2005 (and subsequently in the Hauraki Gulf, Firth of Thames, Lyttelton Port and Marina, Tutukaka Marina, Opua Marina and Nelson Port), this species was added to the list of Unwanted Organisms under the Biosecurity Act 1993 and to the suite of target species for the summer 2005-2006 surveillance surveys.

Determining where to sample

Sampling locations were selected based on proximity to high-volume commercial ports and marinas that are first entry points for international vessels. The previous surveillance surveys (2002-2004) were focussed on eight harbours - Whangarei, Waitemata, Tauranga, Wellington, Nelson, Lyttelton, Otago and Bluff - identified in an earlier report as high risk (Inglis 2001). Picton Harbour (including Shakespeare Bay, Waikawa Marina and Havelock Wharf and Marina) was added to list of sampling locations for the 2005-2006 surveys because of its proximity to the marine-farming areas of the Marlborough Sounds, Tasman and Golden Bays, and the perceived risk posed by *Styela clava* to the marine-farming industry. The nine harbours range in size from 12.7 ha (Havelock Wharf and Marina) to 22578 ha. (Tauranga Harbour) and contain a variety of different marine or estuarine habitats. Therefore a risk-based stratification of environments within each harbour was used to prioritise areas for sampling. These areas were identified by combining spatially explicit descriptions of the species' habitat associations with simulations of likely patterns of establishment in local harbours. As Inglis et al. (2005a) discussed, the results of the previous surveys (2002-2004) showed that the approach had promise, but also highlighted a number of significant impediments to achieving cost-efficient and sensitive surveillance in these environments. One of the most important of these impediments is the general lack of suitable environmental information from which to identify suitable habitats.

Sampling methods

Since the purpose of the surveillance programme is detection rather than enumeration, techniques were selected in which the presence or absence of the target species could be determined rapidly within a sample. This allows a comparatively large number of locations to be sampled on each survey. Baited box traps were used to sample adult crabs (i.e. *Carcinus maenas* and *Eriocheir sinensis*) and Whayman-Holdsworth starfish traps were used to catch asteroids and other large benthic scavengers. Baited traps do not sample juvenile and sub-adult Chinese mitten crabs (*E. sinensis*) effectively because these life stages have a largely herbivorous diet and occupy slack-water habitats where bait odours are not well-dispersed. Therefore, to sample juvenile crabs, we used artificial shelters ("crab condos") that had been designed for surveys of *E. sinensis* in San Francisco Bay. An Ocklemann benthic sled was used to sample soft-sediment habitats for *Potamocorbula amurensis*, *Sabella spallanzanii*, *Asterias amurensis* and *Caulerpa taxifolia*. Divers searched for *S. spallanzanii*, *Undaria pinnatifida, C. maenas* and *A. amurensis* around piles, floating pontoons and other artificial structures in port and marina environments and on intertidal and shallow subtidal reefs that were identified as high risk by the dispersal plume modelling. In addition, timed visual searches for the target species were made of intertidal rocky reefs, sloping sandy shorelines and pontoons and other floating structures in marinas and port areas.

Although the survey methods were chosen to sample the target species they also captured other native and non-indigenous species present in each harbour. To improve knowledge of the distribution within New Zealand of organisms captured in this way, records were kept of easily-identified introduced and cryptogenic organisms, including the portunid crab *Charybdis japonica*, the cryptogenic tube-building polychaete *Chaetopterus* sp, the majid crab *Pyromaia tuberculata*, the semelid bivalve *Theora lubrica*, the nesting bivalve *Musculista senhousia* and the green alga *Codium fragile* ssp. *tomentosoides*.

Field surveys

The field surveys for the target species were undertaken in each of the eight harbours between December 2005 and May 2006. The only target species detected during the surveillance fieldwork were *Undaria pinnatifida* and *Styela clava*. *U. pinnatifida* was recorded within Waitemata, Wellington, Picton, Nelson, Lyttelton, Dunedin and Bluff harbours, where it was already known to be present. *S. clava* was recorded in Waitemata and Lyttelton harbours, where its presence had been reported previously.

Non-target introduced species recorded during the present survey included an introduced species of barnacle, *Austromegabalanus psittacus*, collected from Wellington Harbour and representing a new record for New Zealand (and the first record outside its South American native distribution). The survey also confirmed the persistence of the crab *Pyromaia tuberculata* in Whangarei Harbour, where it was recorded for the first time during the summer 2002/2003 surveys.

Although the present survey detected both of the target species already present in New Zealand (*U. pinnatifida* and *S. clava*) in those harbours where they are known to occur, there remain problems of lack of sensitivity of the methods used. Inglis et al. (2005a) identified the difficulty of achieving sufficiently high probabilities of detection of the target species that would allow effective, early management of new incursions. Low probabilities of detection are likely when the areas of suitable habitat are large relative to the areas that can be searched at acceptable cost. Despite increased levels of replication for 2 of the sampling methods (diver and shore searches) during the present survey, these limitations remain.

The approach used for prioritising sampling within habitats most at risk of incursion by the target species provides a rational way of defining the principal search area and deciding on the appropriate allocation of sampling effort and resources needed to achieve early detection within them. In cases where the search area is large, it may be unreasonable to expect a high probability of detection at an early enough stage for eradication. Nevertheless, as the field surveys show, even with low statistical detection probabilities, there are significant benefits from regular surveillance in detecting both target species and unexpected new arrivals.

1. Introduction

Introduced plants and animals pose a continuing threat to New Zealand's economic, social, and natural ecological values. Preventing the entry into New Zealand and establishment of high impact exotic pests (HIEP) is, therefore, the major priority for biosecurity (NZ Biosecurity Council 2003). Surveillance and incursion response form an important second line of defence, after border-based quarantine, for management of HIEPs (Carter 1989, Nyrop et al. 1999). Although there is a long history of active surveillance for detection and control of exotic plants and agricultural pests (Binns et al. 2000), little consideration has been given to the design and application of pest surveillance in natural ecosystems and, in particular, marine environments.

Public confidence in active surveillance requires it to be efficient with a high likelihood of detecting unwanted species. This can be difficult to achieve in complex natural ecosystems. Founding populations of pests are often sparse and aggregated, making them easy to miss using conventional sampling designs (Binns et al. 2000). In natural ecosystems, these difficulties are compounded by greater heterogeneity in the affected ecological assemblages and, often, by incomplete taxonomic knowledge of native flora and fauna, making identification of incursions by non-indigenous species problematic (Carlton 1996).

Potentially harmful marine species are being transported around the world's oceans at an unprecedented rate (Ruiz et al. 2000, Carlton 2001). Although measures are being taken to reduce the global transport and release of organisms by shipping, aquaculture, the aquarium trade and other vectors, these are not yet sufficiently effective to prevent the introduction of all species to New Zealand waters. New incursions by unwanted species can be expected into the foreseeable future (Hewitt et al. 2004). There is, therefore, a need to develop cost-efficient programmes of surveillance that will allow early detection of new incursions by unwanted marine species so that measures can be taken to contain or eradicate the outbreak (Simberloff 2003). There are significant challenges for surveillance monitoring and early detection in marine environments. High natural biodiversity, difficulty of access, and limited visibility make surveys of marine ecological assemblages labour intensive and expensive, and place significant logistic and cost constraints on the spatial and temporal extent of sampling programmes.

A previous report (Inglis et al. 2005a) described the design and trial of a targeted surveillance programme for seven marine pests that are currently listed on the New Zealand register of Unwanted Organisms under the Biosecurity Act 1993. The methods used were developmental, but drew upon approaches to surveillance design that have been developed for pests in other ecosystems. An important objective of the

programme was to evaluate the efficiency of the approach and its outcomes for the seven marine species.

Sampling locations were selected based on proximity to high-volume commercial ports and marinas that are first entry points for international vessels. The trial surveillance programme was focussed on eight harbours - Whangarei, Waitemata, Tauranga, Wellington, Nelson, Lyttelton, Otago and Bluff - identified in an earlier report as high risk (Inglis 2001). These harbours range in size from 12.7 ha (Havelock Wharf and Marina) to 22578 ha. (Tauranga Harbour) and contain a variety of different marine or estuarine habitats. Therefore a risk-based stratification of environments within each harbour was used to prioritise areas for sampling. This was done: (1) by developing detailed descriptions of the habitat associations of each species, (2) by using hydrodynamic and particle dispersion models for each harbour to simulate likely patterns of dispersal of ballast water or larvae away from high-risk entry points, and (3) developing Habitat Suitability Index (HSI) models for each harbour.

Inglis et al. (2005a) described the results of four surveys undertaken in each of the eight harbours between October 2002 and September 2004 to trial and develop the methodology. Four field surveys for the target species were undertaken in each of the harbours between October 2002 and September 2004. Repeat surveys of each harbour were spaced approximately 6 months apart to encompass summer and winter conditions in each location. The only target species detected during the surveillance fieldwork was the Asian kelp *Undaria pinnatifida* which was recorded within Wellington, Nelson, Lyttelton and Dunedin harbours, where it was already known to be present.

Two new species of portunid crab, previously unknown from coastal New Zealand waters, were discovered during the surveys, namely *Ovalipes elongatus* and a new species in the genus *Nectocarcinus*. The surveys also provided new records of range expansions for the introduced Asian crabs *Charybdis japonica* (in Whangarei) and *Pyromaia tuberculata* (Whangarei), the Japanese sea anemone *Haliplanella lineata* (Nelson) and the introduced red alga *Polysiphonia senticulosa* Harvey (Lyttelton).

The numbers of samples taken in each harbour during the field surveys generally provided low probabilities of detection of manageable-sized incursions (i.e. <1.5 ha.) for most of the target species (see Inglis et al. 2005a for a description of the methods for estimating probabilities of detection). The chance of a sizeable incursion being missed because of statistically low sample numbers, sparse distribution of an incursion and the chance placement of survey locations is amply illustrated by Waitemata Harbour where less than 0.6 % of the total linear distance of the artificial structures could be sampled on each survey.

The present report describes the results of a further survey, undertaken in summer (December-May) 2005-2006, following a gap of a year since the previous survey. This survey followed the methods of previous ones, but included an additional port (Picton, including Havelock), increased numbers of samples for all of the methods used, and an additional target species, the solitary ascidian *Styela clava*.

Styela clava was first collected in New Zealand in August 2005, in the Viaduct Harbour, Waitemata Harbour and was declared an Unwanted Organisms under the Biosecurity Act 1993 in October 2005. It has subsequently been found to occur in Tutukaka Marina (Northland), the Waitemata Harbour, Hauraki Gulf and Firth of Thames, Lyttelton Port and Lyttelton Marina (Gust et al. 2006a). Small numbers of individuals have also been reported on boats from Opua Marina (Gust et al. 2006b), Waikawa Marina, near Picton (Morrisey 2005) and Nelson Port (Morrisey et al. 2006). Further information is given in Appendix 1.

1.1 Target species and locations for surveillance

The eight species that were the focus of the study are described in Table 1 (more detailed information on each species is given in Appendix 1). Each has a prior history of invasion outside New Zealand, is known to have significant impacts on native ecosystems or economic values in the regions it has invaded, is capable of surviving in New Zealand coastal waters, and is currently listed on the New Zealand register of Unwanted Species. Only two of these species - the Asian kelp, *Undaria pinnatifida* and the clubbed tunicate, *Styela clava* - are already present in New Zealand (Forrest et al. 2000, Gust et al. 2006a).

The surveillance programme was focussed on nine harbours – Whangarei, Waitemata (including the Viaduct Harbour, Hobson West, Bayswater and Westhaven Marinas), Tauranga, Wellington, Picton (including Waikawa Marina, Shakespeare Bay and Havelock), Nelson, Lyttelton, Otago and Bluff. These harbours were identified in an earlier report as high risk because of the pattern and volume of international shipping within them, the availability of suitable habitat for the target species, and their history of invasion by non-indigenous species (Inglis 2001). Picton, the Viaduct Harbour, Hobson West, Bayswater and Westhaven Marinas were not included in the first four surveys. Picton and Havelock are close to the marine-farming regions of the Marlborough Sounds and Golden and Tasman Bays. MAF Biosecurity New Zealand has identified the risk posed by *Styela clava* to New Zealand's aquaculture industry as "extreme" (Kluza et al. 2005). This assessment was based on its invasive behaviour in aquacultural regions of eastern Canada and its ability to foul equipment and possibly

to compete for space and food with the cultured stock. *S. clava* has been reported from the Viaduct Harbour, Hobson West, Bayswater and Westhaven Marinas in Waitemata Harbour (Gust et al. 2006a) and Waikawa Marina, Picton (Morrisey 2005).

Table 1 Descriptions of the eight unwanted species that were the subject of the study.

Common name	Scientific name	Native range	Introduced range
Asian kelp	*Undaria pinnatifida* (Harvey) Suringar	Japan, Korea, China	New Zealand, California, Mexico, Argentina, U.K., France, Italy, Tasmania, Victoria
Mediterranean aquarium weed	*Caulerpa taxifolia* (M.Vahl) C. Agardh, 1817	Tropical Pacific, Atlantic and Indian Oceans	Mediterranean, California, New South Wales, South Australia
Northern Pacific seastar	*Asterias amurensis* Lütken, 1871.	North Western Pacific	Tasmania, Victoria
Mediterranean fanworm	*Sabella spallanzanii* (Viviani, 1805)	Mediterranean & Atlantic coasts of Europe	Brazil, Western Australia, South Australia, Victoria
European green crab	*Carcinus maenas* (Linnaeus, 1758)	Europe, North Africa	Atlantic & Pacific coasts of North America, South Africa, Victoria, Tasmania
Chinese mitten crab	*Eriocheir sinensis* H. Milne Edwards, 1853	North Korea, China	U.K., central & western Europe, California
Asian clam	*Potamocorbula amurensis* (Schrenck, 1867)	Russia, China, Korea, Taiwan, Japan	California
Clubbed tunicate	*Styela clava* (Herdman 1882)	Russia, China, Korea, Japan	New Zealand, Atlantic and Pacific coasts of North America, northwest Europe, U.K., Victoria, New South Wales

2. Developing a Sampling Plan

2.1 Determining where to sample

2.1.1 Transportation nodes as high risk entry points

Founding populations of invasive species tend to occur close to major points of entry into the country for the transportation pathways that the species are associated with. Most non-indigenous marine species are introduced into New Zealand unintentionally, in ballast water or as biofouling on the submerged surfaces of international ships (Cranfield et al. 1998). High-volume entry points for international shipping, such as commercial ports and marinas of first entry are, therefore, important foci for early detection surveys.

International shipping is the most likely pathway of introduction for seven of the eight unwanted species that were the subject of this project. The notable exception is the aquarium weed, *Caulerpa taxifolia*, which is thought to have established invasive populations overseas primarily as a result of intentional release of aquarium specimens (Meinesz et al. 2001, Williams 2003, Glasby et al. 2005). Introduced populations of *Eriocheir sinensis* and *Carcinus maenas* on the Pacific coast of the U.S.A. are also suspected of having established as a result of deliberate release and transport of bait species for fisheries, respectively (Cohen & Carlton 1997, Carlton 2001). High risk entry points for these alternative pathways are likely to be more diverse and less predictable than those for species closely associated with shipping. In this project, we focussed predominantly on shipping ports and marinas as the most likely points of entry in each harbour for the eight target species, following the protocol of previous surveys.

2.1.2 Patterns of distribution of founding populations

Because marine species in general, and each of the target species in particular, have dispersal stages that are transported by water movements (e.g. mobile adults, larvae or fragments), founding populations are unlikely to be confined solely to operational areas of ports and marinas, but may be distributed throughout nearby natural environments, within the dispersal range of each species. The nine harbours considered in this study are large embayments or estuaries that range in size from 12.7 ha (Havelock Wharf and Marina) to 22578 ha. (Tauranga Harbour) and contain a variety of different marine or estuarine habitats. Over such large areas, detection and description of rare and clustered founding populations is best achieved by stratifying or clustering most sampling within strata that have the greatest likelihood of containing the species (Christman 2000, Thompson 2004). For this to be achieved, it is

first necessary to define and identify these sample strata. This was done in two ways: (1) by developing detailed descriptions of the habitat associations of each species, and (2) by using hydrodynamic and particle dispersion models for each harbour to simulate likely patterns of dispersal of ballast water or larvae away from high risk entry points. Inglis et al. (2005a) gave details of the methods.

2.2 Methods

2.2.1 Field surveys

The field surveys for the target species were undertaken in each of the nine harbours during the period December 2005 (Picton and Nelson) to May 2006 (Tauranga), representing summer conditions. The four previous field surveys were undertaken between October 2002 and September 2004, spaced approximately 6 months apart to encompass summer and winter conditions in each location.

Sampling strategy

Because marine organisms are typically aggregated in their spatial distribution, they tend to be absent from, or in comparatively low abundance at most locations and in large densities in relatively few places (Gray 2002). This pattern is even more extreme for the small founder populations of introduced species, which, at least initially, are likely to be absent from most areas and to occur in aggregations at relatively few locations (Gaston 1994). For example, during the initial stages of its invasion of Port Phillip Bay, Australia, the seastar *Asterias amurensis* was found at only two out of more 70 locations surveyed in the bay (Garnham 1998). This pattern of distribution – locally abundant, but geographically restricted founder populations – suggests that, in most instances, the probability of detection within locations where the species is present is likely to be greater than its expected rarity among locations. Since eradication and control efforts are likely to be most successful when infestations are relatively localised, surveys that optimise the number of locations surveyed will stand the best chance of detecting founding populations with aggregated distributions (Green & Young 1993). Thus, given limited resources, surveying a relatively large number of discrete locations using rapid sampling techniques is likely to be more effective than intensive searches of a few key locations (although there will be a point at which the survey sensitivity is compromised by under-sampling at each location). This basic assumption - the need to sample a large number of survey locations in each harbour - formed the foundation for our choice of survey methods.

Survey methods

The rationale for selection of survey methods for the target-species surveillance is discussed in the report on the previous surveys (Inglis et al. 2005a). Because the purpose of the surveillance programme is detection, not enumeration, we selected techniques in which the presence or absence of the target species could be determined rapidly within a sample, thereby allowing a comparatively large number of locations to be sampled on each survey.

The methods chosen for the present survey follow those of previous surveys, and Inglis et al. (2005a) gave detailed descriptions and guidelines for their use. Summary descriptions are given below, with notes on any variations from previous surveys, and further details are given in Table 2.

Baited crab traps

Baited box traps were used to sample adult crabs (i.e. *Carcinus maenas* and *Eriocheir sinensis*). These are a light-weight, commercially available, collapsible trap (63 cm x 42 cm x 20 cm) with a 1.3-cm mesh netting (Figure 1). Crabs enter the traps through slits in inward sloping panels at each end. Bait is contained within an internal mesh bag secured to the upper frame.

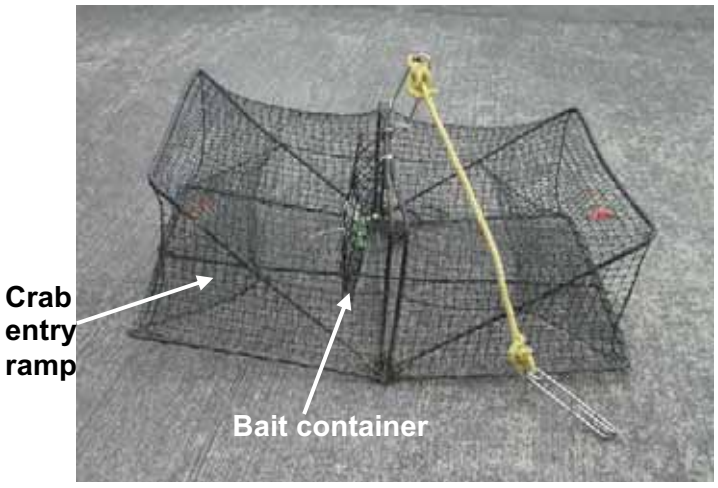

Figure 1 **Baited box trap used to sample adult crabs.**

We deployed the crab traps in shallow subtidal habitats. Most sampling effort was centred near intertidal rocky shores, wharf pilings, breakwalls and other habitats with complex physical structure (e.g. shellfish beds and seagrass meadows). At each survey

location, we deployed 3 baited traps, spaced 3–5 m apart on a single anchored and buoyed trap line. Each trap was baited with 1-2 frozen pilchards. The lines were set on the sea floor and left to soak overnight before retrieval.

For each line deployed we recorded the GPS location, water depth and times of deployment and retrieval at each site. All crabs recovered from the traps were identified in the field. Specimens that resembled the target species or which could not be identified by the field team were retained for formal identification by a specialist taxonomist.

Baited starfish traps

Whayman-Holdsworth starfish traps (Andrews et al. 1996) were used to catch asteroids and other large benthic scavengers. These are circular traps with a basal diameter of 100 cm and an opening on the top of 60 cm diameter (Figure 2). The sides and bottom of the trap are covered with 26-mm mesh and a plastic, screw-top bait holder is secured in the centre of the trap entrance.

Figure 2 **Whayman-Holdsworth starfish trap.**

Starfish traps were set adjacent to wharf pilings, breakwalls and other artificial habitats or near known shellfish beds where *Asterias amurensis* is likely to be present. At each survey location, two starfish traps were deployed on the sea floor clipped to a single buoyed line. The traps were left overnight before being retrieved. Each trap was baited with 2-3 frozen pilchards. For each line we recorded the GPS location, water depth and times of deployment and retrieval. All mobile organisms recovered from the traps were identified in the field. Specimens that resembled *A. amurensis* and which

could not be confidently identified by the field team were retained for formal identification by a specialist taxonomist.

Unbaited crab traps ("crab condos")

Baited traps do not sample juvenile and sub-adult Chinese mitten crabs (*Eriocheir sinensis*) effectively because these life stages have a largely herbivorous diet and occupy slack-water habitats where bait odours are not well-dispersed (Veldhuizen & Stanish 1999). To sample juvenile crabs we used artificial shelters ("crab condos") that had been designed for surveys of *E. sinensis* in San Francisco Bay. The shelters were made of 9 PVC tubes (15 cm long, 5 cm diameter) held together in a plastic mesh basket (Figure 3). The tubes mimic the burrows inhabited by sub-adult crabs (16-42 mm carapace width) in muddy riverbanks and have higher rates of detection than baited traps in these environments (Veldhuizen 2000). Crab condos were set primarily in riverine tributaries or sheltered muddy embayments in brackish areas of each harbour. Locations for deployment were restricted in Wellington, Picton and Otago harbours by the limited numbers of streams of sufficient size. At each survey location we deployed three condos on the seafloor, spaced ~5 m apart and attached to a weighted and buoyed line. The condos were left to soak for at least three days before being retrieved (Veldhuizen 2000). We recorded the GPS location, water depth and times of deployment and retrieval for each line. All mobile organisms recovered from the traps were identified.

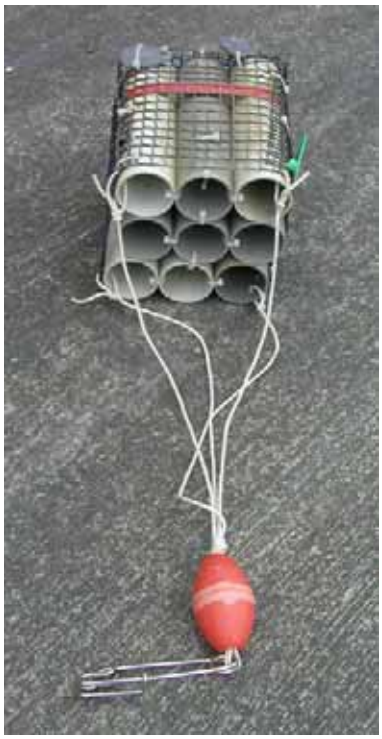

Figure 3 Unbaited crab trap ("crab condo").

Epibenthic sled tows

An Ocklemann sled (hereafter referred to as a "benthic sled") was used to sample soft-sediment habitats for *Potamocorbula amurensis*, *Sabella spallanzanii*, *Asterias amurensis* and *Caulerpa taxifolia*. The sled is approximately one meter long with an entrance width of ~0.7 m x 0.2 m. A short yoke of heavy chain connects the sled to a tow line (Figure 4). The mouth of the sled partially digs into the sediment and collects organisms in the surface layers to a depth of a few centimetres. Runners on each side of the sled prevent it from sinking completely into the sediment so that shallow burrowing organisms and small, epibenthic fauna pass into the exposed mouth. Sediment and other material that enters the sled are passed through a mesh basket (3-mm mesh) that retains organisms larger than about 2 mm.

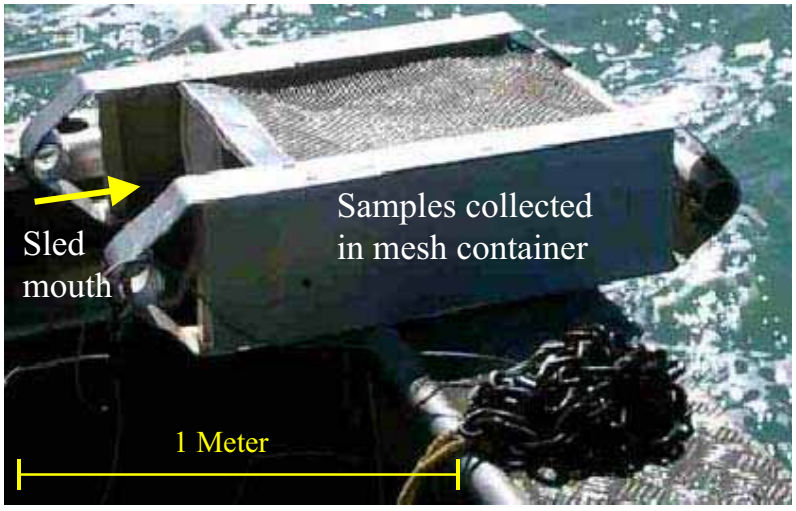

Figure 4 **Benthic Ocklemann sled.**

Sleds were towed for a standard time of 2 minutes at approximately 2 knots. During this time, the sled typically traversed between 80–100 m of seafloor before being retrieved. In muddy sediments, tows were terminated when the sled basket was filled, which sometimes occurred in less than 2 minutes. Sediments and other material recovered in the sled were washed onto a 1-mm sieve and sorted on board the boat. Sorters scanned the washed sample for the target species. Specimens that resembled the target species and which could not be identified confidently by the field team were retained for formal identification by a specialist taxonomist.

The GPS location and water depth at the start of each sled tow were recorded, and observations were made on the sediment type and benthic habitat based on material retrieved in the sled. Sediment was classified visually into one of eight categories represented in Table 3a. The presence of structural habitat (e.g. seagrass, shellfish

beds, macroalgal accumulations, etc) was recorded using the schema presented in Table 3b.

Sled tows were distributed mostly in areas of soft sediments indicated as high risk by the results of the dispersal plume modelling. Where data on the distribution of benthic habitats was available, greater effort was made in muddy sediments (*Potamocorbula amurensis*), shellfish beds (for *Asterias amurensis*) and areas next to public access points (e.g. wharves, boat ramps, marinas; for *Caulerpa taxifolia*, and *Sabella spallanzanii*).

Diver searches

Divers searched for *Sabella spallanzanii*, *Undaria pinnatifida*, *Carcinus maenas*, *Asterias amurensis* and *Styela clava* around piles, floating pontoons and other artificial structures in port and marina environments and on intertidal and shallow subtidal reefs that were identified by the dispersal plume modelling as high-risk. In ports or marinas, the divers searched a sample of 10 pilings (or the equivalent area of pontoon) at each site for the target species. To mitigate the hazard of multiple ascents, the searches entailed a pair of divers swimming at two constant depths (2 m and 4 m) at each site (where visibility permitted). The depths used in the protocol were adopted because the most likely fouling organisms among the target species – *S. spallanzanii*, *U. pinnatifida* and *S. clava* – are typically abundant in the upper 4-5 m of water (Parry et al. 1996, Inglis et al. 2003, Gust et al. 2005).

Visual shoreline searches

In previous surveys, timed (10 minutes) visual searches for the target species were made of intertidal rocky reefs and sloping sandy shorelines, the latter in areas of the harbour where drift material was likely to accumulate[1]. Wherever possible, the searches were made at low tide, but the timing of the search was dictated by the tidal cycle during the week in which the survey was done and by the scheduling of other sampling activities. Two people conducted each search at each site. On areas of rocky reef, the lower shoreline was scanned for the presence of *Undaria pinnatifida* and *Styela clava* and small rocks and cobbles were overturned to look for juvenile *Carcinus maenas*. On sandy shorelines, the searchers walked along the drift line and examined cast material for algae (*U. pinnatifida* and *Caulerpa taxifolia*) and exuviae of moulted crabs (*C. maenas*, *Eriocheir sinensis*).

In the present survey, the number of searches in each port was increased from that in previous surveys (see below), and the extra sampling effort was used to include

[1] The prevailing winds on the preceding days were often a useful guide to where material may accumulate.

pontoons, piles and other structures in marinas and, where access allowed, ports in the visual searches. These represent preferred habitats for *Styela clava* (Gust et al. 2006a). Surface-searches are an effective method for rapid surveys of the distribution of Styela (Gust et al. 2006a), and the inclusion of suitable habitats for *S. clava* in the sampling protocol corresponds with its inclusion in the suite of target species. Shoreline visual searches in previous surveys concentrated on searching beaches and rocky intertidal areas for flotsam.

Non-target non-indigenous or cryptogenic species

Although the survey methods were chosen to sample the target species, they also captured a range of other native and non-indigenous species that were present in each harbour. This provided an opportunity to obtain additional information on the occurrence of other non-indigenous marine species within the samples to improve records of their distribution within New Zealand. A key consideration was that obtaining these additional data should not add significantly to the survey costs by increasing the time required to obtain, sort, or identify the specimens. Thus, additional records were predominantly of species that were easily identified by visual inspection in the field, with verification occurring through identification of voucher specimens by a recognised taxonomist. To ensure consistent collection of information, we provided training to the field teams in identification of the following introduced or cryptogenic species:

- the portunid crab, *Charybdis japonica* (A. Milne-Edwards, 1861)
- the majid crab, *Pyromaia tuberculata* (Lockington, 1877)
- the semelid bivalve, *Theora lubrica* (Gould, 1861)
- the nesting bivalve, *Musculista senhousia* (Benson in Cantor, 1842)
- the cryptogenic, tube-building polychaete, *Chaetopterus* sp. (the Hauraki chaetopterid), and
- the green alga, *Codium fragile* ssp. *tomentosoides* (Sur.) Hariot subsp. (Van Goor) Silva.

Choice of these species was based on: (a) their presence in the initial Summer 1 survey, (b) the relative ease with which they could be identified in the field, and (c) limited existing information on their distribution in the harbours sampled.

In addition, because the field teams consisted of a range of personnel, including some with considerable field experience and skill in taxonomy and parataxonomy, we also instructed the teams to be vigilant to any unusual species in the samples that they had not encountered in previous surveys (within this project or other NIWA surveys) or which they knew to be non-indigenous to New Zealand. The teams were requested to initially check the identification of any such specimens with the team leader on the survey and to retain preserved specimens for formal identification.

Allocation of sampling effort

The total sampling effort in each harbour and survey (i.e. total number of sites surveyed and samples taken) was derived from that used in the previous surveys (Inglis et al. 2005a) which, in turn, was based on average times taken to obtain samples with each method during the first survey in summer 2002/2003 and the number of sites that could be surveyed in the allotted time. The initial estimates of sample time were then used to set targets for the numbers of sites sampled with each technique in subsequent surveys. The allocation of effort among the different survey techniques reflected the relative abundance of each type of habitat in the harbours. For example, most sample effort was allocated to sledding (soft-sediment habitats) and crab trapping (shoreline rocky habitats) because these habitats typically covered larger areas of the harbours than wharves, pilings, and riverine channels.

In the 2002-2004 surveys (excluding the initial Summer 1 surveys), an average of 100 (SD = 9) sites in each harbour was sampled by sled tows, 52 (9) sites by crab trap lines, 10 (5) sites by crab condos, 24 (9) sites by starfish traps, 17 (6) sites by diver searches and 17 (6) sites by shoreline searches.

A summary of the target sampling effort for each survey method in each harbour in the present survey is provided in Table 4. In the present survey, the minimum number of diver searches in each port was increased from 20 to 30 and the minimum number of shoreline visual searches was set at 25. This was done at the request of MAF Biosecurity New Zealand.

Survey plans were developed based on the known distribution of habitat for the target species and outputs from the hydrodynamic modelling. We allocated sampling effort for each sample method and harbour based predominantly on the results of the hydrodynamic modelling and our existing knowledge of the distribution of habitats in each harbour, with highest priority given to suitable habitat for the target species within the predicted dispersion plume. Each harbour was subdivided into large strata (3-4 per harbour) that reflected broad environmental gradients (e.g. head/entrance of the harbour) and the concentrations of particles simulated in the hydrodynamic

modelling. Generally, ~60% of locations surveyed were allocated to the stratum where moderate to high weighted mean concentrations of simulated particles were predicted, with the remainder distributed among the remaining strata. Individual locations surveyed within each habitat type and strata were selected haphazardly on each survey date. The distributions of sampling effort in each harbour in each of the present surveys are presented in Figures 5-92, together with maps of the harbours showing locations mentioned in the text.

Crab condos were not deployed around Picton Harbour because of the lack of suitable rivers entering the harbour and inner Queen Charlotte Sound areas. Condos were deployed in the Pelorus River, west of Havelock (S 41^{o} 17.721' E 173^{o} 37.395'), and the Kaituna River, east of Havelock (S 41^{o} 17.165' E 173^{o} 46.553'). Lyttelton Harbour has very few areas of riverine input and the only significant one lies in the southwest corner of the harbour ("Head of the Bay"). This area is too shallow to allow boat access and access on foot is hazardous because of deep mud. Crab condos were consequently deployed at locations along the northern shore of Lyttelton Harbour and around Quail Island (Figure 70). Inglis et al. (2005a) gave distributions of samples for previous surveys.

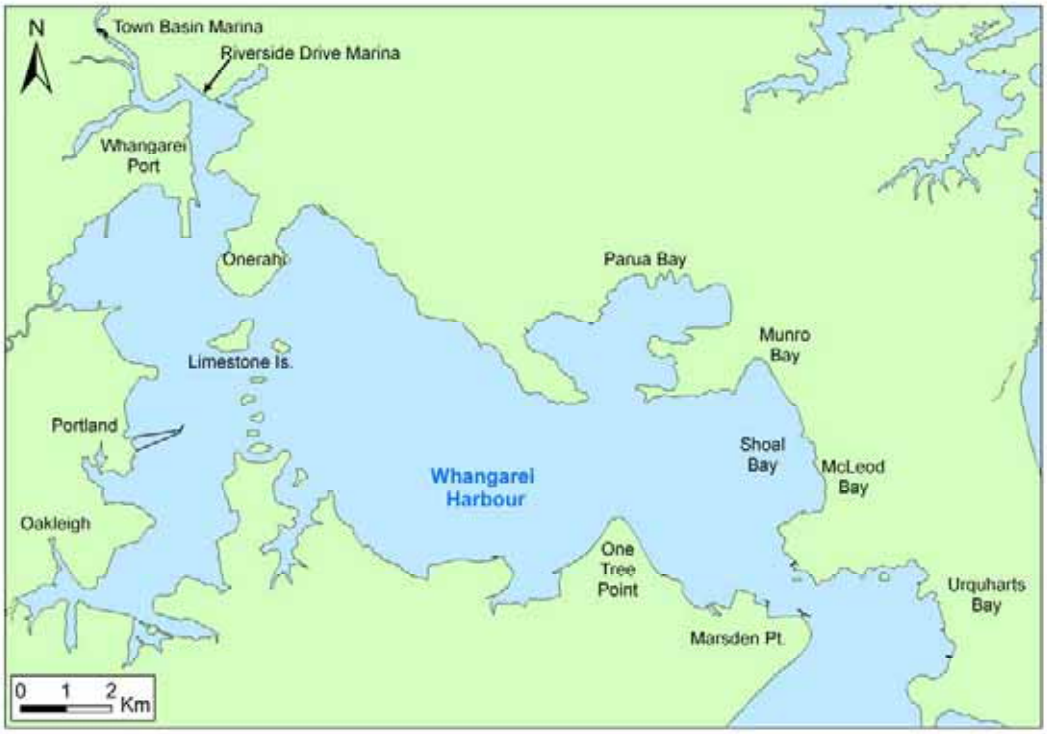

Figure 5 **Map of the sampling area around Whangarei Harbour.**

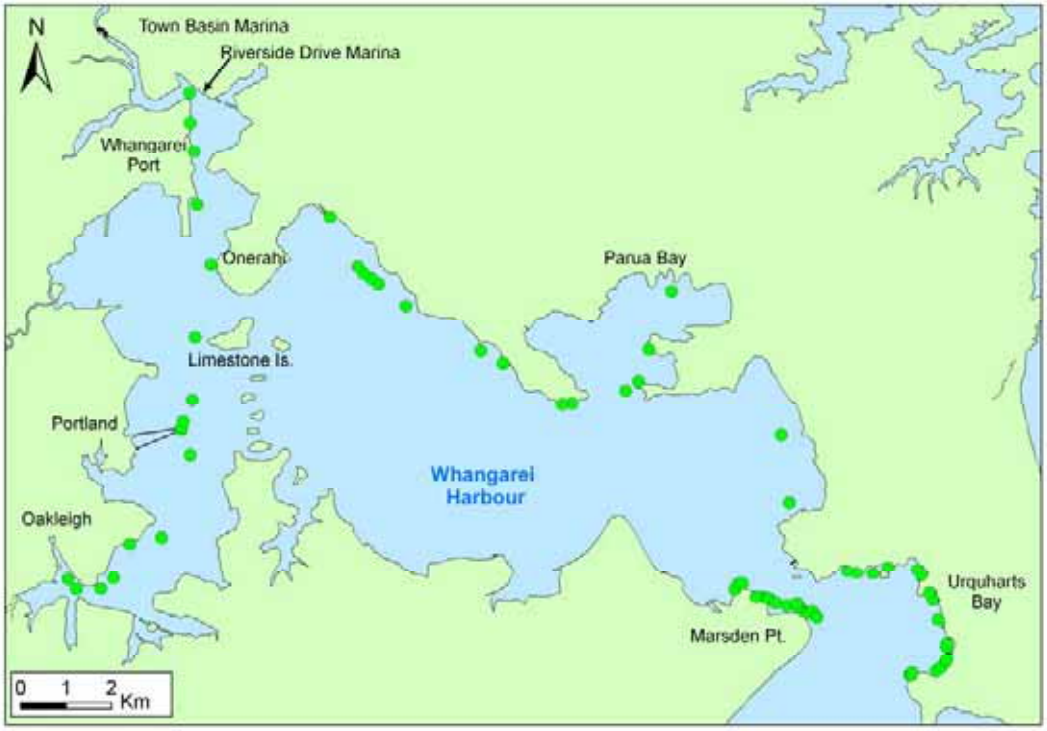

Figure 6 **Distribution of crab traps in Whangarei Harbour, March 2006.**

Figure 7 **Distribution of starfish traps in Whangarei Harbour, March 2006.**

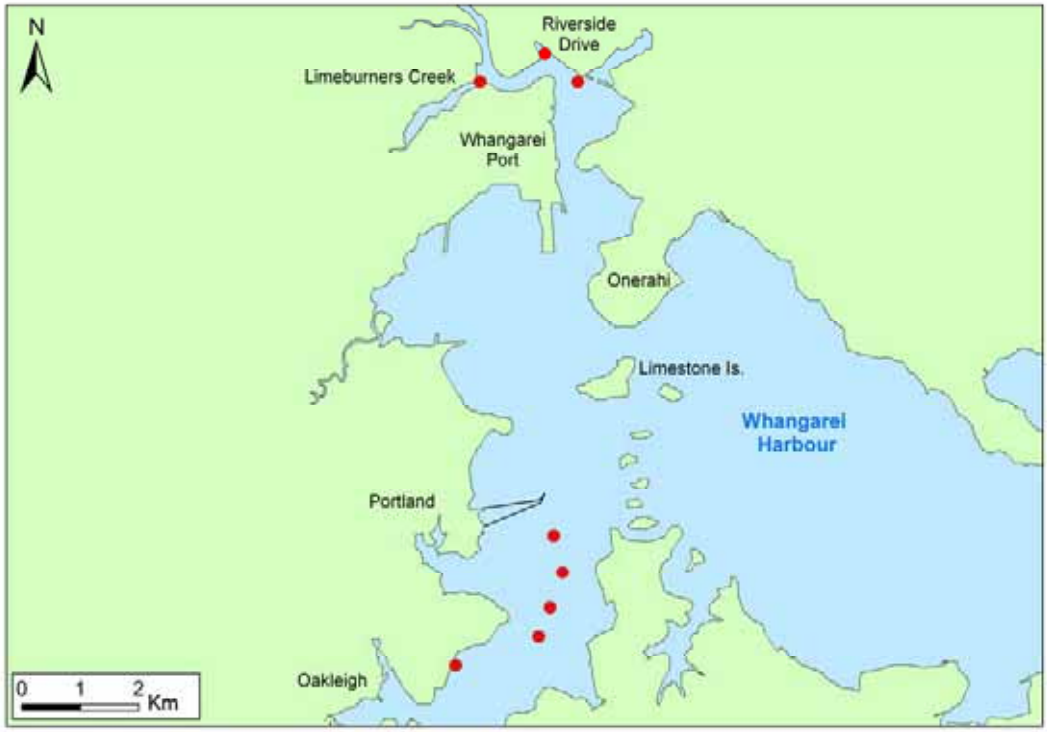

Figure 8 **Distribution of crab condos in Whangarei Harbour, March 2006.**

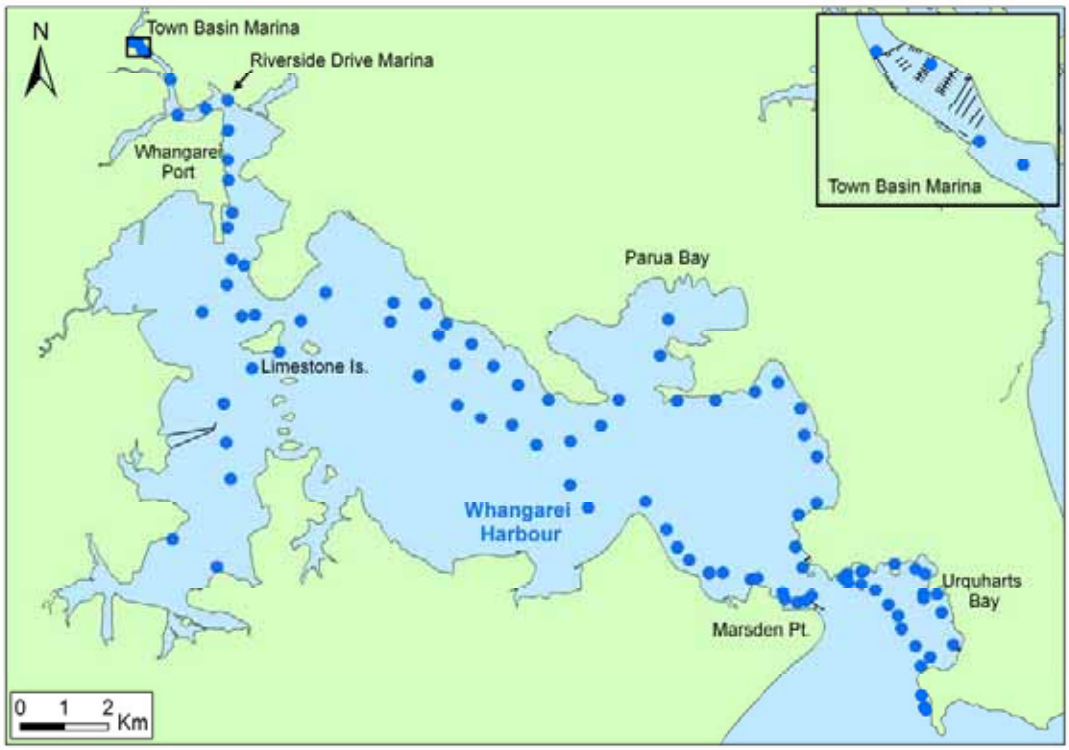

Figure 9 **Distribution of sled tows in Whangarei Harbour, March 2006.**

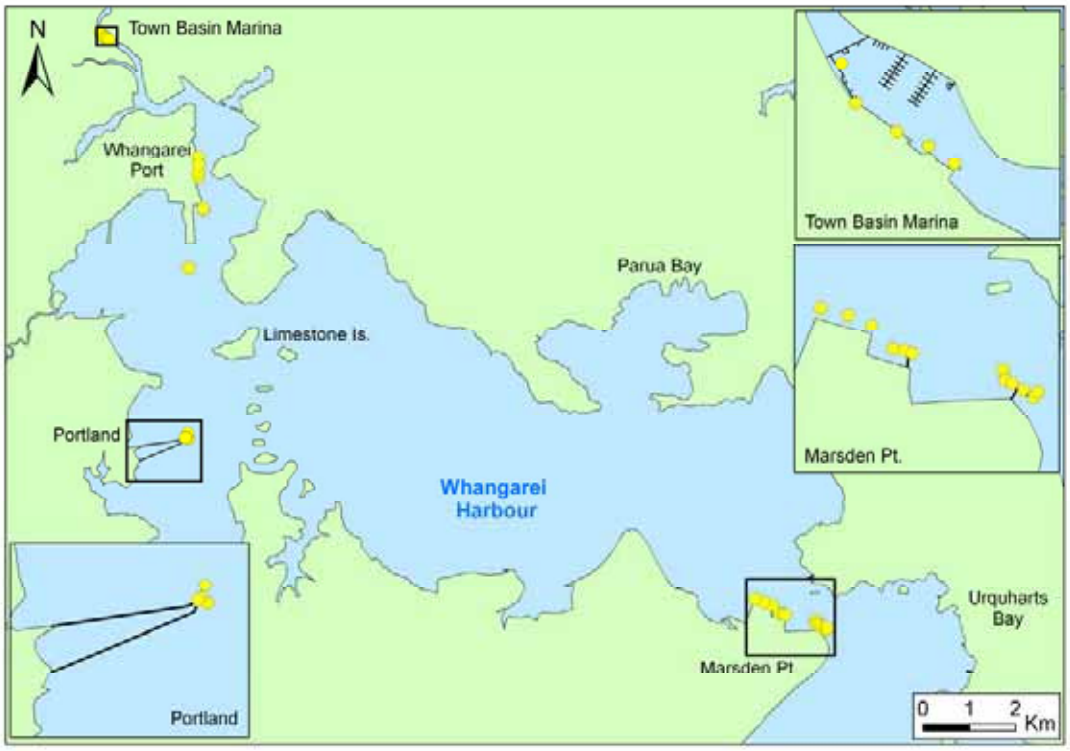

Figure 10 **Distribution of dive searches in Whangarei Harbour, March 2006.**

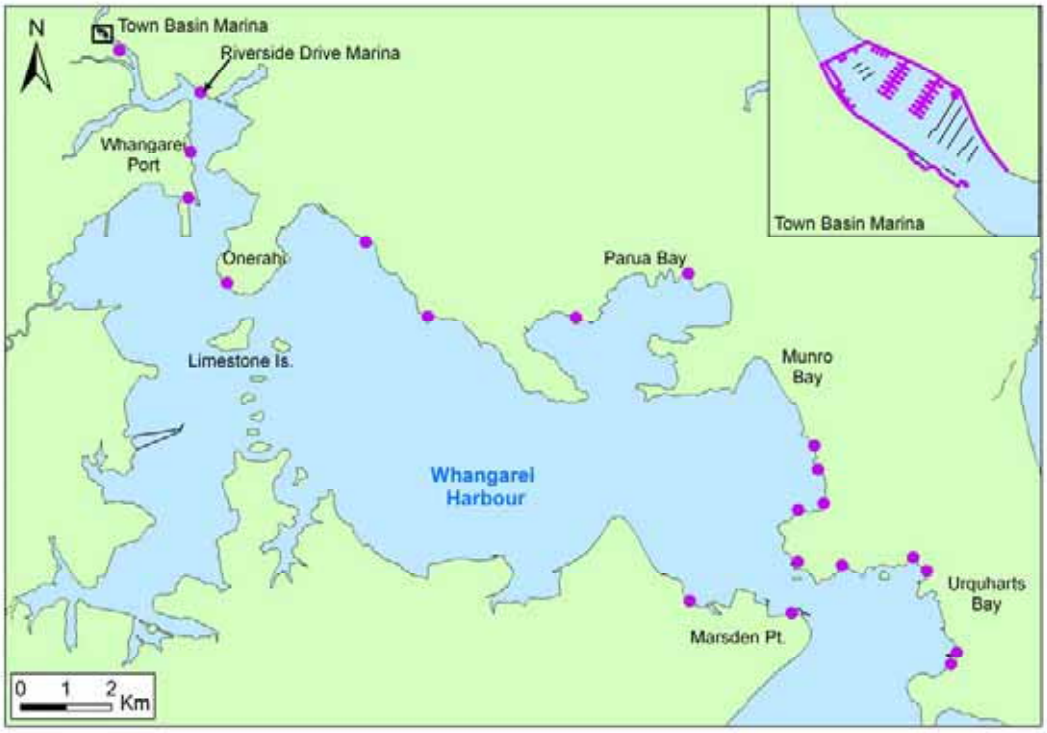

Figure 11 Distribution of shore searches in Whangarei Harbour, March 2006.

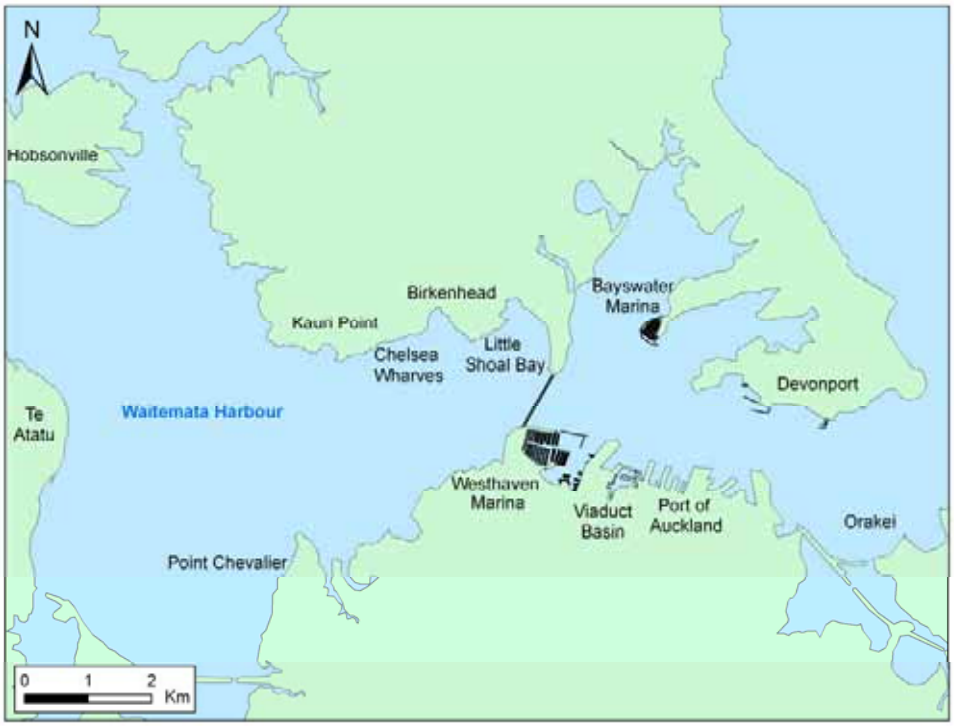

Figure 12 **Map of the sampling area around Waitemata Harbour.**

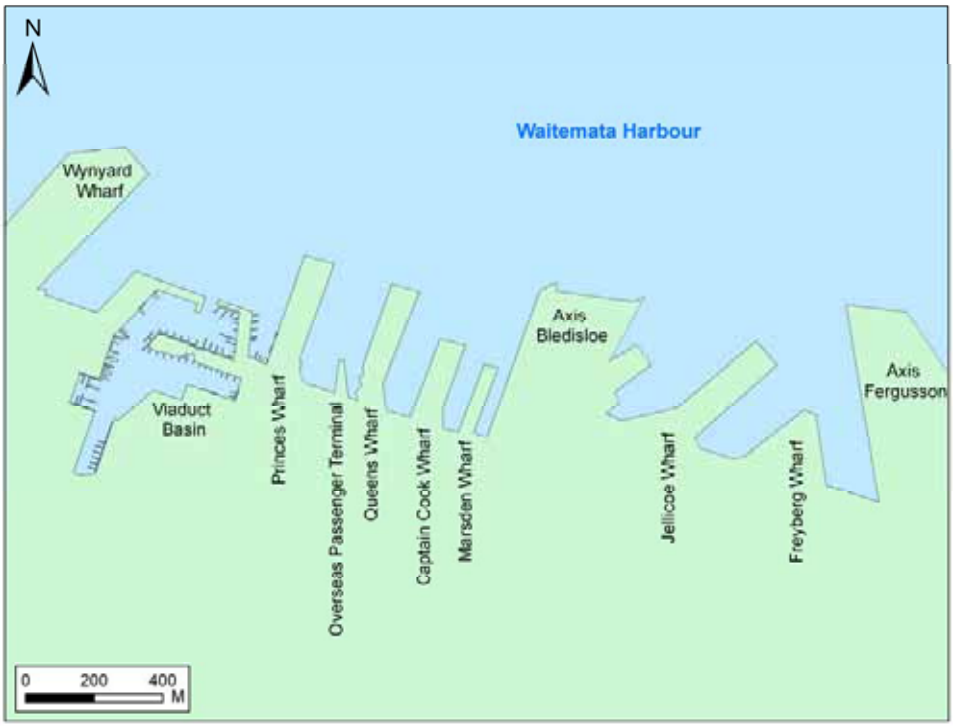

Figure 13 **Detail of the sampling area around Auckland Port.**

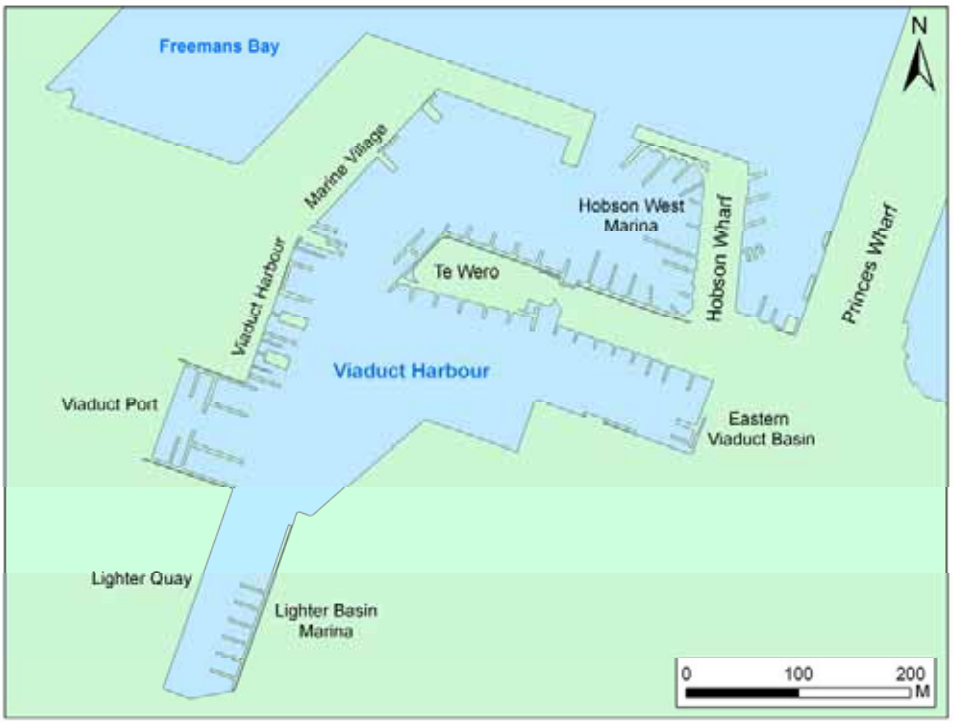

Figure 14 **Detail of the sampling area around Viaduct Harbour.**

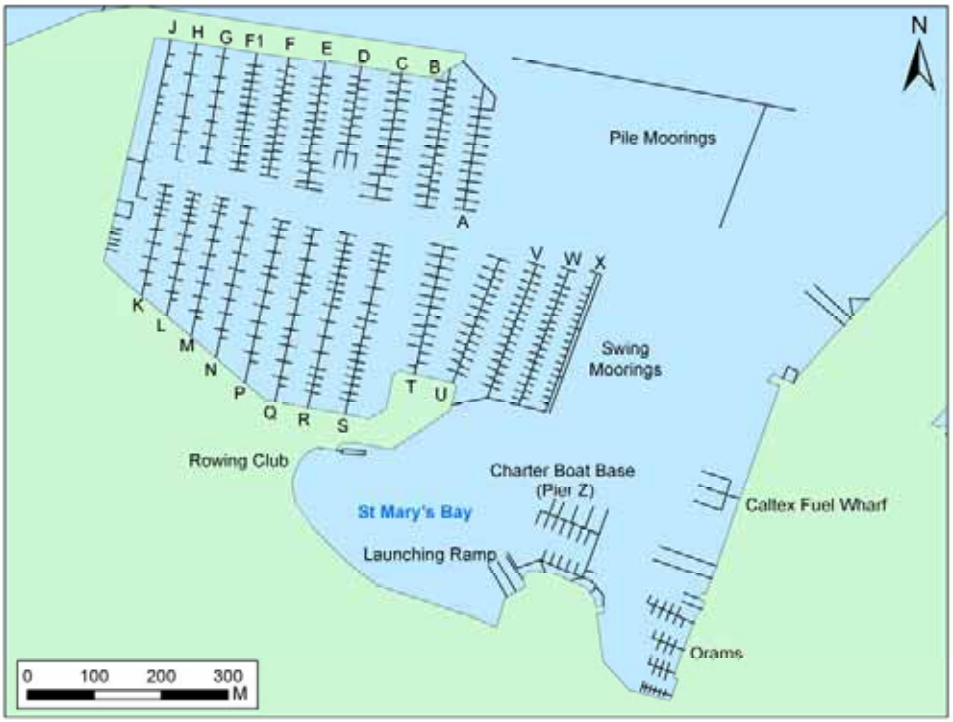

Figure 15 **Detail of the sampling area around Westhaven Marina.**

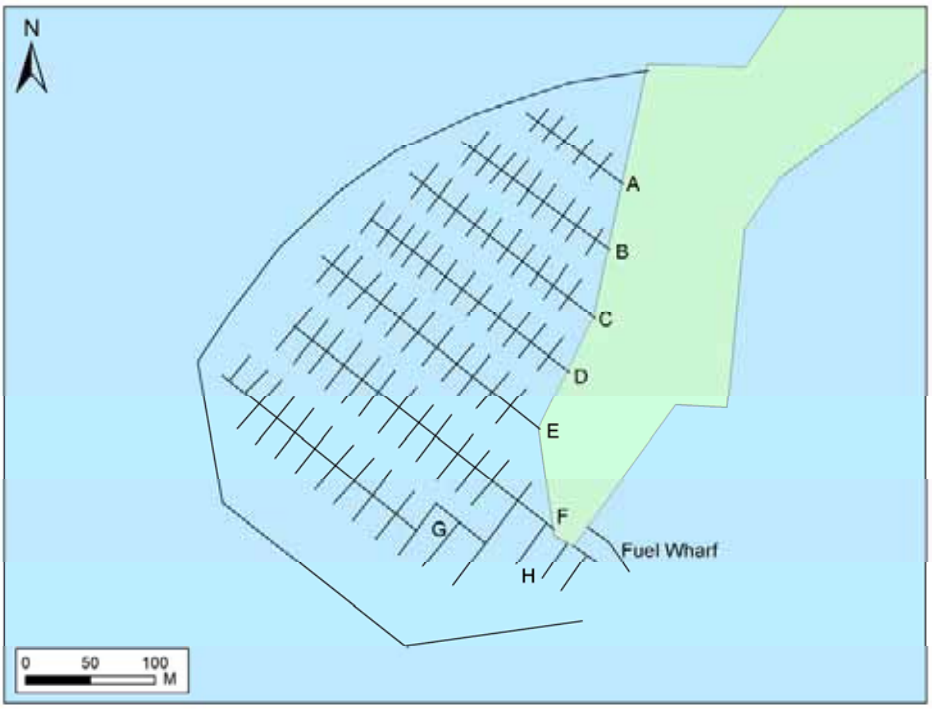

Figure 16 Detail of the sampling area around Bayswater Marina.

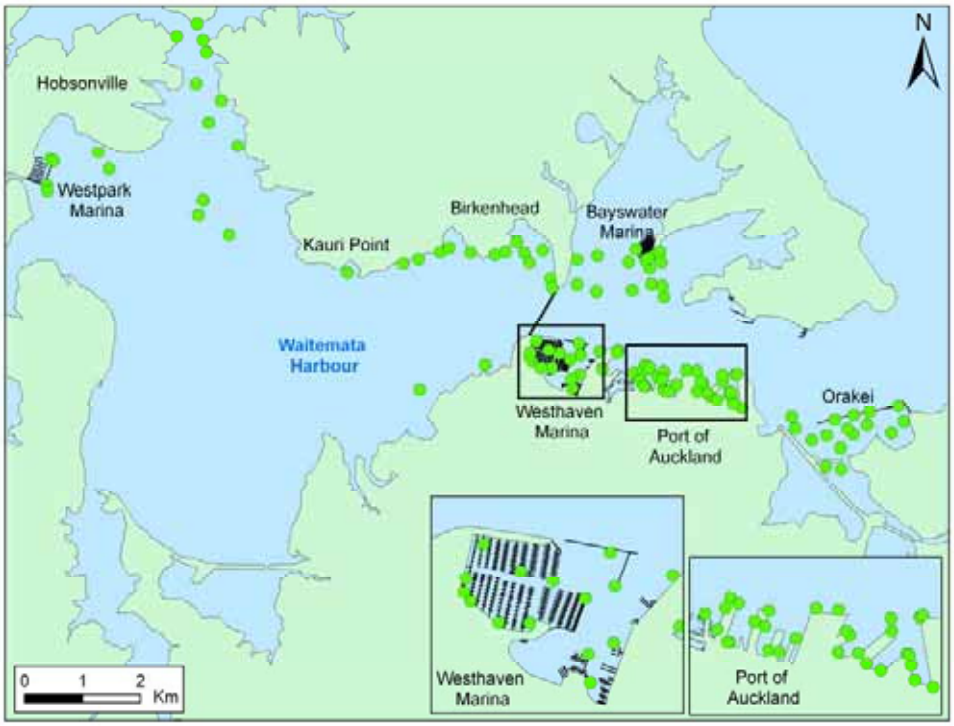

Figure 17 **Distribution of crab traps in Waitemata Harbour, April 2006.**

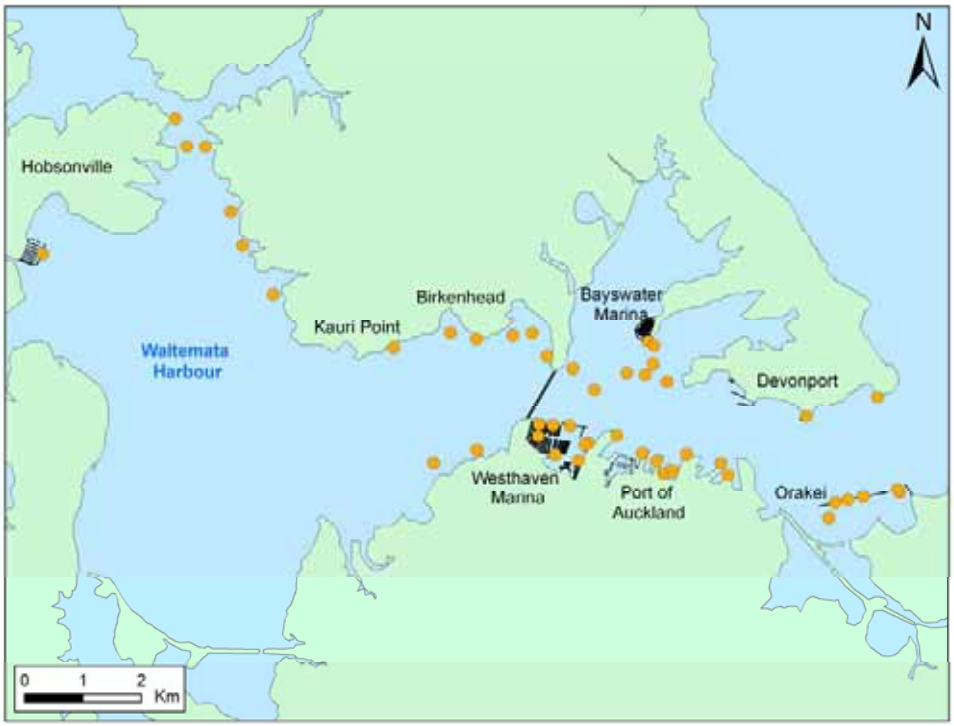

Figure 18 **Distribution of starfish traps in Waitemata Harbour, April 2006.**

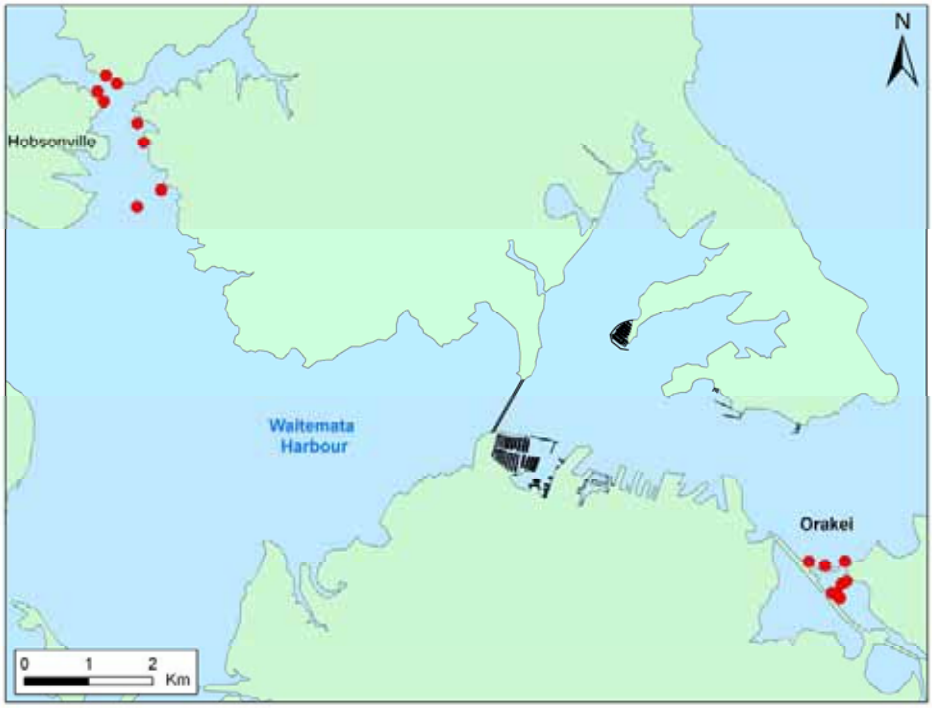

Figure 19 **Distribution of crab condos in Waitemata Harbour, April 2006.**

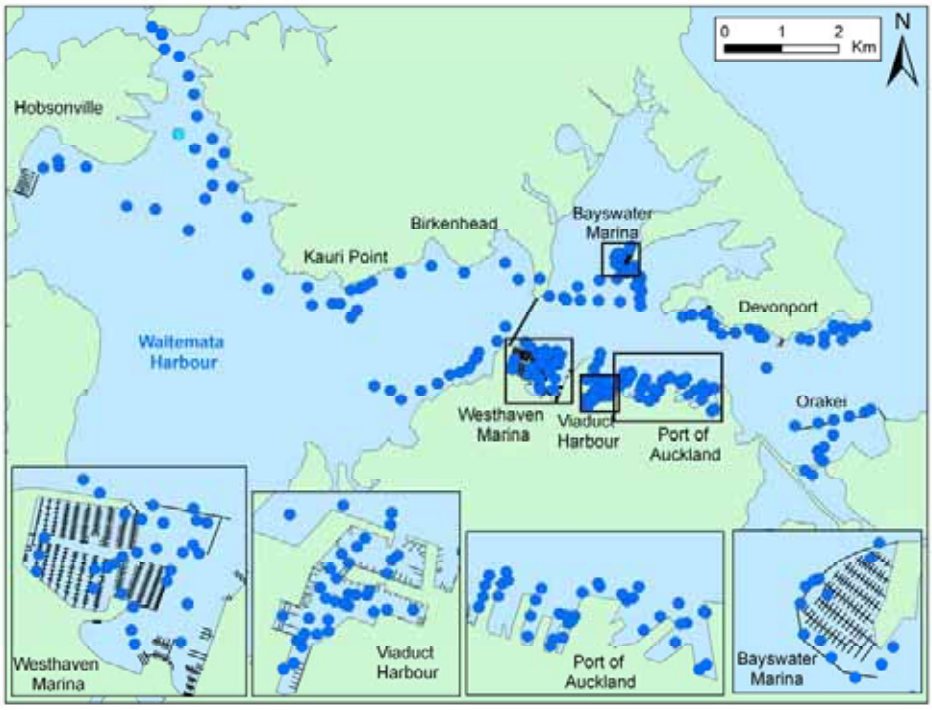

Figure 20 **Distribution of sled tows in Waitemata Harbour, April 2006.**

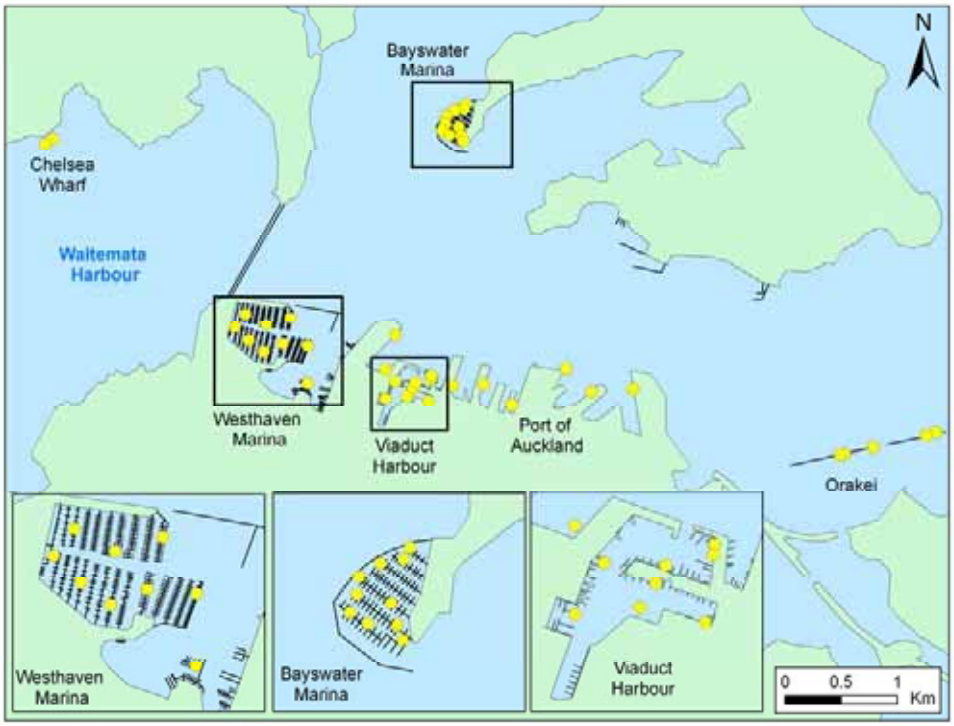

Figure 21 **Distribution of dive searches in Waitemata Harbour, April 2006.**

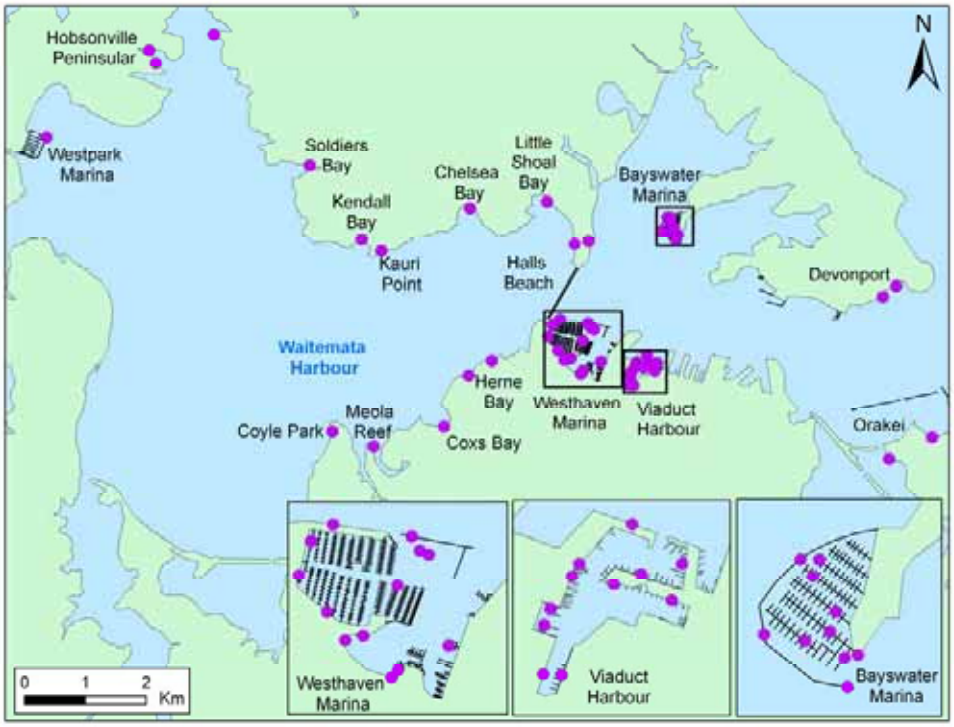

Figure 22 **Distribution of shore searches in Waitemata Harbour, April 2006.**

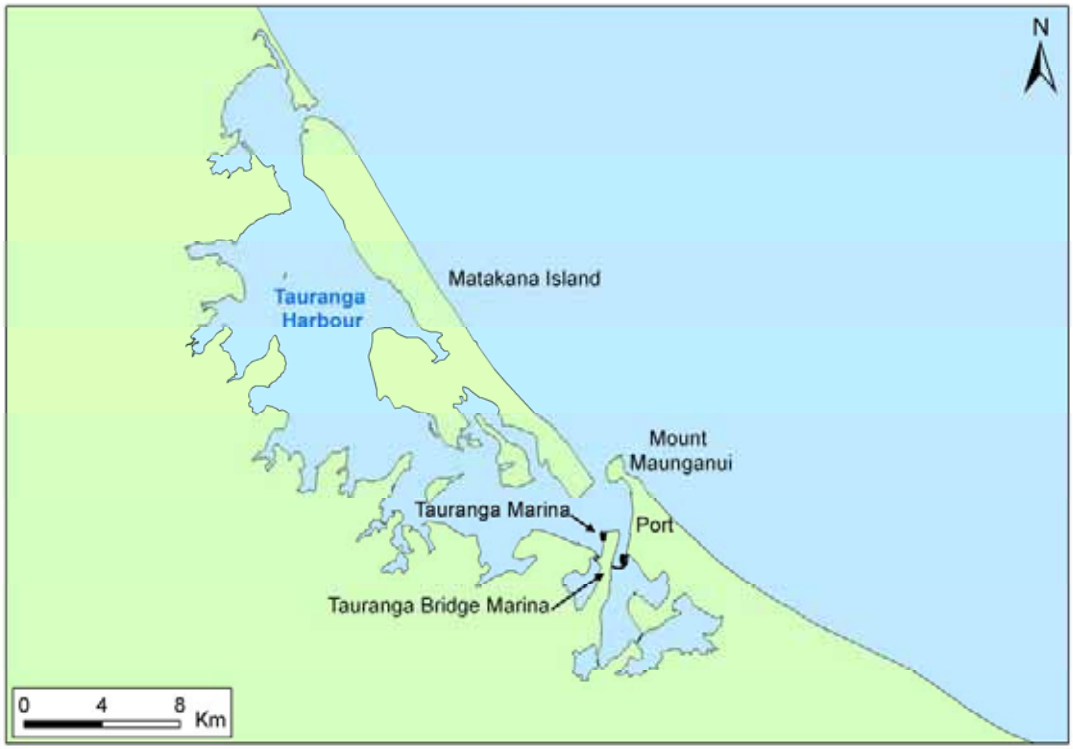

Figure 23 **Map of the sampling area around Tauranga Harbour.**

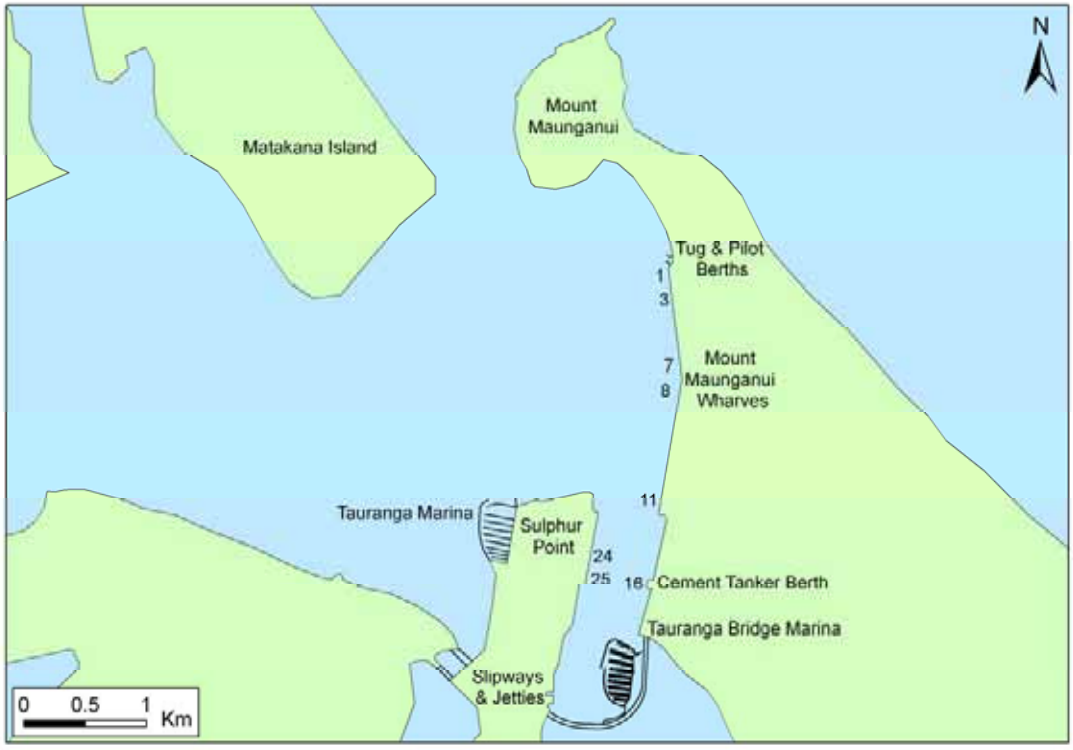

Figure 24 **Detail of the sampling area around Tauranga Port. Numbers identify selected wharves.**

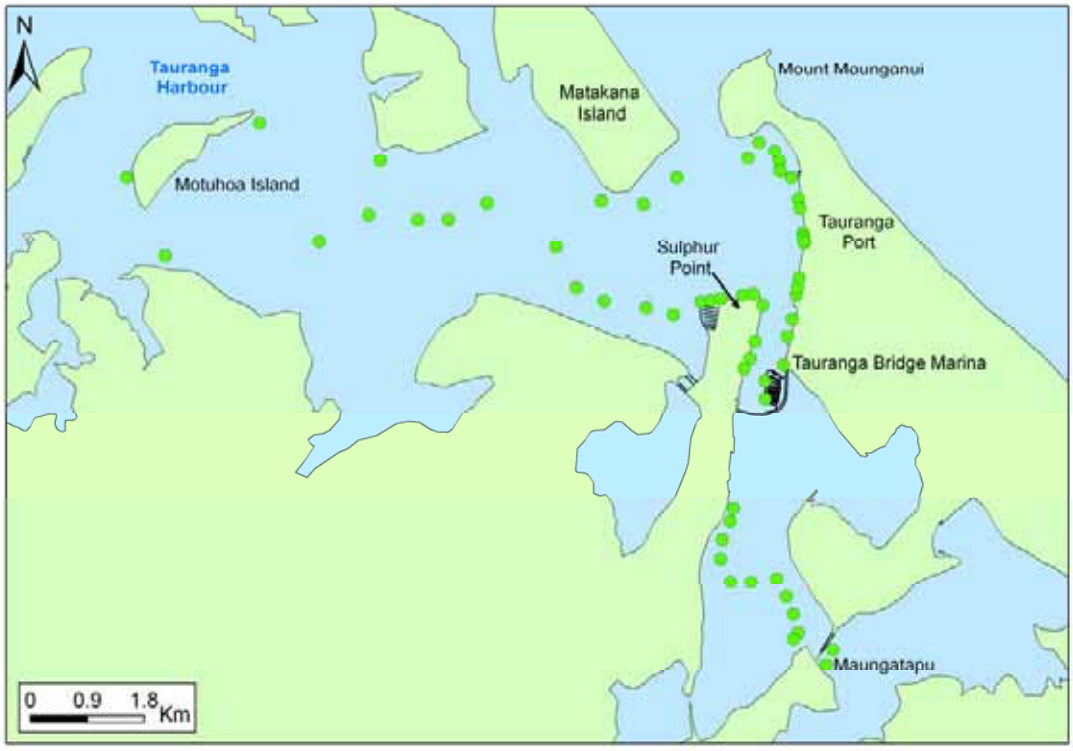

Figure 25 **Distribution of crab traps in Tauranga Harbour, May 2006.**

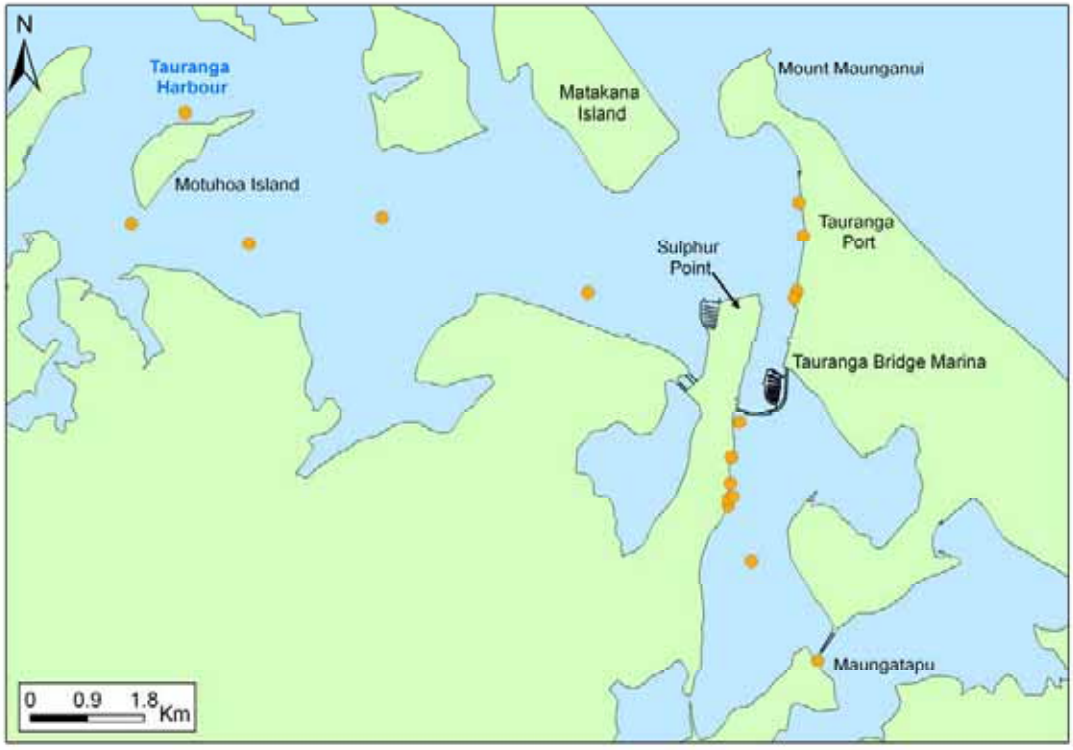

Figure 26 **Distribution of starfish traps in Tauranga Harbour, May 2006.**

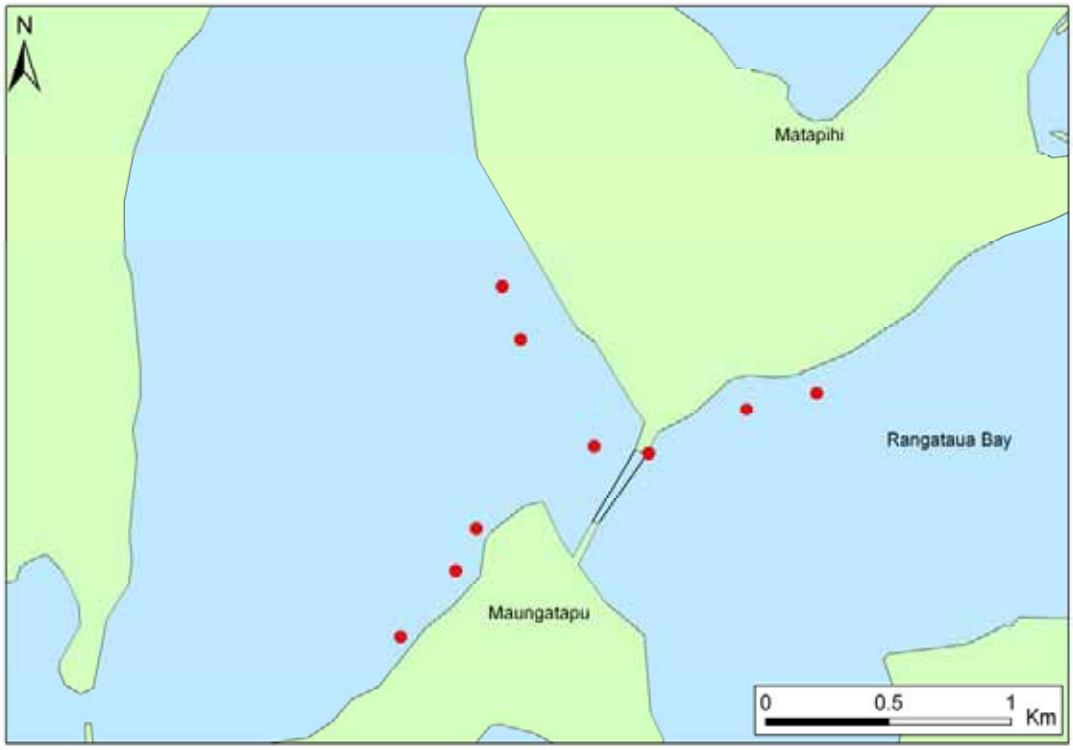

Figure 27 **Distribution of crab condos in Tauranga Harbour, May 2006.**

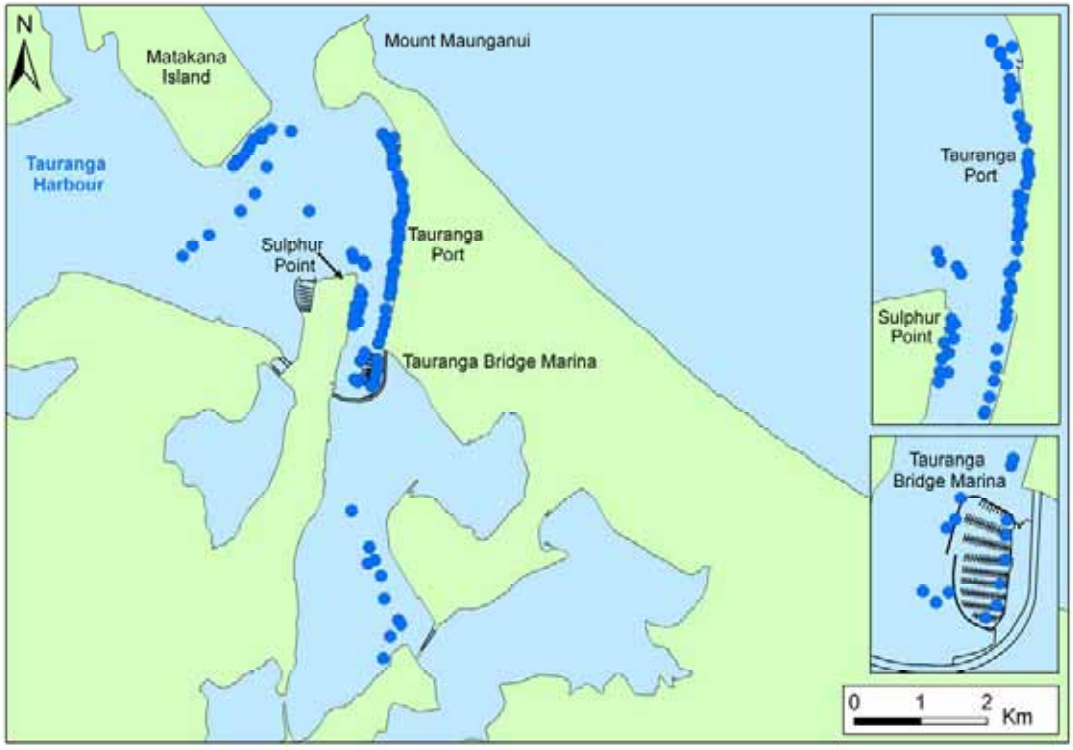

Figure 28 **Distribution of sled tows in Tauranga Harbour, May 2006.**

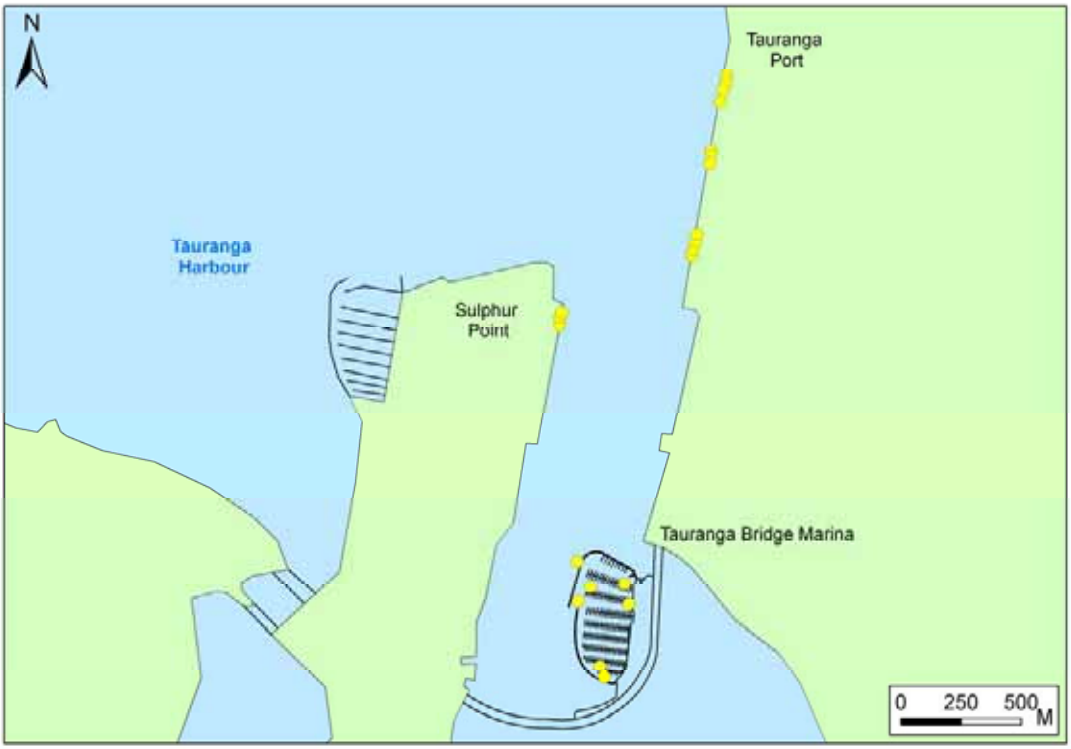

Figure 29 Distribution of dive searches in Tauranga Harbour, May 2006.

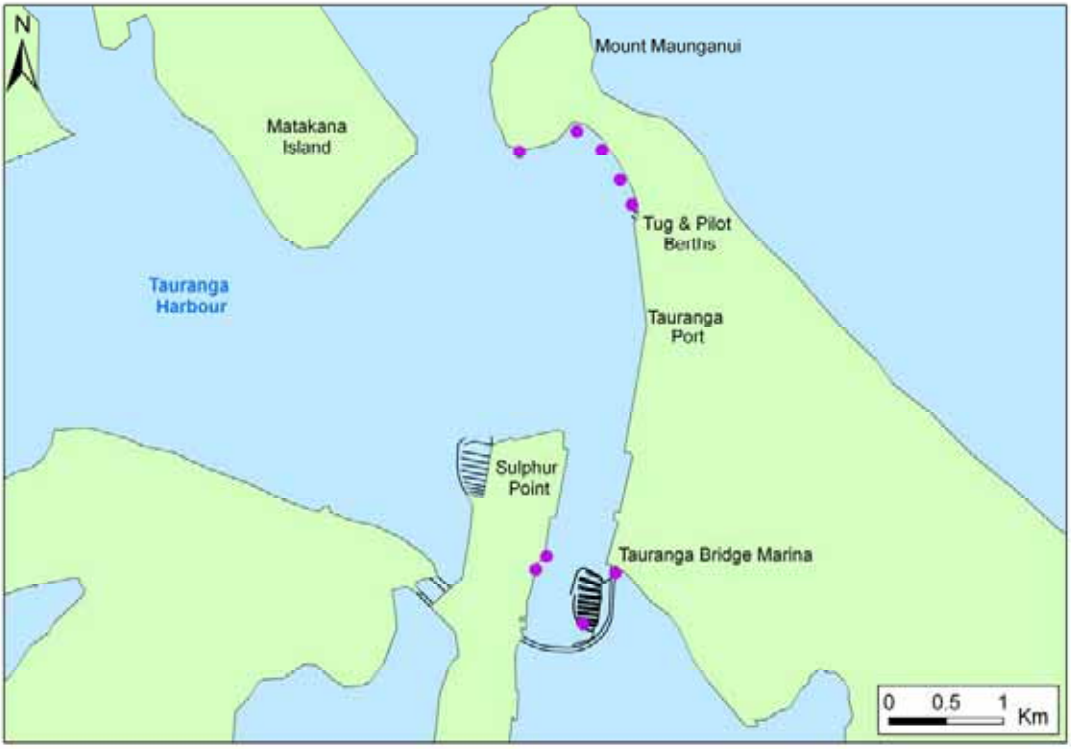

Figure 30 Distribution of shore searches in Tauranga Harbour, May 2006.

Figure 31 **Map of the sampling area around Wellington Harbour.**

Figure 32 **Detail of the sampling area around Wellington Port.**

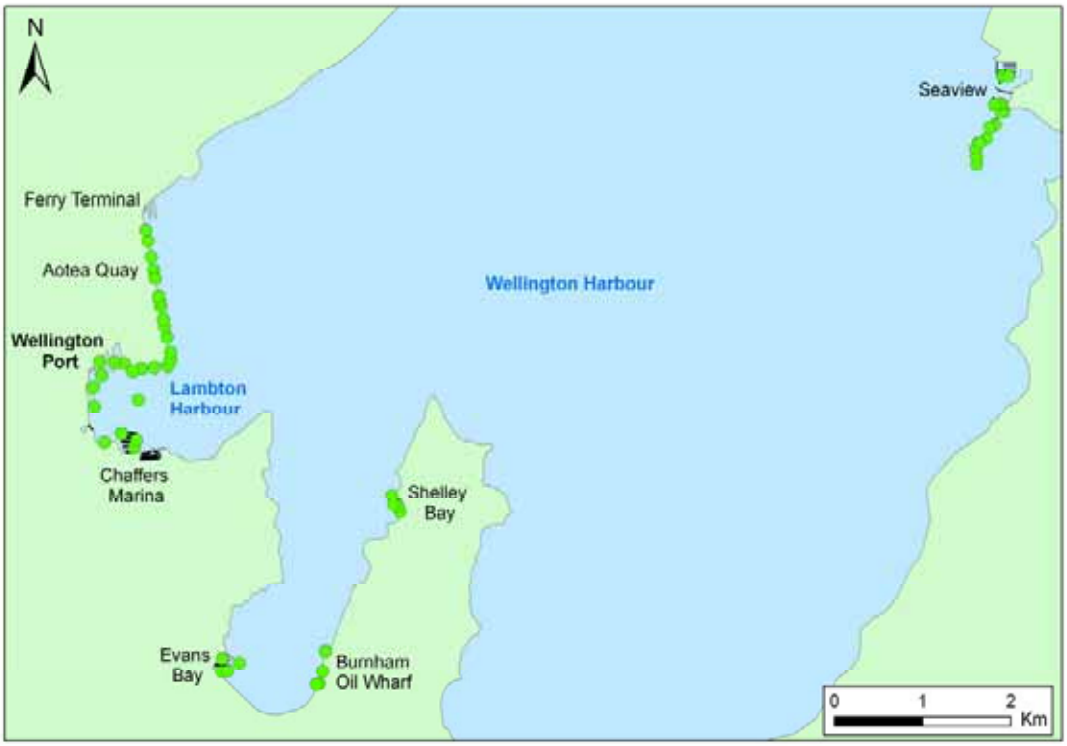

Figure 33 Distribution of crab traps in Wellington Harbour, February 2006.

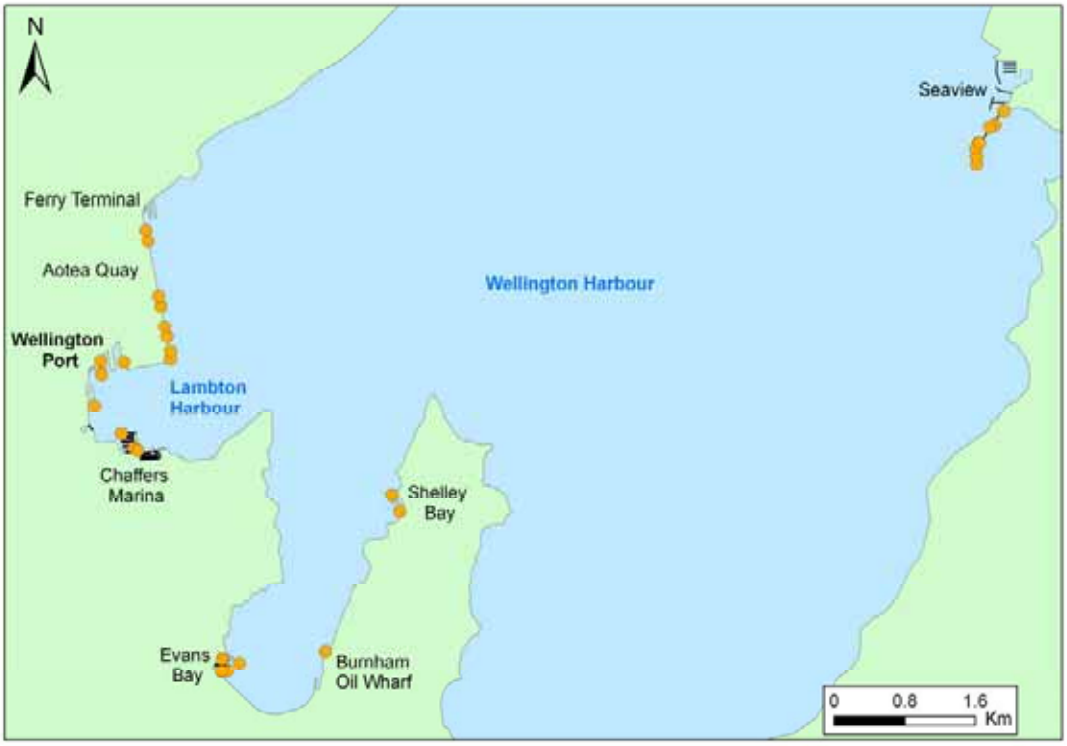

Figure 34 Distribution of starfish traps in Wellington Port, February 2006.

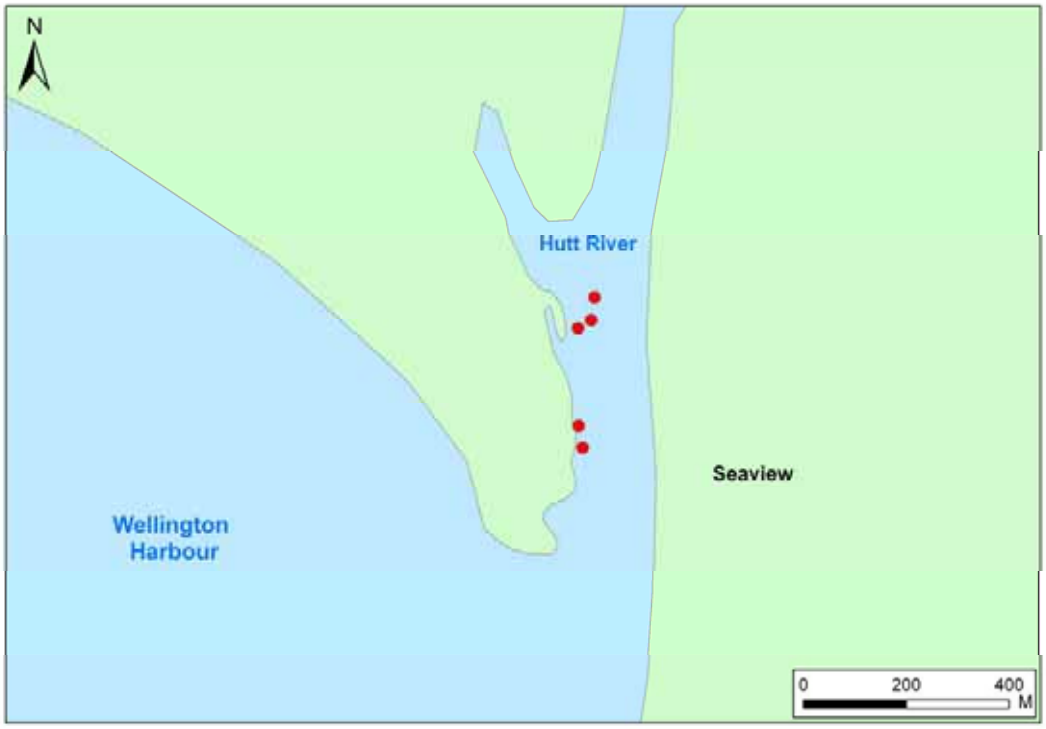

Figure 35 **Distribution of crab condos in the mouth of the Hutt River, northeast Wellington Harbour, February 2006.**

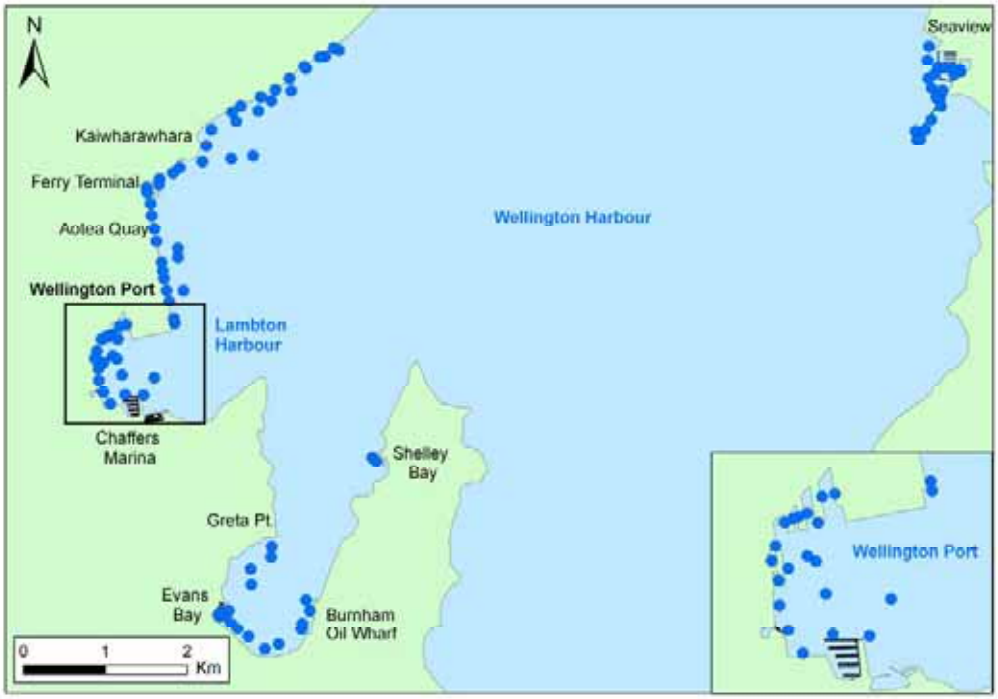

Figure 36 **Distribution of sled tows in Wellington Harbour, February 2006.**

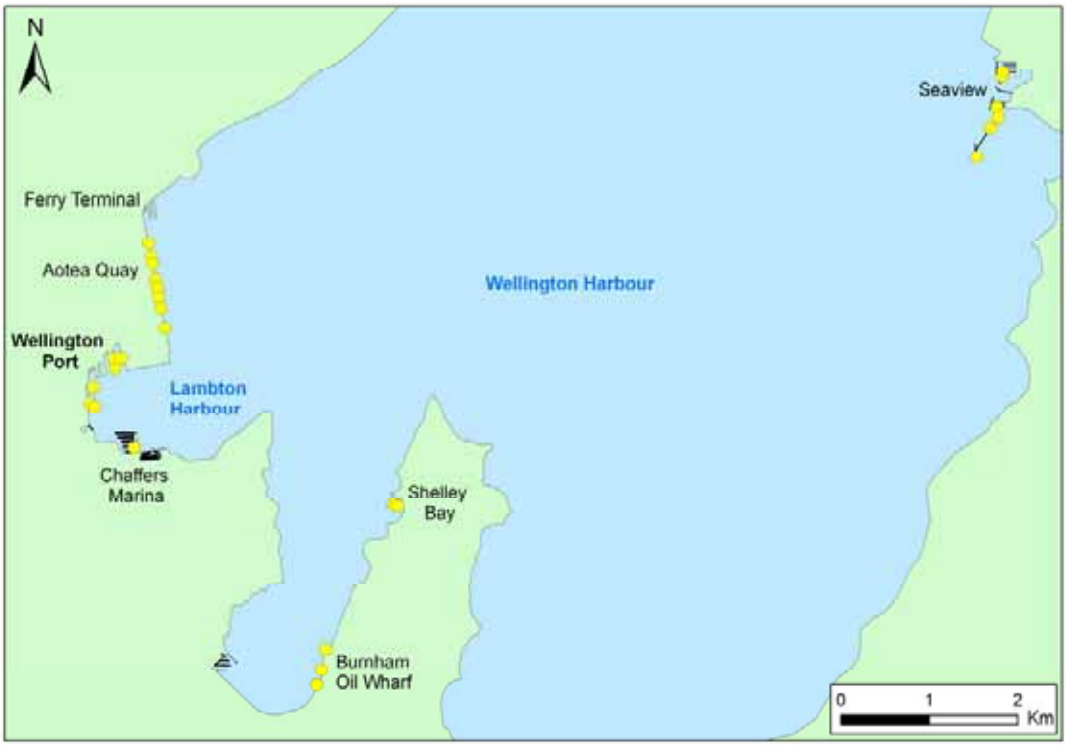

Figure 37 **Distribution of dive searches in Wellington Harbour, February 2006.**

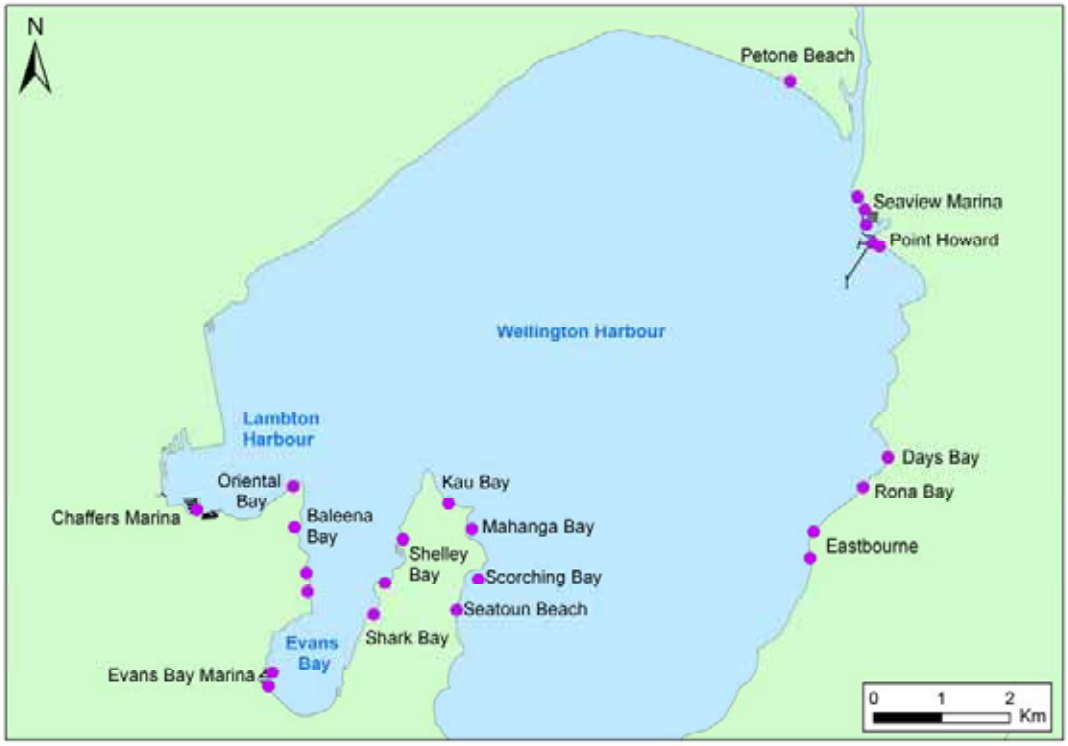

Figure 38 **Distribution of shore searches in Wellington Harbour, February 2006.**

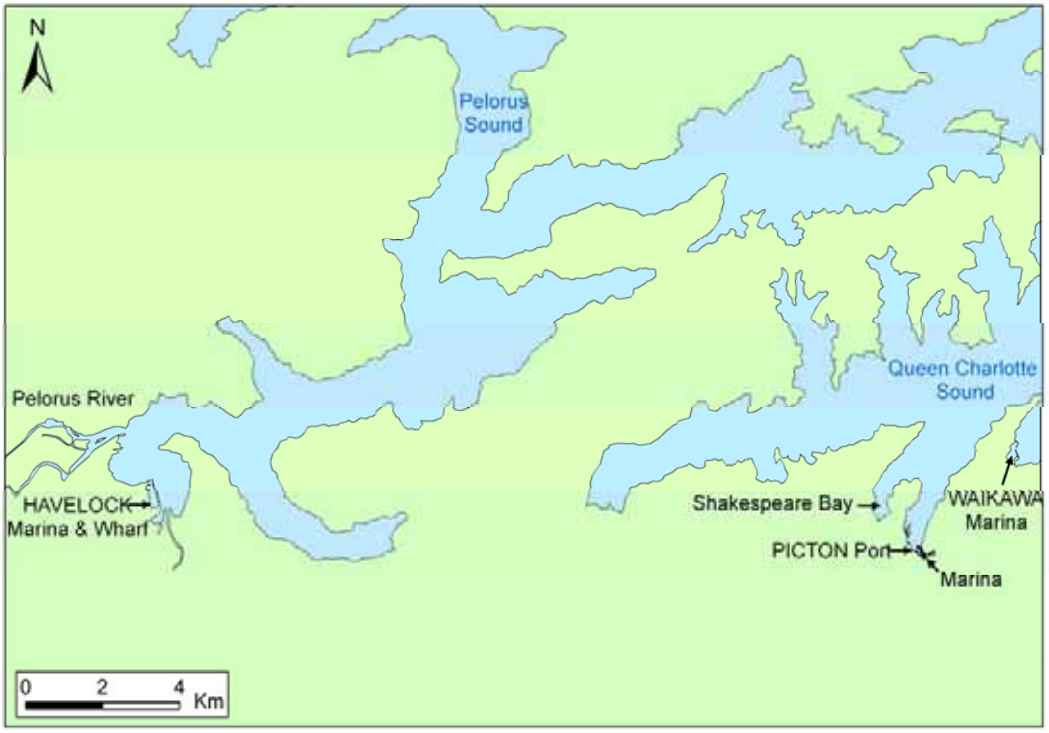

Figure 39 **Map of the sampling area around Picton, Shakespeare Bay, Waikawa Marina and Havelock.**

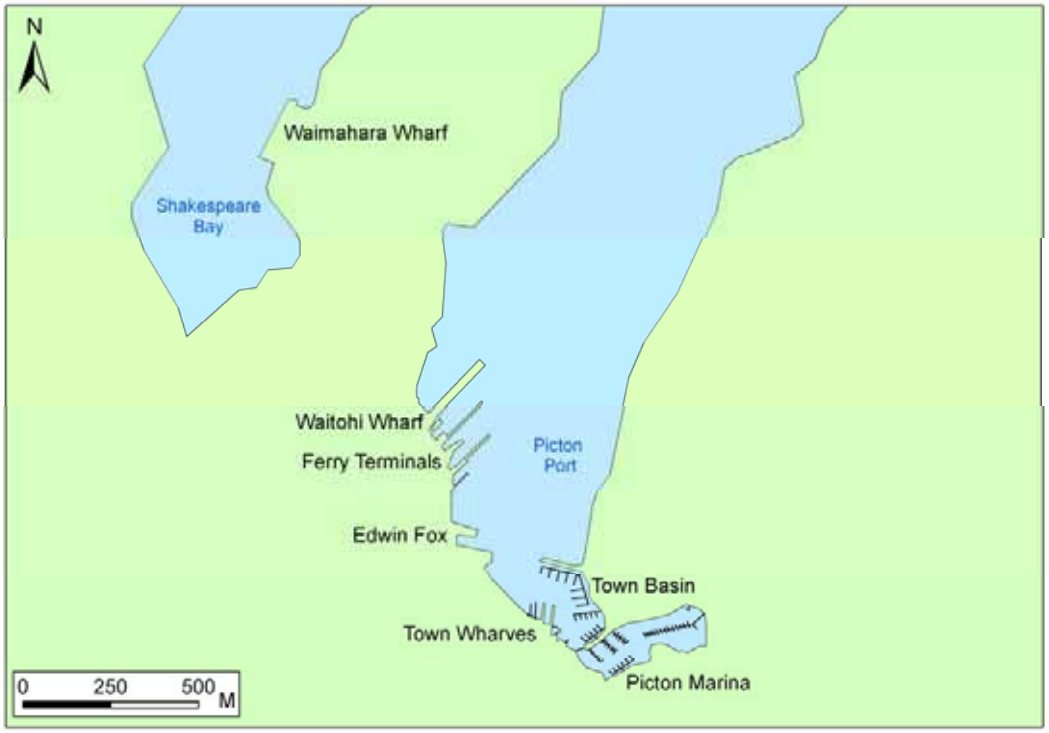

Figure 40 **Map of the sampling area in Picton Port, Picton Marina and Shakespeare Bay.**

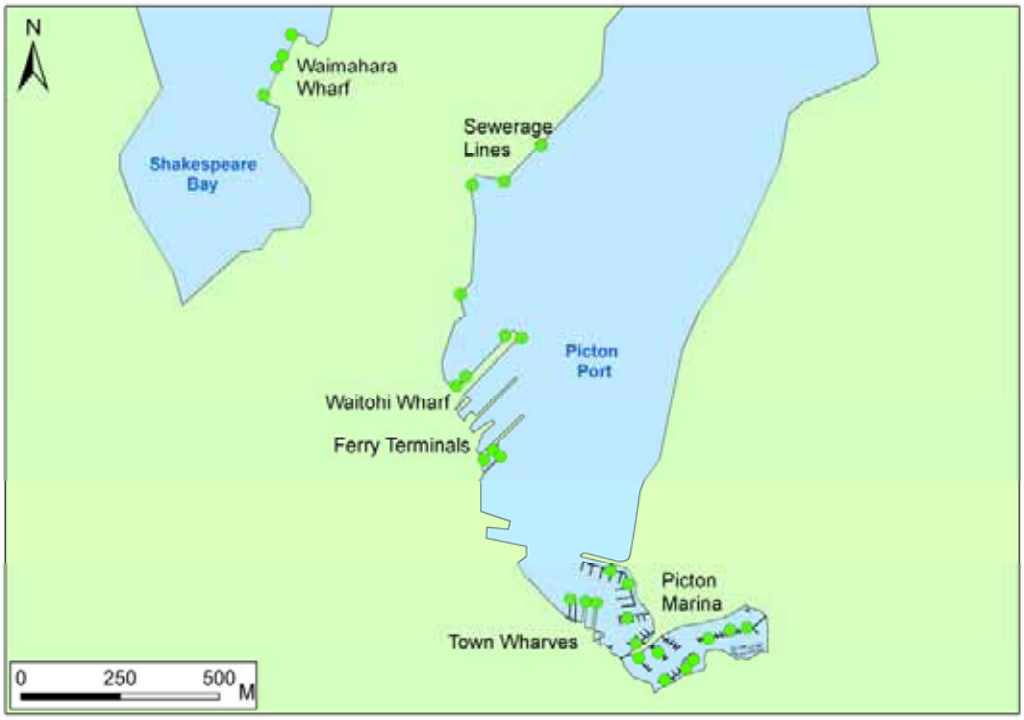

Figure 41 **Distribution of crab traps in Picton Port and Shakespeare Bay, December 2006.**

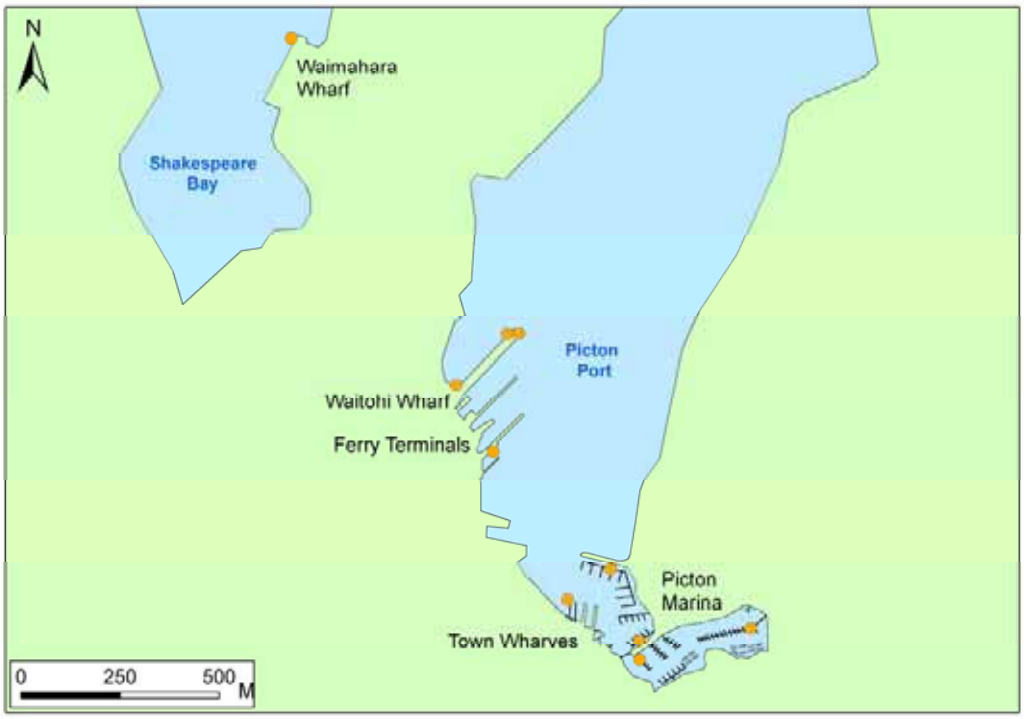

Figure 42 **Distribution of starfish traps in Picton Port and Shakespeare Bay, December 2006.**

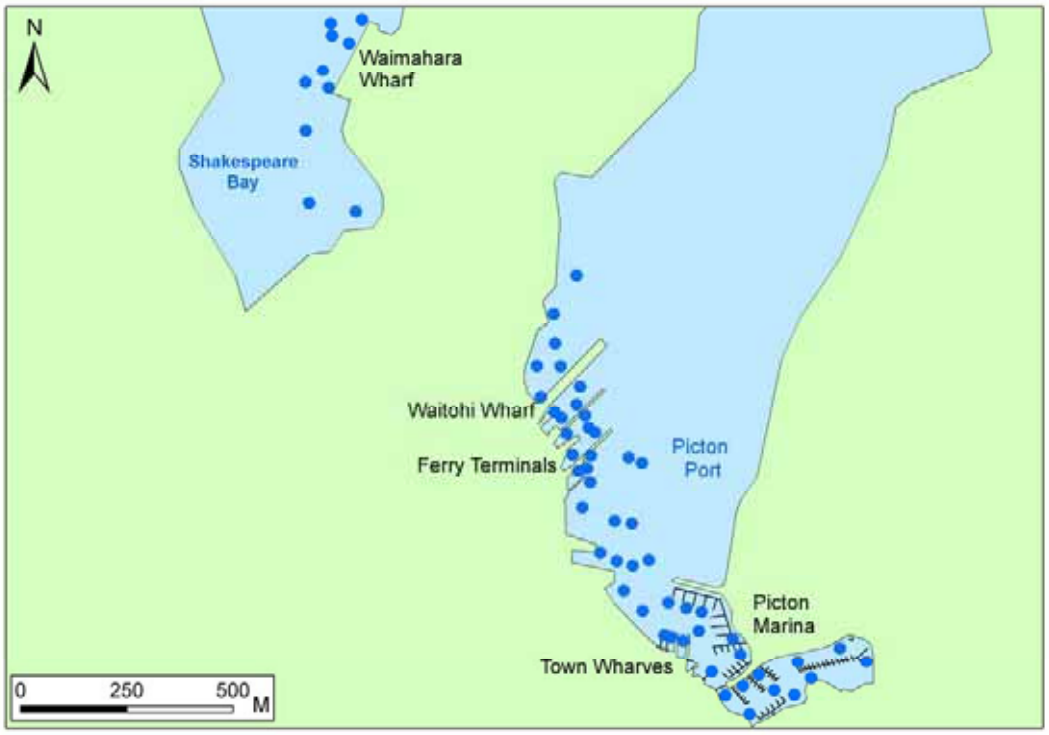

Figure 43 **Distribution of sled tows in Picton Port and Shakespeare Bay, December 2006.**

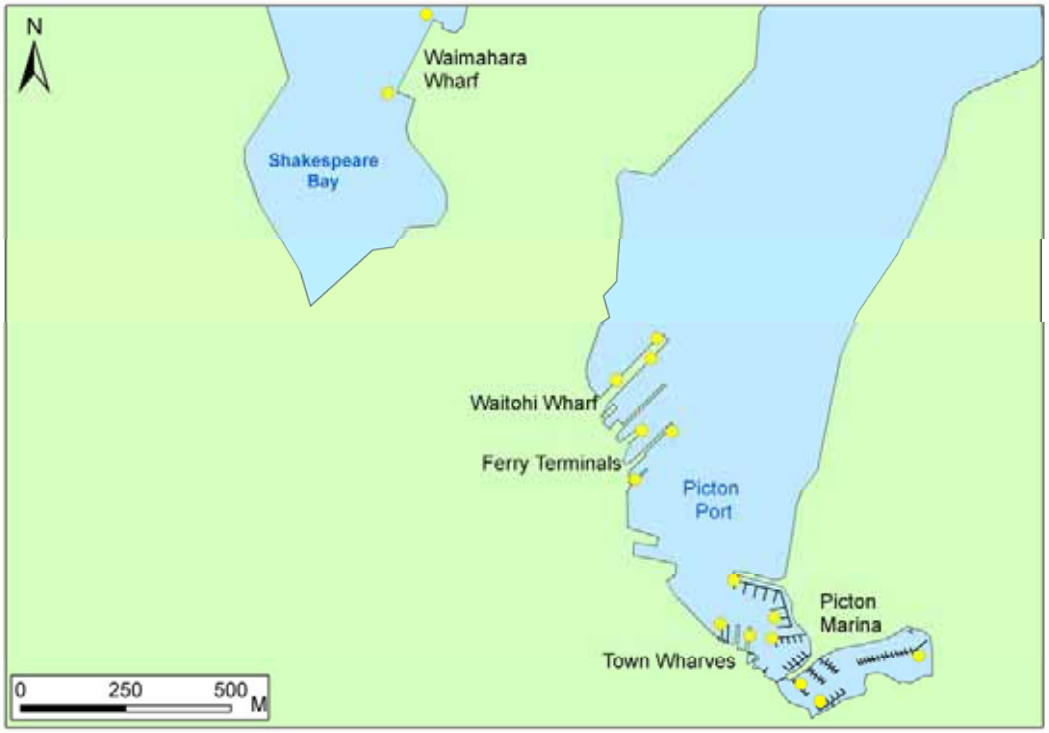

Figure 44 **Distribution of dive searches in Picton Port and Shakespeare Bay, November 2006.**

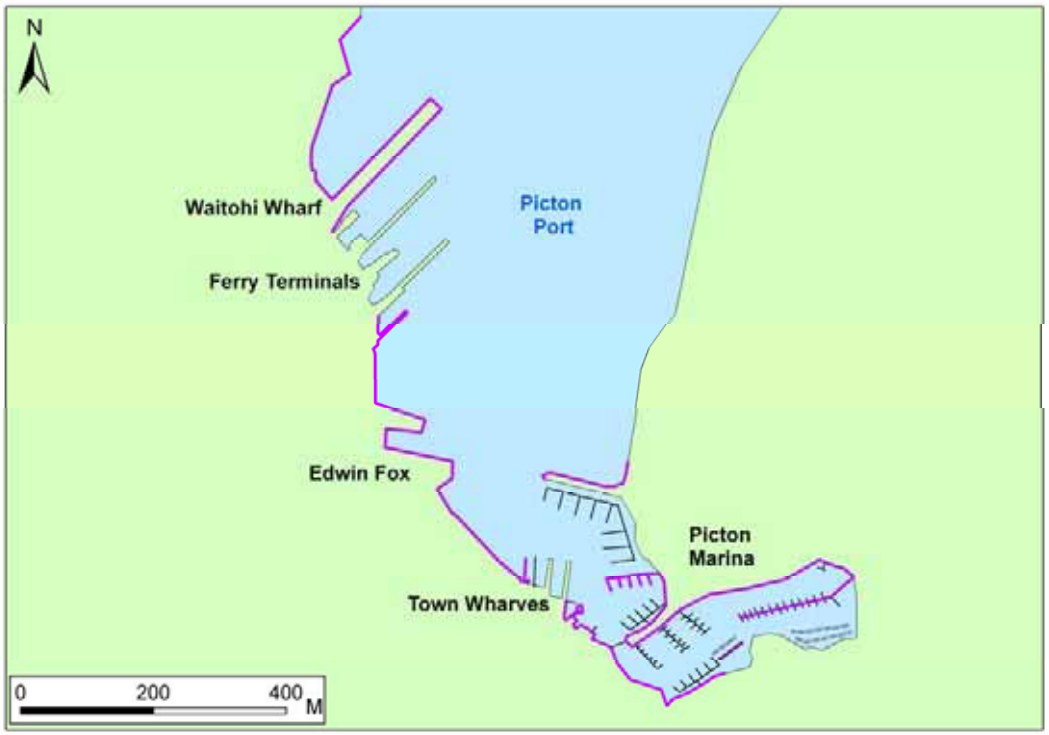

Figure 45 **Distribution of shore searches (purple lines) in Picton Port, December 2006.**

Figure 46 **Distribution of crab traps in Waikawa Marina, December 2006.**

Figure 47 **Distribution of starfish traps in Waikawa Marina, December 2006.**

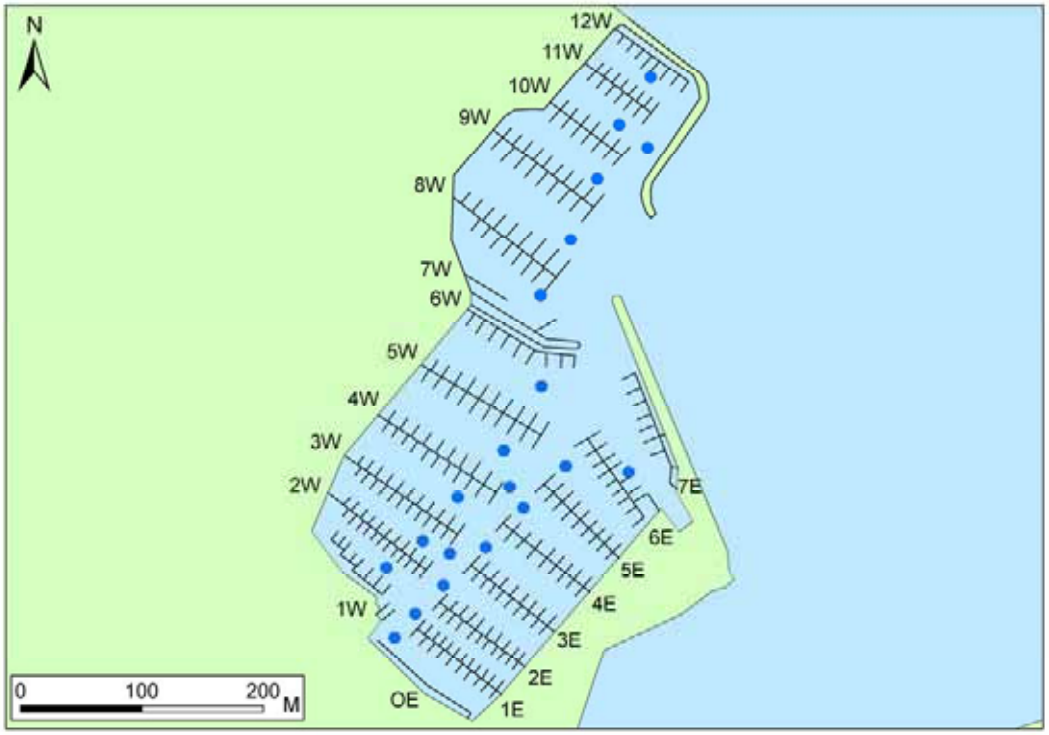

Figure 48 **Distribution of sled tows in Waikawa Marina, December 2006.**

Figure 49 **Distribution of dive searches in Waikawa Marina, November 2006.**

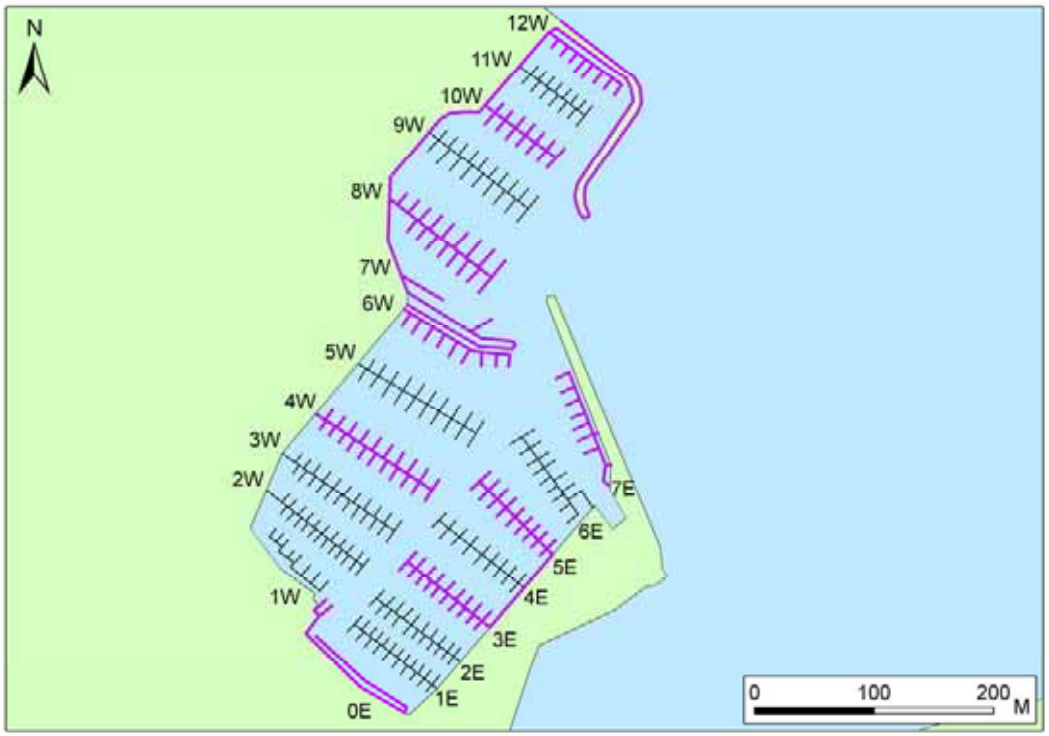

Figure 50 **Distribution of shore searches in Waikawa Marina, December 2006.**

Figure 51 **Distribution of crab traps in Havelock Port and Marina, December 2006.**

Figure 52 **Distribution of starfish traps in Havelock Port and Marina, December 2006.**

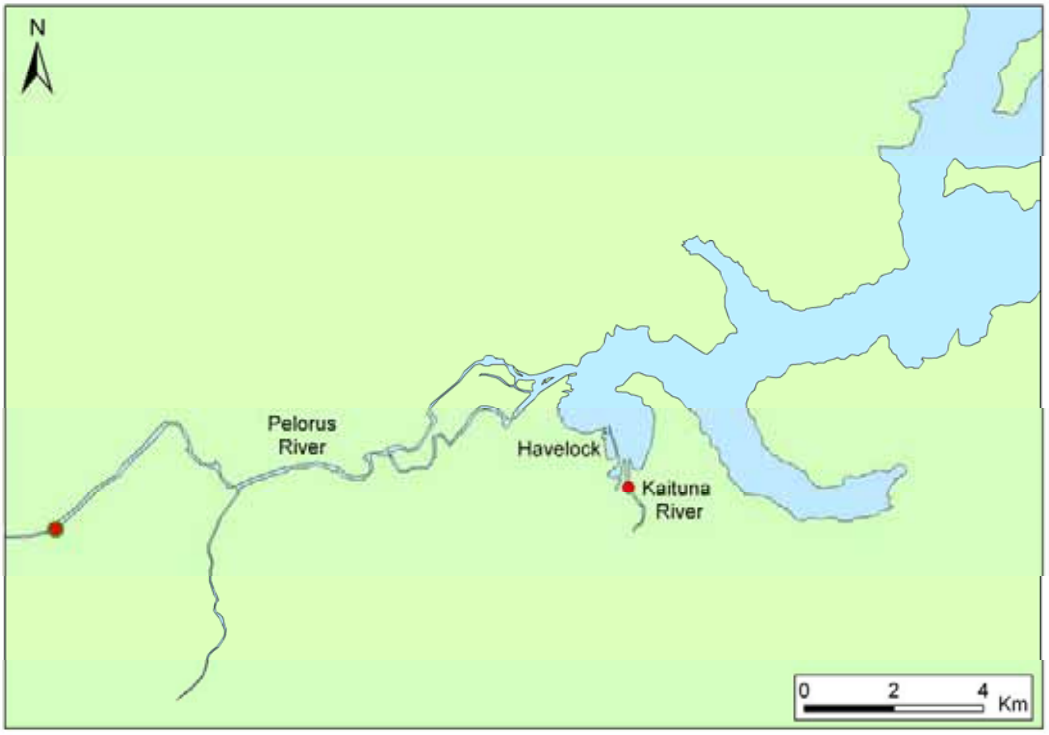

Figure 53 **Distribution of crab condos around Havelock, December 2006.**

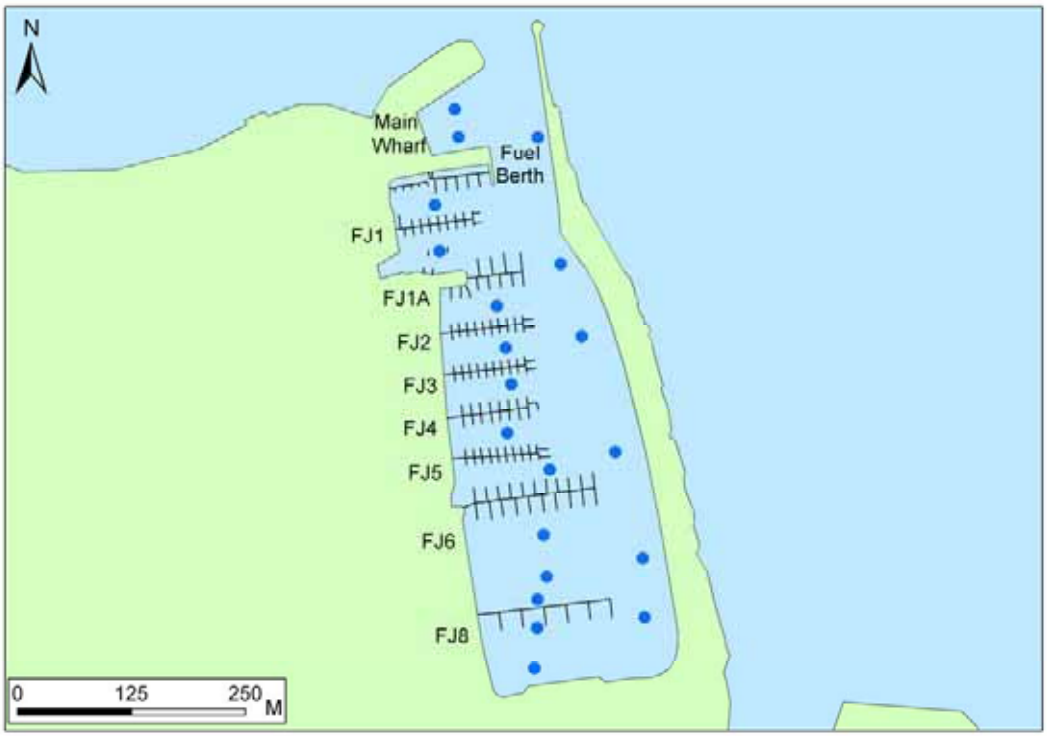

Figure 54 **Distribution of sled tows in Havelock Port and Marina, December 2006.**

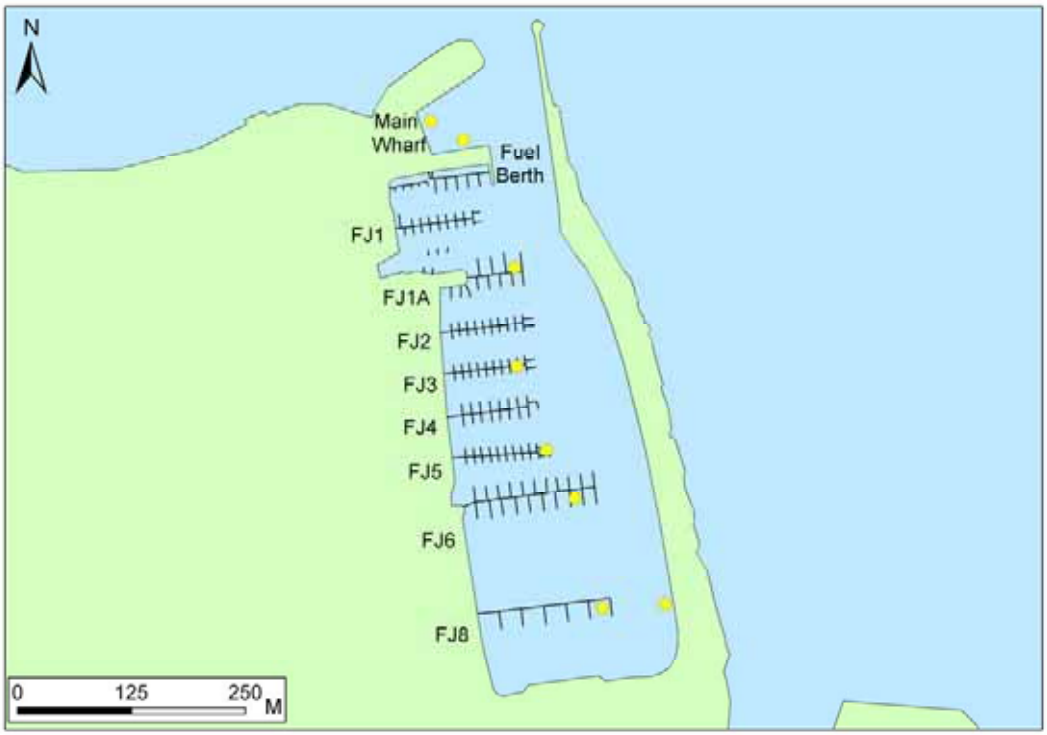

Figure 55 **Distribution of dive searches in Havelock Port and Marina, November 2006.**

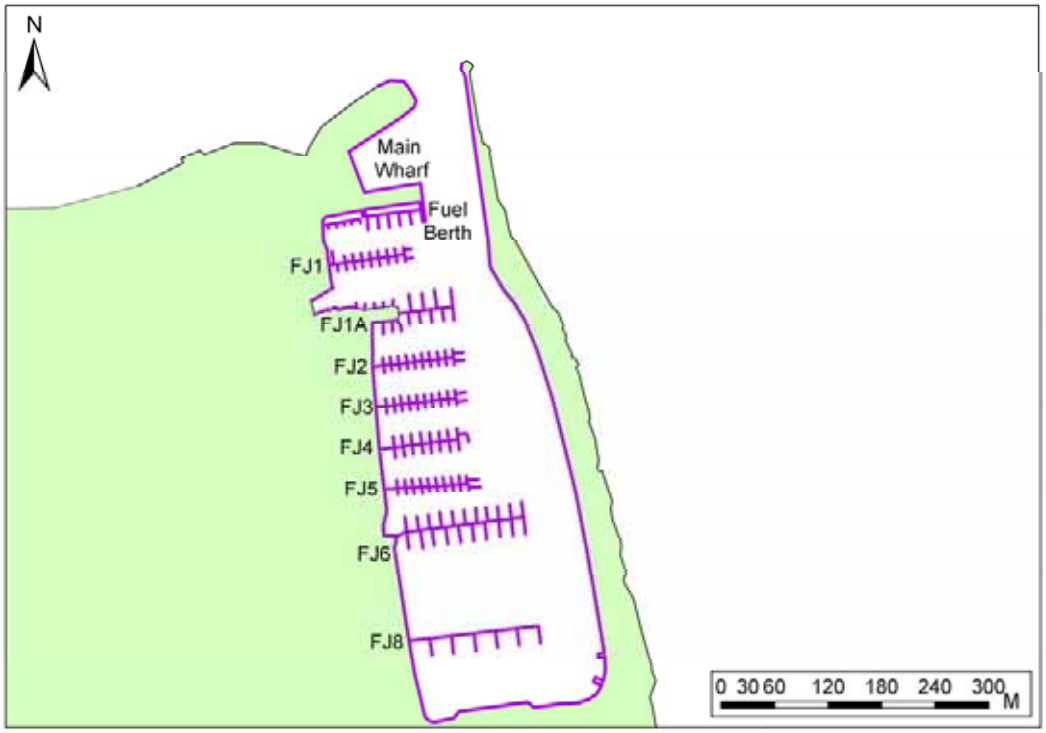

Figure 56 **Distribution of shore searches in Havelock Port and Marina, December 2006.**

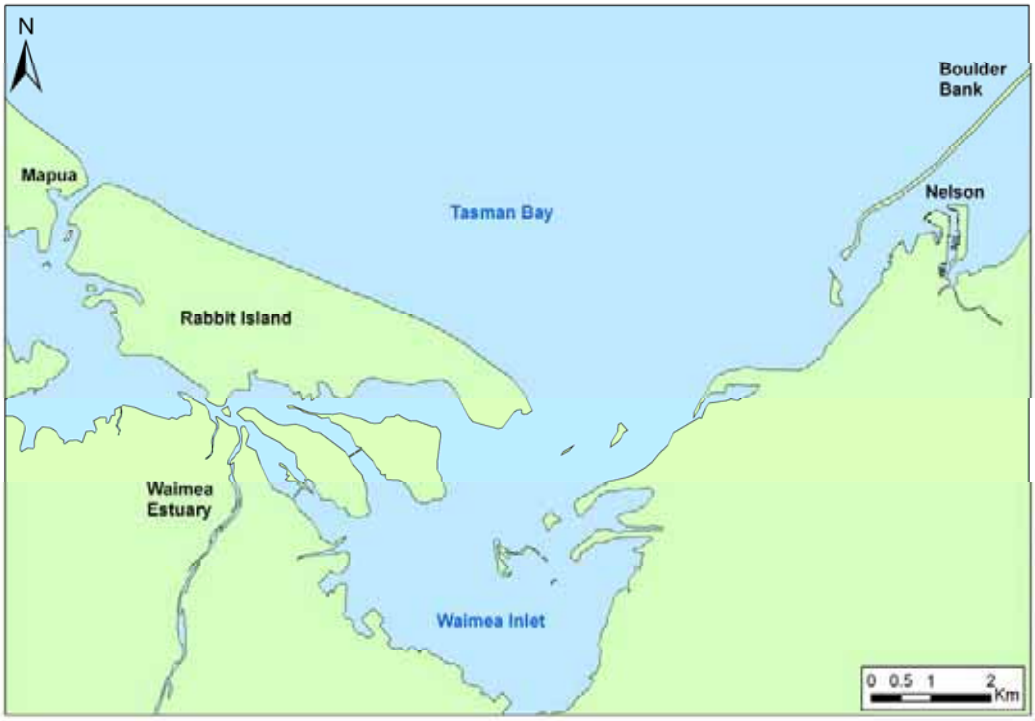

Figure 57 **Map of the sampling area around Nelson Port.**

Figure 58 **Detail of the sampling area around Nelson Port.**

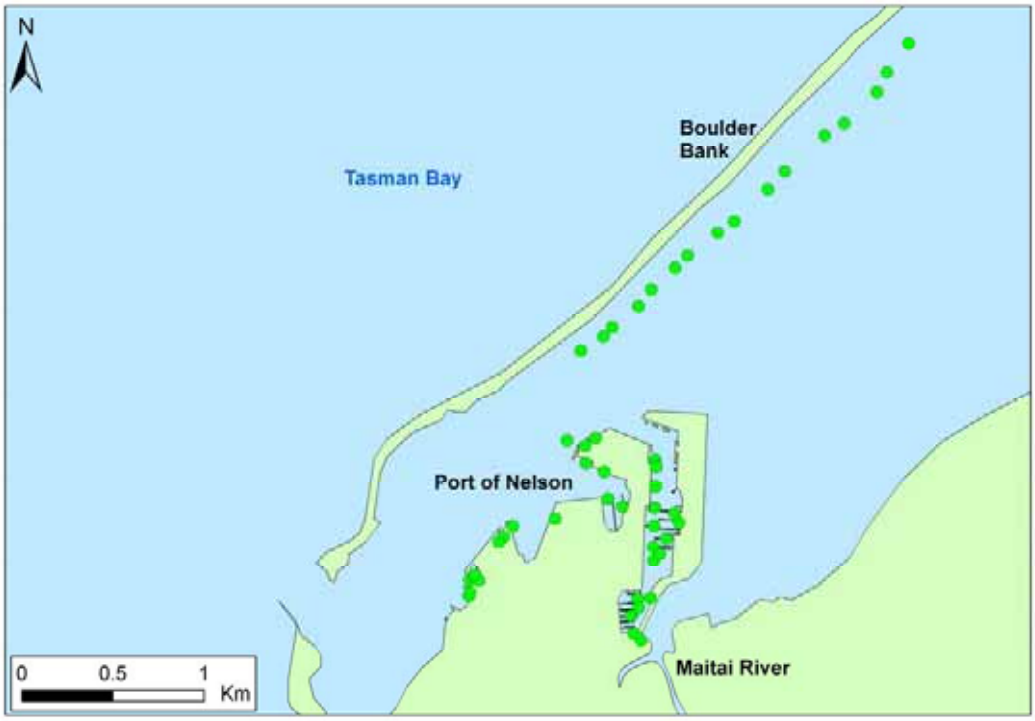

Figure 59 **Distribution of crab traps in Nelson Port and Haven, December 2006.**

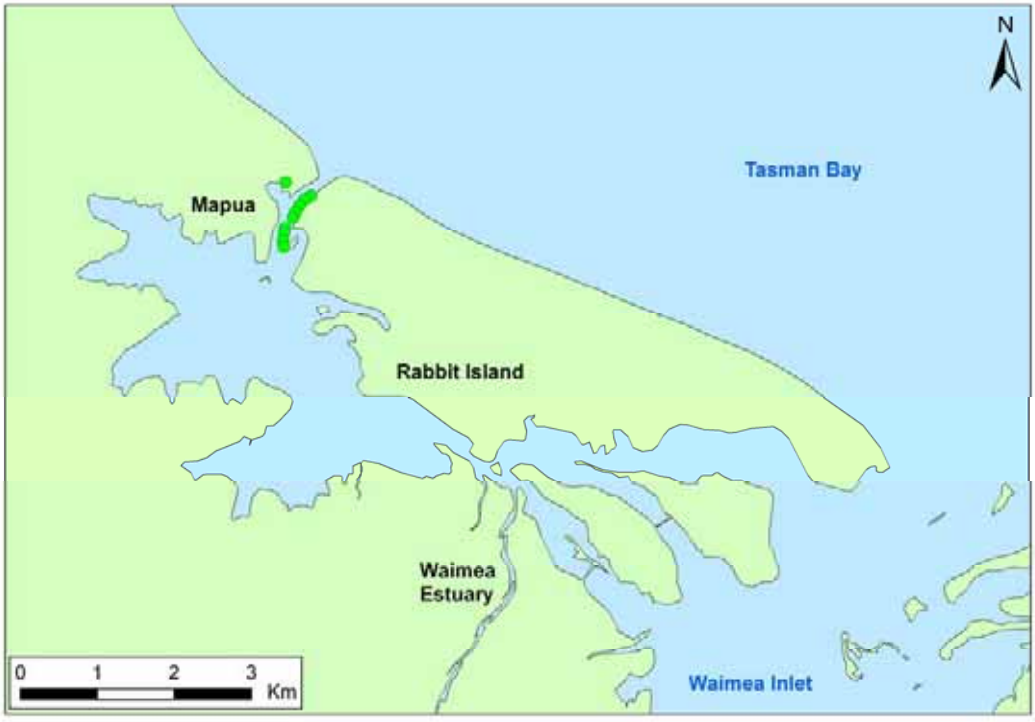

Figure 60 **Distribution of crab traps at the entrance to Waimea Inlet, December 2006.**

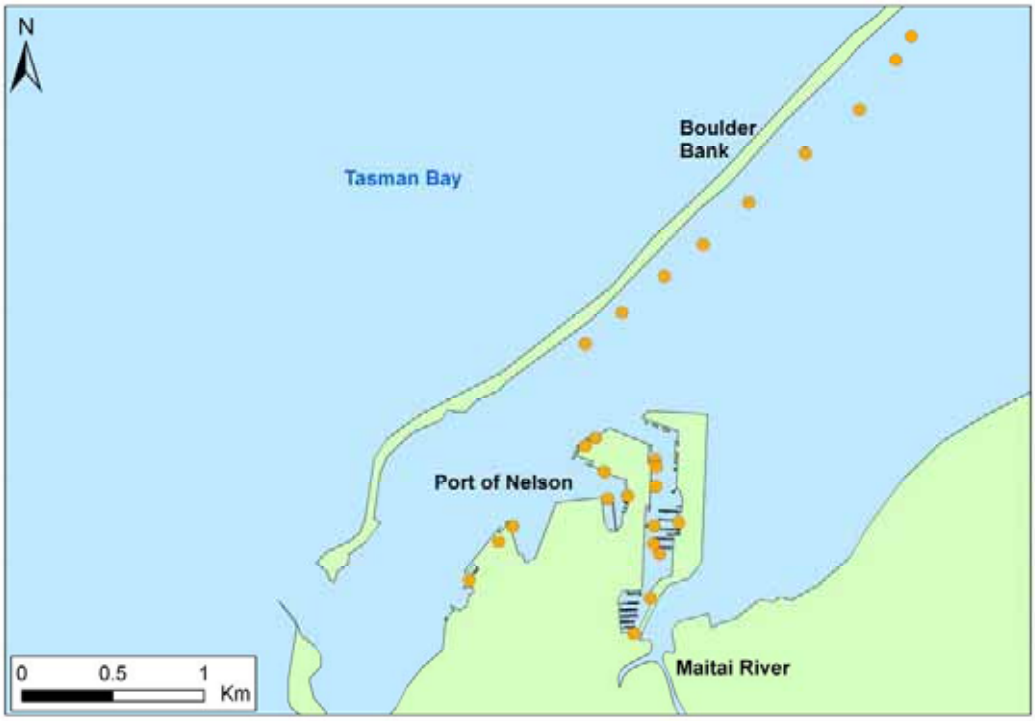

Figure 61 **Distribution of starfish traps in Nelson Port and Haven, December 2006.**

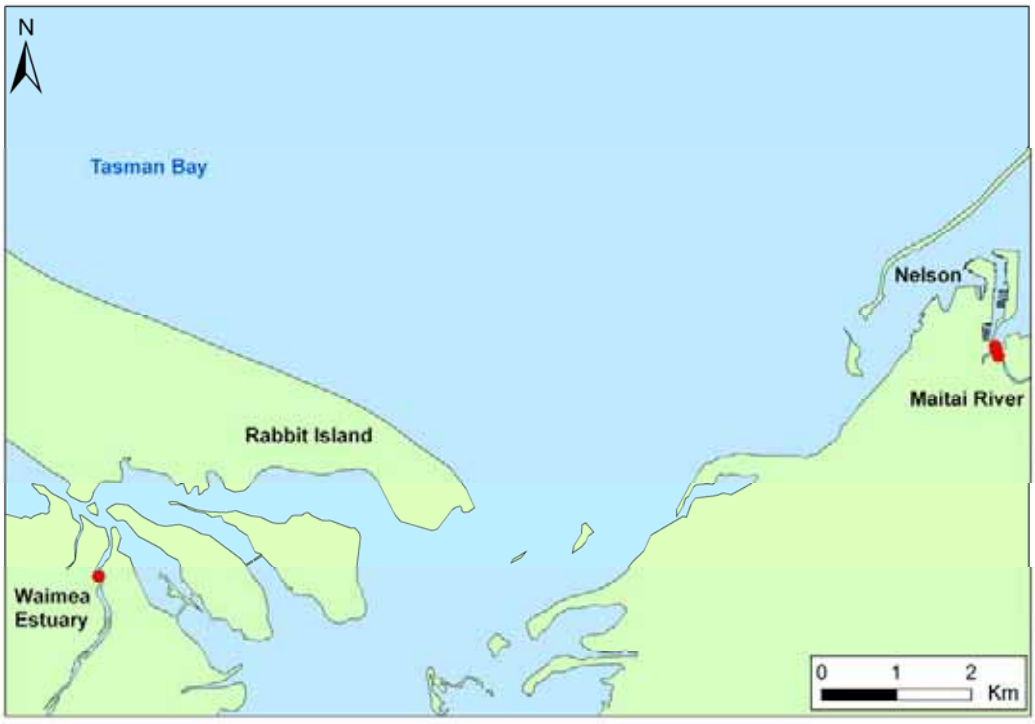

Figure 62 **Distribution of crab condos in the Maitai and Waimea Rivers, Nelson, December 2006.**

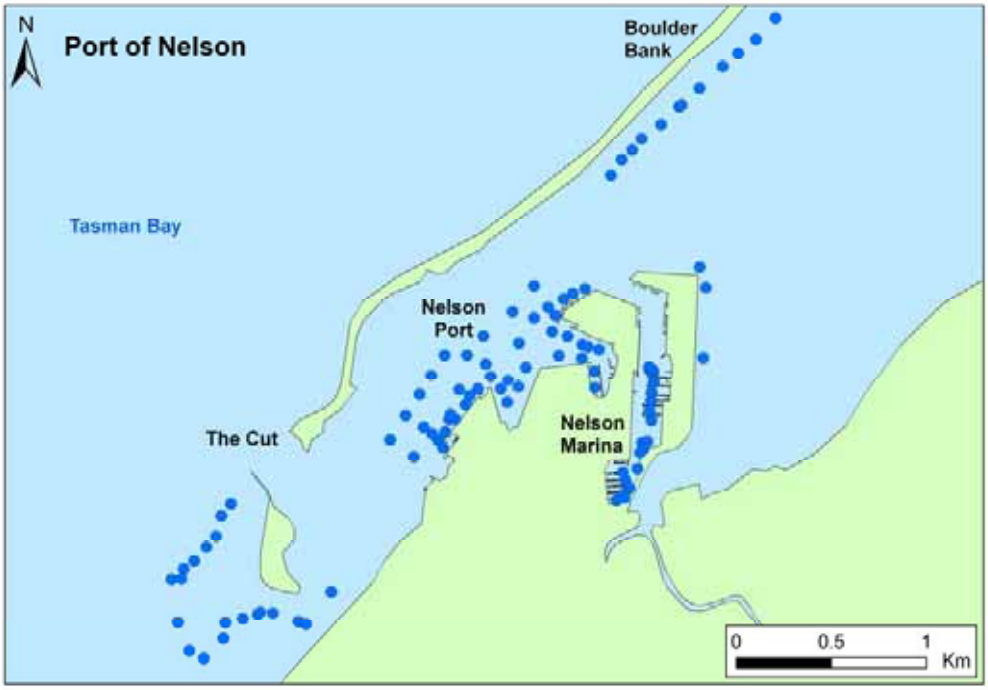

Figure 63 **Distribution of sled tows in the Nelson area, December 2006.**

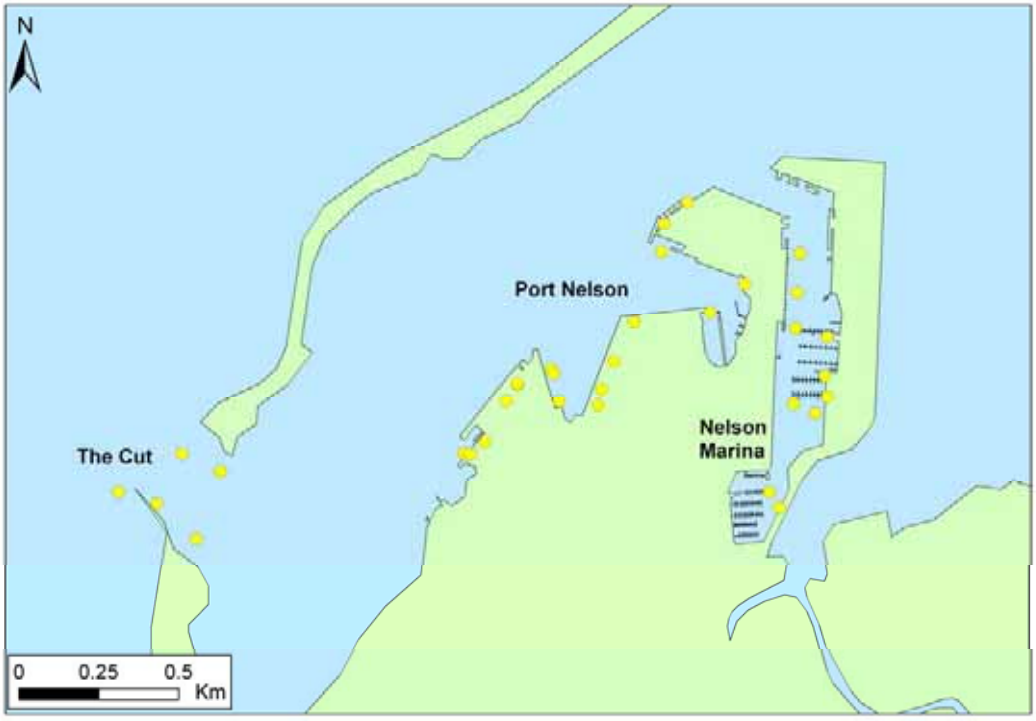

Figure 64 **Distribution of dive searches in the Nelson area, December 2006.**

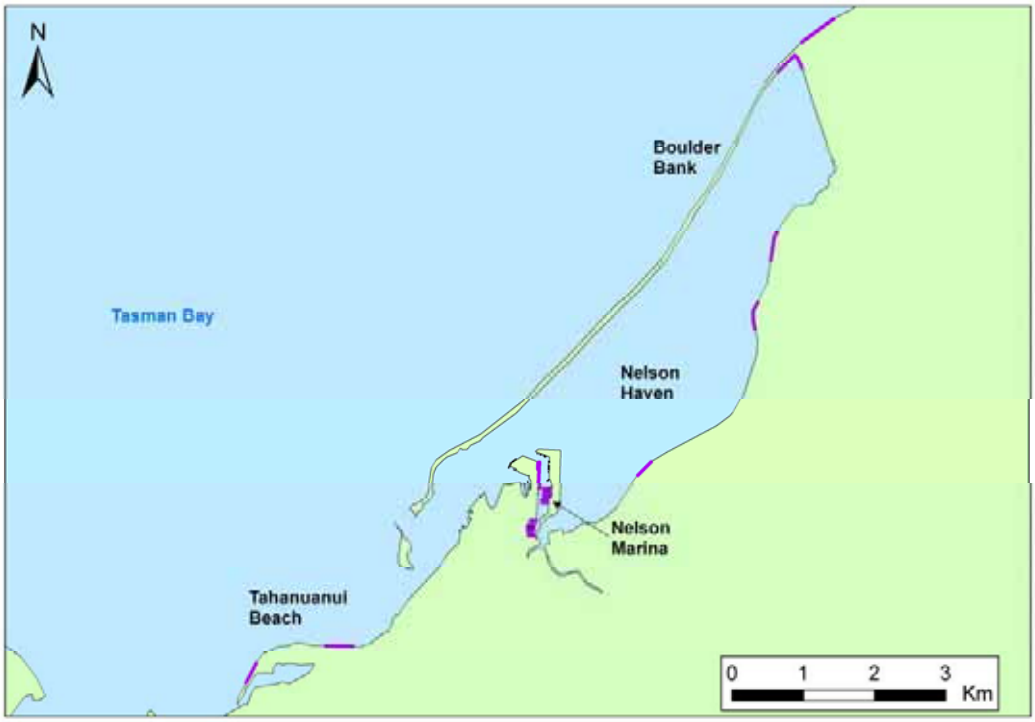

Figure 65 Distribution of shore searches in Nelson Port and Haven, December 2006.

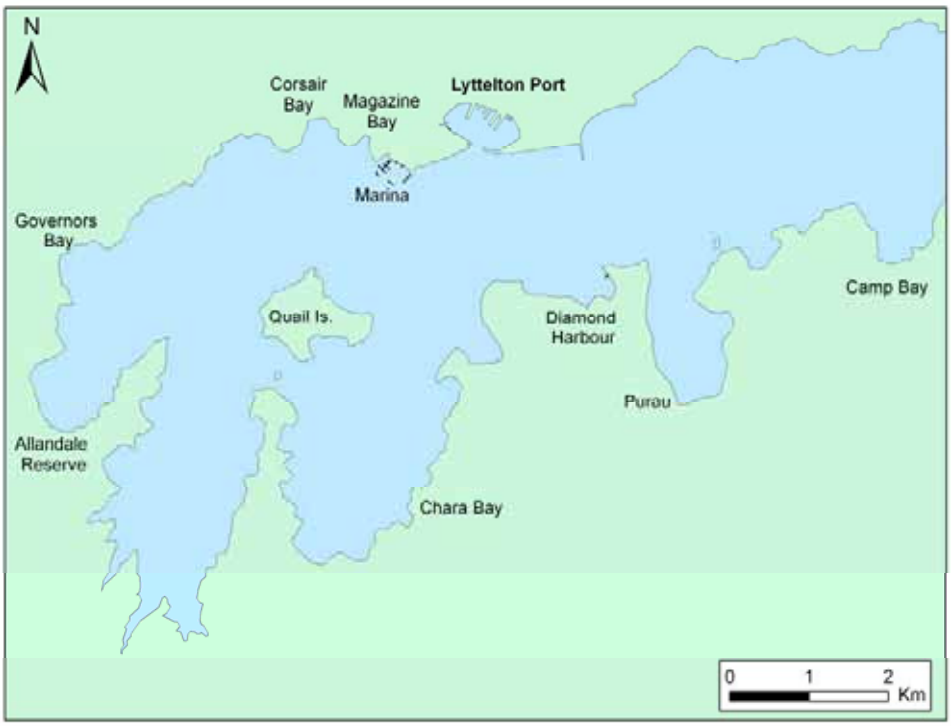

Figure 66 **Map of the sampling area around Lyttelton Harbour.**

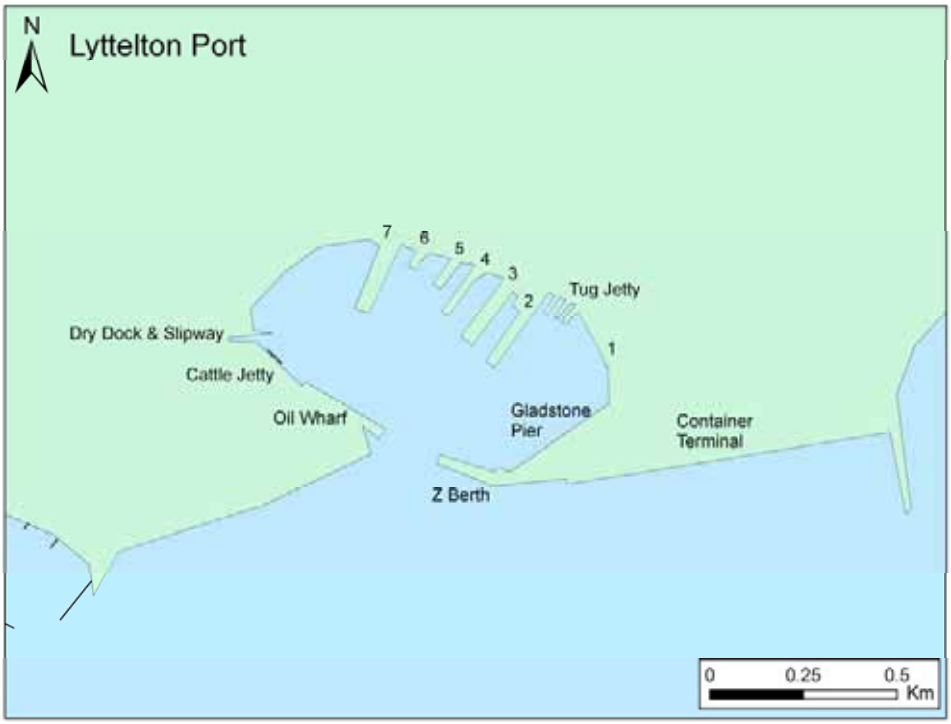

Figure 67 **Detail of the sampling area around Lyttelton Port. Numbers identify wharves.**

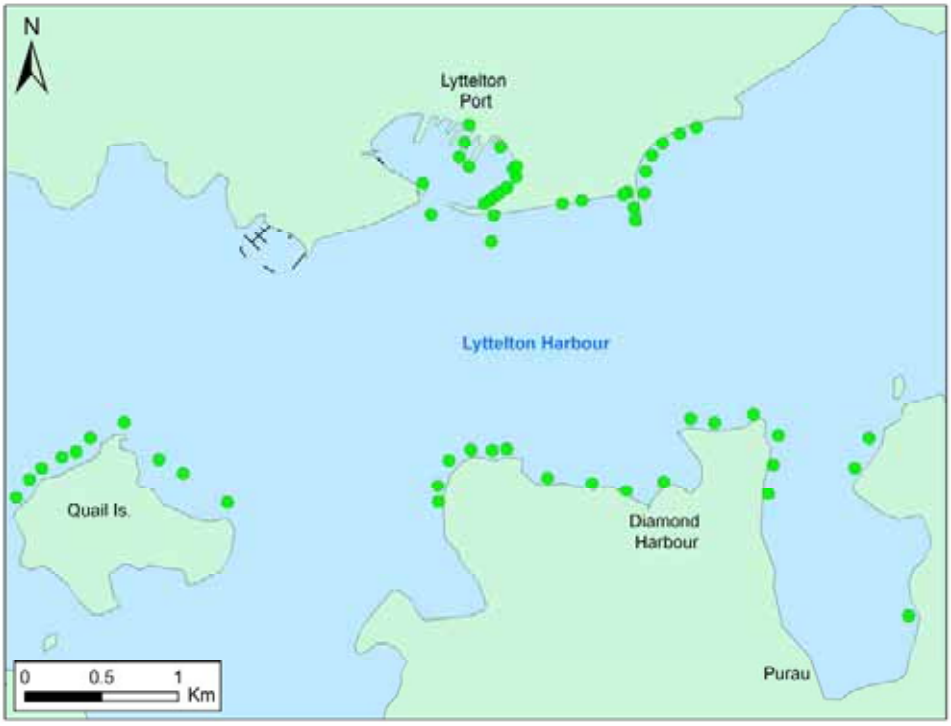

Figure 68 **Distribution of crab traps in Lyttelton Harbour, January 2006.**

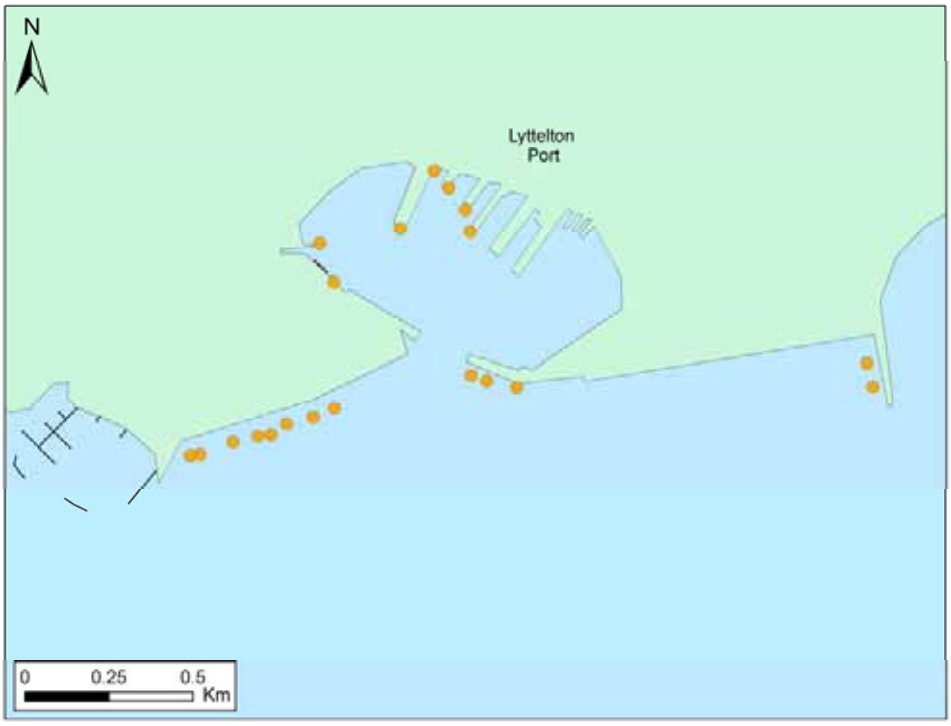

Figure 69 **Distribution of starfish traps in Lyttelton Port, January 2006.**

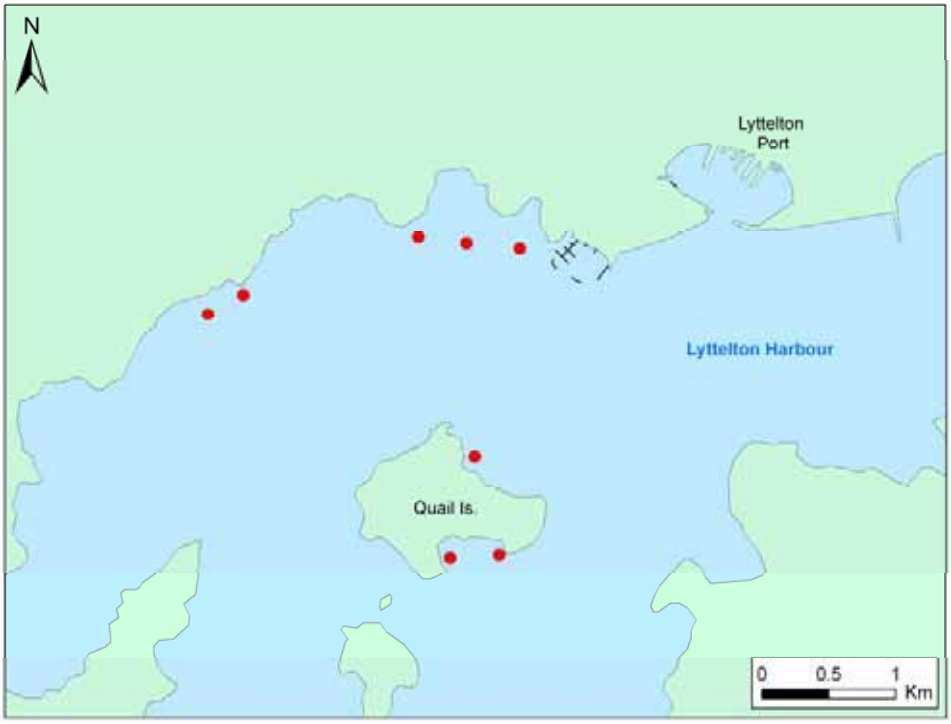

Figure 70 **Distribution of crab condos in Lyttelton Harbour, January 2006.**

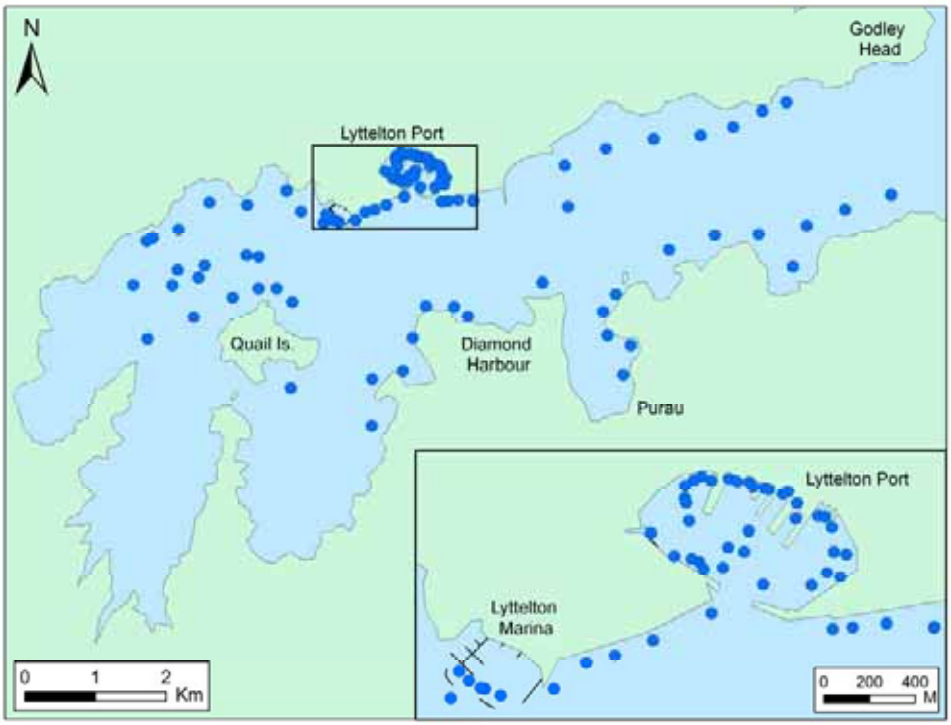

Figure 71 **Distribution of sled tows in Lyttelton Harbour, January 2006.**

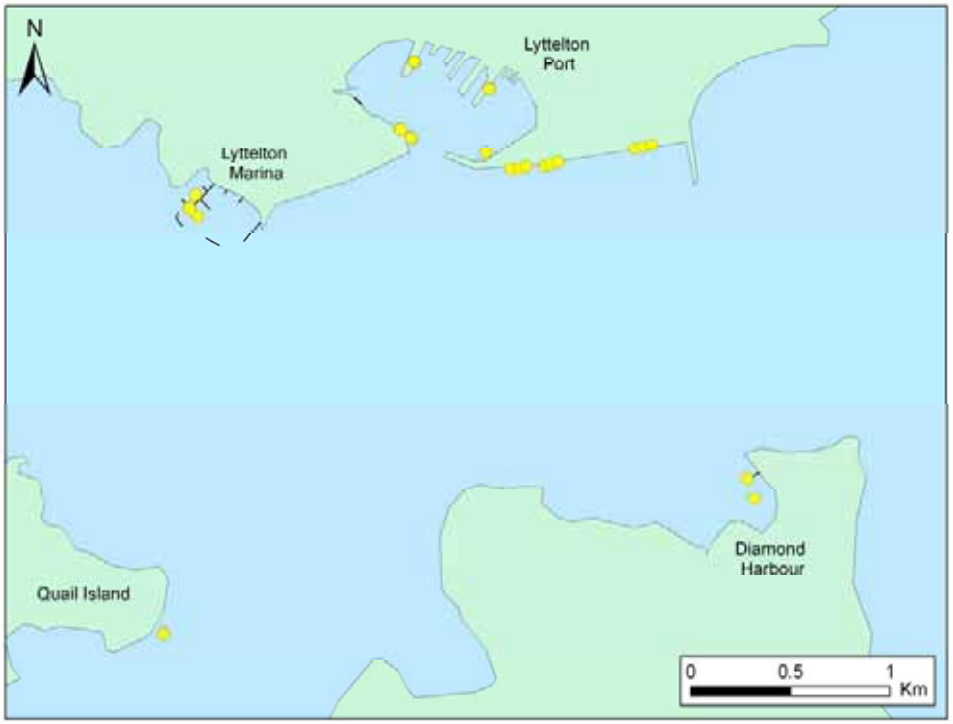

Figure 72 **Distribution of dive searches in Lyttelton Harbour, March 2006.**

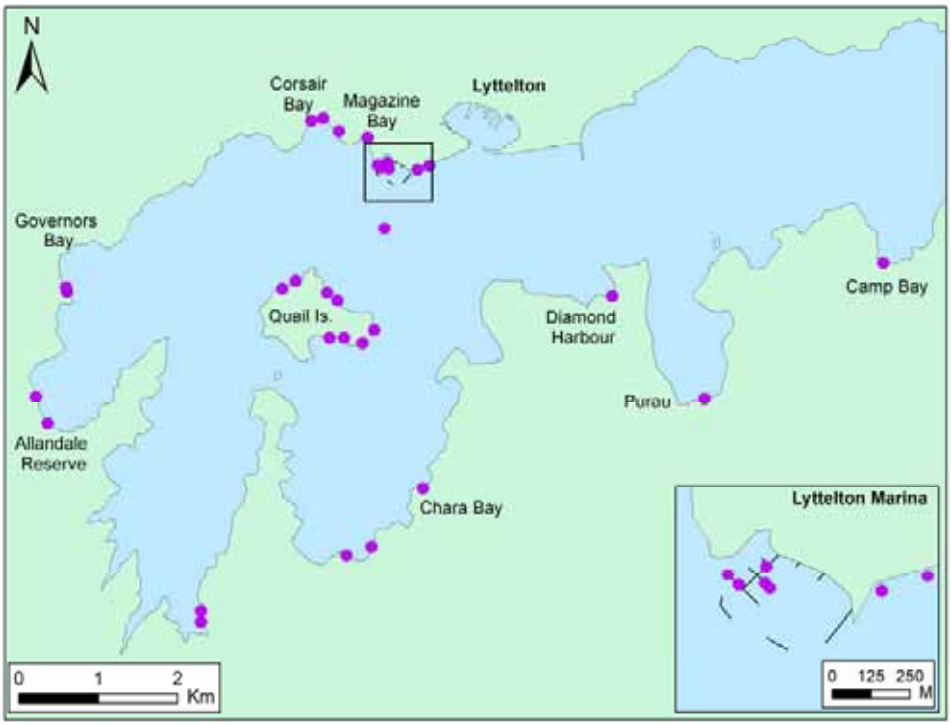

Figure 73 **Distribution of shore searches in Lyttelton Harbour, January 2006.**

Figure 74 **Map of the sampling area around Otago Harbour.**

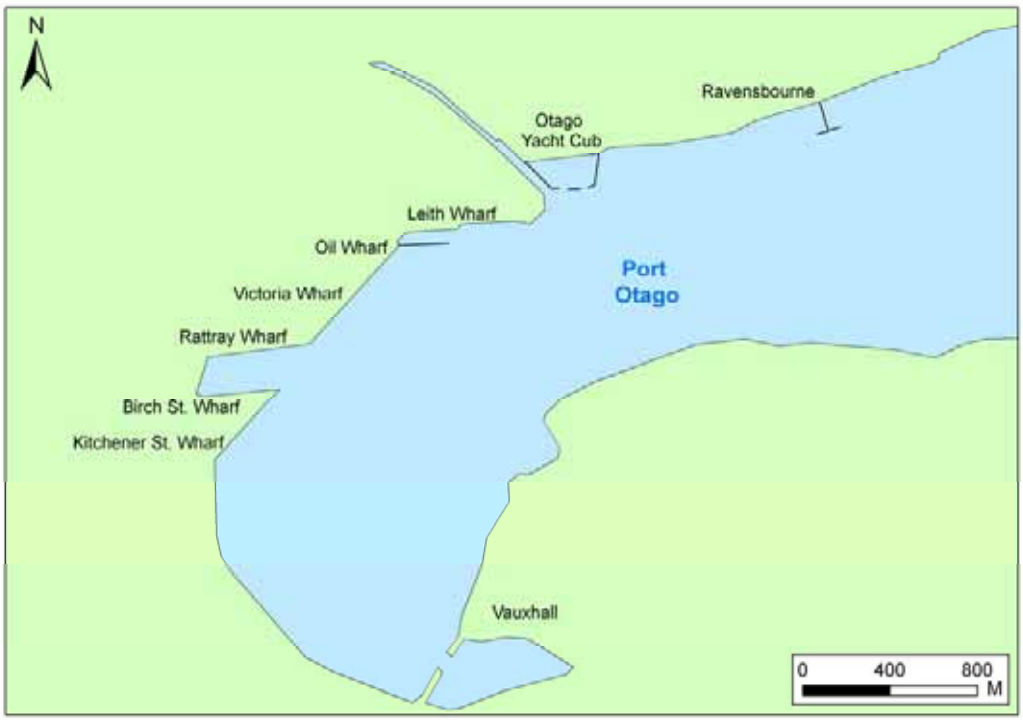

Figure 75 **Detail of the sampling area around Port Otago.**

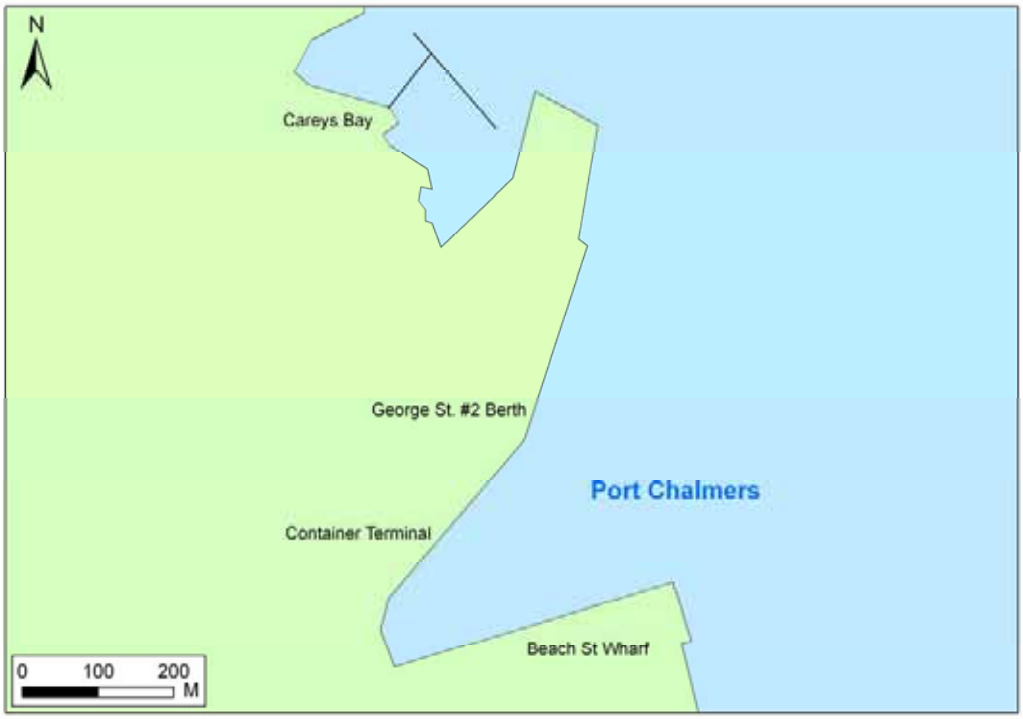

Figure 76 **Detail of the sampling area around Port Chalmers.**

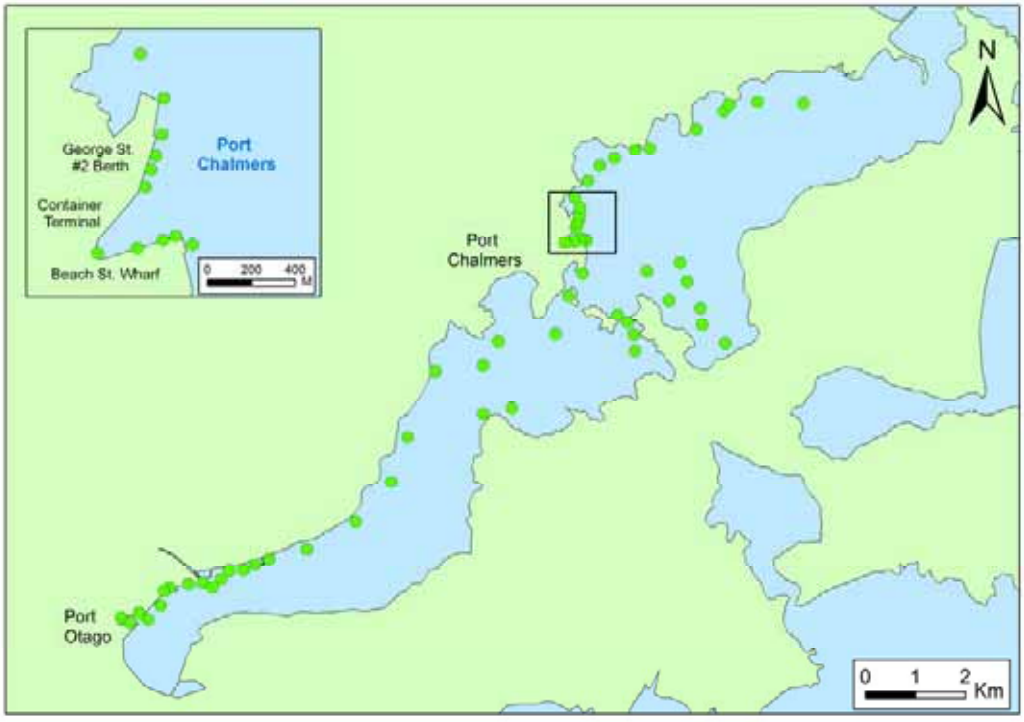

Figure 77 **Distribution of crab traps in Otago Harbour, March 2006.**

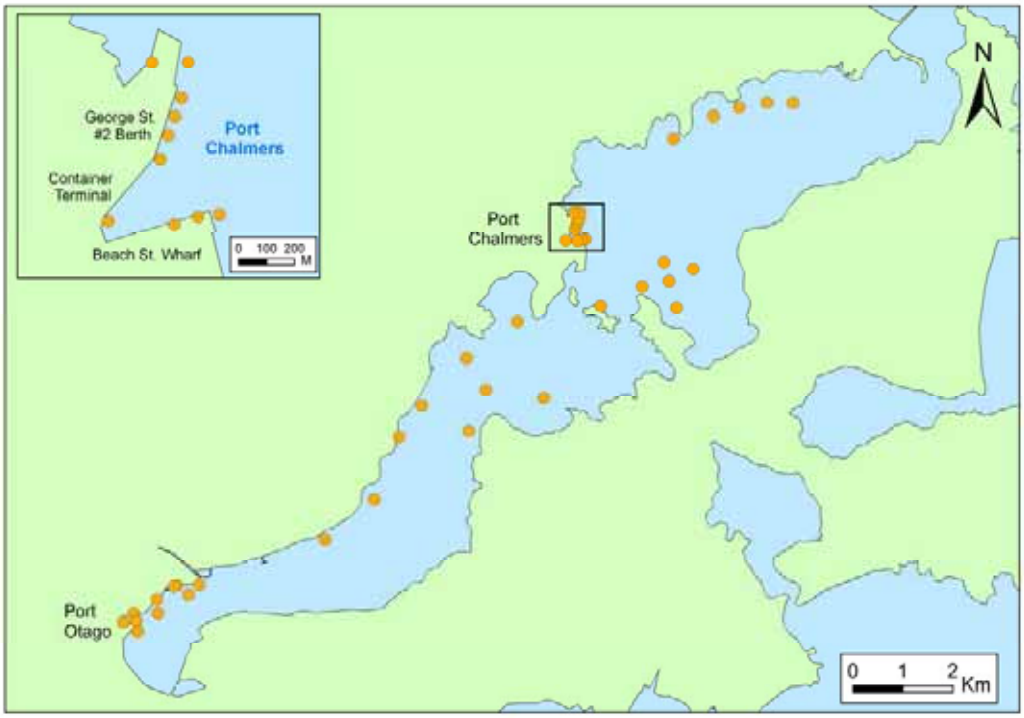

Figure 78 **Distribution of starfish traps in Otago Harbour, March 2006.**

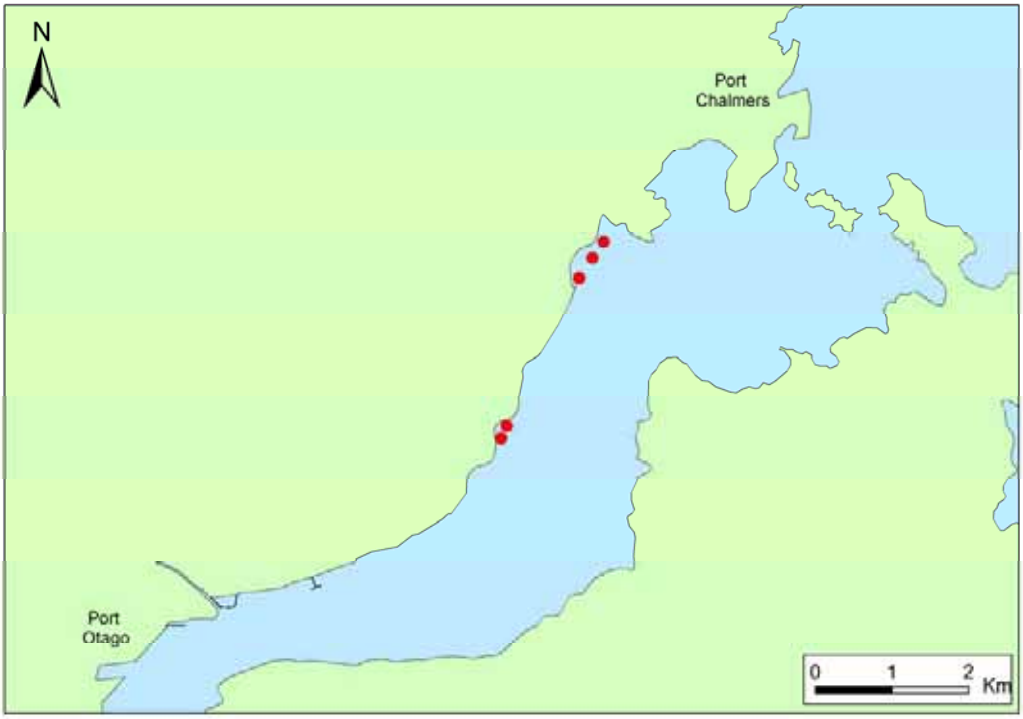

Figure 79 **Distribution of crab condos in Otago Harbour, March 2006.**

Figure 80 **Distribution of sled tows in Otago Harbour, March 2006.**

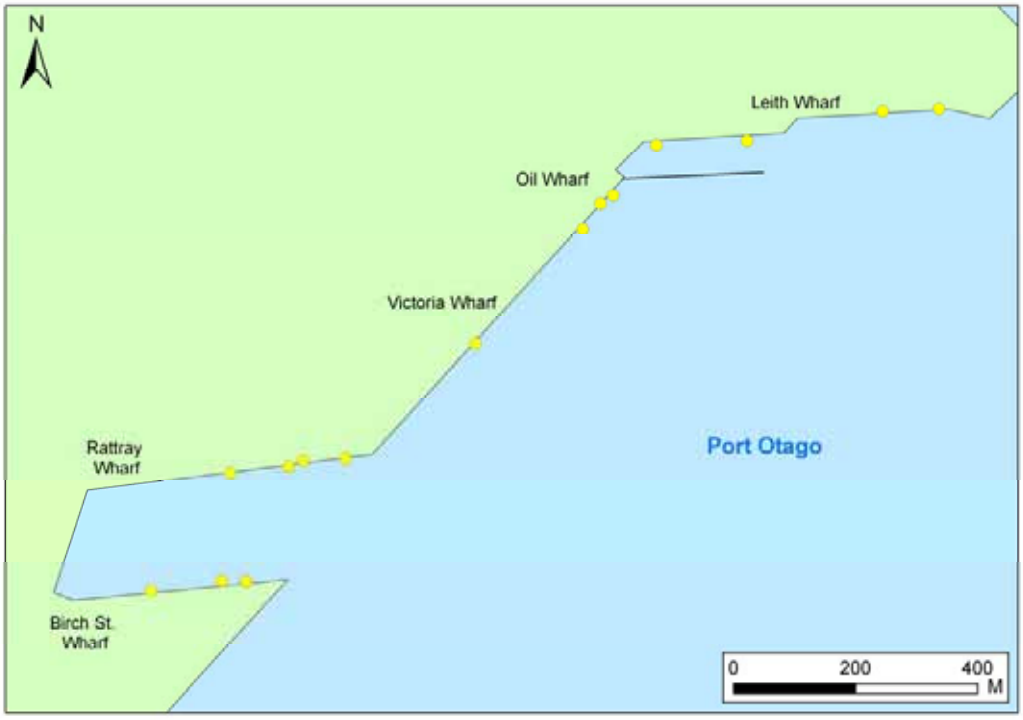

Figure 81 **Distribution of dive searches in Otago Harbour, March 2006.**

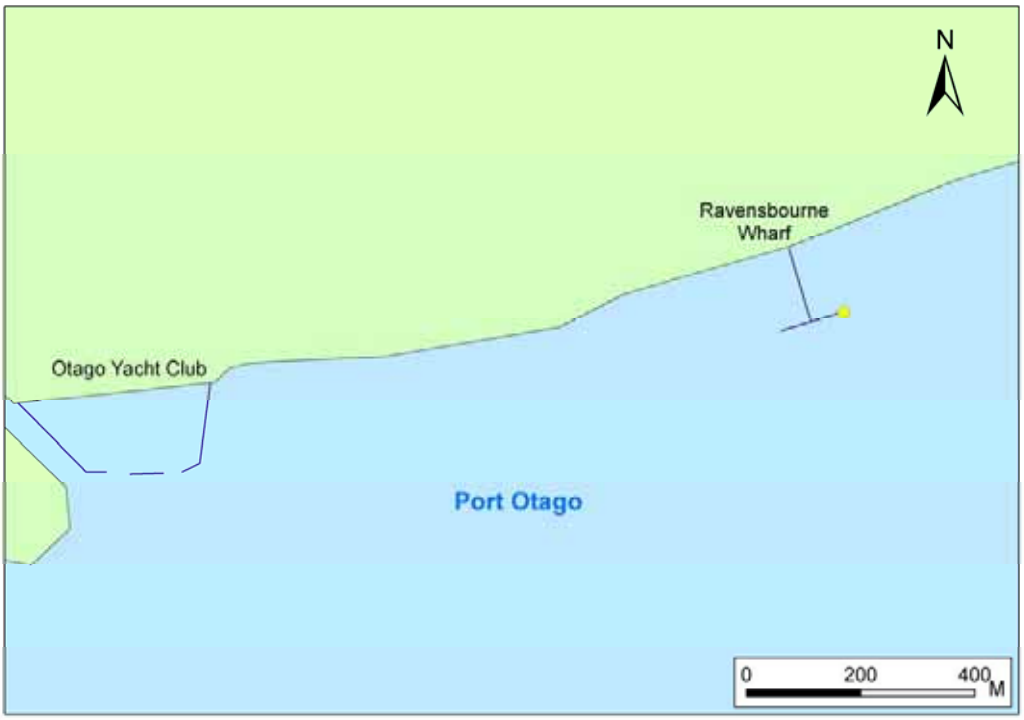

Figure 82 **Distribution of dive searches at the Ravensbourne Wharf, March 2006.**

Figure 83 **Distribution of dive searches in Port Chalmers, March 2006.**

Figure 84 Distribution of shore searches in Otago Harbour, March 2006.

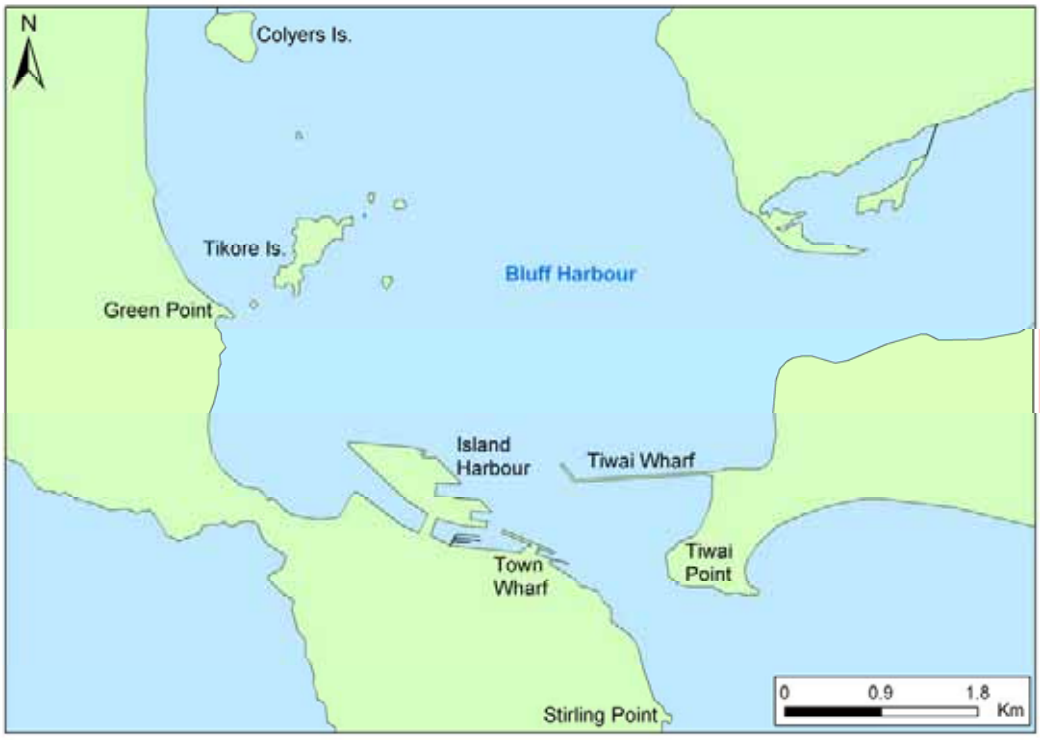

Figure 85 **Map of the sampling area around Bluff Harbour.**

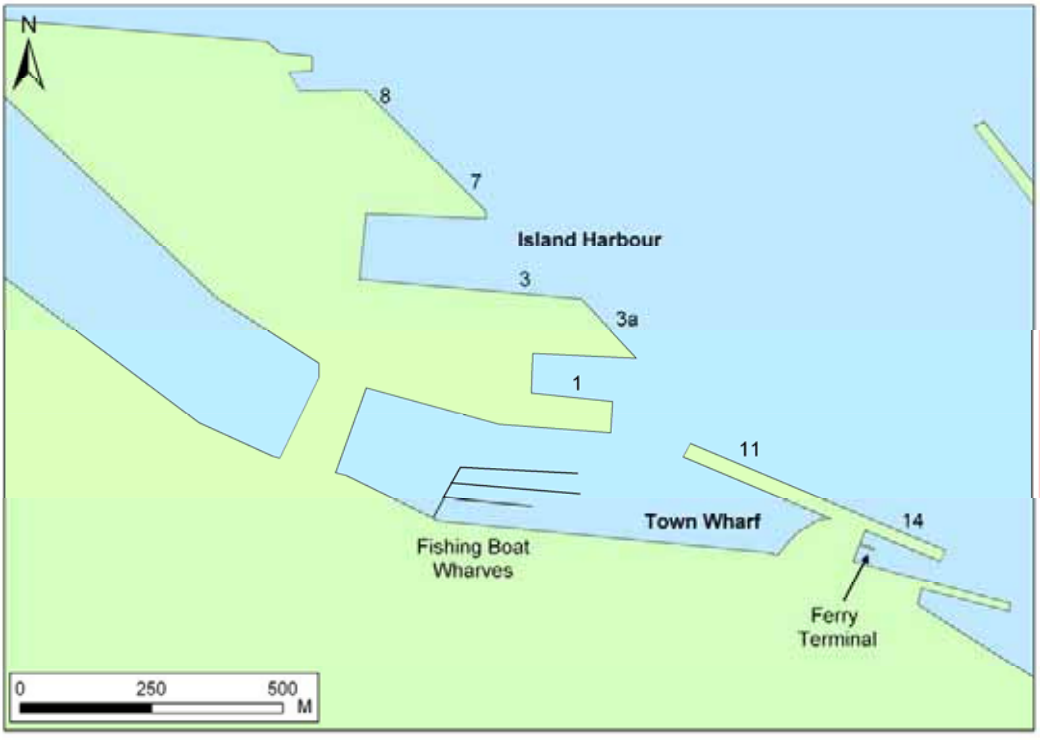

Figure 86 **Map of the sampling area around Bluff Town Wharf and Island Harbour.**

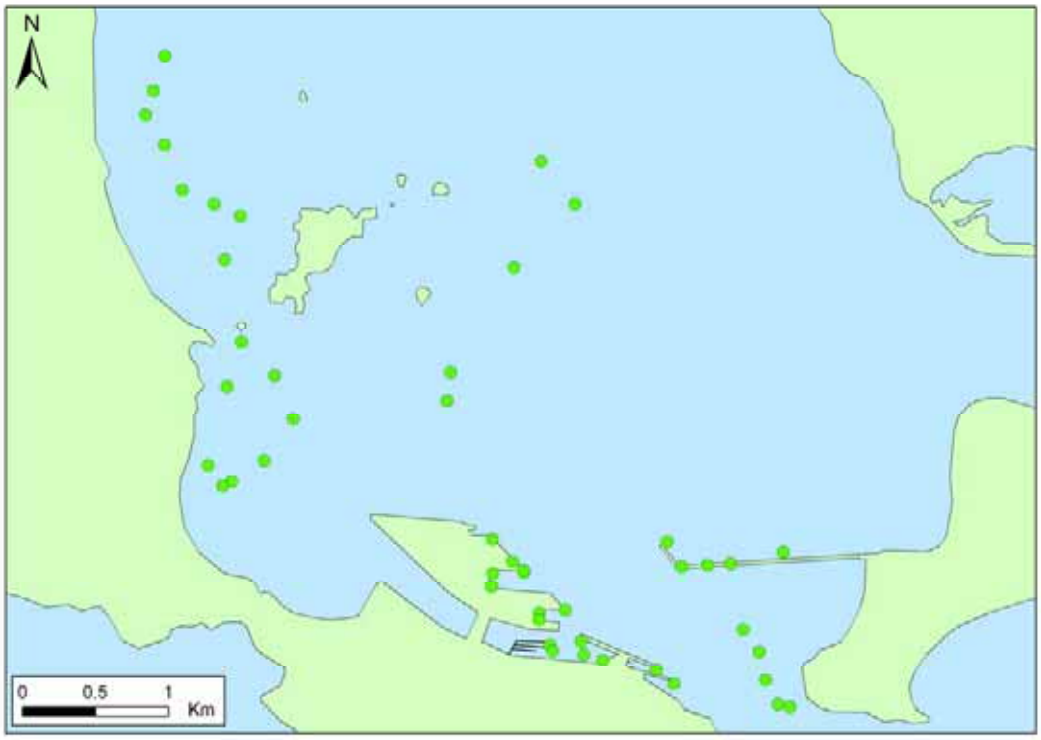

Figure 87 **Distribution of crab traps in Bluff Harbour, February 2006.**

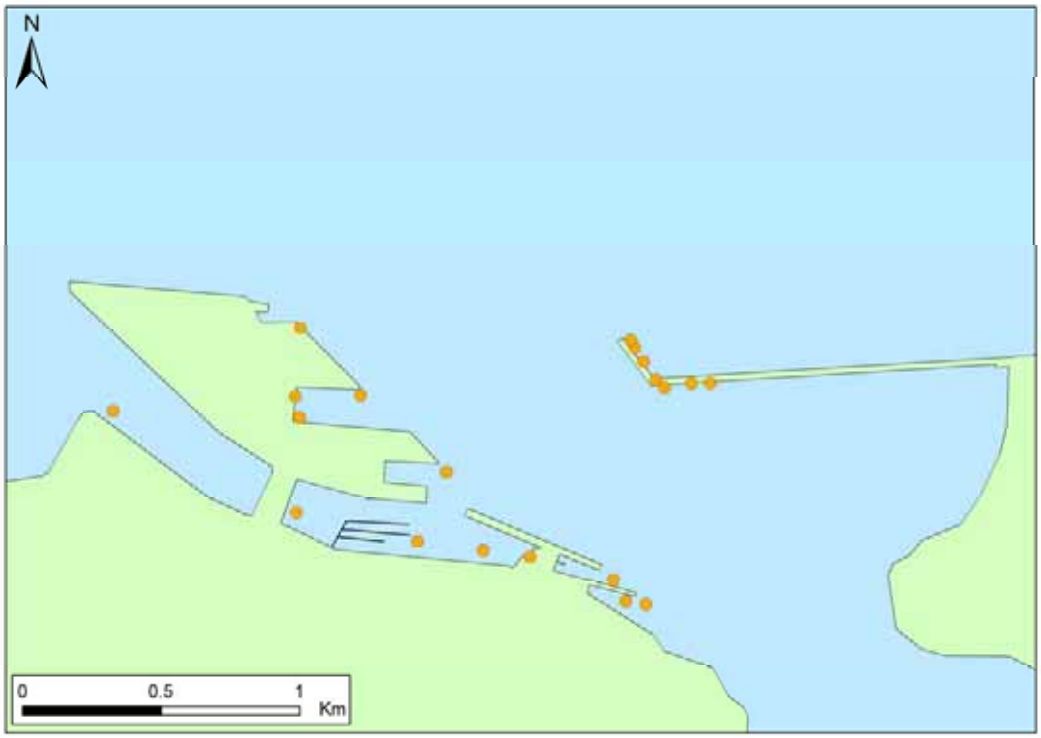

Figure 88 **Distribution of starfish traps in Bluff Harbour, February 2006.**

Figure 89 **Distribution of crab condos in Bluff Harbour, February 2006.**

Figure 90 **Distribution of sled tows in Bluff Harbour, February 2006.**

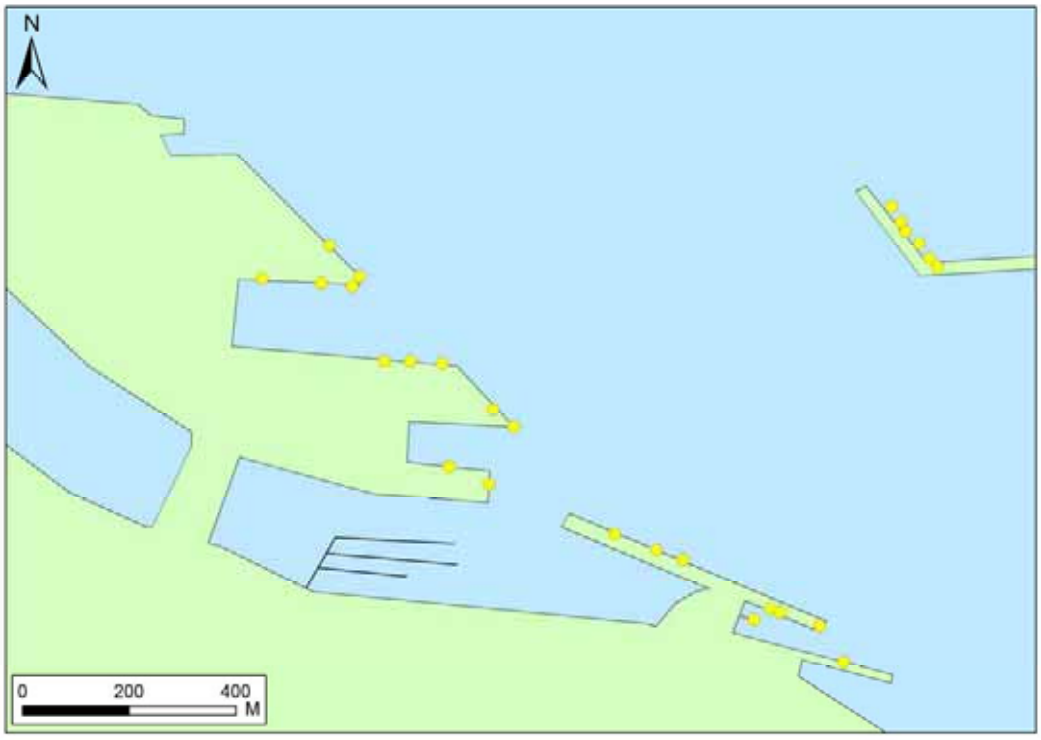

Figure 91 **Distribution of dive searches in Bluff Harbour, February 2006.**

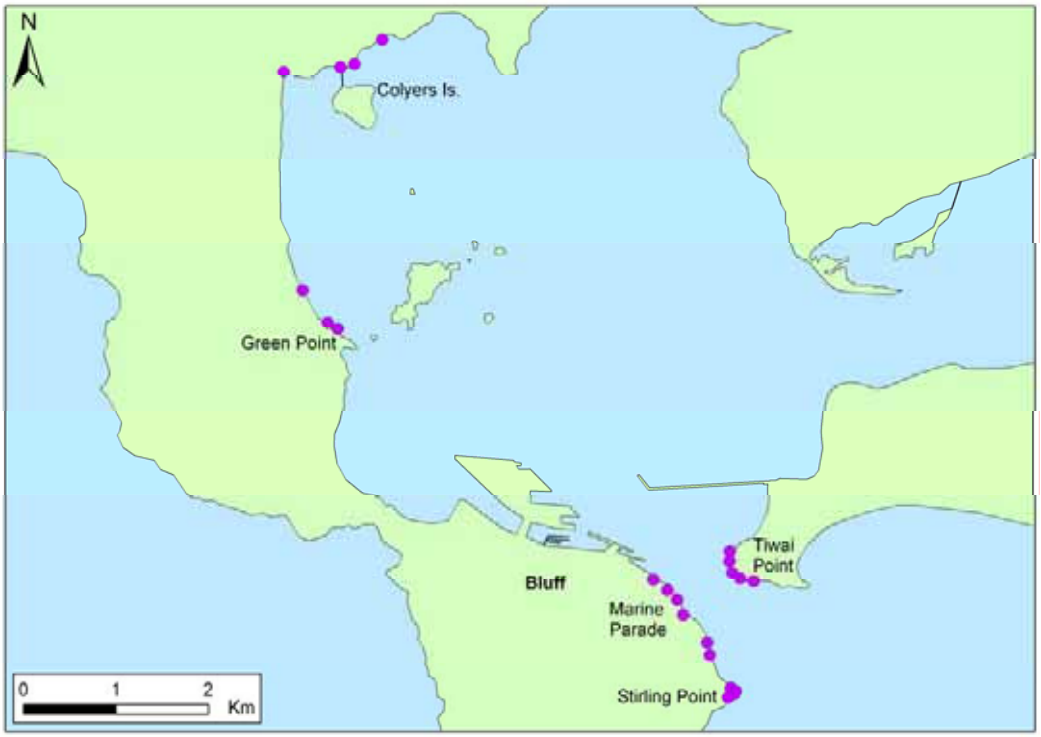

Figure 92 **Distribution of shore searches in Bluff Harbour, February 2006.**

2.2.2 Training of field personnel and QA/QC

Each field survey was led by a PhD-level scientist with experience of previous surveys or, in the case of the Waitemata Harbour survey, a PhD-level scientist was present on site during the survey (participating in the concurrent baseline survey, ZBS2005/18) and the field work was led by a NIWA staff member with experience of previous target-species surveys. Each survey included NIWA scientific divers, and staff with a range of experience in marine field surveys and parataxonomy. Prior to beginning the field programme, the team leaders were trained in field identification of the target species. The Project Leader (in consultation with the field-team leaders) was responsible for developing the sample plan for each survey and for scheduling and coordinating the field work.

Immediately prior to beginning each survey, the team leader gave the field team a briefing that described:

- the survey plan,
- key diagnostic features for identifying the eight target species and the habitats where they could be expected to occur,
- use of the survey methods,
- health and safety considerations (including procedures for liaison with local harbour control and diving), and
- protocols for data recording and preservation of samples.

Field teams were supplied with waterproofed identification guides for the target species developed by MFish (http://www.fish.govt.nz/sustainability/biosecurity/pests/index.html, accessed 27/07/05) and with copies of Tables 3 and 4 describing the sampling protocols. We also supplied the teams with pictorial guides for a range of native species that were commonly retrieved in the traps and sled tows. Where possible, the same personnel were used for repeat surveys of each harbour so the team developed a level of familiarity with species commonly encountered in the samples.

Specimens identified by the field team as a target species or which resembled the target species in some way were retained, registered, and preserved in appropriate fixative. They were then sent to the Marine Invasives Taxonomic Service (MITS) at NIWA, Wellington for registration and identification by a specialist taxonomist.

3. Results

3.1 Field surveys in the nine harbours

The only target species detected during the surveillance fieldwork were the Asian kelp *Undaria pinnatifida* and the clubbed tunicate *Styela clava* (Table 5). *U. pinnatifida* was recorded in Waitemata, Wellington, Picton, Nelson, Lyttelton, Dunedin and Bluff harbours, where it was already known to be present. It has not been recorded in Bluff during previous surveillance surveys, nor during the port baseline survey (ZBS2000/04) in March 2003, but MAF Biosecurity New Zealand was already aware of its presence there when informed of the record during the present survey (Brendan Gould, MAF BNZ, pers. comm.). Control of *U. pinnatifida* in Bluff Harbour by the Department of Conservation ceased toward the end of the previous surveillance work. *S. clava* was recorded in Waitemata and Lyttelton harbours, where its presence had been reported previously (Gust et al. 2006a).

Non-target introduced species are also listed in Table 5. They include an introduced species of barnacle, *Austromegabalanus psittacus*, collected from Wellington Harbour and representing a new record for New Zealand (Hosie & Ahyong, 2006).

Although most identification was done by the field-team, 159 specimens were retained and sent to specialist taxonomists for formal identification via MITS during the course of the present series of field surveys. This contrasts with more than 570 specimens retained in the 2002-2004 surveys. This reduction probably represents an increase in the confidence with which field teams were able to distinguish specimens of the target species as a result of previous experience, in addition to the smaller number of surveys represented (1 survey at each of 9 ports compared with 4 surveys of each of 8 ports). The total number retained from the present surveys included 52 ascidians (mostly *Styela clava*), 35 algae (mostly *Codium* and *Undaria pinnatifida*), 20 polychaetes, 17 crustaceans (mostly decapods), 15 molluscs (mostly bivalves), 13 fish and 4 asteroids.

In addition to *Styela clava*, the ascidians retained were the known cryptogenic species *Aplidium phortax* (Michaelson, 1924) and *Styela plicata* (Lesueur, 1823), the known introduced *Ciona intestinalis* (Linnaeus, 1767) and the natives *Corella eumyota* Traustedt, 1882 and *Synoicum otagoensis* Millar, 1982.

Of the 35 algal specimens kept for specialist identification, 24 specimens were species of *Codium*, and 5 of these were thought to be the invasive subspecies *Codium fragile* ssp. *tomentosoides* (van Goor) P.C. Silva (from Waitemata and Tauranga harbours). The known introduced species *Hydroclathrus clathratus* (C.Agardh) M.Howe was also recorded (in Whangarei).

Several species of tube-building polychaetes were retained for formal identification, either to discount them as *Sabella spallanzanii* or to confirm them as the known introduced species *Ficopomatus engimaticus* or the known cryptogenic *Chaetopterus* sp. Specimens retained included: *Chaetopterus* sp. (Tauranga, Waitemata and Whangarei harbours) and *F. engimaticus* (Whangarei); native species (the onuphid *Onuphis aucklandensis*, the sabellids *Branchiomma* sp. Nov., *Demonax* sp., *Megalomma suspiciens* (Ehlers, 1904) and *Pseudobranchiomma grandis* Baird, 1865, and the terebellid *Streblosoma toddae* Hutchings & Smith 1997); the known cryptogenic *Branchiomma curtum* (Ehlers, 1907); and the known introduced serpulid *Hydroides elegans* Haswell 1883 (Nelson).

In addition to the barnacle *Austromegabalanus psittacus*, another, known introduced species of crustacean, the crab *Pyromaia tuberculata* (Lockington, 1877), was collected from Waitemata Harbour. Numerous specimens of the introduced paddle crab *Charybdis japonica* were also collected from Waitemata Harbour and preserved but were not registered with MITS, being easily identified in the field.

The bivalves retained included individuals of the native species of *Corbula*, *C. zelandica*, and two known introduced species, *Limaria orientalis* (Adams & Reeve, 1850) and *Theora lubrica* Gould, 1861 (the former from the Waitemata Harbour, the latter from Tauranga Harbour). *T. lubrica* was also recorded from Whangarei, Waitemata, Wellington, Picton, Nelson and Lyttelton harbours, but specimens were identified *in situ*.

Several native New Zealand starfish were regularly caught in sled tows and traps, the most numerous of which were the cushion star *Patiriella regularis* and 11-arm sea-star, *Coscinasterias muricata*. However, specimens of the native 5-armed starfish *Allostichaster insignis* (Farquhar, 1895) and *Pentagonaster pulchellus* Gray 1840 were occasionally retained for specialist identification.

Of the fish retained for identification, 4 were individuals of a known introduced species of goby, *Acentrogobius pflaumii* (Bleeker, 1853), collected in Waitemata Harbour. The native range of this species is the northwest Pacific from Russia to Taiwan (Froese & Pauly 2006) but it has also been introduced into southeast Australia. It may have arrived in New Zealand in ballast water from Australia or from its native range (Francis et al. 2003). It has previously been recorded in Waitemata Harbour and Whangapoua Harbour on the Coromandel Peninsula (Francis et al. 2003). Other individuals of this genus were retained, but it was not possible to identify them to species because of their size or condition.

3.2 Target species

3.2.1 Undaria pinnatifida

Undaria pinnatifida was present in Waitemata, Wellington, Picton, Waikawa Marina, Nelson, Lyttelton, Dunedin and Bluff harbours (Figures 93-100: maps show records of *U. pinnatifida* from sleds, dive searches and shore searches). It was widely distributed on artificial habitats and sheltered hard shorelines in all of these harbours except Bluff, where it was only recorded at one location (Tiwai Wharf). It was most frequently encountered in diver surveys of artificial habitats (wharf pilings, breakwalls and floating pontoons) and shoreline searches. Drifting sporophytes were also occasionally recorded in sled tows and traps. During previous surveys (2002-2004), *U. pinnatifida* was recorded from Wellington, Nelson, Lyttelton and Dunedin (Figures 101-105 display the composite distribution of *U. pinnatifida* in each harbour derived from the four surveys during 2002-2004). Its presence in Waitemata Harbour was first reported in October 2004 from the Viaduct Harbour, and it was subsequently found in St Mary's Bay, Westhaven Marina, Wynyard Wharf, Quarantine Wharf and Freyberg Wharf (Stuart & McLary 2004).

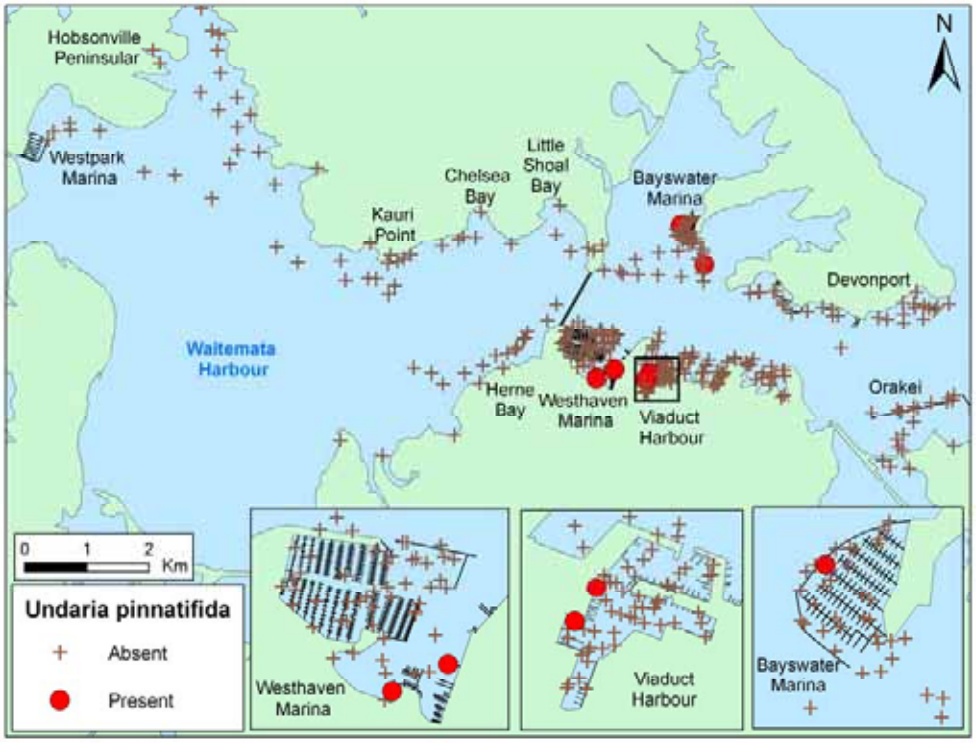

Figure 93 **Distribution of *Undaria pinnatifida* in Waitemata Harbour in the present survey.**

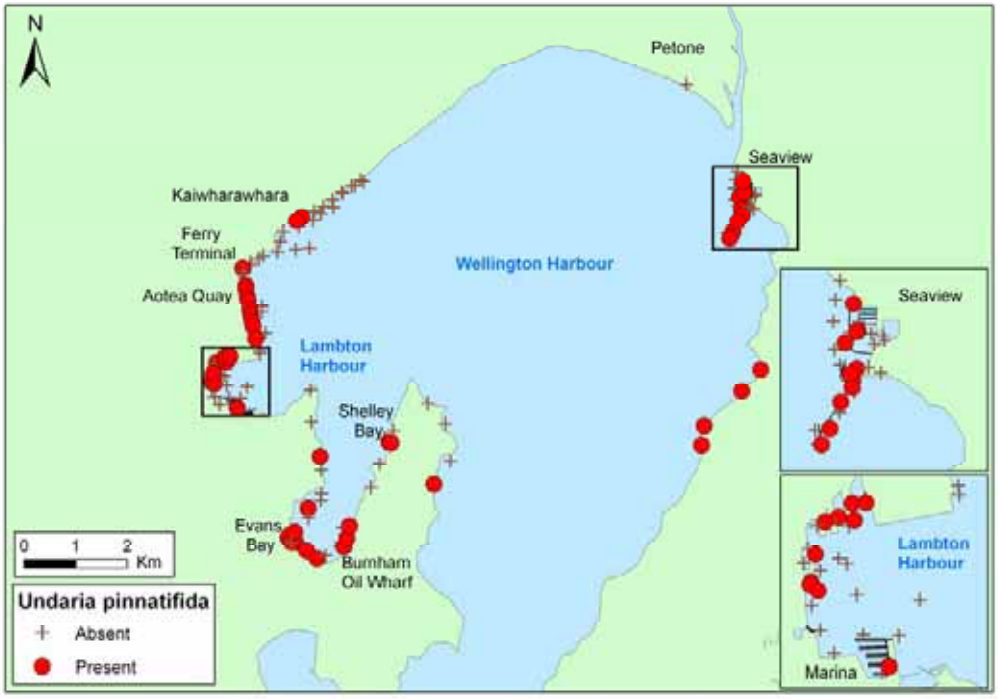

Figure 94 **Distribution of *Undaria pinnatifida* in Wellington Harbour in the present survey.**

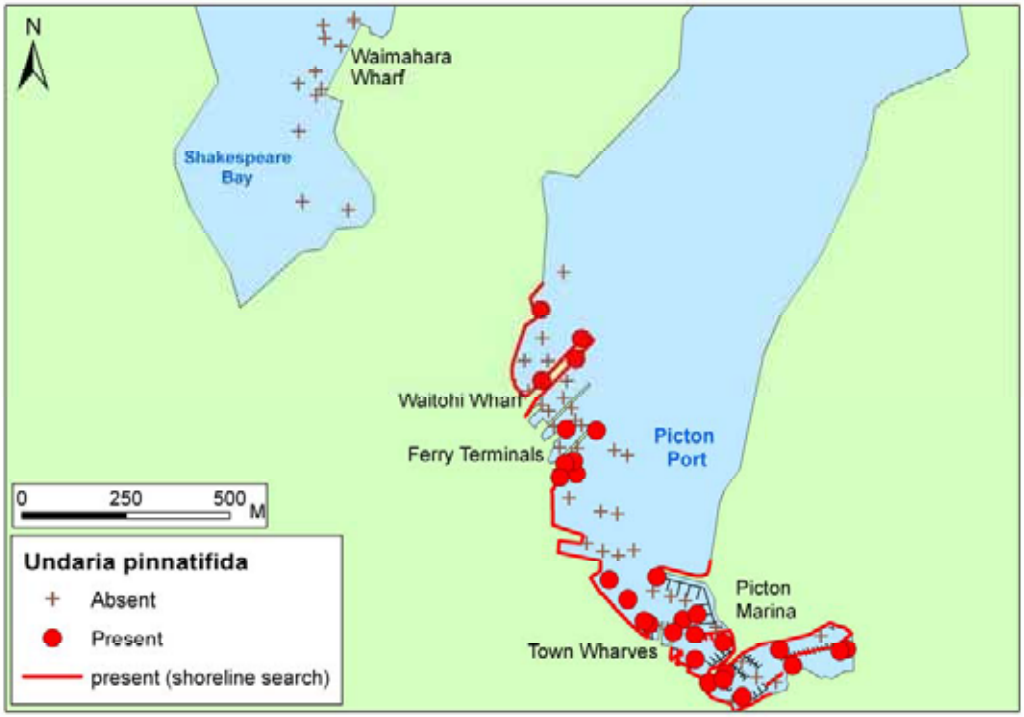

Figure 95 **Distribution of *Undaria pinnatifida* around Picton in the present survey.**

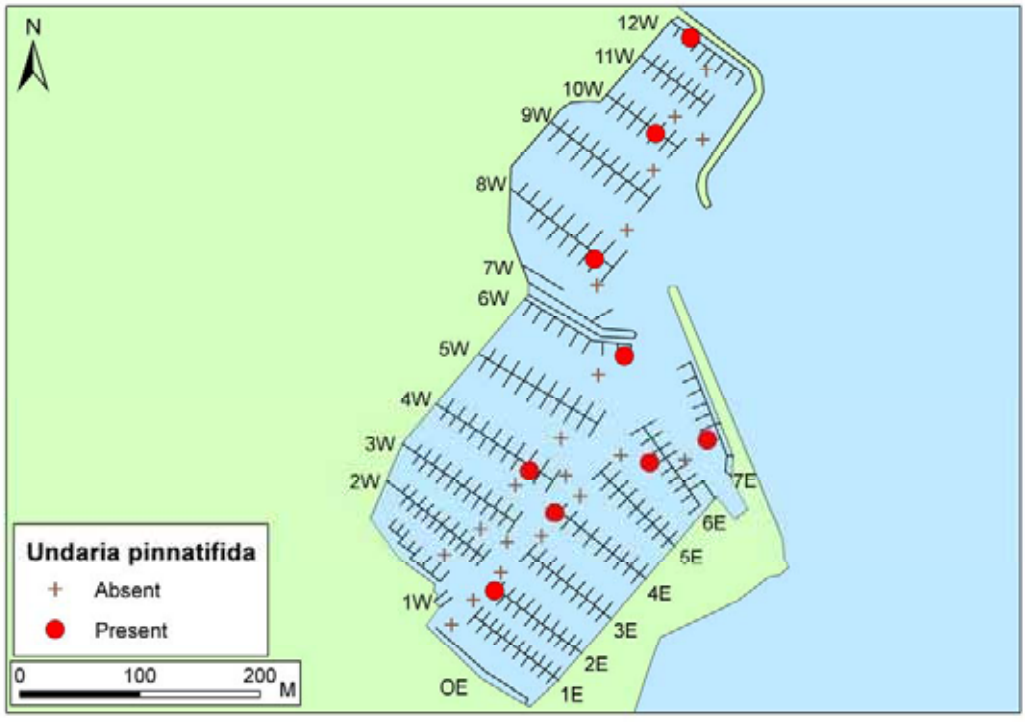

Figure 96 **Distribution of *Undaria pinnatifida* around Waikawa Marina in the present survey.**

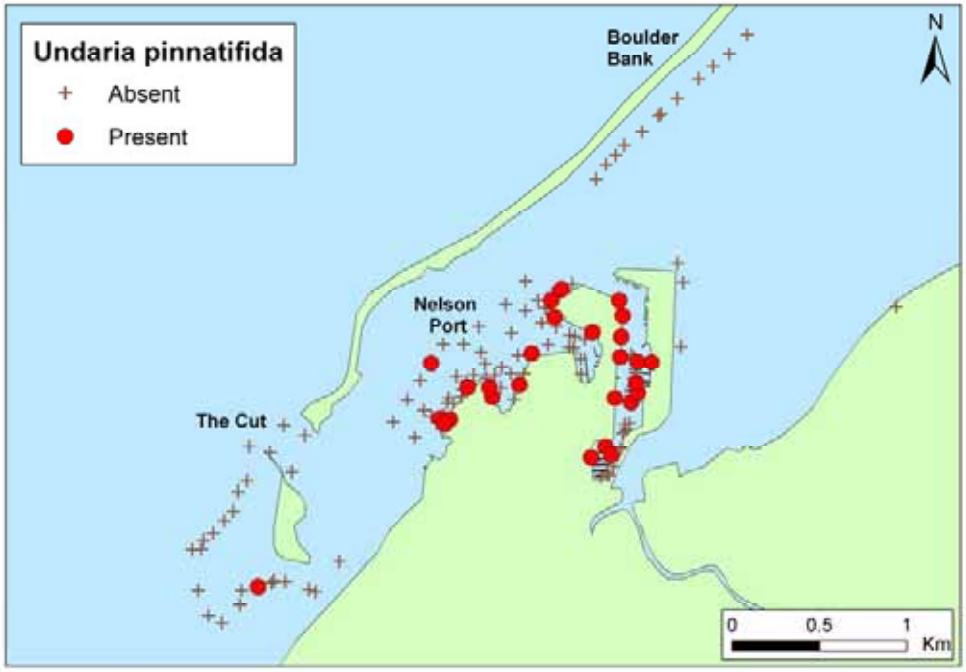

Figure 97 **Distribution of *Undaria pinnatifida* around Nelson Port in the present survey.**

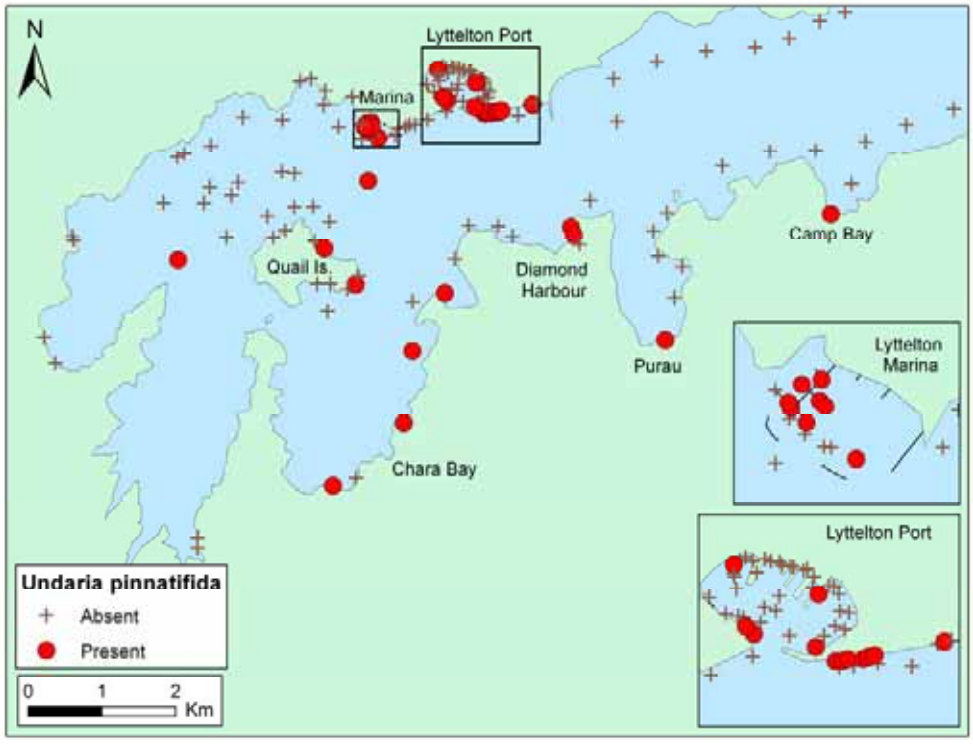

Figure 98 **Distribution of *Undaria pinnatifida* in Lyttelton Harbour in the present survey.**

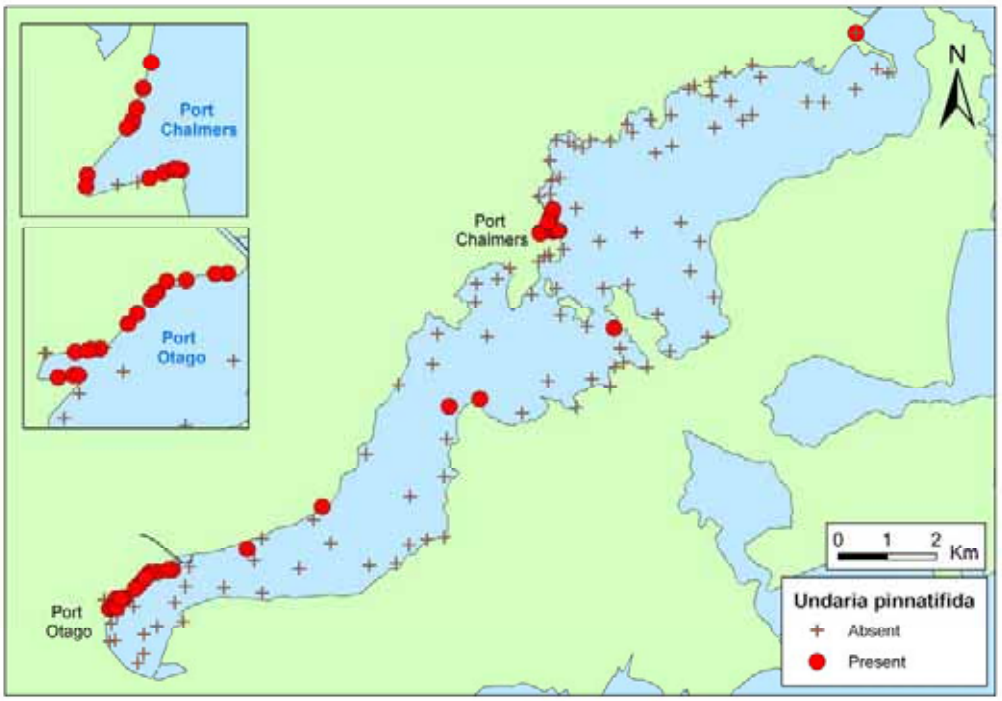

Figure 99 **Distribution of *Undaria pinnatifida* around Otago Harbour in the present survey.**

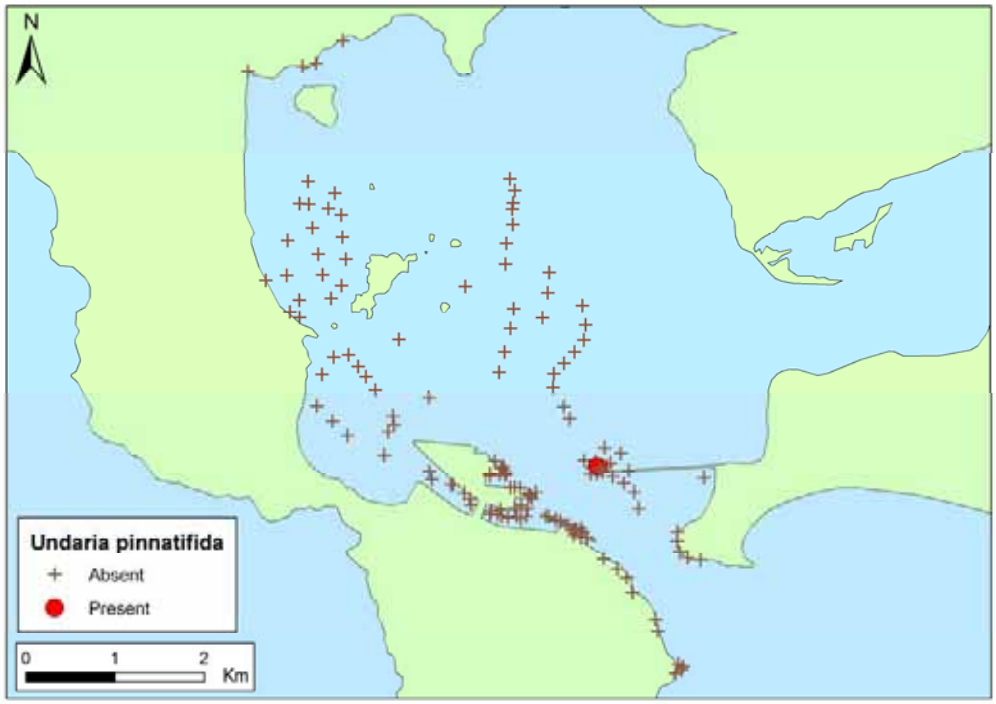

Figure 100 **Distribution of *Undaria pinnatifida* around Bluff Harbour in the present survey.**

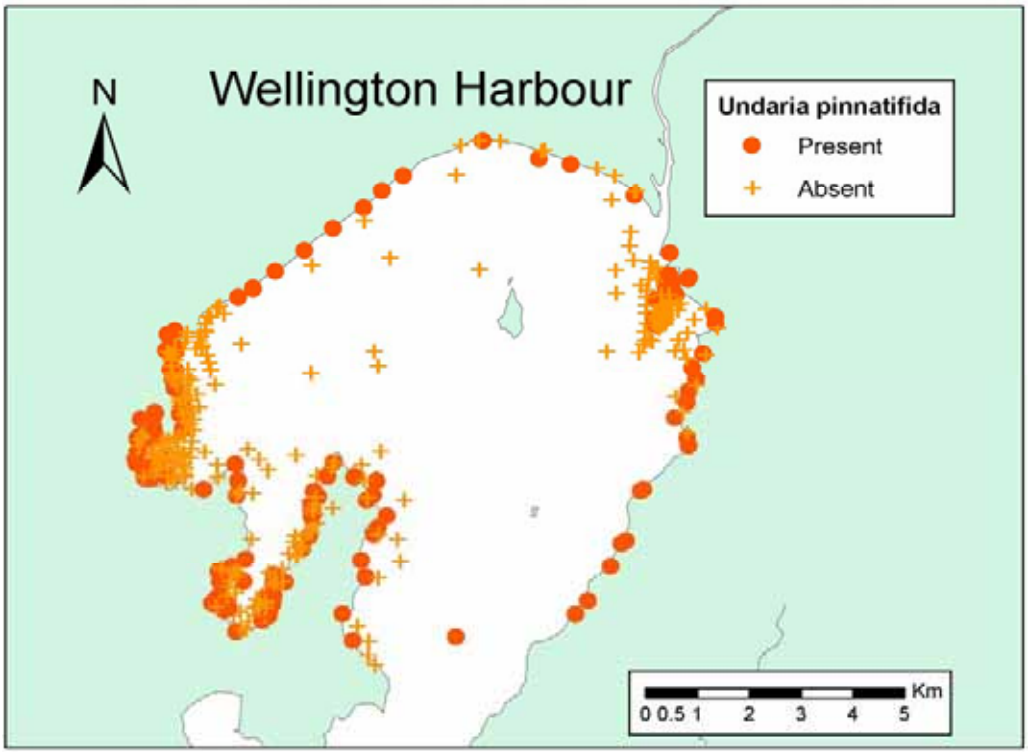

Figure 101 **Distribution of *Undaria pinnatifida* in Wellington Harbour 2002-2004.**

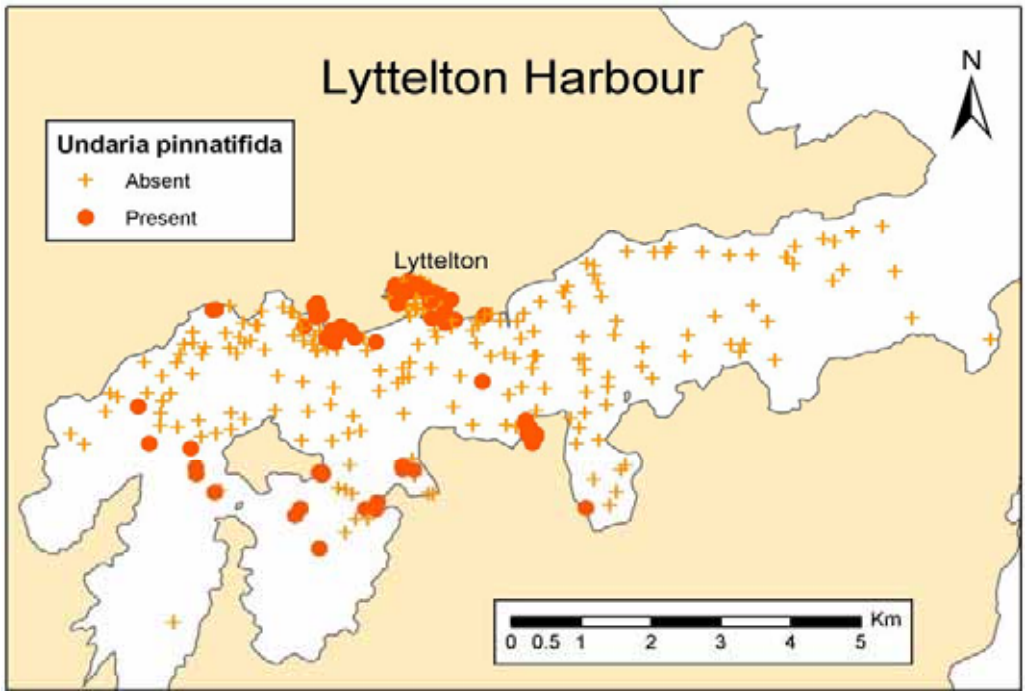

Figure 102 **Distribution of *Undaria pinnatifida* in Lyttelton Harbour 2002-2004.**

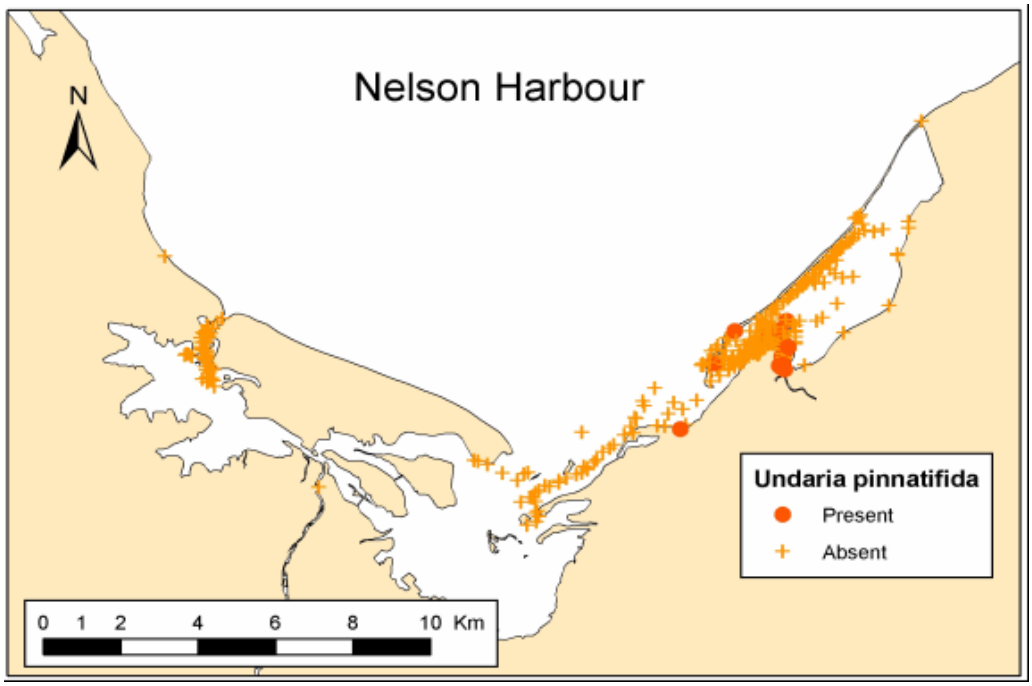

Figure 103 **Distribution of *Undaria pinnatifida* around Nelson 2002-2004.**

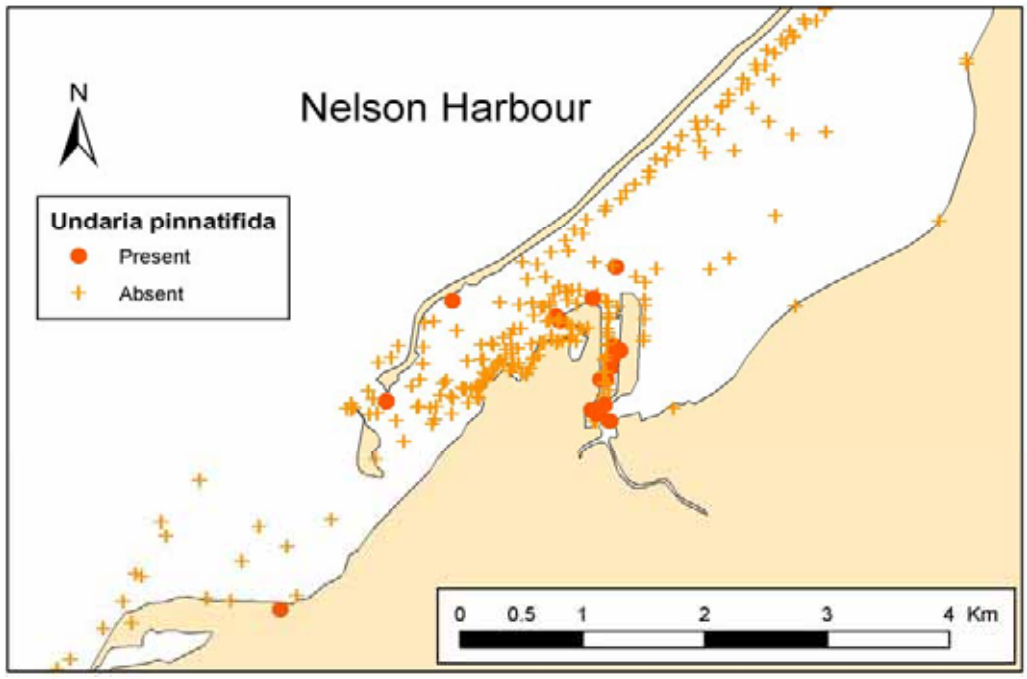

Figure 104 **Detail of the previous map showing the port area and Nelson Haven.**

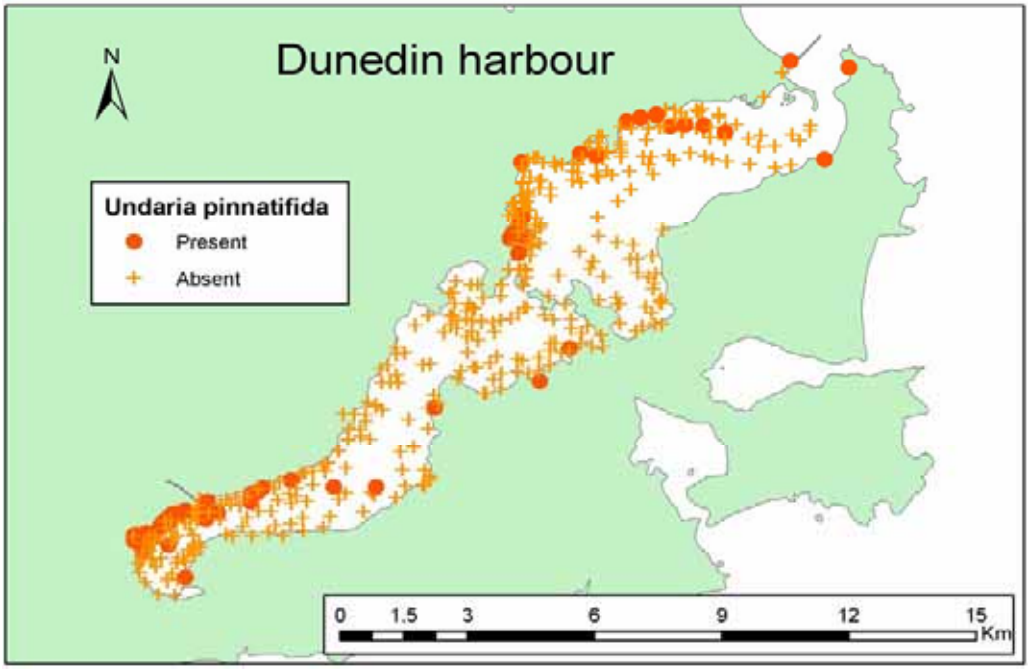

Figure 105 **Distribution of *Undaria pinnatifida* around Otago Harbour 2002-2004.**

In Nelson Port, *Undaria pinnatifida* appears to have extended its distribution westward since the previous surveys, to include the port area between Kingsford Quay and the Main Wharf (the isolated record southwest of the port was probably a detached plant). It also appears to occur at more sites in the outer part of the marina than in previous surveys. These apparent changes in distribution should, however, be treated with caution because *U. pinnatifida* exhibits annual cycles of growth and senescence (Forrest et al. 2000, Thompson 2004) and its detectability is likely to vary with season. It is also interesting to note that during the port baseline survey (ZBS2000/04) in December 2004, *U. pinnatifida* was only found in the marina and on the inside of the Boulder Bank, and not at sampling sites in the port. This also lends support to the suggestion that *U. pinnatifida* had expanded its distribution during the period 2004 to late 2005.

In Wellington Harbour, the distribution of U*ndaria pinnatifida* was similar in the present survey and the 2002-2004 surveys. In contrast to previous surveys, however, the present survey did not detect *U. pinnatifida* north of Kaiwharawhara, at Petone Beach or at several sites around the peninsula between Evans Bay and Seatoun. Again, these absences may relate to differences in the time of sampling relative to the annual cycle of growth and senescence of *U. pinnatifida* (previous surveys occurred in July, November and December).

Similarity between distribution in 2002-2004 and 2006 surveys is also evident in Lyttelton Harbour. New records in Chara and Camp Bays were at locations not searched in previous surveys.

Undaria pinnatifida was abundant in Port Otago and Port Chalmers in previous and present surveys, but was less widely distributed in the outer part of Dunedin in the present study, being found at only one site (the northern side of the harbour entrance). Previous surveys took place in January, March, July and August.

Undaria pinnatifida was not detected in Waitemata and Bluff harbours in previous surveys. In the present survey it was recorded in the entrance to Ngataringa Bay (outer harbour), Bayswater and Westhaven Marinas and the Viaduct Harbour. *U. pinnatifida* may have been present at the time of previous surveys (this is known to have been the case with the September 2004 survey of Waitemata Harbour) but was not detected.

3.2.2 Styela clava

Styela clava was not detected in previous surveys, largely because it was not a target species at that time, and was not known to occur in New Zealand. In the Waitemata Harbour, *S. clava* occurred at Bayswater Marina, Westhaven Marina, the Viaduct Harbour (including Hobson West Marina), the port area and the breakwater off Orakei/Hobson Bay (Figure 106: maps show records from dive searches and shore searches and, in the case of Waitemata Harbour, box traps because an individual was found in a trap, presumably dislodged from a pile or pontoon while the trap was being recovered). Its wide distribution in the Westhaven Marina and Viaduct Harbour areas reflects the findings of an earlier, more intensive survey (Gust et al. 2005). Bayswater Marina was not surveyed in the earlier study but 2 vessels that had been brought from Bayswater Marina to the Viaduct Harbour for slipping and cleaning were inspected and found to have *S. clava* on their hulls (Gust et al. 2005). These vessels had not left Bayswater Marina for several months prior to moving to the Viaduct Harbour.

Individuals found in the Waitemata Harbour were attached to piles, floating pontoons and other structures, and the breakwater off Orakei/Hobson Bay. Because of the large numbers of individuals present throughout the area surveyed, it was not practical to collect all those seen, but samples were taken from most locations where they were found.

Styela clava was found at 3 locations in Lyttelton Port: the eastern end of the Oil Wharf, the western end of Gladstone Pier and Number 2 Wharf (Figure 107). A total of 25 individuals were found, all during dive searches (on 16 March 2006) and all those found were removed. MAF BNZ was informed of the finds on 21 March. Individuals on the Oil Wharf were attached to ropes and piles and occurred on 3 of the

10 piles inspected. On Gladstone Pier they were attached to a concrete pontoon, fenders and to 15 of the total of 20 piles inspected at 2 sites on the Pier. Three sites were searched on Number 2 Wharf and *S. clava* was found at each, on 3 of 10 piles at one site, 1 of 10 at the second and 4 of 10 at the third. Population densities were in the range 1-10 individuals m^{-2}.

Although *Styela clava* was previously known to occur in Lyttelton (Gust et al. 2006a), the individuals found on Number 2 Wharf represent a new site within the harbour, but this may be because this particular site was not searched in previous surveys. A single specimen was collected in a pile-scrape sample from Gladstone Pier as part of the baseline port survey (ZBS2000/04) in November 2004 (Inglis et al. 2006a). In October-November 2005, 15 specimens were collected from Lyttelton Port and 5 from Lyttelton Marina (Gust et al. 2006a). In the port, individuals were found attached to the underside of a floating pontoon at the western end and on piles in the central part of Gladstone Pier, on piles at 2 locations on Number 7 Wharf and the end of the Oil Wharf. All individuals collected in the marina were on submerged ropes. Population densities at all sites were in the range 1-10 individuals m^{-2} and all specimens found were adults (7-13 cm).

Specimens of *Styela clava* collected during the present study were sent to Sharyn Goldstien at the University of Canterbury (with the approval of MAF BNZ) for inclusion in a study of genetic relationships among *S. clava* populations in New Zealand.

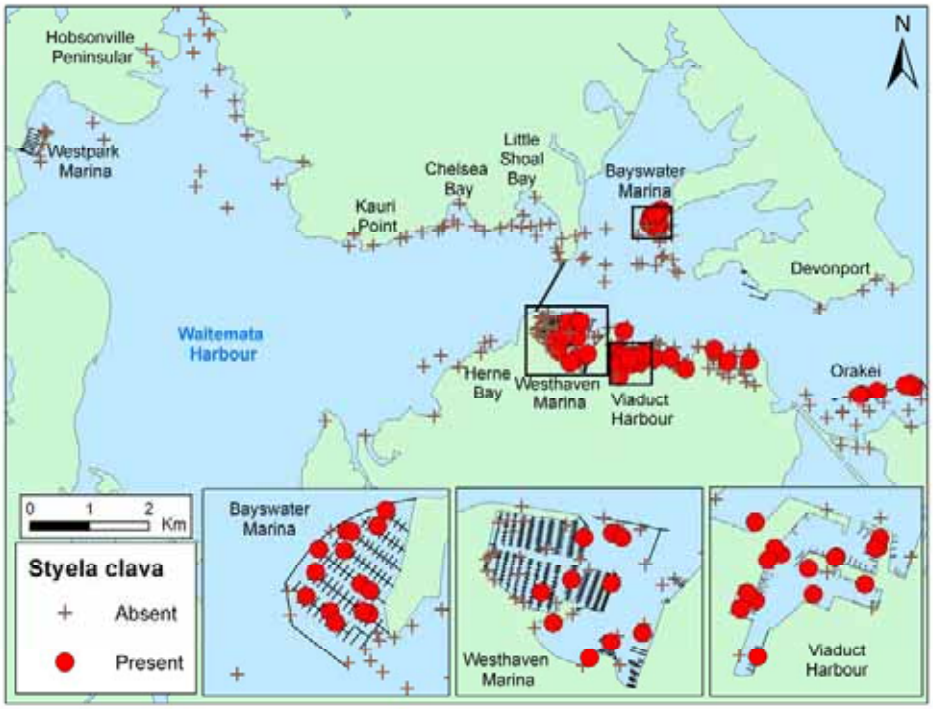

Figure 106 Distribution of *Styela clava* in Waitemata Harbour.

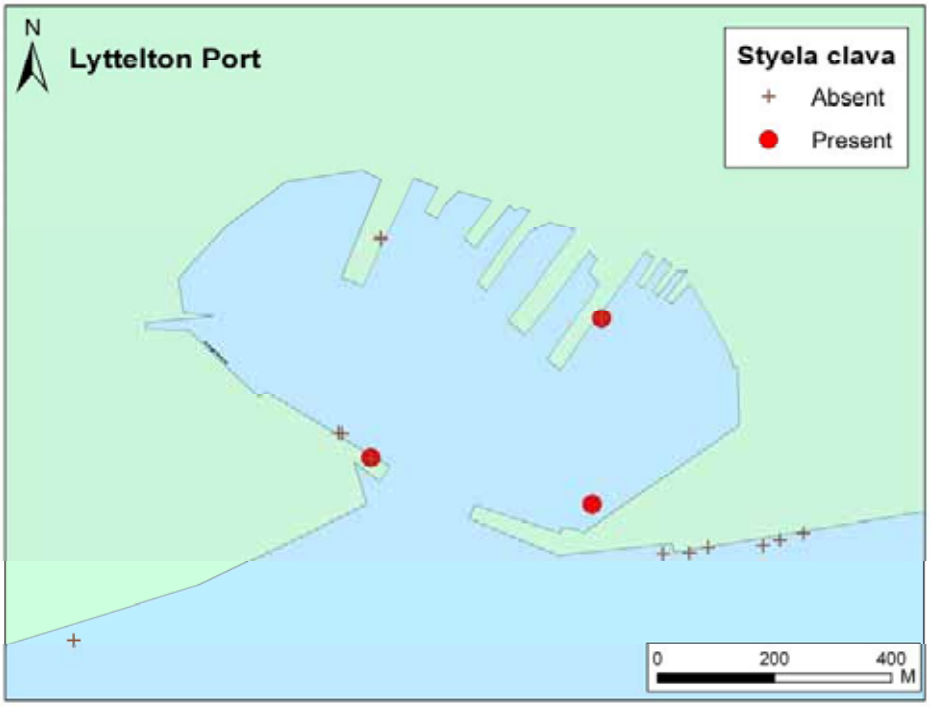

Figure 107 Distribution of *Styela clava* in Lyttelton Harbour.

3.2.3 Non-target species

Chaetopterus sp.

The cryptogenic parchment tubeworm, *Chaetopterus sp*., was recorded from the three northern harbours of Whangarei (Figure 108), Waitemata (Figure 109) and Tauranga (Figure 110: maps show records from sleds, dive searches and shore searches). In Whangarei the species was most frequently sampled on sandy sediments located near the harbour entrance. In the Waitemata Harbour it was recorded on the breakwater off Orakei/Hobson Bay and on pontoons in Bayswater Marina. In Tauranga it was found on piles and a sled tow in the port area.

In Whangarei Harbour, *Chaetopterus* sp. was present on sandy-mud or sandy subtidal sediments, from the harbour entrance to One Tree Point on the southern shoreline, Parua Bay on the northern shoreline, and along Tamaterau Reach in the middle harbour (Figure 108). It was abundant on wharf pilings at Marsden Point in previous surveys (2002-2004: Inglis et al. 2005a) but in the present study, although empty tubes were present at most sites in this location, few live individuals were recorded. In previous surveys *Chaetopterus* sp. was present in the major shellfish beds in Urquharts Bay, Taurikura, and Shoal Bay, including Snake Bank and MacDonald Bank (these areas were not sampled during the present survey).

Few living *Chaetopterus* sp. were usually captured during previous (2002-2004) surveys of Tauranga and Waitemata Harbours. Samples obtained through epibenthic sledding and intertidal visual searches often consisted of empty tubes. During the earlier (March 2002) baseline port survey of the port of Tauranga, dense coverings of *Chaetopterus* sp. occurred at the base of pilings (10-12 m depth) on berths 1-3 at the port in Mount Maunganui (Inglis et al. 2005b). Only occasional records of *Chaetopterus* were made from wharf pilings in the target-species surveillance programme because of the shallow depths (2-4 m) surveyed by divers.

Chaetopterus sp. came to the attention of New Zealand scientists in 1997 when commercial scallop fishers reported dense tube mats in scallop grounds at Little Barrier Island. It was subsequently reported from other areas of northern New Zealand that are fished by scallop boats. Surveys undertaken for MFish by Auckland Uniservices Ltd in 2001 showed that its distribution extended from Bream Head, in the north, to the Motiti Islands in the south (Tricklebank et al. 2001). There is some uncertainty about the taxonomy of this species, since museum specimens and holotypes are often poorly preserved making comparisons with other species difficult. As a result, it has not been possible to determine whether this species is endemic to the

New Zealand region or has been introduced from overseas (G. Read pers. comm.). At present it is considered cryptogenic (of uncertain origin).

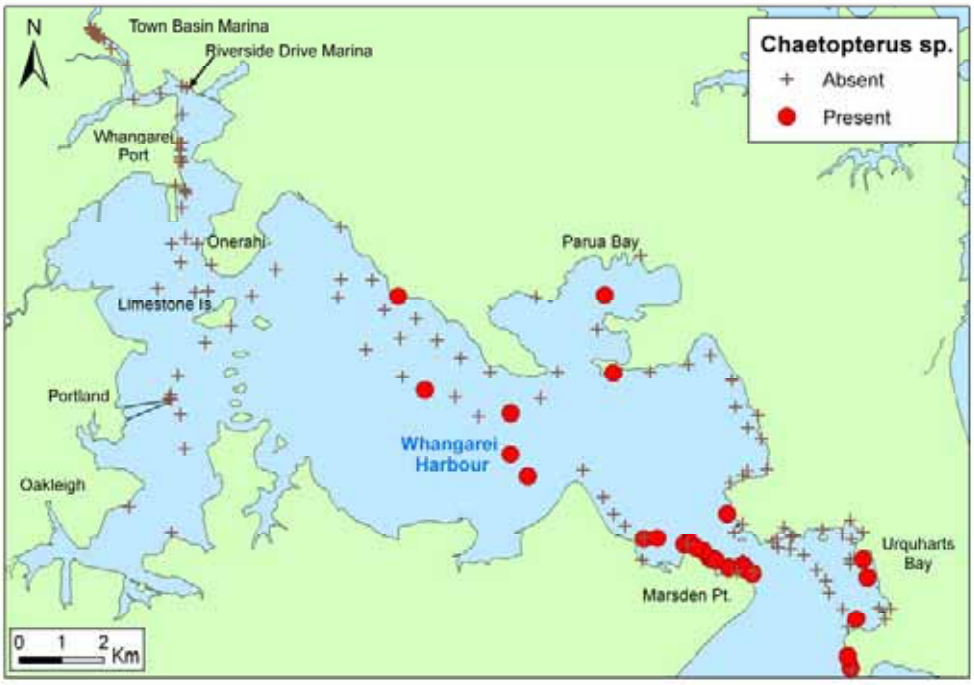

Figure 108 Distribution of *Chaetopterus* sp. in Whangarei Harbour.

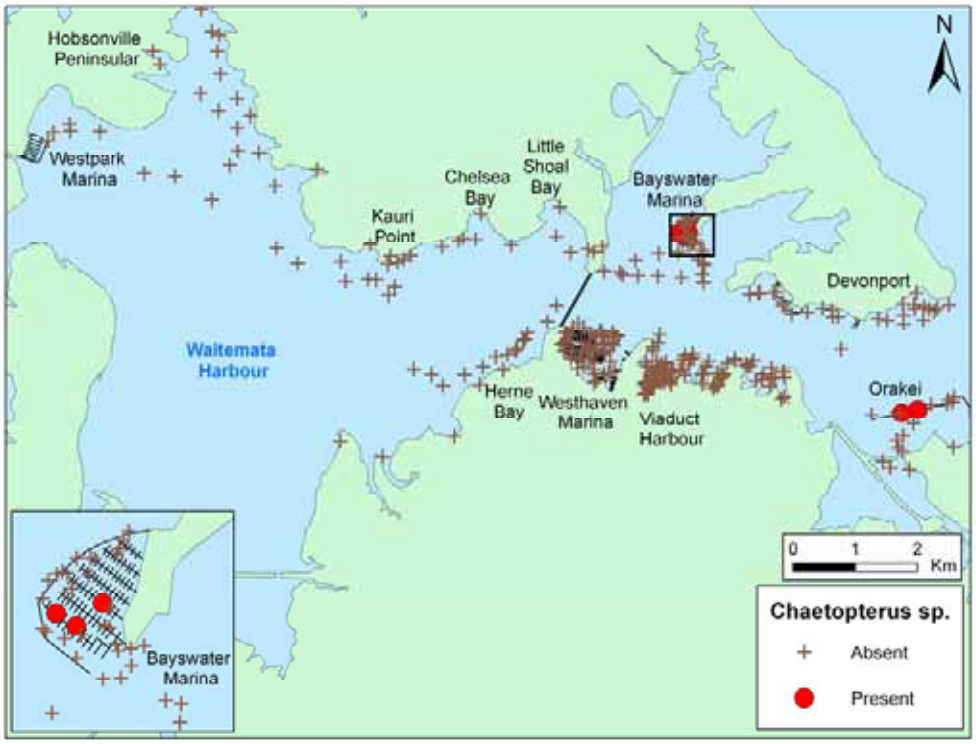

Figure 109 Distribution of *Chaetopterus* sp. in Waitemata Harbour.

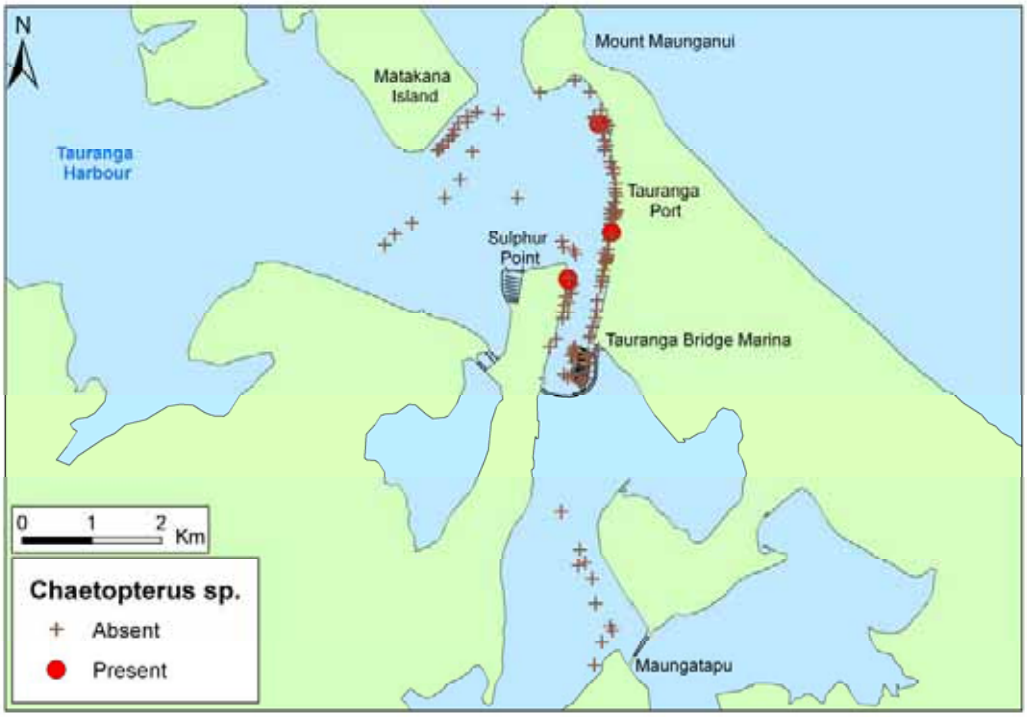

Figure 110 Distribution of *Chaetopterus* sp. in Tauranga Harbour.

Musculista senhousia

The Asian date mussel, *Musculista senhousia*, was recorded from Whangarei Harbour (Figure 111) and Waitemata Harbour (Figure 112: maps show records from sleds). It has been found previously in both harbours. *M. senhousia* was first reported from Waitemata Harbour and Te Puru in the Firth of Thames in 1980 (Willan 1987). Although it has previously been a dominant component of the fauna of intertidal and subtidal sediments in Waitemata Harbour and the nearby Tamaki Estuary (Hayward et al 1997), specimens were found in only seven of the >200 sled tows in the present study, at sandy or shelly-gravel sites in the upper and outer harbour (Figure 112). During the four previous surveys of Waitemata Harbour (2002-2004), *M. senhousia* was found in a total of 4 sled tows (<1% of the total), over muddy subtidal and intertidal sediments between Orakei Basin and Point Chevalier in the Summer 1 and Summer 2 surveys (April 2003 and April 2004, respectively). The high fecundity, rapid growth and short life span of this species mean that its distribution and abundance is notoriously patchy in space and time (Crooks 1996, Creese et al. 1997).

In Whangarei Harbour, *Musculista senhousia* occurred in 17 of the 99 sled tows, over sediments ranging from shelly sand, muddy sand to mud in the upper and middle harbour, including the Portland Arm (Figure 111). It was also found in each of the 4 previous surveys, and occurred in a broad range of environmental conditions from the Town Basin Marina to One Tree Point and McLeod Bay. In previous and present surveys it was most abundant in muddy sediments near Limestone Island, Portland reach and the Town Basin, but also occurred in muddy sands, and shell grit that contained little mud. Large numbers were also caught in sled tows from muddy sediments within Parua Bay in previous surveys but not in the present survey. *M. senhousia* was first reported from Whangarei Harbour in 1985, when dead shells were washed ashore at Manganese Point. In 1986 a large living population was recorded near Takahiwai, in the main shipping channel (Willan 1987).

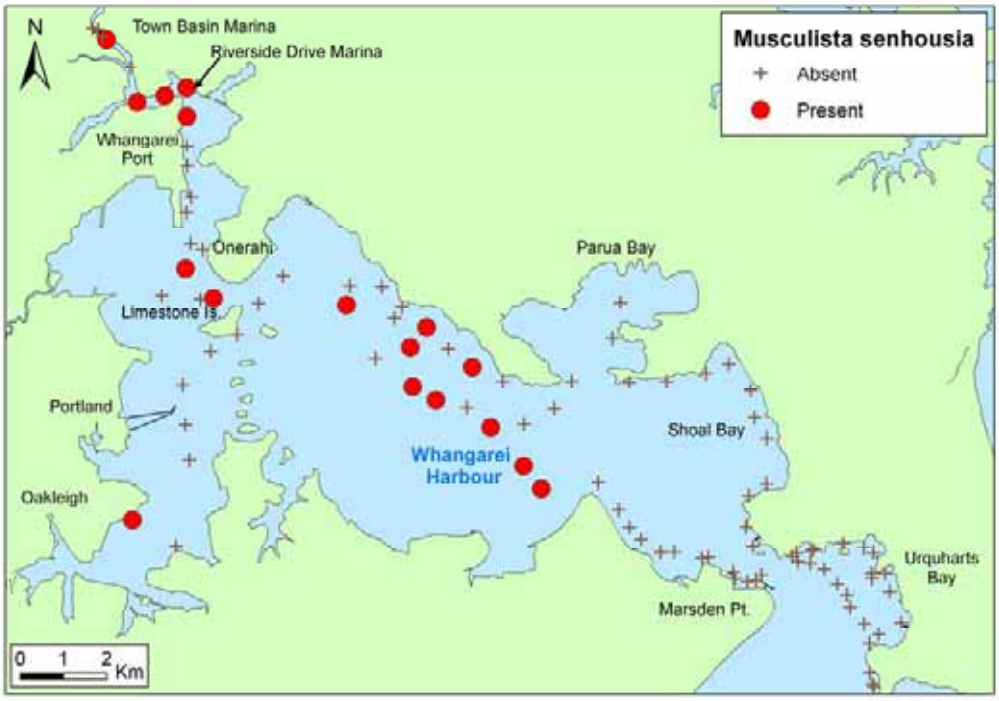

Figure 111 Distribution of *Musculista senhousia* in Whangarei Harbour.

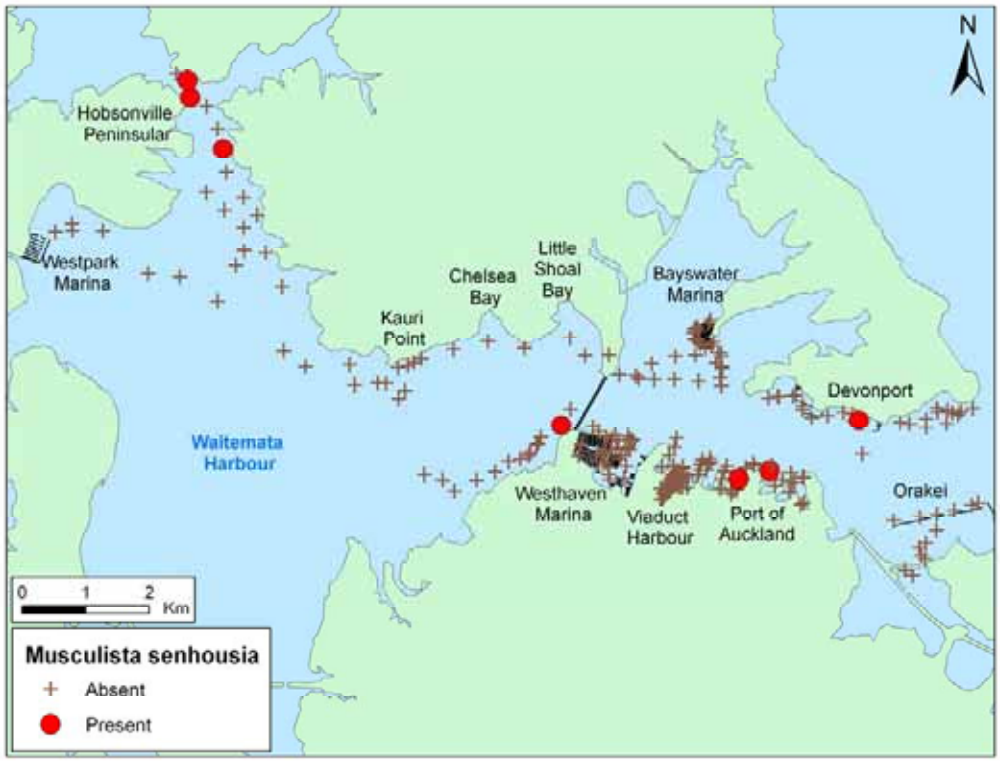

Figure 112 Distribution of *Musculista senhousia* in Waitemata Harbour.

Theora lubrica

The introduced semelid bivalve *Theora lubrica* occurred in large densities in Whangarei (Figure 113: maps show records from sleds), Waitemata (Figure 114), Tauranga (Figure 115), Wellington (Figure 116), Havelock (Figure 117), Nelson (Figure 118) and Lyttelton harbours (Figure 119) in the present survey. It was also less abundantly present in Picton Port (Figure 120) and Waikawa Marina (Figure 121). *T. lubrica* had not been reported from Tauranga in previous target-species surveys (2002-2004) but was known to occur there (Roper 1990). A native of Japan, *T. lubrica* was first recorded in New Zealand in 1972 (Hayward 1997). During the surveys it was recorded predominantly from fine-muddy, subtidal sediments. Epibenthic sled tows from these environments often collected many hundreds of individuals. Although it was widely distributed in each of the harbours that it was recorded from, greatest densities and occurrences were often in fine muds within the immediate vicinity of the commercial ports (see Figures 113-121).

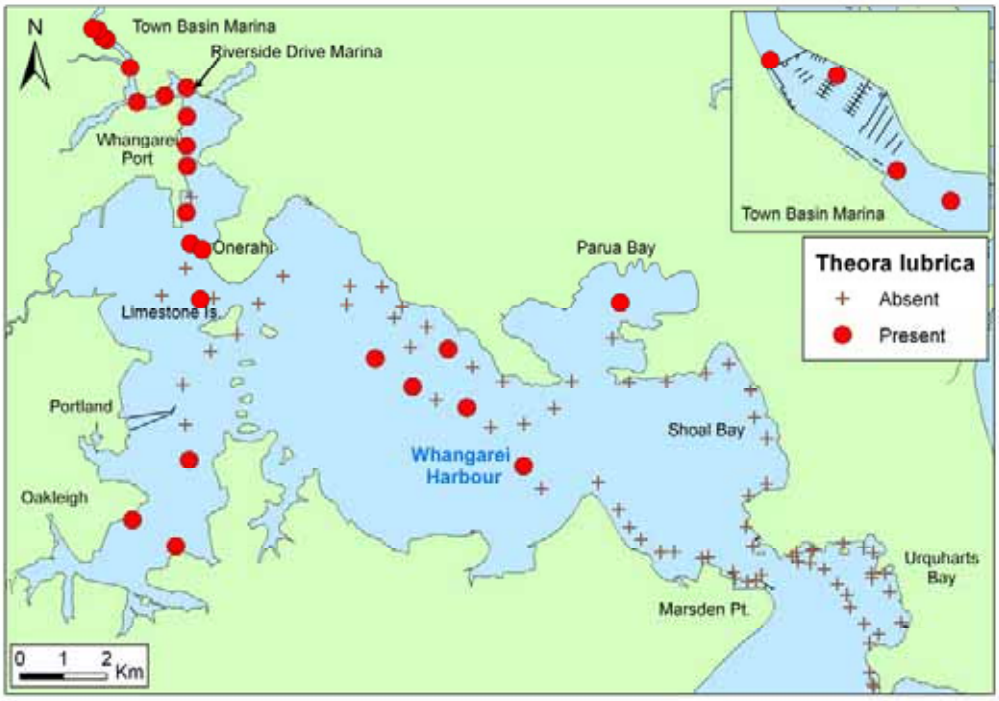

Figure 113 Distribution of *Theora lubrica* in Whangarei Harbour.

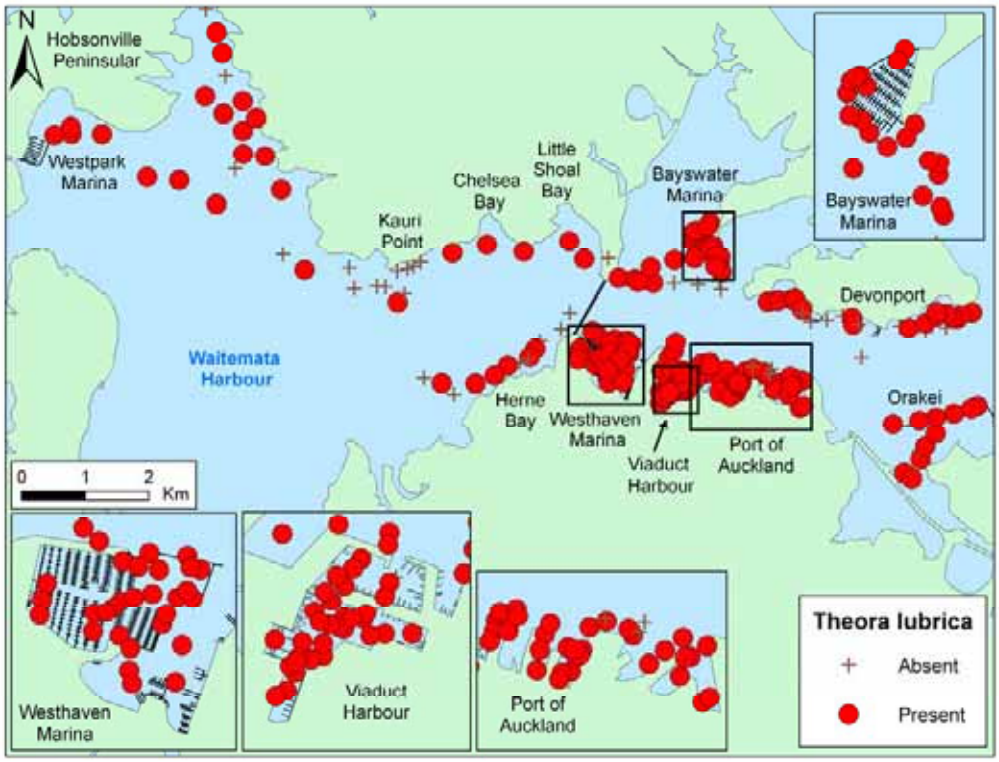

Figure 114 Distribution of *Theora lubrica* in Waitemata Harbour.

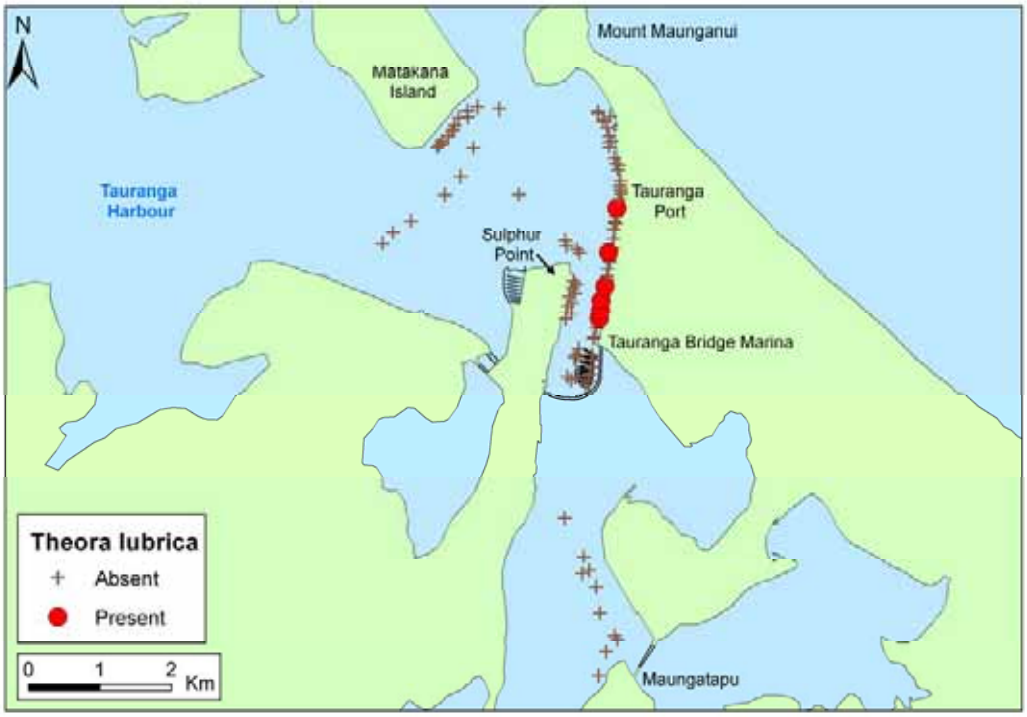

Figure 115 Distribution of *Theora lubrica* in Tauranga Harbour.

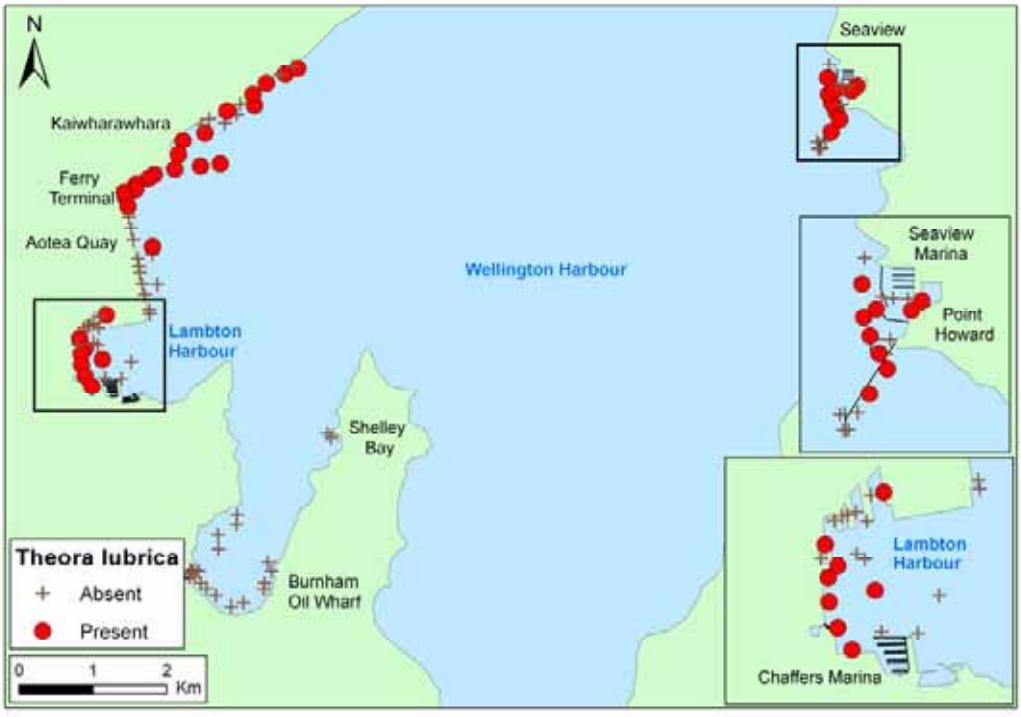

Figure 116 Distribution of *Theora lubrica* in Wellington Harbour.

Figure 117 **Distribution of *Theora lubrica* in Havelock Wharf and Marina.**

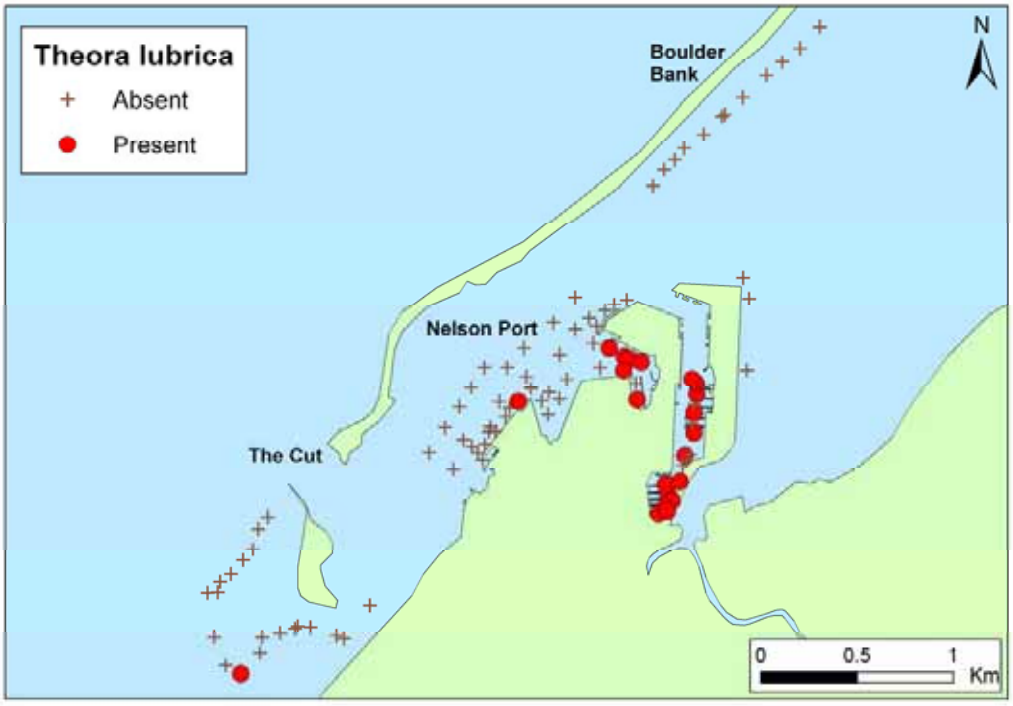

Figure 118 **Distribution of *Theora lubrica* around Nelson.**

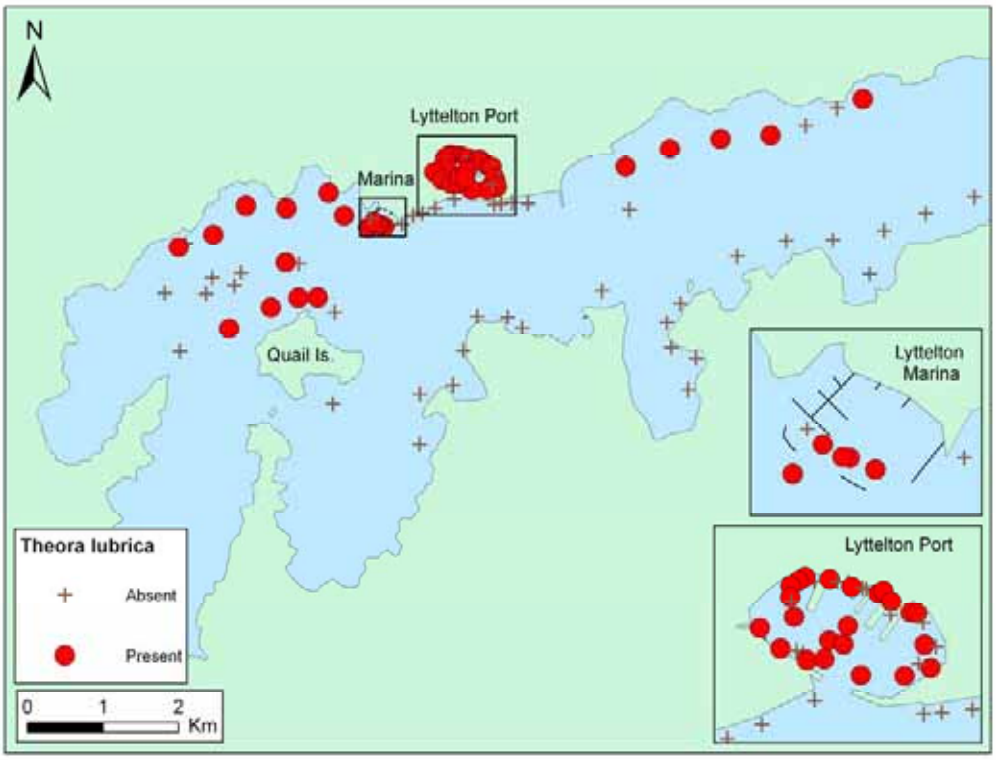

Figure 119 Distribution of *Theora lubrica* in Lyttelton Harbour.

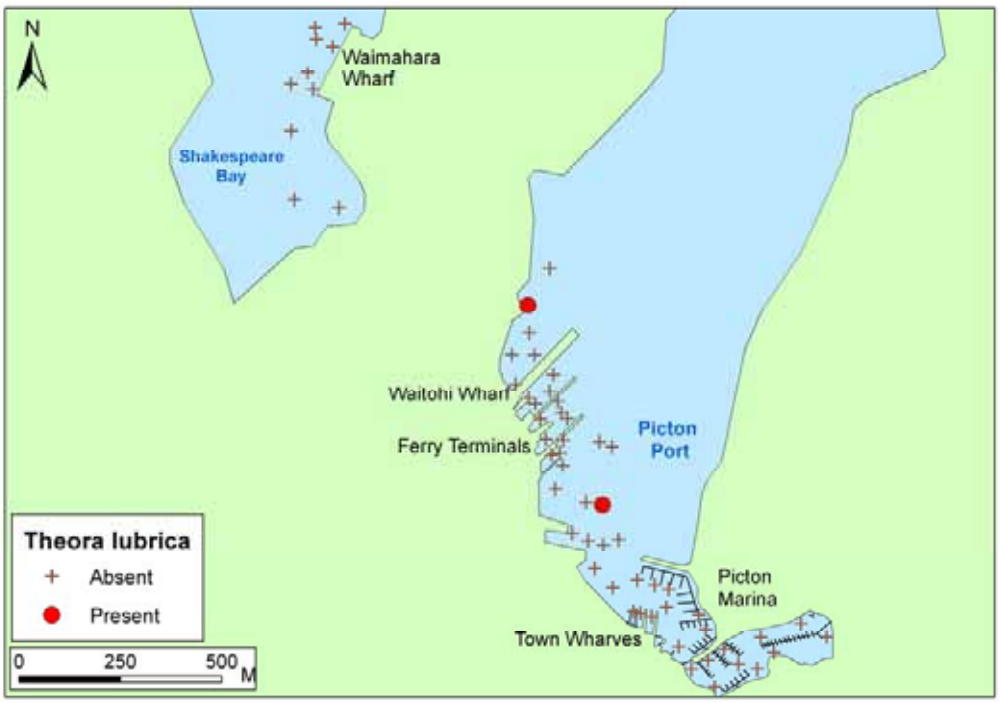

Figure 120 Distribution of *Theora lubrica* in Picton Harbour.

Figure 121 Distribution of *Theora lubrica* in Waikawa Marina.

Charybdis japonica

The introduced portunid crab, *Charybdis japonica*, was captured throughout Waitemata Harbour during the present survey (Figure 122: map shows records from box and starfish traps and shore searches) and in each of the four previous surveys. In the present survey it was recorded around Westpark Marina and Hobsonville in the upper harbour, at several locations in the middle harbour between Kauri Point and the Harbour Bridge, and in Bayswater and Westhaven Marinas, the port area and Orakei/Hobson Bay in the outer harbour.

Although it was widely distributed throughout Waitemata Harbour in the previous studies, the mean number of crabs caught per trap declined over the course of the studies (see Inglis et al. 2005a for details). In the first survey of this population initiated by NIWA in April 2002, *C. japonica* occurred at 60% of the locations trapped, at an average of 0.479 (± 0.046 S.E.) crabs per trap (Table 6: Gust et al. 2003, Gust & Inglis 2006). By September 2004, the average catch per unit effort had dropped to 0.079 (± 0.019) crabs per trap and *C. japonica* was captured at 20% of trapped locations. Size-frequency distributions of the crabs over this time comprised a broad, single cohort of animals whose modal size changed little between surveys. There was no evidence of younger year-classes recruiting into the trapped population over the 2 years that the study was in place. Shoreline searches of intertidal reefs (the preferred habitat of juveniles) also did not recover any juvenile *C. japonica*. In the present survey, the mean number of crabs caught per trap had increased to 0.169 (± 0.027) from the value in September 2004, and *C. japonica* was present at just over 30% of the 158 locations trapped throughout the harbour (Table 6). Analyses of the size-frequency data were not available at the time of writing.

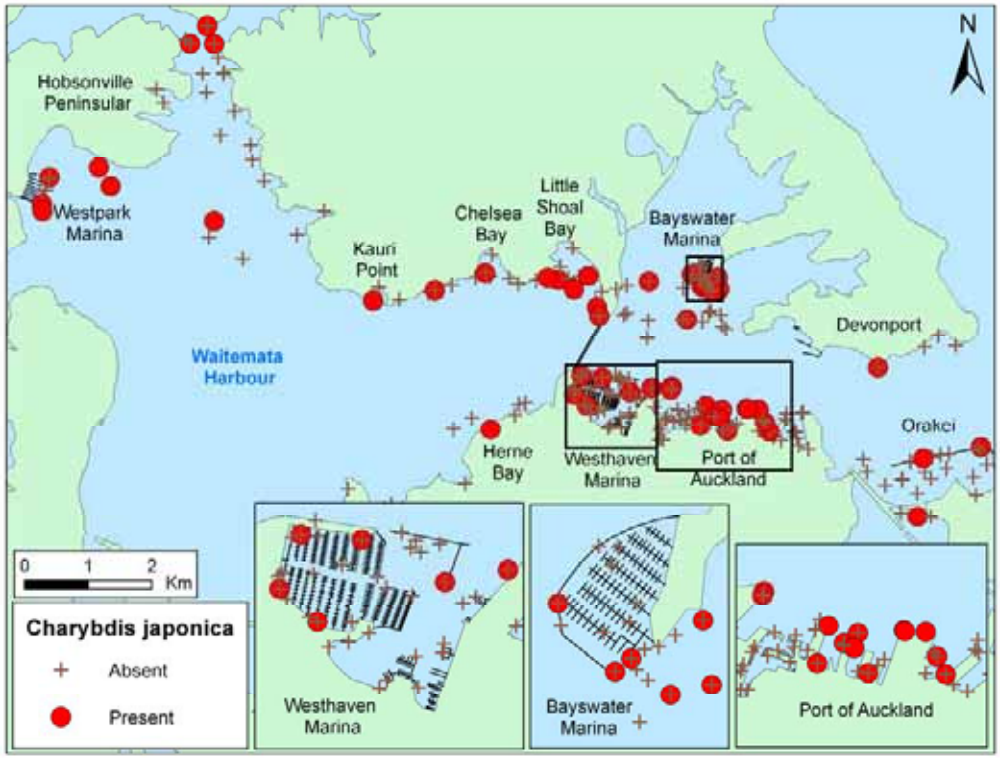

Figure 122 Distribution of *Charybdis japonica* in Waitemata Harbour.

During the September-October 2003 survey of Whangarei Harbour, a single, mature female *Charybdis japonica* was captured in a starfish trap at the Main Wharf at Port Whangarei (Inglis et al. 2005a). This was the first record of *C. japonica* outside the central Hauraki Gulf and represented a significant range extension from the Waitemata Harbour population ~140 km south. No further specimens were recovered during that survey, despite deployment of more than 180 baited traps at 69 locations throughout Whangarei Harbour. Subsequent surveys in March and September 2004 trapped a total of 136 locations throughout Whangarei Harbour (285 box trap hauls and 82 starfish trap hauls), but did not recover any additional specimens. In April 2004, trapping surveys and shoreline searches for *C. japonica* in the Whangarei region were extended to four smaller estuaries (Ngunguru, Pataua, Ruakaka and Mangawhai) within 100 km of Whangarei Harbour (Research Project ZBS2001/01 – Objective 3). A total of 55 locations (165 box trap hauls) were trapped in these estuaries, but again, no additional specimens were recovered (Inglis et al. 2004). This finding was reinforced by discussions with commercial crab fishermen in the region, none of whom had recovered *C. japonica* from their catches (Inglis et al. 2004).

Because of its size, location of capture, and the absence of other animals in Whangarei Harbour or surrounding estuaries, the specimen captured in September 2003 does not appear to have recruited through natural planktonic dispersal, but may have been transported to Whangarei on shipping from the population in Waitemata Harbour. *Charybdis japonica* was not found in any of the other six harbours surveyed in this project. No specimens were collected in Whangarei Harbour during the present survey (during which a total of 158 locations were sampled with baited traps), supporting the suggestion that the individual found in 2003 was transported to Whangarei as an adult.

Pyromaia tuberculata

The introduced majid crab *Pyromaia tuberculata* was recorded from Whangarei Harbour and Waitemata Harbour (Figure 123: map shows records from sleds) during the present survey. In Whangarei Harbour the single specimen found was probably collected in a crab trap deployed at Marsden Point (the specimen was found in the boat and is likely to have fallen through the mesh of the trap). In the Waitemata Harbour a specimen was collected in a sled sample east of the Harbour Bridge.

Pyromaia tuberculata has been present in the Hauraki Gulf and Firth of Thames since 1975 (Cranfield et al. 1998). During the Summer 1 target-species survey (November 2002) and the port baseline survey (ZBS2000/04, also carried out in November 2002) specimens were collected in Whangarei Harbour, representing the first known records of this species outside these two areas (Inglis et al. 2005a). More than 40 specimens were subsequently recorded from traps and sled samples taken during the Winter 1

survey of Whangarei Harbour (September 2003), two specimens from the Summer 2 survey (March 2004), and an additional eight specimens were captured in the Winter 2 survey (October 2004). These were mostly located in muddy sands in the mid-reaches of the harbour, near Limestone Island and Manganese Point. In Waitemata Harbour a total of six individuals was recorded from sled and trap samples during the Summer 1 (April 2003) and Winter 1 (October 2003) surveys.

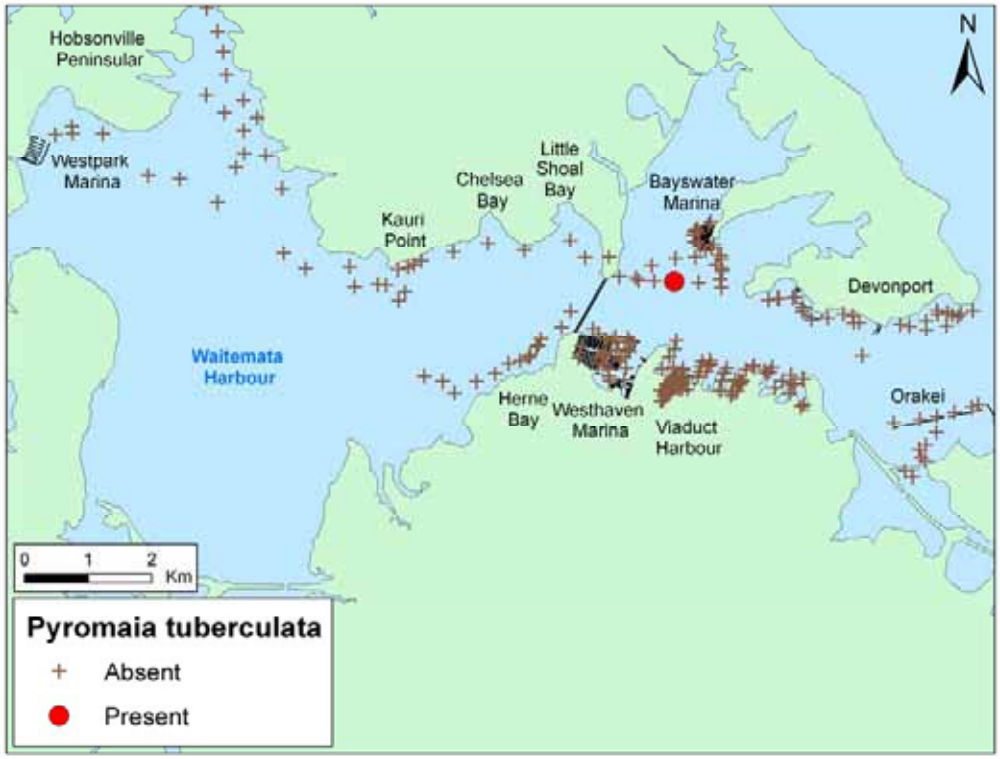

Figure 123 Distribution of *Pyromaia tuberculata* in Waitemata Harbour.

3.2.4 Opportunistic identifications

A number of other non-indigenous species were sampled opportunistically (Table 5).

Codium fragile* ssp. *tomentosoides

Codium fragile ssp. *tomentosoides* was collected in the Waitemata and Tauranga harbours (Figures 124 and 125: maps show records from dive searches and shore searches). In the Waitemata Harbour it was collected during visual searches of the fuel barge in Bayswater Marina and on the shore at 2 bays in the middle harbour. In Tauranga Harbour specimens were collected in a crab trap in the upper harbour and a sled tow over shelly gravel off Matakana Island at the southern entrance to the harbour. It was also recorded during the baseline port survey of Tauranga Harbour in March 2002. It is a large, dichotomously branching green alga that can attain almost 1 m in length and weigh up to 3.5 kg. This *C. fragile* subspecies probably originated in east Asia, including Japan, and is regarded as one of the most invasive of all algal species (NIMPIS 2002a). It is utilised as a food in some Asian countries. It has spread to Africa, the northeast and northwest Atlantic, North and South America, the Mediterranean, eastern Pacific, Australia and New Zealand. There is some evidence that this subspecies can prevent the re-establishment of native algal species, but not competitively exclude them. It is found in a wide variety of areas, from very protected through to intermediately wave exposed in both intertidal and subtidal habitats. It grows profusely on any hard substrate including rocky reef, boulders, cobbles, shellfish, wharf pylons and marine farming equipment. This species has wide environmental tolerances (estuarine to fully marine), occurs in intertidal and subtidal habitats, growing profusely over natural and artificial substrates.

Hydroclathrus clathratus

The introduced alga *Hydroclathrus clathratus* was collected in a sled tow over sandy mud in Urquharts Bay near the entrance to Whangarei Harbour. It is a cosmopolitan species found in tropical and warm-temperate waters (Johnson & Dromgoole 1977). It has been present in New Zealand since at least 1974 and has been recorded from the Kermadec Islands, Northland (Whangarei Harbour, Motukaroro Island, Whangaroa Harbour and Pekapeka Bay) and the Auckland region (Johnson & Dromgoole 1977, Cranfield et al. 1998). It has also been reported Port Phillip Bay, Victoria (Johnson & Dromgoole 1977).

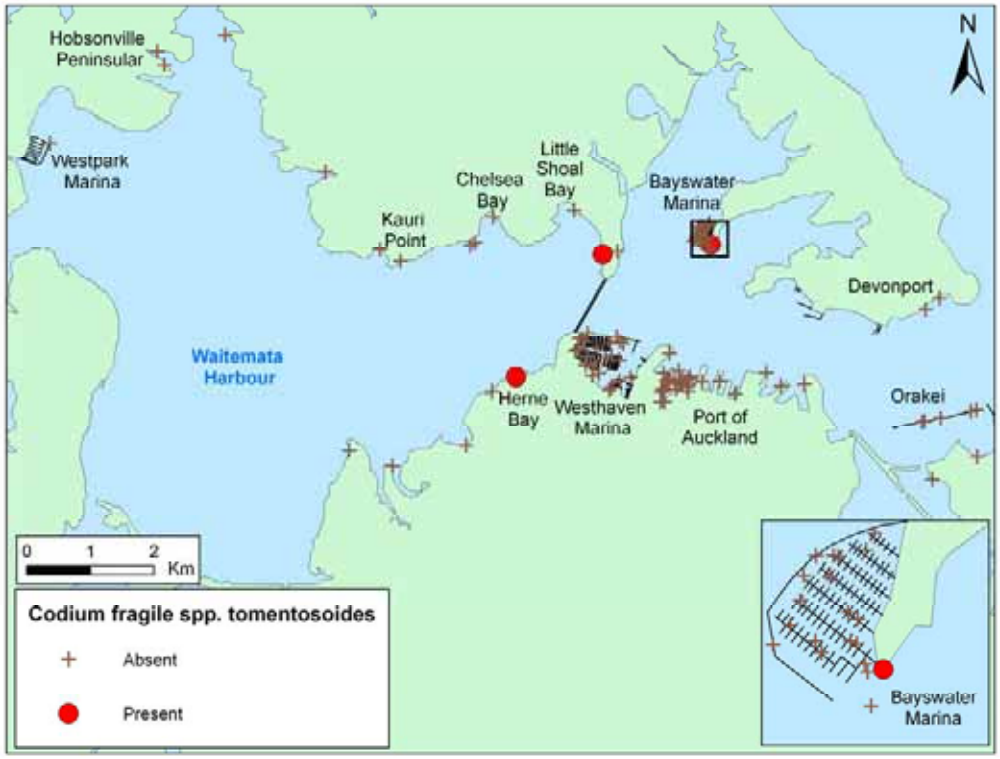

Figure 124 **Distribution of *Codium fragile* ssp. *tomentosoides* in Waitemata Harbour.**

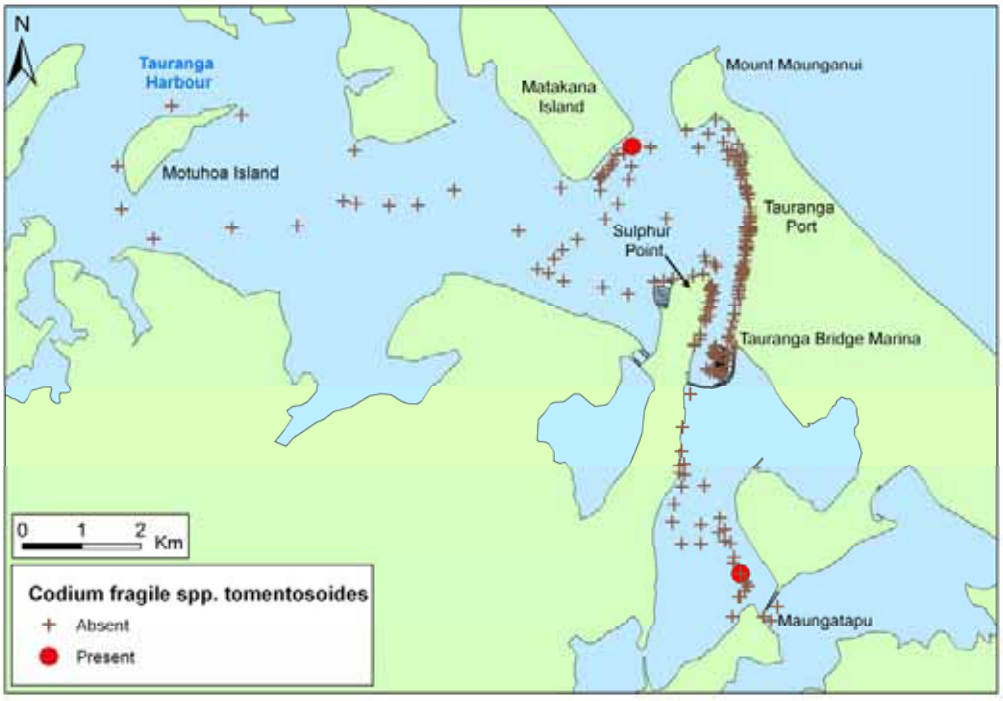

Figure 125 **Distribution of *Codium fragile* ssp. *tomentosoides* in Tauranga Harbour.**

Ectopleura larynx

The introduced colonial hydroid *Ectopleura larynx* grows up to 5 cm tall. It was collected in Tauranga Port (Mount Maunganui side) during one of the dive searches in the present survey. It has previously been recorded in Wellington and Otago Harbours (Ralph 1953, as *Tubularia larynx*). The species is probably native to the Atlantic coast of Europe, northeast and northwest America and northeast Asia (the type locality is the British Isles) but it has been widely distributed, probably by shipping (Schuchert 1996).

Pennaria disticha

The introduced hydroid *Pennaria disticha* forms large colonies up to 30 cm tall on artificial and natural hard substrates where there is some water movement. It is a very common fouling organism in harbours and commonly found on reefs usually in more protected areas or in cracks and crevices (Eldredge & Smith 2001). The native range of *P. disticha* is thought to be the northeast Atlantic, but it now also occurs in tropical and subtropical seas around the world (Cranfield et al. 1998). Its impacts on native organisms are unknown. In the present survey it was found by dive searches at the Marsden Wharf, Whangarei Port (Town Wharf) and Portland Wharf in Whangarei (Figure 126: map shows records from sleds and dive searches). It has been present in Waitemata Harbour since at least 1928 (Cranfield et al. 1998. During the port baseline surveys (ZBS2000/04) it was recorded in pile scrape samples taken from Auckland Port.

Branchiomma curtum

This cryptogenic, sabellid polychaete was recorded in a dive search in the inner part of Port Chalmers, Otago Harbour during the present study. It has also been collected during port baseline surveys (ZBS2000/04) of Lyttelton Port.

Ficopomatus enigmaticus

As in each of the previous surveys, divers reported encrustations of the tube-building serpulid polychaete *Ficopomatus enigmaticus* from the Whangarei Town Basin Marina during the present survey (Figure 127: map shows records from sleds and dive searches). Tubes were found on piles and pontoons at 5 of the 6 sites searched. It was not reported from the other eight harbours surveyed. *F. enigmaticus* causes nuisance growths of calcareous tubes on piles, pontoons and pleasure craft. It was first reported

from the Town Basin Marina in the summer of 1967/68 (Read and Gordon 1991) and has remained abundant in the inner harbour from the marina to Limeburners Creek. It is a habitat-forming species that has been found in association with the introduced bryozoan *Conopeum seurati* and the introduced spionid polychaete *Polydora cornuta* in the Whangarei Town Basin (Read and Gordon 1991). It has also been reported from Waitemata Harbour and Hawkes Bay (Cranfield et al. 1998).

Hydroides elegans

The introduced serpulid polychaete *Hydroides elegans* was recorded during dive searches of Nelson Marina during the present survey (Figure 128: map shows records from sleds and dive searches). It has also been recorded during port baseline surveys (ZBS2000/04) in Auckland Port, Gulf Harbour Marina and Nelson Port and has been reported from Lyttelton Harbour (Cranfield et al. 1998). *Hydroides elegans* is a small, tube dwelling polychaete worm that grows up to 20mm in length. It constructs hard, sinuous, calcareous tubes and is a fouling species on both natural and artificial structures. It is found subtidally and is highly tolerant of contaminated waters (NIMPIS 2002b). Although the type locality is Sydney Harbour, Australia, the native range of *H. elegans* is unknown as it may have been introduced to Australia prior to 1883 (Australian Faunal Directory 2005). Its present distribution includes in the Caribbean, Brazil, Argentina, northwest Europe, the Mediterranean, northwest and southeast Africa, Japan and New Zealand. It is able to grow in high densities, particularly in tropical and sub-tropical ports, sometimes heavily fouling any newly immersed structure. It creates microhabitat for some species and competes with others for food and space. In Japan it has reportedly had adverse effects on farmed oysters (NIMPIS 2002b). *H. elegans* has been present in New Zealand since at least 1952 (Cranfield et al. 1998).

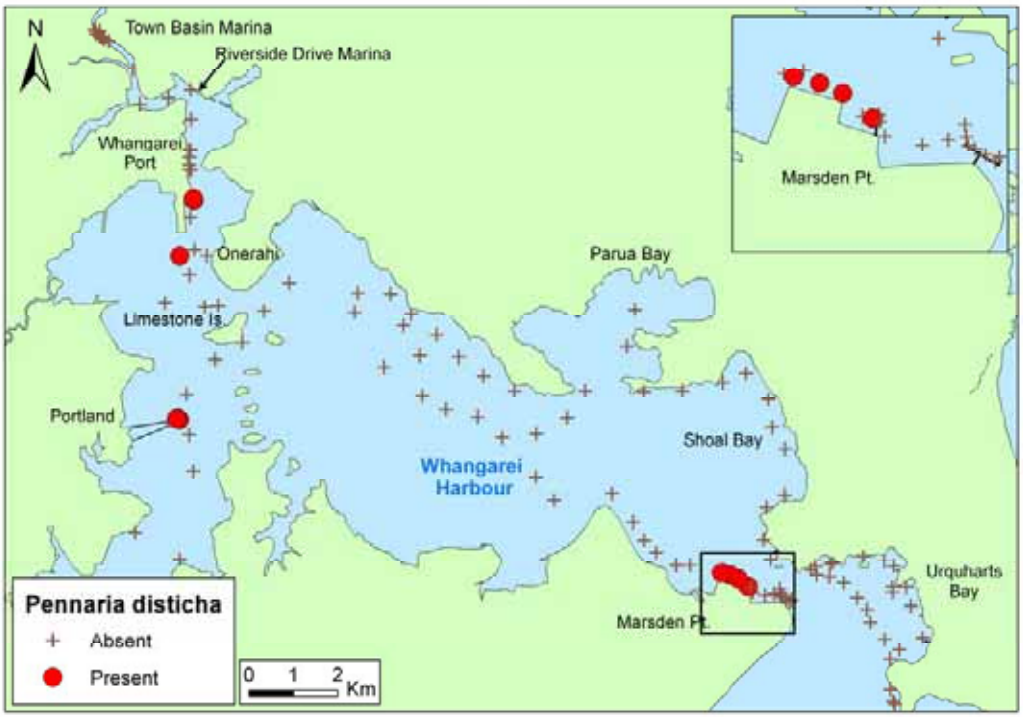

Figure 126 Distribution of *Pennaria disticha* in Whangarei Harbour.

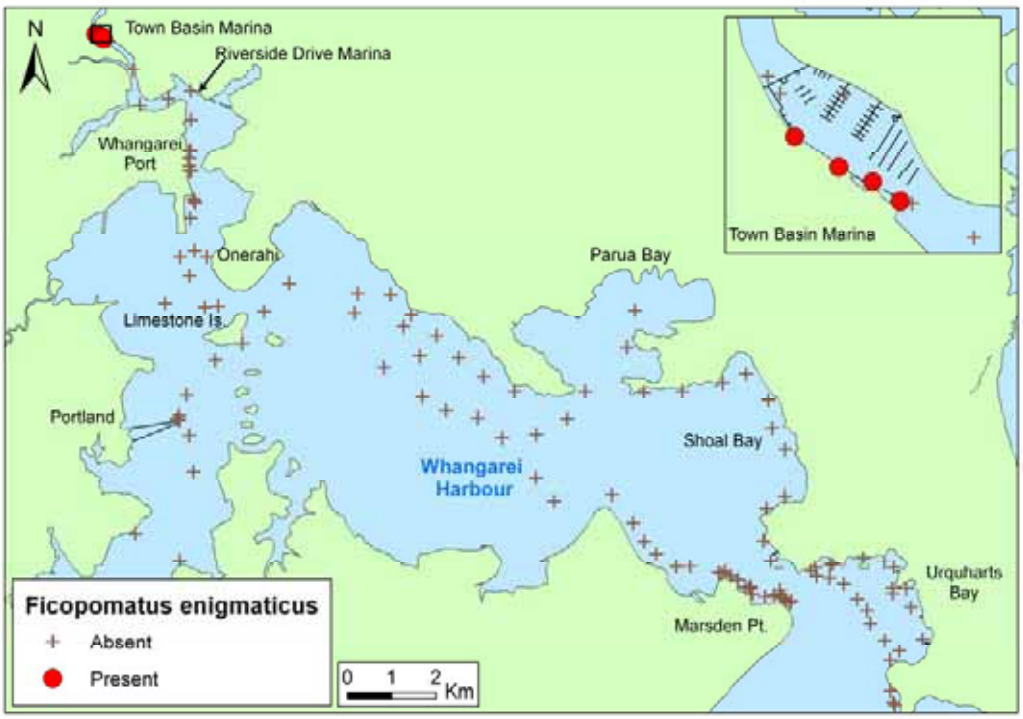

Figure 127 Distribution of *Ficopomatus enigmaticus* in Whangarei Harbour.

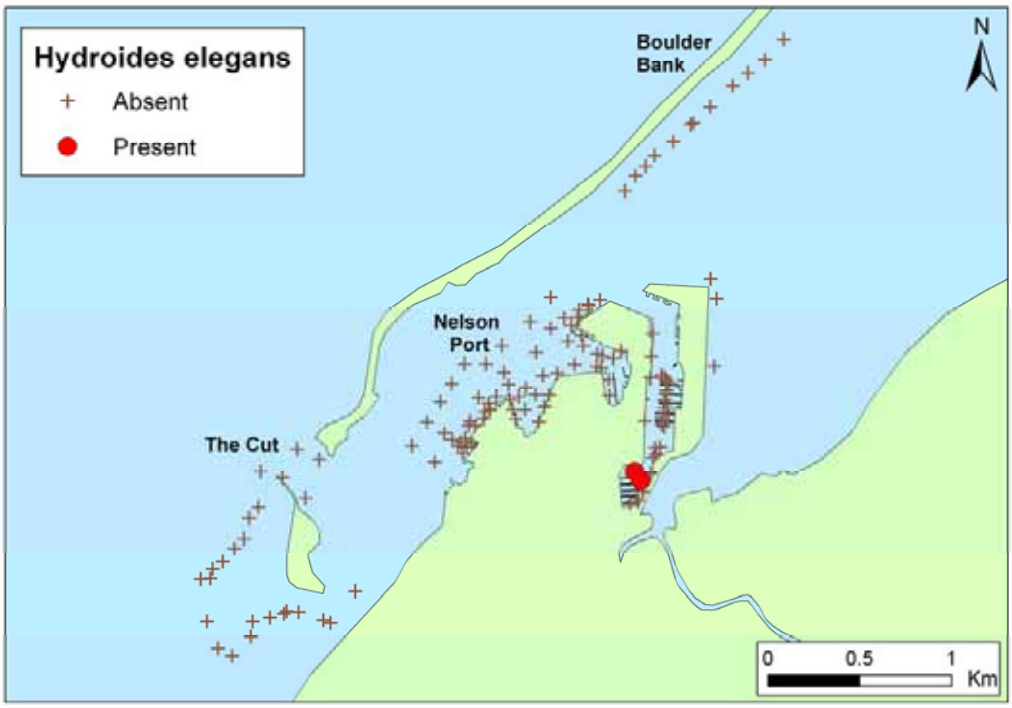

Figure 128 Distribution of *Hydroides elegans* around Nelson Marina.

Limaria orientalis

The small Indo-Pacific bivalve *Limaria orientalis* was recorded from shelly gravel in the upper and middle Waitemata Harbour (Figure 129: map shows records from sleds, shore searches, box and starfish traps) and in Picton Harbour (a single specimen was found in a sled tow from the inner part of Picton Marina). It was widespread in the Waitemata Harbour, from the upper harbour, off Hobsonville, to the port area. In previous target-species surveys it has also been collected from Whangarei Harbour, where 18 specimens were recovered from four sled tows taken over sandy shell and bryozoan gravels near McLeod's Bay and Urquharts Bay during the Summer 1 and Summer 2 surveys (November 2002 and March 2004, respectively). In Waitemata Harbour, three specimens were recorded from a single sled tow near the Bledisloe Terminal in the commercial port during the Winter 1 survey (October 2003). In Wellington Harbour, a single specimen was recovered from a sled sample taken in Evans Bay during the Summer 1 survey (December 2002).

Limaria orientalis was first recorded in New Zealand in 1972 from the Hauraki Gulf and Waitemata Harbour. It has since been recorded from the Bay of Islands and Coromandel (Cranfield et al. 1998) and is common around mussel farms in the Marlborough Sounds (NIWA, unpublished data). Although it does not reach the same densities as *Musculista senhousia* and *Theora lubrica*, *L. orientalis* can be a dominant member of benthic assemblages in muddy shell gravels (Hayward 1997).

Austromegabalanus psittacus

A clump of 9 individuals of the barnacle *Austromegabalanus psittacus* was collected by sled from shelly gravel at a depth of 10 m in the inner port area of Wellington Harbour (Figure 130: maps show records from sleds, dive searches and shore searches). This barnacle, which grows to 20 cm high and 8 cm diameter, is native to South America from the Pacific coast of Peru to Patagonia on the Atlantic coast. This is the first record of the species outside its native range. The specimens were probably attached to a rock or other hard material. Although it is possible that they were knocked off the hull of a vessel, the shape of the underside of the specimens indicates that this is unlikely (Hosie & Ahyong, 2006).

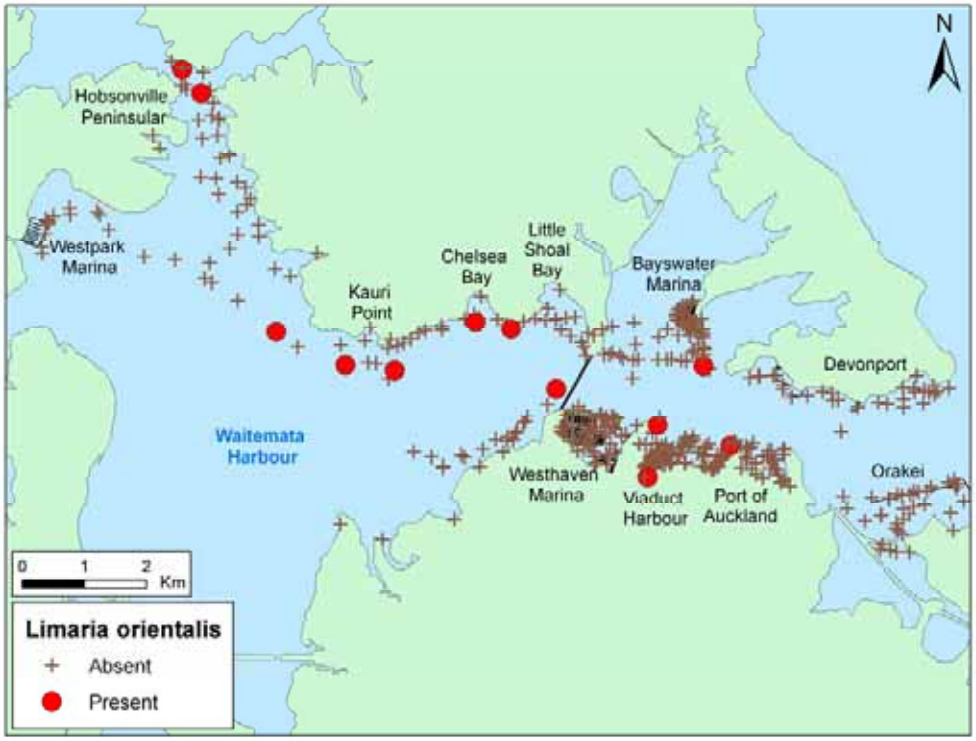

Figure 129 Distribution of *Limaria orientalis* in Waitemata Harbour.

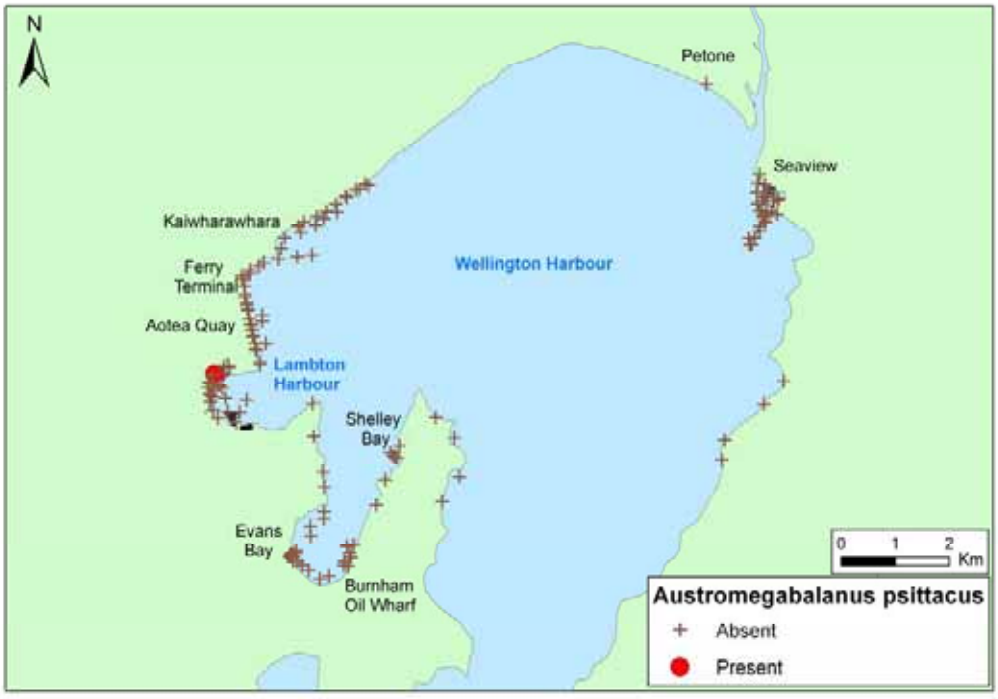

Figure 130 Distribution of *Austromegabalanus psittacus* in Wellington Harbour.

Aplidium phortax

The cryptogenic, colonial ascidian *Aplidium phortax* was collected during visual searches of marina pontoons in the Viaduct Harbour, Auckland in the present study. It was collected during port baseline surveys (ZBS2000/04) of Whangarei, Tauranga, Auckland, Gulf Harbour, Gisborne, Napier, Wellington, Lyttelton, Timaru, Dunedin and Bluff. It has also been reported from D'Urville Island, Stewart Island and the Chatham Islands (Miller 1982). The species is probably native to northeast Australia and the Solomon Islands (Cranfield et al. 1998).

Ciona intestinalis

The introduced ascidian *Ciona intestinalis* was recorded during dive searches of pontoons in Nelson Marina and a sled tow adjacent to Number 6 Wharf in Lyttelton Port in the present study. It has also been recorded during port baseline surveys (ZBS2000/04) of Napier, Nelson, Lyttelton and Timaru.

Ciona intestinalis occurs on stones, piles, algae and other hard substrates from the lower shore to depths of at least 500 m (Millar 1970). The first published record of the species in New Zealand was from wharf piles in Lyttelton Harbour in 1950 (Millar 1982, Cranfield et al. 1998). It is probably native to Europe (Cranfield et al. 1998) but is now widely distributed in temperate, warm and cool waters around the world (Millar 1982). The body of *C. intestinalis* is cylindrical, soft and colourless to greenish, with yellow-rimmed siphons.

***Didemnum* sp.**

The colonial ascidian *Didemnum vexillum* (Kott, 2002) was first described from Whangamata Harbour in 2001, when it appeared suddenly and formed nuisance growths on ship's hulls, wharf piles and other submerged structures (Kott 2002). It has subsequently been reported from several other port environments in the Bay of Plenty and upper South Island (Port Nelson and Shakespeare Bay, Picton) and a local control programme has been trialled in the Marlborough Sounds to prevent its spread to aquaculture sites (Coutts 2002). The appearance of *D. vexillum* in New Zealand was followed closely by reports of other nuisance species in this genus from the Atlantic coast of the USA, Mediterranean, North Sea and English Channel, but these now appear to be different species. Kott (2004) concluded that, "since it [*D. vexillum*] is only known from New Zealand and, in the absence of data to the contrary, the simplest explanation of its occurrence is that it is indigenous". No further, contrary evidence has emerged since Kott's (2004) paper (Mike Page, NIWA, pers. comm.).

In this survey, species of *Didemnum* with colonial growth forms resembling those of *D. vexillum* were observed by divers in Whangarei Harbour (Marsden Wharf), Picton (Waimahara Wharf in Shakespeare Bay) and the Main Wharf in Nelson Port. In previous target-species surveys (2002-2004) it was also recorded at Waitemata Harbour, Tauranga Harbour and at Port Chalmers and the Ravensbourne terminal in Otago Harbour. The specific identity of these colonies was not confirmed, since formal identification of *Didemnum* species requires dissection and microscopic examination (Kott 2004), which was beyond the scope of this study. In the associated baseline port survey project (ZBS2000/04), *D. vexillum* was recorded from the ports of Tauranga (March 2002) and Nelson (January 2002), while the related *D. incanum* was present in Bluff (March 2003), Picton (December 2001) and Tauranga (March 2002) (MAF Biosecurity New Zealand, unpubl. data). Unidentified species of *Didemnum* were recorded from Tauranga (March 2002 and April 2005), Wellington (November/December 2001 and February 2005), Picton (December 2001 and January 2005), Nelson (January 2002 and December 2004) and Timaru (February 2002 and November 2004).

Styela plicata

The solitary ascidian *Styela plicata* has a cosmopolitan distribution and is considered cryptogenic in New Zealand (NIMPIS 2002c), where the first published record was in 1948 (Cranfield et al. 1998). It grows up to 9 cm tall with an oval, opaque, white-tan body and "cobblestone" appearance created by deep grooves and rounded lumps (NIMPIS 2002c). It occurs from the low intertidal to 30 m in tropical to warm-temperate areas and is tolerant of salinity variation. It lives on hard substrata in protected embayments and harbours, and has been reported to foul aquaculture structures (NIMPIS 2002c). The native range of *S. plicata* appears to be unknown and it is cryptogenic on the west and northeast coasts of North America, the Caribbean, the Mediterranean and southeast Asia. It is presumed to have been introduced to the east coast of South America and west, south and northeast Australia (NIMPIS 2002c).

During the present survey *Styela plicata* was detected in a sled tow adjacent to Marsden Wharf in Whangarei Harbour and by visual searches of marina pontoons in the Viaduct Harbour, Hobson West Marina and Westhaven Marina, Auckland. It has previously been collected during port baseline surveys (ZBS2000/04) of Whangarei (November 2002), Opua Marina (November 2002), Gulf Harbour Marina (April 2003), Auckland (April 2003), Tauranga (March 2002), Taranaki (April 2002), Wellington (November/December 2002), Nelson (January 2002) and Lyttelton (March 2002).

Acentrogobius pflaumii

This introduced goby was collected in sled tows mud or sandy/shelly mud in the middle Waitemata Harbour (off Herne Bay), the Viaduct Harbour and Hobson West Marina during the present study (Figure 131: maps show records from sleds). A total of 4 individuals was collected for identification (and a further individual identified on site), and another 4 specimens were identified as belonging to the same genus but their specific identity could not be determined. Further specimens were noted in the field as probably belonging to the same species but were not retained (since the objective of these surveys was to determine presence/absence rather than abundances).

Although *Acentrogobius pflaumii* has not been reported in previous target-species surveys, it was first recorded in New Zealand in 4 locations the upper Waitemata Harbour (February 2001) and 2 locations in Whangapoua Harbour (Coromandel Peninsula, March-April 2002) (Francis et al. 2003). All specimens were taken by beach-seine tows over mud or sand. It is native to the northwest Pacific (Japan, Korea, Taiwan and the Philippines) but has been introduced to Australia, first appearing in Port Phillip Bay, Victoria in 1996. It has subsequently become abundant in that location, but its ecological impact is unknown (Francis et al. 2003). It has also been reported from Botany Bay and Sydney Harbour, New South Wales. Francis et al. (2003) suggested that *A. pflaumii* probably arrived in New Zealand in ships' ballast water.

Another introduced goby, *Arenigobius bifrenatus*, was collected during the March 2004 target-species survey of Whangarei (Inglis et al. 2005a) but was not recorded during the present survey. This species is native to Australia but has been recorded from 5 harbours on the east coast of the North Island, including Waitemata Harbour.

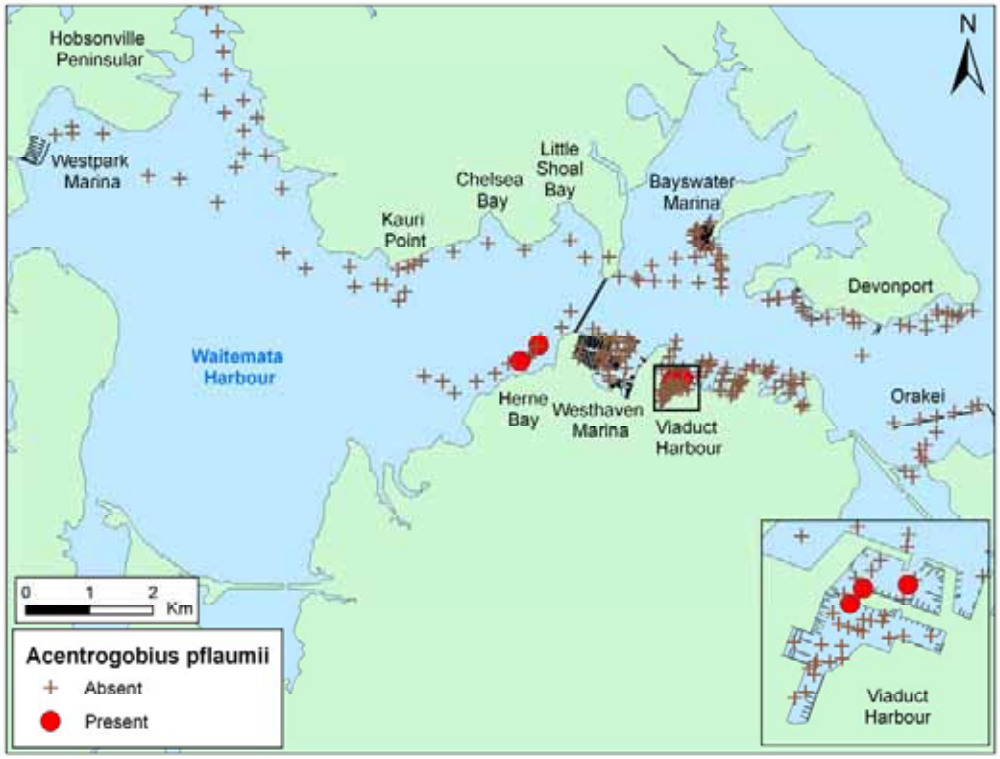

Figure 131 Distribution of *Acentrogobius pflaumii* in Waitemata Harbour.

4. Discussion

This report describes a survey for 8 species of unwanted, non-indigenous marine species in 9 ports around New Zealand. It continues the application of the methods for surveillance for unwanted marine species developed by Inglis et al. (2005a).

Two of the target species, the alga *Undaria pinnatifida* and the ascidian *Styela clava*, were detected. These detections confirmed earlier reports of the former in Waitemata, Taranaki, Wellington, Picton, Nelson, Lyttelton, Dunedin and Bluff harbours, and the latter in Waitemata and Lyttelton harbours. There were no new detections outside the ports where these 2 species were already known to occur, and no other target species were detected.

Previous surveys in this programme (ZBS2001/01) and the port baseline survey programme (ZBS2000/04), conducted in April and October 2003 and April and September 2004, did not detect *Undaria pinnatifida* in the Waitemata Harbour. The species was, however, opportunistically collected from this harbour in October 2004. A subsequent delimitation study (Stuart & McLary 2004) found it growing on floating structures in parts of the Viaduct Harbour, St Mary's Bay, Westhaven Marina, Wynyard Wharf, Quarantine Wharf and Freyberg Wharf. These locations overlapped with those of earlier shore and diver searches but *U. pinnatifida* was not detected at that time.

Inglis et al. (2005a), however, also identified a number of other factors that may have contributed to the earlier failure to detect this incursion by *Undaria pinnatifida*. First, probability of detection is likely to be low as a result of the large area of available, suitable habitat for the species relative to the area that could be searched within the cost and logistical constraints of the survey. Second, previous searches of locations where *U. pinnatifida* was subsequently found were conducted in late summer (March-April), when adult sporophytes are likely to be absent or in senescence (Forrest et al. 2000, Thompson 2004). Third, the visual search profile used by divers was not optimised to detect just *U. pinnatifida* but, rather, was a compromise to permit sampling of several target species while maintaining safe diving practices (Inglis et al. 2005a).

The fact that *Undaria pinnatifida* was detected in Waitemata Harbour (and Bluff Harbour, where previous target-species surveys had not reported it) during the present survey may be due to subsequent expansion of the range, or increase in population density, of the species in these harbours and/or the increased search effort allocated to diver and surface searches. This additional effort was focussed on fixed and floating

structures in port and marina areas because these are preferred habitats for *U. pinnatifida* and *Styela clava*.

Subsequent to the present survey of Nelson (and a rapid delimitation survey for *Styela clava* in November 2005: Gust et al. 2006a), three individuals of *S. clava* were found on two boats in the Slipway Basin in the inner harbour in July 2006 (Morrisey et al. 2006). Both vessels had been moored in the harbour for at least 2 years and the size of the individuals indicates that they would have been present at the time of the target-species survey. Although the Slipway Basin was visually searched from the surface during the present survey, there were no dive searches on either vessel nor in the area immediately around them. All 3 specimens were attached to the vessel hulls in positions that could not be seen from the surface. The inner Slipway Basin was intensively searched for *S. clava* in July 2006 but no other individuals were found (Morrisey et al. 2006). This suggests that the population of *S. clava* in the Slipway Basin is small and limited in its distribution. This combination of limited distribution and the relatively small area of suitable habitat (including moored vessels) searched by divers in relation to the total available area probably explains the failure to detect the species during previous surveys.

The present surveys confirmed the presence of a number of non-target, introduced species in several of the harbours. In the cases of *Theora lubrica* in Tauranga Harbour and *Acentrogobius pflaumii* in the Waitemata Harbour these were first records for the target-species surveillance programme (their presence in these harbours has been reported in other studies: Roper 1990, Francis et al. 2003). An introduced species not previously recorded in New Zealand, the barnacle *Austromegabalanus psittacus*, was recorded from Wellington Harbour.

Picton Harbour and Marina, Waikawa Marina, Waimahara Wharf (Shakespeare Bay) and Havelock Harbour and Marina were included in the target-species surveys for the first time. The survey confirmed the presence of *Undaria pinnatifida* in these locations except for Havelock Harbour and Marina (where the presence of a low-salinity layer in the upper water column [pers. obs.], derived from the inputs of the Pelorus and Kaituna Rivers, may prevent its colonisation or persistence). The survey also confirmed the presence of *Didemnum* sp. at Waimahara Wharf and identified the presence of *Theora lubrica* in Picton and Havelock and *Limaria orientalis* in Picton.

The sampling approach taken in the target-species surveys targets sampling effort on areas of the surveyed harbours that are at relatively high risk of containing the target species (Inglis et al. 2005a). These areas were identified by combining spatially explicit descriptions of the species' habitat associations with simulations of likely patterns of establishment in local harbours. As Inglis et al. (2005a) discussed, the results of the previous surveys (2002-2004) showed that the approach had promise,

but also highlighted a number of significant impediments to achieving cost-efficient and sensitive surveillance in these environments. One of the most important of these impediments is the general lack of suitable environmental information from which to identify suitable habitats. These can then be used to make more specific predictions of suitability, using qualitative models (Inglis et al. 2006b).

Although the present survey detected both of the target species already present in New Zealand (*Undaria pinnatifida* and *Styela clava*) in those harbours where they are known to occur, there remain problems of lack of sensitivity of the methods used. Inglis et al. (2005a, 2006b) identified the difficulty of achieving sufficiently high probabilities of detection of the target species that would allow effective, early management of new incursions. Low probabilities of detection are likely when the areas of suitable habitat are large relative to the areas that can be searched at acceptable cost. Despite increased levels of replication for 2 of the sampling methods (diver and shore searches) during the present survey, these limitations remain.

To quote the concluding paragraphs of the previous target-species surveillance report (Inglis et al. 2005a), "public confidence in, and continued funding for, surveillance requires that it be efficient, with a high likelihood of detecting nuisance species. For this to be achieved, more critical evaluation is needed of the technical requirements for surveillance in marine environments and, in particular, more efficient survey techniques are required to locate and identify unwanted species. The approach we have outlined in this report for prioritising sampling within habitats most at risk of incursion by the target species provides a rational way of defining the principal search area and deciding on the appropriate allocation of sampling effort and resources needed to achieve early detection within them. In cases where the search area is large, it may be unreasonable to expect a high probability of detection at an early enough stage for eradication. Nevertheless, as the field surveys show, even with low statistical detection probabilities, there are significant benefits from regular surveillance in detecting both target species and unexpected new arrivals".

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Table 2 Description of sampling methods used in the targeted surveillance programme.

Sampling method	Method of deployment	Habitat	*Target species*	Additional data recorded
Epibenthic sled tows	Deploy the sled with sufficient line to allow the chain bridle to lay flat. Motor forward slowly pulling the sled for a fixed time of 2 minutes. Retrieve the sled and wash contents in the sled mesh. Empty remaining material onto 1 mm sieve, wash and examine for target species. Because of its small size, particular attention should be made for *Potamocorbula*. Any similar (e.g. *Corbula gibba*) or suspicious specimens should be labelled and kept for identification.	Subtidal soft-sediments Particular focus should be made on known shellfish beds (for *Asterias*) and areas next to public access (e.g. wharves, boat ramps, marinas, etc *Caulerpa, Sabella*)	*Asterias amurensis* *Potamocorbula amurensis* *Caulerpa taxifolia* *Sabella spallanzanii*	Record sediment type recovered in the sled (see guide) and any other noticeable habitat features (e.g. accumulations of drift algae). Record easily identifiable large species (e.g. 11-armed seastar, *Patiriella*, crabs, etc). .
Starfish traps	Bait with 2-3 partially crushed pilchards. 2 traps per line ~ 5 m apart. Deploy overnight with a marked buoy to mark its position, away from shipping lanes.	Adjacent to wharf pilings & other artificial habitats	*Asterias amurensis*	Record easily identifiable large species (e.g. 11-armed seastar, *Patiriella*, crabs, etc). .
Box crab traps	Bait each trap with 1-2 partially crushed pilchards. 3 traps per line~ 5 m apart. Line needs to be weighted and marked by a single buoy. Deploy overnight with a buoy to mark its position, away from shipping lanes.	Intertidal and shallow subtidal rocky shores, breakwalls and saltmarsh, particularly habitats with complex physical structure (e.g. mussel beds, seagrass beds)	*Carcinus maenas* *Eriocheir sinensis* *Charybdis japonica*	Record easily identifiable large species (e.g. 11-armed seastar, *Patiriella*, crabs, etc).
Crab condos	These traps are not baited, they are artificial shelters for juveniles. Clip 3 traps per line. Line weighted and marked by a single buoy. Deploy for a minimum of 3 nights.	Intertidal and shallow subtidal banks of rivers, particularly brackish water habitats with complex structure (e.g. saltmarsh or fringing riparian vegetation)	*Eriocheir sinensis* *Carcinus maenas* *Charybdis japonica*	Record easily identifiable large species (e.g. 11-armed seastar, *Patiriella*, crabs, etc).

Table 2 Continued.

Sampling method	Method of deployment	Habitat	*Target species*	Additional data recorded
Shoreline searches	Walk along the driftline at low tide for 10 min. Examine cast algal material for algae (*Undaria* and *Caulerpa*) and exuviae of moulted crabs (*Carcinus, Eriocheir, Charybdis*). In areas of rocky reef, scan shoreline at low tide for *Undaria* sporophytes and overturn cobbles to find juvenile crabs.	Sloping sandy shorelines and areas where drift material is likely to accumulate (note that prevailing winds on the preceding days are a useful guide to where material may accumulate) Intertidal rocky reefs	*Eriocheir sinensis* *Carcinus maenas* *Charybdis japonica* *Undaria pinnatifida* *Caulerpa taxifolia*	
Diver searches	At each station in ports or marinas, divers are to search 10 pilings on SCUBA for the target species. Searches are done by two divers swimming at depths of approximately 2 m and 4 m along the row of pilings or equivalent area of rock wall or rip-rap. Divers should retrieve specimens of any organisms that resemble the target species.	Wharf piles, marina piles and floating pontoons Intertidal and shallow subtidal reefs	*Carcinus maenas* *Undaria pinnatifida* *Asterias amurensis* *Sabella spallanzanii*	Record secchi disc depth

Table 3a **Visual classification scheme for sediments retrieved in the benthic sled (after Tricklebank et al. 2001) (≤ 63 µm = muds, silts & clays; ≥ 125 µm and < 2 mm = sands; ≥ 2 mm = gravel & coarse shell).**

Code	Sediment category	% Mud	% Sand	% Gravel	Notes
1	Sandy mud	> 50%	< 50%	-	
2	Muddy sand	<50%	>50%	-	
3	Sand	< 10%	> 90%	-	
4	Sandy gravel	-	> 20%	< 80%	Coarse sand with large fragments of shell material dispersed throughout
5	Shelly gravel	-	< 20%	> 80%	Very coarse shell and gravel – little or no sand apparent
6	Sand foul	-	-	-	Large areas of rock and/or boulders interspersed among patches of sand
7	Sand reef	-	-	-	Interface between subtidal rocky reefs and sand
8	Reef	-	-	-	Rocky reef

Table 3b **Habitat classification schema based on observations of contents of the benthic sled or direct observation of sea floor.**

Code	Habitat type
1	Seagrass (*Zostera*) meadow
2	Oyster bed 2.1 Pacific oysters 2.2 Flat oysters
3	Horse mussels (*Atrina* sp.)
4	Scallops (*Pecten* sp.)
5	Large bivalves 5.1 Cockles (Austrovenus) 5.2 Pipi / tuatua (Paphies sp.) 5.3 Other
6	Macroalgae
7	Sponge bed
8	Nothing

Table 4 **Numbers of sites and sample units (in brackets) surveyed in each harbour in the present survey. The target minimum numbers of samples are shown in the first row of the table. Target numbers of box traps and dive searches were not achieved in Bluff because of bad weather.**

Harbour	Survey date	Crab: Box traps[2]	Crab: Condos[2]	Starfish traps[2]	Benthic sled tows	Dive searches[3]	Visual searches
Target number[1]		60	8	15	100	30	25
Whangarei	27-31/3/06	66	8	30	99	30	22
Waitemata	3-13/4/06	113 (325)	17 (51)	47 (94)	232	54 (170)	53
Tauranga	1-5/5/06	60 (180)	9 (26)	17 (34)	100	21 (146)	10
Wellington	20-24/2/06	60 (180)	5 (13)[4]	33 (66)	100	30 (280)	25
Picton (including Havelock)	8-9 and 20-22/12/05	57 (171)	8 (24)	18 (36)	100	32 (222)[5]	40
Nelson	12-16/12/05	60 (178)	8 (24)	27 (51)	100	33 (136)	28
Lyttelton	23-27/1/06	59 (170)	8 (23)	20 (40)	100	31 (310)[6]	34
Otago	6-10/3/06	60 (180)	5 (15)[4]	44 (87)	98	30 (250)	25
Bluff	20-24/2/06	47 (141)	8 (23)	20 (40)	102	26 (200)	22

[1] Target numbers for Waitemata were twice those shown for other harbours.
[2] Data in each cell represent the number of sites sampled and the total number of traps set (in brackets).
[3] Data in each cell represent the number of sites sampled and the total number of piles searched (in brackets, but not including pontoons or breakwalls that were sampled at some sites).
[4] Suitable locations for deployment of condos were limited in Wellington and Otago harbours by the small number of suitably-sized streams.
[5] Dive searches in Picton and Havelock were done 9-24 November.
[6] Dive searches in Lyttelton were done on 16 March.

Table 5 **Presence (✓) or absence (×) of the eight target species, five non-target species, and opportunistic identifications on each survey, in each harbour. [1]Species identity determined as a result of opportunistic collection. Blank cells do not necessarily indicate absences.**

	Undaria pinnatifida	***Caulerpa taxifolia***	***Potamocorbula amurensis***	***Asterias amurensis***	***Sabella spallanzanii***	***Carcinus maenas***	***Eriocheir sinensis***	***Styela clava***	*Charybdis japonica*	*Chaetopterus* sp.	*Musculista senhousia*	*Theora lubrica*	*Pyromaia tuberculata*	[1]*Ficopomatus enigmaticus*	[1]*Acentrogobius pflaumii*	[1]*Arenigobius bifrenatus*	[1]*Austromegabalanus psittacus*	[1]*Nectocarcinus* sp. nova	[1]*Ovalipes elongatus*	[1]*Limaria orientalis*	[1]*Didemnum* sp.	[1]*Haliplanella lineata*	[1]*C. fragile* ssp. *tomentosoides*	[1]*Hydroclathrus clathratus*	[1]*Polysiphonia senticulosa*
Whangarei																									
21-25/11/02	×	×	×	×	×	×	×		×	✓	✓	✓	✓	✓					✓	✓					
29/09–4/10/03	×	×	×	×	×	×	×		✓	✓	✓	✓	✓	✓							✓				
15–20/03/04	×	×	×	×	×	×	×		×	✓	✓	✓	✓	✓		✓				✓					
6–10/09/04	×	×	×	×	×	×	×		×	✓	✓	✓	✓	✓											
27-31/3/06	×	×	×	×	×	×	×	×	×	✓	✓	✓	✓	✓							✓			✓	
Waitemata																									
5-9/04/03	×	×	×	×	×	×	×		✓	✓	✓	✓	✓								✓				
13-18/10/03	×	×	×	×	×	×	×		✓	×	×	✓	✓							✓					
29/03–2/04/04	×	×	×	×	×	×	×		✓	✓	×	✓	×												
20–24/09/04	×	×	×	×	×	×	×		✓	×	✓	✓	×								✓				
3-13/4/06	✓	×	×	×	×	×	×	✓	✓	✓	✓	✓	✓		✓					✓			✓		

Table 5 Continued.

	Undaria pinnatifida	***Caulerpa taxifolia***	***Potamocorbula amurensis***	***Asterias amurensis***	***Sabella spallanzanii***	***Carcinus maenas***	***Eriocheir sinensis***	***Styela clava***	*Charybdis japonica*	*Chaetopterus* sp.	*Musculista senhousia*	*Theora lubrica*	*Pyromaia tuberculata*	[1]*Ficopomatus enigmaticus*	[1]*Acentrogobius pflaumii*	[1]*Arenigobius bifrenatus*	[1]*Austromegabalanus psittacus*	[1]*Nectocarcinus* sp. nova	[1]*Ovalipes elongatus*	[1]*Limaria orientalis*	[1]*Didemnum sp.*	[1]*Haliplanella lineata*	[1]*C. fragile* ssp. *tomentosoides*	[1]*Hydroclathrus clathratus*	[1]*Polysiphonia senticulosa*
Tauranga																									
17-21/02/03	×	×	×	×	×	×	×		×	×	×	×	×								✓				
14-19/09/03	×	×	×	×	×	×	×		×	✓	×	×	×												
23-27/02/04	×	×	×	×	×	×	×		×	✓	×	×	×								✓				
23–27/08/04	×	×	×	×	×	×	×		×	✓	×	×	×					✓	✓		✓				
1-5/5/06	×	×	×	×	×	×	×	×	×	✓	×	✓	×										✓		
Wellington																									
9-14/12/02	✓	×	×	×	×	×	×		×	×	×	✓	×												
21-25/07/03	✓	×	×	×	×	×	×		×	×	×	✓	×												
24-28/11/03	✓	×	×	×	×	×	×		×	×	×	✓	×					✓		✓					
26-30/07/04	✓	×	×	×	×	×	×		×	×	×	✓	×												
20-24/2/06	✓	×	×	×	×	×	×	×	×	×	×	✓	×				✓								
Picton																									
8-22/12/05	✓	×	×	×	×	×	×	×	×	×	×	✓	×							✓	✓				

Table 5 Continued.

	Undaria pinnatifida	***Caulerpa taxifolia***	***Potamocorbula amurensis***	***Asterias amurensis***	***Sabella spallanzanii***	***Carcinus maenas***	***Eriocheir sinensis***	***Styela clava***	*Charybdis japonica*	*Chaetopterus* sp.	*Musculista senhousia*	*Theora lubrica*	*Pyromaia tuberculata*	[1]*Ficopomatus enigmaticus*	[1]*Acentrogobius pflaumii*	[1]*Arenigobius bifrenatus*	[1]*Austromegabalanus psittacus*	[1]*Nectocarcinus* sp. nova	[1]*Ovalipes elongatus*	[1]*Limaria orientalis*	[1]*Didemnum* sp.	[1]*Haliplanella lineata*	[1]*C. fragile* ssp. *tomentosoides*	[1]*Hydroclathrus clathratus*	[1]*Polysiphonia senticulosa*
Havelock																									
8-22/12/05	×	×	×	×	×	×	×	×	×	×	×	✓	×								×				
Nelson																									
10-14/02/03	✓	×	×	×	×	×	×		×	×	×		×								✓				
1-5/09/03	✓	×	×	×	×	×	×		×	×	×	✓	×								✓				
15-20/12/03	✓	×	×	×	×	×	×		×	×	×	✓	×								✓				
9-13/08/04	✓	×	×	×	×	×	×		×	×	×	✓	×								✓	✓			
12-16/12/05	✓	×	×	×	×	×	×	×	×	×	×	✓	×								✓				
Lyttelton																									
21-25/10/02	✓	×	×	×	×	×	×		×	×	×	✓	×												
7-12/07/03	✓	×	×	×	×	×	×		×	×	×	✓	×												
3–7/11/03	✓	×	×	×	×	×	×		×	×	×	✓	×												
14-18/06/04	✓	×	×	×	×	×	×		×	×	×	✓	×												✓
23-27/1/06	✓	×	×	×	×	×	×	✓	×	×	×	✓	×												

Table 5 Continued.

	Undaria pinnatifida	***Caulerpa taxifolia***	***Potamocorbula amurensis***	***Asterias amurensis***	***Sabella spallanzanii***	***Carcinus maenas***	***Eriocheir sinensis***	***Styela clava***	*Charybdis japonica*	*Chaetopterus* sp.	*Musculista senhousia*	*Theora lubrica*	*Pyromaia tuberculata*	*[1]Ficopomatus enigmaticus*	*[1]Acentrogobius pflaumii*	*[1]Arenigobius bifrenatus*	*[1]Austromegabalanus psittacus*	*[1]Nectocarcinus* sp. nova	*[1]Ovalipes elongatus*	*[1]Limaria orientalis*	*[1]Didemnum* sp.	*[1]Haliplanella lineata*	*[1]C. fragile* ssp. *tomentosoides*	*[1]Hydroclathrus clathratus*	*[1]Polysiphonia senticulosa*
Otago																									
1-5/03/03	✓	×	×	×	×	×	×		×	×	×	×	×												
2-8/08/03	✓	×	×	×	×	×	×		×	×	×	×	×												
19-23/01/04	✓	×	×	×	×	×	×		×	×	×	×	×												
28/06–2/07/04	✓		×	×	×	×	×		×	×	×	×	×								✓				
6-10/3/06	✓	×	×	×	×	×	×	×	×	×	×	×	×												
Bluff																									
22-26/03/03	×	×	×	×	×	×	×		×	×	×	×	×												
18-22/08/04	×	×	×	×	×	×	×		×	×	×	×	×												
1-5/02/04	×	×	×	×	×	×	×		×	×	×	×	×												
12-16/07/04	×	×	×	×	×	×	×		×	×	×	×	×												
20-24/2/06	✓	×	×	×	×	×	×	×	×	×	×	×	×												

Table 6 ***Charybdis japonica*** **catch characteristics in baited traps (box and starfish traps) set overnight in Waitemata Harbour.**

Survey	Date	Total no. captured	Mean catch per trap ± 1 S.E	Total no. locations trapped	No. (%) locations present
Initial NIWA Survey	April 2002	114	0.479 ± 0.046	120	72 (60 %)
ZBS2001/01 Summer 1	April 2003	58	0.291 ± 0.045	126	41 (33 %)
ZBS2001/01 Winter 1	Oct. 2003	62	0.290 ± 0.036	78	43 (70 %)
ZBS2001/01 Summer 2	March-April 2004	17	0.079 ± 0.021	81	14 (17 %)
ZBS2001/01 Winter 2	Sept. 2004	19	0.079 ± 0.019	84	17 (20 %)
ZBS2001/01 Summer 2005-6	April 2006	70	0.169 ± 0.027	158	49 (31%)

7. Appendix 1: Summaries of the habitat associations and life histories of the target species.

7.1 Northern Pacific seastar (*Asterias amurensis*)

7.1.1 General information

The northern Pacific seastar, *Asterias amurensis*, naturally inhabits the northern coast of China, the coasts of Korea and Japan, and along the Russian coast to the Bering Strait. It is also found occasionally in Alaska and northern Canada (Morrice, 1995). Its distribution has since increased to several other countries, including Australia. (http://www.fish.wa.gov.au/hab/broc/marineinvader/marine01.html).

Fully-grown seastars reach sizes of 40-50 cm in diameter, with reproduction possible at 10cm, when the seastar is around one year old (CRIMP, 2000). The seastar can increase its diameter by 8cm each year. (http://www.fish.wa.gov.au/hab/broc/marineinvader/marine01.html). Increasing size is also a response to food. When food is short the seastars shrink: their sexual organs also shrink which reduces fertilisation success (http://www.marine.csiro.au/PressReleasesfolder/99releases/seastar4jun99/backgrnd.html#gaps).

7.1.2 Timing of reproduction and recruitment

In the southern hemisphere, spawning occurs during winter (July-October) when temperatures are around 10 to 12 ^{o}C. Fertilisation takes place externally (http://www.parliament.vic.gov.au/enrc/ballast/Ballast-30.htm). Small eggs of approximately 150μm in diameter hatch, and develop into free-swimming larvae through a series of stages - coeloblast, gastrula, bipinnaria and brachiolaria (Bruce, 1998). A single adult female seastar can produce 10-20 million eggs each year for about 5 years. Both the eggs and larvae are planktonic, drifting in the ocean for up to two months before they settle and metamorphose into juvenile seastars. (http://www.parliament.vic.gov.au/enrc/ballast/Ballast-30.htm). Based on this 60-day larval period, settlement in Australian waters has been shown to occur during mid-September (Parry et al. 2001). The northern Pacific seastar lives for up to five years. It is known to reach outbreak proportions that occur in three to ten year cycles, and which last two to three years (http://www.fish.wa.gov.au/hab/broc/marineinvader/marine01.html).

7.1.3 Habitat and biology

Morrice (1995) suggests that in Tasmanian studies, it is unclear whether the northern Pacific seastar is present in areas due to specific habitat requirements or whether their location is dependant on their rate of spread.

Substratum type

The preferred substrata for *A. amurensis* are mud, sand or pebbles (http://www.fish.wa.gov.au/hab/broc/marineinvader/marine01.html), extending to a mixture of rock, algae and seagrass (Morrice, 1995). It is rarely found on reefs or places subject to high wave action. However, a benthic habitat is not essential - in Tasmania, both adults and juveniles have been recorded attached to scallop longlines, mussel and oyster lines, salmon cages and spat bags (http://www.fish.wa.gov.au/hab/broc/marineinvader/marine01.html). Research has shown that substratum seems important for the induction of settlement and metamorphosis - brachiolara have shown high rates of settlement on non-geniculate coralline algae, followed by rock and mud. Sand and mussel shell did not induce settlement well. Bacterial cover on mussel lines, accompanied by the fine algae that grows on the ropes, may also provide a very attractive settlement surface (Morris & Johnson, 1998).

7.1.4 Food preferences

The seastar is a predator of many organisms but has a particular preference for shellfish (http://www.fish.wa.gov.au/hab/broc/marineinvader/marine01.html). Other prey include sponges, crustaceans, polychaetes and fish (http://www.parliament.vic.gov.au/enrc/ballast/Ballast-30.htm), as well as tunicates, bryozoans and echinoderms (Morrice, 1995).

7.1.5 Physiological tolerances (range and preferences)

Temperature

The seastars prefer water temperatures of between 7 and 10 ^{o}C in their natural range (http://www.fish.wa.gov.au/hab/broc/marineinvader/marine01.html), but can tolerate a range of 5 – 20^{o}C. In Japan, water temperatures above 20^{o}C limit the seastars' range, with adults losing weight and larvae dying above this temperature (Morrice, 1995, Bruce, 1998). The survival of larvae is temperature dependant, with the optimal range

being between 8 to 16 ^{o}C (Bruce, 1998). However, adult seastars have been shown to adapt to warmer temperatures of up to 22 ^{o}C in countries outside their natural range, such as Australia (http://www.fish.wa.gov.au/hab/broc/marineinvader/marine01.html).

Depth

The seastar is mainly found in sublittoral to subtidal areas, but can also be present at depths of up to 200m

(http://www.fish.wa.gov.au/hab/broc/marineinvader/marine01.html). In Australia, it occurs in the intertidal zone down to a depth of 25m (CRIMP, 2000). Parry & Cohen (2001) have observed that in some parts of Port Philip Bay, the density of the seastar decreases at depths of <15 metres. Morrice (1995) states that in the northern Pacific, the seastar inhabits deeper water in the summer and moves into shallower water in the winter. This may be to survive summer temperatures and to move between areas.

Salinity

Little research appears to have been conducted on salinity tolerances of the northern Pacific seastar, but adults seem to be restricted to salinities above 28 psu (Morrice, 1995). In general, the seastar is sensitive to any changes in salinity and as a result is unlikely to tolerate fluctuating salinities
(http://www.fish.wa.gov.au/hab/broc/marineinvader/marine01.html).

Optimal salinity for larval survival is 32 psu. The larvae become adversely affected by 10 minute exposures to salinities <17.5 psu and do not survive exposures to salinities <8.55 psu, when extensive cellular damage has been found to occur (Bruce, 1998).

7.1.6 Route of introduction

The most likely route is as seastar larvae contained in the ballast water of international vessels, although research suggests that 'sea chests' are another potential method of transport (Dodgshun & Coutts, 2002). Juvenile seastars found on mussel lines in Port Philip Bay, Australia, indicate a further risk of spread (Garnham, 1998).

7.1.7 Methods of sampling

- Parry & Cohen (2001) used a 2.7 m wide peninsula scallop dredge, covered by 25mm mesh to sample *Asterias*. Estimates of field densities were based on the number of seastars collected in a 60 second tow at a speed of 5.7 +_ 0.3 knots. The average tow length was around 170 m (Parry & Cohen, 2001).

- Whayman/Holdsworth seastar traps have been designed to catch *Asterias*. Traps with a mesh size of 26mm catch more seastars than larger mesh (65mm) traps. Most seastars are caught within the first 24-48 hours. Pilchards are the more attractive bait but only for short soak times (24-48 hours). The traps effectively fish an area of approximately $30m^2$ (Martin, 1998).

- Vertical distribution of larval asteroids can be measured using vertical tows of a 100μm free-fall plankton net with a 500mm diameter mouth and 5m in length. A choking bridle closes the net when hauled. Vertical tows are undertaken to depths of 5m, the depth at which the net completely submerges, or 15m. A small float can be tied to the end of the plankton net by 10m of fine line – when this submerges, the net has reached the appropriate depth of 15m (Parry et al, 2001).

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7.2 Asian clam (*Potamocorbula amurensis*)

7.2.1 General information

The Asian clam, *Potamocorbula amurensis*, is a native of estuaries from southern China (22^{o} N latitude) to southern Siberia (53^{o} N) and Japan (Cohen & Carlton, 1995). However, it has extended its range to establish abundant populations in California, USA, particularly San Francisco Bay. Asian clams are euryhaline at all stages of development, and reach settlement 17-19 days after fertilisation (Nicolini & Penry, 2000).

7.2.2 Timing of reproduction and recruitment

Field studies in San Francisco Bay suggest that the clam spawns throughout the year, although site-specific seasonal reproduction appears to be related to food supply (Parchaso & Thompson 2002). The eggs are negatively buoyant, so fertilisation and initial development occur in more saline bottom waters. It takes 48 hours for development to the straight hinged larval stage through several life phases – fertilised egg, two-cell stage, four-cell stage, blastula, trochophore). At 17 – 19 days after fertilisation, the bivalve settles at a shell length of approximately 135 μm. Newly settled clams can reproduce within a few months (Nicolini & Penry, 2000). Juvenile clams studied in San Francisco Bay had a mean shell length of 1.7 mm. By the time they were under a year old, shell length was approximately 11 mm (Cohen & Carlton, 1995). Adults generally reach a length of 20 – 30 mm (NZ Ministry of Fisheries, 2001).

Studies in San Francisco Bay have shown that the clam displays a complex picture of patchy recruitment in space and time, which is expected for an invasive eurytopic species (Carlton et al. 1990). The zone of greatest recruitment shifts dramatically with changes in flow - high riverine outflow conditions may reduce clam densities, but the clams are quick to repopulate brackish water habitats when high flows abate (Peterson, 1998).

7.2.3 Habitat and biology

Substratum type

The Asian clam is pervasive with regard to habitat. It can invade environments which are nearly freshwater, creeks and sloughs, intertidal sand-mud flats, and on a wide

range of subtidal soft bottomed substrata - flocculant mud, coarse sand, peat and hard clay (Carlton et al, 1990). It typically sits with one-third to one-half of its length exposed above the sediment surface. (Cohen & Carlton, 1995). It has been found in very high densities in the benthic layer in the majority of San Francisco Bay estuary, at up to 48,000 individuals.m^{-2} (Peterson, 1998). Research in laboratory aquaria has shown that its behaviour can lead to the formation of depressions in the underlying substrate, which can significantly disturb sediment layers to a depth of about 1cm. The highly altered, complex surface left behind may cause difficulties for other mobile and sedentary infauna, thus allowing the clam to dominate (Carlton et al, 1990).

Feeding

Potamocorbula amurensis is an efficient suspension feeder (Thompson et al, 1991). Examination of faeces from specimens collected in San Francisco Bay show that the clam ingests both planktonic and benthic diatoms. It also filters bacterioplankton as well as phytoplankton, though at lower efficiency, and assimilates both with high efficiency. Laboratory experiments have shown that the bivalve can also readily consume certain copepod nauplii (Kimmerer et al 1994). Other research suggests it may feed on the larvae of other benthic organisms (Cohen & Carlton, 1995).

7.2.4 Physiological tolerances (range and preferences)

The Asian clam is one of the few species of bivalves able to tolerate virtually any salinity, withstand tropical or cold temperate waters and survive in polluted environments. Research in San Francisco Bay suggests that the Asian clam has spread rapidly, irrespective of sediment type, water depth and salinity (Thompson et al. 1991). The following information highlights the wide range of physiological tolerances that this species displays.

Temperature

Their latitudinal range in Asia suggests that Asian clams can survive a temperature range of 0 – 28^{o}C (Cohen & Carlton, 1995). There is very little information for *P. amurensis*, but data for the similar Chinese corbulid *P. laevis* (found at approximately the same latitude as San Francisco Bay) suggest that gametogenesis requires water temperatures ranging from 12 – 23^{o}C. Reproductively active *P. amurensis* have been seen in San Francisco Bay in water temperatures ranging from 6 – 23^{o}C (Parchaso and Thompson 2002). Fertilised eggs of *P. laevis* are shed at temperatures of between 16 and 20^{o}C. Growth rates are greatest when water

temperatures are between 22 and 28°C. Growth rates decline below 17°C, and growth ceases below 11.8°C (Carlton et al, 1990).

Depth

The clams live both subtidally and intertidally (Cohen & Carlton, 1995), but primarily subtidally (Carlton et al, 1990).

Salinity

The Asian clam can survive in a range of salinities from almost freshwater (< 1 psu) to full-strength seawater (32 – 33psu) (Cohen & Carlton, 1995, Carlton et al, 1990, http://www.fish.wa.gov.au/hab/broc/marineinvader/marine08.html) but long-term survival of adults is highest at salinities from 5 to 25 psu (Nicolini & Penry, 2000). Spawning and fertilisation can occur at salinities from 5 – 25 psu, with a maximum at about 10 – 15 psu. Eggs and sperm can tolerate at least a 10-psu step increase or decrease in salinity. Studies have shown that fertilisation and initial development tend to occur in the more saline bottom waters of San Francisco Bay. Embryos of two hours old have been shown to tolerate salinities from 10 – 30 psu, and at 24 hours old they can tolerate the same wide range of salinities that adult clams can. However, any *rapid* changes in salinity may adversely affect larval growth (Nicolini & Penry, 2000).

7.2.5 Route of introduction

The initial introduction of the Asian clam to San Francisco Bay seems to have been as veliger larvae transported in ballast water by trans-Pacific cargo ships. The clams' ability to tolerate wide changes in salinity suggests it can survive incomplete oceanic exchanges of ballast water (Nicolini & Penry, 2000). The infaunal habitat of the clam suggests that it did not arrive as a fouling organism (Carlton et al, 1990).

7.2.6 Methods of sampling

- Carlton et al. (1990) described a combination of sampling devices that were used to sample Potamocorbula, including a modified Van Veen grab, a Ponar grab and a Van Veen grab, that sampled between 0.05 and 0.1 m^2 of sediment. Samples were sieved through screens of 0.5 mm to 1mm mesh size. Between 3 to 5 replicate grabs were taken at each sampling station.

- Peterson (1998) describes an extensive survey for Potamocorbula in San Francisco Bay using a Ponar grab.

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7.3 Chinese mitten crab (*Eriocheir sinensis)*

7.3.1 General Information:

The Chinese mitten crab *Eriocheir sinensis* is a burrowing crab native to mainland China and coastal rivers and estuaries of the Yellow Sea. It is a palm-sized greyish-brown grapsid crab with small white pincers protruding from hairy brown claws. The native range of the mitten crab extends from the southern border of North Korea (40^{o}N latitude) to Hong Kong (22 ^{o}N). It has established introduced populations in Vietnam, northern Europe and the west coast of America. The first specimens to be found in Europe were reported from near Hamburg in Germany 1912 (Panning 1939). Since then, mitten crabs have spread from Finland to the Atlantic coast of southern France and to the UK, Russia, Holland, Belgium, the Czech republic, Denmark, Sweden, France, Poland and Portugal and Spain. The first reported occurrence of the mitten crab in North America was in the Detroit River in 1965 by the city of Windsor, Canada. Later, in 1973, commercial fishermen netted several crabs in Lake Erie near Erieau and Port Stanley, Ontario, Canada (Nepszy & Leach, 1973). In June 2006 a specimen was caught in Chesapeake Bay (SERC 2004). On the west coast, it was first reported from San Francisco Bay in 1992 where it has since become well-established (Halat & Resh 1996). Ballast water introductions have been blamed, but speculation also exists about possible deliberate release into the U.S.A.

Eriocheir sinensis is a catadromous species that lives most of its life in freshwater environments. Mature males and females migrate during late summer to tidal estuaries where they mate and spawn. Adults (Maximum body size 10-cm carapace width, but more commonly between 5 and 8 cm) are capable of very long distance migrations e.g. over 1000km in the Yangtze River (Cohen & Carlton 1995). After mating the females are thought to continue seaward, over-wintering in the deeper water and returning to brackish water in the spring to hatch their eggs (Panning 1939). The movement of crabs to deeper water and the timing of egg hatching/larval release is temperature dependent. Winter temperatures are much colder in Europe than San Francisco, which is probably why crabs there move to deeper water and why hatching is delayed until spring. In the San Francisco Estuary, preliminary data indicate that the adult crabs remain in the spawning areas (~ 20ppt) and hatching occurs in November/December and again in March. The timing of hatching varies yearly depending upon winter water temperatures. Settled juvenile crabs gradually move upstream into brackish (1-5 psu) and fresh water to complete the life cycle.

Mitten crab ‘plagues’ of extreme numbers have been reported from Germany in the mid 1930’s (Panning 1939) and in the Netherlands in 1981 (Ingle, 1986). Adults are capable of emerging from water and crossing dry land when migrating.

7.3.2 Timing of reproduction and recruitment

Crabs mature at different ages according to locality. Maturity has been reported at ages of 3 to 5 years in Europe (Panning 1939), 1 to 2 years in China (Cohen & Carlton 1995) and 2 to 3 years in California (Veldhuizen and Stanish 1999. Each female produces from 250,000 to 1 million eggs, which hatch in late spring or early summer. In laboratory culture, the larval period lasts for 1 –2 months and the larvae develop through five zoeae and a megalopa stage (Kim & Hwang 1995). After the final larval moult the juvenile crab settles to the bottom in late spring and begins its migration upstream (Panning, 1939; Ingle, 1986; Anger, 1991). Experiments indicate that complete development of larvae is not possible in rivers or in brackish estuarine conditions (Anger 1991).

7.3.3 Habitat and Biology:

Substratum type

The normal habitat of the juveniles is the bottoms and banks of brackish and freshwater rivers and estuaries, individuals prefer hard bottoms and areas covered with submerged plants (Nepzy & Leach 1973). Older juveniles are found in a diversity of habitats including silt, gravel, and open unvegetated stream channels. In freshwater habitats of San Francisco Bay, *E. sinensis* is most common in areas with steep, vegetated banks that are high in clay content. Burrows are concentrated underneath the root profile of the aquatic macrophytes lining the banks, which mainly consists of *Scirpus* (Halat & Resh 1996). Submerged aquatic vegetation is an important component to the habitat. It provides cover and high concentrations of invertebrates (Veldhuizen 2000).

In Asia and Europe mitten crabs live in burrows dug in river banks or in rice paddies in coastal areas (Cohen & Carlton 1995). Young mitten crabs are found in tidal freshwater areas and usually burrow in banks and levees between high and low-tide marks. Optimal rearing habitat for juveniles is areas with still or slow velocity water, a stable water depth, low turbidity, and warm temperatures (ranging from 20^{o}C to 30^{o}C, with optimal growth at 24^{o}C to 28^{o}C) (Veldhuizen 2000). Mitten crabs apparently do not burrow as extensively in non-tidal areas. Older juveniles are found further upstream than young ones and both adults and juveniles can move hundreds of km.

In China, recently settled juvenile mitten crabs are harvested during spring tides in late May and June when they congregate over sandy bottom areas in water of 1 to 3 $^{0}/_{00}$ (Hymanson et al. 1999)

Food preferences

The mitten crab is known to be predominantly an omnivorous, opportunistic feeder, although feeding habits change as they mature. Juvenile crabs mainly eat vegetation (Halat & Resh 1996) primarily filamentous algae (Veldhuizen & Stanish 1999). As they mature they also prey on small invertebrates, especially worms and clams so that adults and juveniles are considered omnivorous (http://www.wsg.washington.edu). Gut content analysis of crabs in the San Francisco Bay area revealed a high proportion of vegetative matter, with low amounts of invertebrates, regardless of the size of the crab or the habitat from which it was captured (Rudnick et al. 2000).

Vegetation type

Juveniles were observed taking cover in floating vegetation, especially water hyacinth in the USA (Hieb & Veldhuizen 1998). An ongoing study by Veldhuizen is currently assessing habitat associations for this crab in the San Joaquin Delta, but results are presently unavailable. In Asia, the juveniles can be associated with rice paddies (Panning 1939). An attempt to characterise habitat associations of mitten crabs in the San Joaquin River in 2000 failed to capture any individuals (May & Brown 2000).

7.3.4 Physiological Tolerances (range and preferences):

Temperature

Adult mitten crabs exhibit a wide range of temperature tolerances. Growth ceases only at temperatures below 7°C and above 30°C (Rudnick et al. 2000). All larval stages of the Chinese mitten crab show a clear preference for warm water, however (15° to 18°C), and temperatures below 12°C do not allow any development beyond the first zoeal stage in the laboratory (Anger 1991). Adults can tolerate temperatures as low as 0 °C for a week and temperatures up to 31 °C are suitable for juveniles (Veldhuizen & Stanish 1999).

Depth

Juvenile mitten crabs appear to occur mostly in shallower waters (i.e. < 10m) (Veldhuizen & Stanish 1999, preliminary results), with largest densities found in areas with an average depth of 2 m, which corresponds to the depth of submerged aquatic vegetation (Veldhuizen 1999). However, through the winter sexually mature females are thought to move to “deep” water to develop their fertilised eggs. Adult mitten

crabs are highly tolerant of desiccation and are able to remain on land for several hours without mortality. Veldhuizen (pers. comm.) compared the relative abundance of juvenile mitten crabs among six different habitat types - shallow (0-2.4 m) vegetated natural substrate, shallow unvegetated natural substrate, shallow vegetated rock substrate, shallow non-vegetated rock, mid-depth channels (2.5 – 4.9 m), and deep channels (5 - 10 m) - that occurred in a tidal freshwater marsh. Crabs occurred in all habitat types, but were overall more abundant in shallow (0 to 2.4 m) vegetated areas with natural substrate. Most of the crabs ranged in size from 20 to 38 mm, average size was 28 mm.

Salinity

Juvenile and adult Chinese mitten crabs are extremely euryhaline (i.e. high range of tolerated salinities) and its osmoregulatory abilities appear well developed (Onken 1996). By hyper-regulating the ionic content of their body fluids, the crabs can quickly adapt from high to low salinity environments (Welcomme & Devos 1991 cited in Rudnick et al. 2000). Different larval stages are known to vary in their salinity tolerances. The first zoeal stage, which occurs in seawater, is strongly euryhaline, but successive zoeal stages become increasingly stenohaline (low range of tolerated salinities) and prefer more typical marine salinities (e.g. >30 psu). The megalopa, which migrates to freshwater, is euryhaline, with an optimal growth response in brackish waters (5-25 psu) (Anger 1991). Salinities in the areas where *E. sinensis* has been found range from 0-5 psu in San Francisco Bay (Halat & Resh 1996). It cannot spawn in fresh water and larval growth cannot go to completion in rivers or brackish waters (Anger 1991). Mating and fertilisation in the San Francisco estuary occur in late autumn and winter, generally at salinities of 15- 20 psu. In China, most mating occurs in brackish water (10 – 16 psu) (Hymanson et al. 1999). A large increase in the abundance of this species in England coincided with a drought and a large change in the salinity of the estuaries they occupied (Atrill & Thomas 1996).

7.3.5 Methods of Sampling:

Methods of sampling mitten crabs need to differ between adults and juveniles to reflect their different diets and habitats. Adults migrate downstream in late summer to spawn. These crabs are sexually mature. Only juveniles migrate upstream. Juveniles are found in creeks, rivers, and tidal freshwater and brackish marshes and sloughs. Juveniles burrow and occupy burrows but also remain in the subtidal zone.

- Panning (1938) found that because juveniles are mostly vegetarian, capturing them with baited traps didn't work and they had to be excavated from their

burrows during low tides. Capturing juveniles in the USA has involved intertidal searches at low tide where all cavities such as burrows and root tunnels were excavated and all debris, driftwood and small puddles were examined. Juveniles were also successfully captured in 'crab condos', submerged artificial structures of PVC tube used for shelter.

- A comparison of trapping techniques by Veldhuizen et al (1999) suggested that traditional crab sampling techniques are not very effective for this species due to the change in diet between juveniles and adults, the diversity of habitats occupied, and their escape tendencies. For juvenile crabs she recommends using artificial shelter substrates ("crab condos") made of 12 vertical PVC tubes (6 in long, 2 in diameter) and burrow searches for juveniles in the banks of silty, tidally influenced streams. Crab condos are typically submerged for 48 hours to allow the crabs to enter (Veldhuizen 2000), but significant increases in catch are achieved with longer soak times (3, 5and 9 days).

- Beach seining for adults was possible in shallow intertidal areas and subtidal areas. Baited traps were not recommended for juvenile mitten crab, or for monitoring and detection programs where adult densities may be very low.

- Various other baited traps, snares and ring nets have also been trailed, with variable success. Ring nets are most successful when densities of crabs are high. The crabs appear to be most active in the two weeks surrounding the full moon.

7.3.6 Impacts

The crab has caused numerous problems in Europe when found in extremely high densities. The burrows that it excavates can destabilise river banks and lead to accelerated bank erosion. The sharp claws of *E. sinensis* cut up commercial fish nets, increasing operating costs of fishing operations. The most widely reported economic impact of mitten crabs in Europe has been damage to commercial fishing nets and the catch when the crabs are caught in high numbers. Because of the severe problems the crab has caused in European waters, *E. sinensis* recently has been listed as a federally injurious species in the United States.

The ban on importing live Chinese mitten crabs to the USA was enacted due to concern over potential damage from its burrows to levees or rice fields in the Central Valley, and because the crab is a second intermediate host of a human parasite, the oriental lung fluke *Paragonimus westermanii* (Cohen & Carlton 1995). The Chinese

mitten crab has been widely reported to be an intermediate host for the Oriental lung fluke, a parasite that uses a snail as its primary host, freshwater crayfish and crabs as intermediate hosts, and a variety of mammals, including humans, as final hosts in its life cycle (Chandler & Read 1961; Lapage 1963). Humans can become infected with the parasite through ingestion. The fluke settles in the lungs and other parts of the body, and can cause significant bronchial or, in cases where it migrates into the brain and/or muscles, neurological illnesses. It is believed that no species of snail that is in the family of the primary host currently occurs in Europe, and no appropriate snail host has been found in the San Francisco Bay-Delta system (Clark et al. 1988; Veldhuizen & Stanish 1999). Armand Kuris and Mark Torchin of U. C. Santa Barbara found no parasites of any kind in 25 mitten crabs from San Francisco Bay (A. Kuris, pers. comm., 1995).

The potential ecosystem impacts of large numbers of crabs invading new areas are unknown but authors have often speculated on possible effects to benthic invertebrate communities. There is concern that the crab will consume benthic invertebrates, salmon and trout eggs and may affect other species through direct predation or competition for food resources. In England there is some concern that it may compete with the native crayfish in fresh water (Clarke et al 1998). In China and Korea Juvenile mitten crabs have been reported to damage rice crops by consuming the young rice shoots and burrowing in the rice field levees. Since *E. sinensis* often inhabit areas that may contain high levels of contaminants, bioaccumulation of contaminants could also be transferred to predators or humans.

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7.4 European green crab (*Carcinus maenas*)

7.4.1 General information

The European green crab, *Carcinus maenas*, is native to the Atlantic, Baltic and North Sea coasts of Europe, but has established populations outside this range on the Atlantic and Pacific coasts of North America, in South Africa, and Australia. Green crabs produce planktonic larvae that pass through six developmental stages – a prezoea, 4 zoeal stages, and a megalopa – before metamorphosis to the benthic, juvenile crab phase. The crabs themselves grow through 18 to 20 moult cycles before reaching maximum size and terminal anecdysis (Parry et al. 1996). In its native range, the green crab can live up to 5 years and males reach a size of 86 mm carapace width. In western North America, adult males can be up to 92 mm carapace width within 2 years (Grosholz & Ruiz 1996).

7.4.2 Timing of reproduction and recruitment

Green crabs mate after the females moult, usually between spring and autumn. In warmer waters, females carry eggs for around four months. Egg-bearing females tend to migrate into deeper water during winter and prezoeae hatch from the eggs predominantly in spring (http://www.wa.gov/wdfw/fish/ans/greencrab.htm). The prezoeae pass through four zoeal stages in the plankton before moulting into the megalopal stage. Megalopae appear in early-mid summer and metamorphose and settle into the juvenile crab phase in late summer (Parry et al. 1996). The average development time for *C. maenas* larvae varies with temperature. At 10^{o}C development takes around 75 days, and at 25^{o}C it can take as little as 13 days (Parry et al. 1996). The timing of settlement is related to the number of months in which water temperatures are below 10^{o}C. In cooler waters, settlement occurs in late summer. In warmer waters, megalopae can begin to settle in late autumn (Yamada et al. unpubl. ms). Settlement occurs predominantly at night around the time of high tide (Zeng et al. 1997).

7.4.3 Habitat and biology

Substratum type

In its native range, the Green Crab, *Carcinus maenas*, occurs on both hard (rocky) and soft intertidal and shallow subtidal habitats in semi-exposed soft-sediment bays (Moksnes 2002). In Europe, eastern North America, Australia and South Africa, green

crabs occur in protected embayments and on moderately exposed rocky shores. In western North America green crabs occur only in sheltered embayments and only in soft-sediment environments (Grosholz & Ruiz 1996). A recent survey of the distribution of *C. maenas* in southern Australia found crabs in a range of soft-sediment habitats in low energy embayments. Substratum type, depth and water quality were all poor predictors of its presence and abundance in traps set in these habitats (Thresher et al. 2003).

Post larvae (megalopae) settle and metamorphose predominantly in shallow (< 1 m) sheltered or semi-exposed areas that have some form of structured habitat that provides shelter from predators (e.g. seagrass, macroalgae, mussels, shell debris, etc). Small crabs are often found in close proximity to vegetation such as beach grass, reeds, and eelgrass, although they also occur in exposed areas such as bare mud. Larger crabs do not need vegetative cover. In Sweden, young crabs are concentrated in greatest densities within structurally complex habitats, such as mussel beds, shell debris, seagrasses and filamentous algae. Much smaller densities occur in adjacent sand or mud. Densities of juvenile crabs (2^{nd} – 9^{th} instar) are significantly greater in mussel beds and shell habitats (mean = 206 crabs.m^{-2}) than in eelgrass (45 crabs.m^{-2}), filamentous green algae (24 crabs.m^{-2}) or sand (13 crabs.m^{-2}). Settlement of megalopae occurs predominantly to structurally complex habitats such as filamentous algae (231 settlers.m^{-2}), eelgrass (159 settlers.m^{-2}) and mussel beds (114 settlers.m^{-2}), rather than to open sand (4 settlers.m^{-2}), but larger animals redistribute themselves among these habitats. Indeed, adult crabs are highly mobile and are capable of foraging over large areas (km to 10's km).

Food preferences

Green crabs are omnivorous. Adult crabs feed predominantly on bivalves (rank = 1), small crustaceans (rank = 2) and smaller numbers of polychaetes and green algae (rank = 3 to 4) (Grosholz & Ruiz 1996).

7.4.4 Physiological tolerances (range & preferences)

Temperature

Carcinus maenas can tolerate a wide range of temperatures. In its native and introduced ranges, animals can tolerate average summer water temperatures of 22°C and average winter temperatures of 0 °C, although adult mortality has been recorded at sustained winter temperatures of 0 °C or below (Cohen et al. 1995). Crabs stop

moulting and drastically reduce their activity below 10°C, and stop feeding when temperatures are below 7°C (Yamada et al. unpubl. MS). Successful embryonic development occurs at temperatures between 11 and 25 °C.

Depth

Green crabs are found predominantly in the mid-intertidal zone, between about 1.3 m to 1.7 m above datum, and shallow subtidal, although adults have been recorded as deep as 60 m (Cohen et al. 1995). Juveniles (0-1+ age, 1-20 mm carapace width) are found mainly < 1 m water depth (Moksnes 2002). In Bodega Harbour, California, green crabs were caught between +0.7m and 1.4 m above mean lower low-water, with crabs being most abundant at +1.2 m (Grosholz & Ruiz 1995: see Figure 3). Parry et al. (1996) and Thresher et al. (2003) report greatest catches of adult *C. maenas* in water depths < 10 m. However, in Sweden, subadults and adults are found commonly between 0.1 to 20 m depth (occasionally to 60 m).

Salinity

Green crabs tolerate a wide range of salinity, but appear to prefer more saline areas (Proctor 1997). Adults reside in water from 4 psu to 34 psu. Populations breed successfully at salinities down to at least 13 psu, although larvae may only settle at salinities above 17 psu (Cohen et al. 1995). Survival of eggs to larval stages occurs at salinities between 26 and 39 psu and larval development may be prevented at < 13 psu (http://www.wa.gov/wdfw/fish/ans/greencrab.htm*)*. In the laboratory, adult *Carcinus* prefer salinities of 22-41 psu, but can tolerate maximum salinities of up to 54 psu (Cohen et al. 1995).

7.4.5 Methods of sampling

- Standard baited minnow traps (cylindrical with inverted cone entrances of ~ 57 mm) are set near the edge of vegetation or along mud/peat banks, generally far from the low tide drainage channels. Set 5-10 traps with openings perpendicular to the incoming tide with a rock in the trap to hold it in place, and possibly a rock "cradle" made in the substrate to keep the traps from being moved by wave action (http://www.pac.dfo-mpo.gc.ca/ops/fm/shellfish/Green_Crab/FIND.HTML).

- Shore searches along the high tide wrack line where storm driven vegetation accumulates for exuviae of molting crabs. This is most profitable in areas with

some vegetation intertidally or subtidally, as molting crabs prefer to have cover available during this vulnerable process (http://www.pac.dfo-mpo.gc.ca/ops/fm/shellfish/Green_Crab/FIND.HTML).

- Yamada et al. (2001) compared 4 types of traps for catching *Carcinus*: unbaited pitfall traps, minnow traps, fish traps and box traps deployed in intertidal and shallow subtidal environments. In high intertidal areas, pitfall traps were successful for sampling crabs < 45 mm carapace width. Folding traps and box traps successfully caught crabs > 40 mm. The box traps typically yielded larger catches than other types and caught crabs in their second or third summer.

- Thresher et al. (2003) used collapsible box traps (62 cm x 42 cm x 20 cm) to survey populations of *C. maenas* in southern Australia. Traps were typically baited with oily fish and deployed over night for 15-24 hours. Average catch rates from a single overnight set were occasionally as high as 44 crabs.trap^{-1}.

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7.5 Mediterranean fanworm (*Sabella spallanzanii*)

7.5.1 General information

Sabella spallanzanii is a large (up to 70 cm length) tube-building polychaete that is native to the Mediterranean and Atlantic coasts of Europe. Introduced populations of *S. spallanzanii* have been recorded in Brazil, and in the southern states of Australia (Western Australia, South Australia and Victoria) where it occurs in large densities attached to a variety of substrata. The worm's tubes are constructed of a tough but flexible material with the outer layer often incorporating deposits of silt and mud. The base of the tube is usually secured to hard substrata such as rocks, jetty pilings or shell fragments (Clapin & Evans 1995), but they may inhabit soft sediments where there are some solid particles (e.g. shell fragments, pebbles) on which the tubes can attach.

7.5.2 Timing of reproduction and recruitment'

Sabella spallanzanii is a gonochoric broadcast spawner that releases strings of mucus containing eggs or sperm into the water column (Giangrande et al. 2000). Worms attain sexual maturity at around 50 mm length after 6 months of growth. Spawning is thought to occur in autumn and winter in Victoria (Currie et al. 2000), although there is some evidence for summer spawning in Western Australia (Clapin & Evans 1995). Females are highly fecund and can produce >50 000 eggs which appear to be fertilised either internally or in situ (Giangrande et al. 2000). The fertilised egg masses are negatively buoyant and sink rapidly to the bottom (Giangrande et al. 2000). As the egg membrane disappears, free-swimming trochophore larvae emerge. These larval stages have a planktonic life of up to 21 days before they settle to the adult habitat. Settling larvae are gregarious and new recruits often occur in dense clusters. In Victoria, small worms (10-14 cm length) have been recorded in late November (Parry et al. 1996). Larvae spend about 2 weeks in the plankton before they settle and metamorphose (CRIMP 2001), but appear to travel only short distances (<20 km) from their parent stock prior to settlement (Parry et al. 1996).

7.5.3 Habitat and biology

Substratum type

Sabella spallanzanii grows preferentially in sheltered, nutrient enriched waters that are not subject to waves (Currie et al. 2000). In its native range it occurs predominantly on hard substrata and, in Port Phillip Bay, Australia, it is particularly abundant on man-

made hard surfaces such as wharf pilings, channel markers, marina piles, etc. It is not common on the hulls of ships (Giangrande et al. 2000). Largest densities occur on hard surfaces between 2 m and 7 m depth (Currie et al. 2000). In unconsolidated sediments, *Sabella* occurs in areas where suitable attachment substrata (rocks, concrete, wood, steel, bivalves, ascidians, etc) are present and tends to be aggregated in smaller densities. Although it has become established in most subtidal habitats in Port Phillip Bay, Currie et al. (2000) suggest that the larger densities on pilings and artificial hard surfaces reflect a preference for settlement on vertical surfaces.

Feeding

Sabella spallanzanii is a filter feeder that traps suspended food particles using its fan-shaped crown of tentacles. It has apparently been reared in the laboratory on a variety of food, but few details of actual diets are available (Parry et al. 1996).

7.5.4 Physiological tolerances (range and preferences)

Temperature

Spawning of *S. spallanzanii* occurs when seawater temperatures range between 11^{o}C and 14^{o}C **(**Giangrande et al. 2000). Optimum conditions for growth are at temperatures of between 10-19^{o}C.

Depth

Sabella spallanzanii has been recorded in water depths of 1 m to 30m (Parry et al. 1996). In soft sediments, densities tend to be larger at depths of < 7 m, but decline significantly at greater depth (17 to 22 m) (Currie et al. 2000). Densities on hard surfaces in Port Phillip Bay generally increased with depth, but were largest between 2 m and 9 m depth (Currie et al. 2000).

Salinity

There are few data on the salinity preferences of *S. spallanzanii*. In its native and introduced ranges, it is abundant in sheltered harbours and ports that are subject to fluctuations in salinity, but most studies have been of populations in relatively saline (> 32 psu) waters.

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7.6 Aquarium weed (*Caulerpa taxifolia*)

7.6.1 General information

Caulerpa taxifolia is a green single-celled alga (Chlorophyta: order Caulerpales, family Caulerpaceae) native throughout many areas of the tropical Pacific and Caribbean (GISP 2002). It is a popular aquarium plant, and prolonged breeding in aquaria and associated exposure to chemicals and UV light are thought to have produced a hardier strain that differs from native plants genetically and has a higher tolerance to cold water temperatures (Jousson et al. 1998). *C. taxifolia* has been introduced to at least three geographical regions outside its native range: the Mediterranean Sea on the coasts of Croatia, France, Italy, Monaco, and Spain, (2) the southern Californian coast near San Diego, and (3) parts of the coasts of New South Wales and South Australia (Meinesz 1999; Campbell & Tebo 2001). However, the "aquarium hypothesis" has been challenged by recent work on the temperature tolerance of native populations in eastern Australia (Chisholm et al. 2000; see below).

The basic morphology consists of a thallus with horizontal stolons that give off rhizoids and erect feather-like branches, with pinnately arranged pinnules (GISP 2002). In its native range, *C. taxifolia* occurs mostly in small isolated clumps that reach an average height of 25 cm. In the Mediterranean Sea, however, introduced *C. taxifolia* forms dense "astroturf-like" mats with a height of up to three feet, and up to 213 m of stolon growth and 5,000 emerging fronds per square metre (Meinesz 1999; Anderson & Keppner 2001; Yip 2001). *C. taxifolia* produces several types of secondary metabolites (caulerpenyne) that are toxic to potential competitors or grazers belonging to a range of taxa.

7.6.2 Timing of reproduction and recruitment

Little information exists on the reproduction of *C. taxifolia*. Reproduction in native tropical populations can occur sexually during a short period of the year by synchronised (light intensity) release of anisogamous gametes and formation of zygotes (Zuljevic & Antovic 2000). However, Mediterranean and other introduced populations appear to be able to produce only male gametes, and are thus not capable of sexual reproduction. Therefore, reproduction and dispersal of *C. taxifolia* in the introduced range appear to be solely vegetative (asexual) or by fragmentation (Smith & Walters 1999; Anderson & Keppner 2001; Ramey 2001). *C. taxifolia* is pseudoperennial, with highest rates of stolon growth (up to 8 cm day^{-1}) in summer and autumn, followed by a short resting period from January to April (GISP 2002; Neill 2002). Successful recruitment of dispersed fragments of *C. taxifolia* (as small as 10

mm) can occur throughout the year, but establishment probabilities are highest during summer (Ceccerelli & Cinelli 1999).

7.6.3 Habitat and biology

Substratum type

Caulerpa taxifolia occurs on all types of substrata in both native and introduced range. The alga flourishes equally well on rocky, sandy, mud or clay substrata, both in sheltered and exposed conditions, and in polluted and pristine waters (Meinesz *et al.* 1993; Williams & Grosholz 2002). Dense mats of *C. taxifolia* in the Mediterranean smother other benthic biota, including corals, sponges, and other seaweeds (Meinesz 1999; Neill 2002). *C. taxifolia* can adjust its growth strategy to suit the type of substratum available. For example, in the San Diego population, upright fronds developed adventitious rhizoids and stolons when lying on sediments, and stolons when entwined within existing algal canopy (Williams & Grosholz 2002).

Food preferences

Caulerpa taxifolia occurs in both polluted and nutrient-poor (e.g. the Mediterranean) habitats (Meinesz 1993). The rhizoid system is used to take up major nutrients from the substratum (Anderson & Keppner 2001), and the extensive biomass of *C. taxifolia* mats acts as a vast nutrient trap (P and N) (Yip 2001). Non-native populations of *C. taxifolia* lack severe nutrient (P and N) limitation (Delgado *et al.* 1996), which may be an important factor enabling it to out-compete native macrophytes.

7.6.4 Physiological tolerances (range & preferences)

Temperature

Mediterranean (introduced) populations of *C. taxifolia* have a temperature range of 9 – 32.5 ^{0}C. Some reports claim observation of live plants at 5 ^{0}C (Makowka 2000). Survival without growth occurs at temperatures of 10 – 12.5 ^{0}C; frond and stolon development commence at 15 and 17.5 ^{0}C, respectively, with optimum growth occurring at 25 ^{0}C (Gillespie *et al*. 1997; Komatsu *et al.* 1997). The lower temperature tolerance limit is thought to occur only in introduced strains, and to have developed during decades of aquarium-breeding. It is common opinion that *C. taxifolia* within the native range do not grow in water colder that 20 ^{0}C (Meinesz & Boudouresque 1996). However, recent research from eastern Australia showed that native

populations are able to survive temperatures of 11 ^{0}C for a period of four weeks, and that a temperature of 13 ^{0}C is sufficient to maintain existing tissue biomass (Chisholm *et al.* 2000). Maximum growth occurs at > 20 ^{0}C (Komatsu *et al.* 1997).

Depth

Dense mats of *C. taxifolia* commonly occur at depths of 1 – 30 m, but the alga is known to occur down to a depth of ~ 100 m (Meinesz 1999; Anderson & Keppner 2001; Yip 2001). See "*Light*" (below) for more information.

Salinity

No specific information on *C. taxifolia*'s salinity tolerance range exists in the literature. Populations in the San Diego area were sampled at 34 psu (Williams & Grosholz 2002). Congeners of *C. taxifolia* are able to grow at salinities of 10 – 40 psu (*C. racemosa*; Carruthers *et al.* 1993) and 15 – 50 psu (*C. lentillifera*; Liao & Cheng 1989).

Light

Stolon and frond growth occur at very low light levels (27 μmol m^{-2} s^{-1}); the optimal light intensity ranges from 88 to 338 μmol m^{-2} s^{-1} (Mediterranean population; no upper irradiation limit established; Komatsu *et al.* 1997). Other studies report highest growth rates at an irradiance of 75 μmol m^{-2} s^{-1} (Gillespie et al. 1997). *C. taxifolia*'s annual productivity pattern is less affected by fluctuations in light and temperature than what has been reported from endemic seaweeds (Gacia *et al.* 1996). Photosynthetic assays suggest depth limits for colonisation at 80 m (clear water) and 50 m (turbid water) (Gacia *et al.* 1996). Mediterranean *C. taxifolia*'s maximum photoautotrophic growth limit was determined as 24 m during winter. Although this correlates reasonably with the distribution of dense populations on the Monaco coastline, the limit is greatly inferior to the maximum reported depth of ~ 100 m, and implies significant heterotrophic carbon acquisition at depths much greater than 24 m (Chisholm & Jaubert 1997).

7.6.5 Methods of sampling

There appears to be no single "best" sampling method for *C. taxifolia* due to its occurrence on a range of substrata. Sampling methods that have been used to detect *Caulerpa* and estimate its abundance include visual transects, video transects, quadrat

surveys (hard and soft substrata), grab samples (soft bottom) or sled samples (soft bottom).

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7.7 Japanese kelp (*Undaria pinnatifida*)

7.7.1 General information

Undaria pinnatifida is a brown seaweed (order Laminariales) that is native to the Japan Sea and the northwest Pacific coasts of Japan and Korea (CRIMP, 2000). In its native region, *U. pinnatifida* is a popular food item ("wakame") and is cultivated on submerged rope lines. Introduced populations of *U. pinnatifida* occur on the Mediterranean and Atlantic coasts of France, Spain and Italy, on the south coast of England, on parts of the coastline of Tasmania and Victoria (Australia), off southern California, and around most of New Zealand and parts of Argentina (K.A. Miller - www.sdoceans.org/news/undaria.htm; Fletcher & Farrell 1999).

Adult *U. pinnatifida* plants reach an overall length of 1-3 m, and are easily distinguished from other kelp species by a convoluted spore-bearing structure (the sporophyll) that forms on the stem of the mature plant, between the holdfasts and the blade (Centre for Research on Introduced Marine Pests 2000). *Undaria's* life cycle alternates between microscopic stages (spores and gametophytes) and the visible kelp stage known as the sporophyte (Sinner *et al.* 2000).

7.7.2 Timing of reproduction and recruitment

Within its native range, *U. pinnatifida* exhibits a strongly defined annual growth cycle. Sporophytes grow through winter, mature in early/mid spring (at a length of about 400 mm - Campbell & Burridge 1998), reach maximum size in late spring/early summer, and die back as spores are released (up to 100 million per plant) in late summer. Spore viability ranges from 5 to 14 days (Forrest *et al.* 2000). In contrast to native Asian populations, those from New Zealand have no autumnal hiatus between sporophyte generations: sporophytes are always present (Hay & Villouta 1993). Following settlement, spores form microscopic male and female gametophytes. As seawater temperatures drop, fusion of egg and sperm produced by the gametophytes give rise to the next season's sporophytes. The maximum life span recorded in the field is 7.5 mo, with the majority of plants reaching an age of 6 mo (Castric-Fey *et al.* 1999). Highest growth rates occur both in the spring rise and autumnal fall of temperature and illumination. Density of plants can be high: 140-150 plants m^{-2} were observed in Port Phillip Bay, Australia, and New Zealand (Hay & Villouta 1993; Campbell & Burridge 1998). Differences in seasonality occur between habitats in the northern and southern hemispheres, caused by latitudinal differences in annual fluctuations in sea temperature (Sinner *et al.* 2000).

7.7.3 Habitat and biology

Substratum type

Undaria pinnatifida requires a hard surface for recruitment and growth. It is able to grow well on rocky shores and reefs, rock/shell rubble, and artificial surfaces such as vessel hulls and the vertical sides of pontoons in ports and marinas (Castric-Fey *et al.* 1993; Floc'h *et al.* 1996; Campbell & Burridge 1998; Fletcher & Farrell 1999a). *U. pinnatifida* can also grow well on small and patchy hard substrata: populations discovered near Santa Catalina Island (offshore of Los Angeles, U.S.A.) grew epibiotically on the tubes of *Chaetopterus* worm tubes in soft sediment (K.A. Miller; www.sdoceans.org/news/undaria.htm).

7.7.4 Physiological tolerances (range & preferences)

Temperature

Undaria pinnatifida gametophytes have one of the highest upper survival temperatures (28 – 30^{o}C) of members of the Laminariales (Dieck 1993). Optimal temperatures for the vegetative growth of sporophytes are < 12 ^{o}C, but growth occurs up to 20 ^{o}C. Death of sporophytes occurs at temperatures greater than 23 ^{o}C, and gametophytes die within 10-40 days at 30 ^{o}C (Kim & Nam 1997; Sinner *et al.* 2000). During summer, the optimum water temperature for photosynthesis in *U. pinnatifida* is approximately 23 ^{o}C (Matsuyama 1983). During winter, photosynthetic maximum values are independent of temperatures from 5–20^{o}C (Oh & Koh 1996).

Depth

Introduced populations of *U. pinnatifida* along the European coast occur from the low water mark at neap tides down to a depth of 15 m below the low water mark at spring tides (Floc'h *et al.* 1996). *U. pinnatifida* plants off the coast of southern California (Santa Catalina Island) occur to a depth of 26 m, with highest abundances (0.47 ind. m^{-2}) at 24 m (K.A. Miller; www.sdoceans.org/news/undaria.htm). In New Zealand, highest densities of *U. pinnatifida* (150 ind. m^{-2}) occur from the low intertidal fringe to about 3 m depth (Hay & Villouta 1993; Sinner *et al.* 2000). Generally, the depth at which *U. pinnatifida* occurs around New Zealand seems limited mainly by light and substrate availability.

Salinity

No detailed information exists on the salinity tolerance of *U. pinnatifida*. However, because *U. pinnatifida* does not occur in Riverton (at the mouth of the Mataura River) is has been suggested that it is likely to be excluded also from other areas with a high freshwater input (Sinner *et al.* 2000). This seems to be supported by the CSIRO Centre for Research on Introduced Marine Pests (CRIMP) in Australia, who suggest freshwater immersion as an effective method for killing *U. pinnatifida* (CRIMP, 2000).

Light

Photosynthetic maxima at a range of ambient temperatures (5 – 20 ^{O}C) occur in March (Oh & Koh 1996). Maximum growth and rapid maturation of gametophytes occurs at a light:dark cycle of 12:12 h and 60 μmol m^{-2} s^{-1}. At 17^{o}C, gametophytes can survive complete darkness (0:24 h L:D) for 210 days without any growth and maturation (Kim & Nam 1997). For sporophytes, lower saturated light requirement and compensation points were observed in spring compared to summer plants. Lower saturated light requirement values of juvenile sporophytes compared to adult sporophytes were found during spring. Spring and summer compensation points in this study mostly ranged from 7.63 to 15.49 μmol m^{-2} s^{-1} (Campbell *et al.* 1999).

7.7.5 Methods of sampling

Quantitative sampling of *U. pinnatifida* sporophytes can be achieved using traditional techniques such as diver or video transects or quadrats. Intertidal populations can be accessed and sampled during spring low tides without entering the water. However, the detection of *U. pinnatifida* on hard substrata, in particular ship hulls, is less straightforward. While an observer may not notice macroscopic stages of *U. pinnatifida* (i.e. sporophytes), microscopic stages (spores and gametophytes) may nevertheless be present at the site or on the vessel (Hay 1990).

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7.8 Clubbed tunicate (*Styela clava*)

7.8.1 General information

The clubbed tunicate, *Styela clava*, is a solitary ascidian native to the northwest Pacific from the Sea of Okhotsk, southern Siberia, Japan, Korea and the coast of China south to Shanghai (Millar 1970, Cohen 2005). It has a club-shaped body, up to 160 mm long, with a distinct stalk and basal disc with which it attaches to the substratum. Small individuals (<30 mm) may lack a stalk (Lützen 1999). The body wall (test) is leathery and variable in colour (commonly brown-white, yellow-brown or red-brown), with conspicuous tubercles on the upper part and longitudinal ridges on the stalk.

Like all ascidians, *Styela clava* is hermaphroditic (but not self-fertile) and gametes are shed into the water column. The "tadpole" larvae peculiar to ascidians are planktonic and hatch from the eggs after ca 12 hr, although the duration of this period varies with egg size and water temperature (Svane & Young 1989, cited in Bourque et al. 2005). The larvae are active for a similar period before settling to the substratum (Holmes 1969, cited in Holmes 1976, Minchin et al. 2006). The larvae do not feed and at first tend to swim upwards, though this behaviour later reverses (Millar 1970).

In those species of ascidians that have been studied, life-spans are generally 12-20 months, although some may live for several years (Millar 1970). Minchin et al. (2006) stated that the size of individual *Styela clava* (75-180 mm) collected in Ireland "suggests that they were between one and two years old", although they did not give any reason for this conclusion. Individuals that settled in the Limfjord, Denmark in mid-August grew to 17-48mm by the end of October (Lützen 1999), after which growth ceased during the colder months. Considerable mortality of smaller individuals also occurred during winter. Survivors reached lengths of 50-75 mm by June and became fully mature, spawning in July and August at 75-95 mm. Many small (12-40-mm-long) individuals were also present in early and mid-summer, representing late settlers from the previous year. These, and some of the larger individuals, probably survive a second winter to reach a length of 110-120 mm and reproduce for a second time aged 1.75-2 years old. The lifespan of individuals in southern England was found to be shorter, only rarely exceeding 15 months (Holmes 1969, cited in Lützen 1999). Death may results from senescence, predation or adverse environmental conditions. Reported predators of juvenile *Styela clava* include gastropods (*Mitrella lunata* in eastern North America) and fish (NIMPIS 2002).

The first recorded occurrences of *Styela clava* outside its native range were at Newport Bay (1932) and Elkhorn Slough (1935, a single specimen and no longer present at this site), both in California (Cohen 2005). It subsequently spread along the Pacific coast of North America, north as far as Puget Sound (collected in 1998) and Vancouver Island (collected in 1994) and south as far as Baja California (collected at Ensenada in 2000). On the east coast of North America, it was collected in Massachusetts in 1970, New York in 1972, Connecticut, Maine, New Hampshire and Rhode Island in the 1980s and, more recently, in New Brunswick and Prince Edward Island (1998) (Cohen 2005).

Styela clava was recorded in southwest England in 1953 (Carlisle 1954, Houghton & Millar 1960, both cited in Eno et al. 1997) and has since spread to northwest England, southwest Scotland and southern Ireland (collected 1972: Minchin & Duggan 1988). It has also been found in France (1968), the Netherlands (1974), Denmark (1978-1979), Germany (1997), Portugal (2003) and Spain (2004) (Lützen 1999, Cohen 2005, Davis & Davis 2005).

The first record of *Styela clava* in Australia was in 1972 in Port Phillip Bay, Victoria (Holmes 1976) and in 1977 it was reported from Sydney Harbour, New South Wales (Cohen 2005). It was first recorded in New Zealand in the Viaduct Harbour, Auckland in August 2005 and there appear to be well-established populations in the Waitemata Harbour, Hauraki Gulf and Firth of Thames (Gust et al. 2006a). More localised populations have also been found in Lyttelton Port, Lyttelton Marina, Tutukaka and Opua Marinas (Northland) (Gust et al. 2006a and b) and Nelson Port (Morrisey et al. 2006).

7.8.2 Timing of reproduction and recruitment

Reproduction is usually restricted to warmer seasons in ascidians living in temperate and cold seas (Millar 1970). Holmes (1969, cited in Holmes 1976) reported that *Styela clava* bred throughout all but the coldest 2-3 months in southern England, with a marked peak of settlement in mid-late summer (late July-early September). A similar pattern of settlement was observed in the Limfjord, Denmark (Lützen & Sørensen 1993, cited in Lützen 1999). Monthly sampling of *S. clava* in southern Ireland (Parker et al. 1999) showed gametogenesis (presence of ripe gametes in the gonads) from February-November, with a peak in August-October, and spawning in September-October (when average water temperatures were 15.2^{o}C (±0.4 SD) – 14.1^{o}C (±1.3 SD)).

Spawning in ascidians generally occurs in response to a period of light following a period of darkness (Svane & Young 1989, cited in Bourque et al. 2005). The rapidity of response to this period of light varies among species and, therefore, not all species spawn at the same time of day. Time of spawning may also vary among populations of the same species from different locations (Bourque et al. 2005). In *Styela plicata*, the duration of the light period required to stimulate spawning decreases with increase in the preceding period of darkness (West & Lambert 1976, cited in Bourque et al. 2005). Light intensity may also affect the duration of the light period prior to spawning (Forward et al. 2000, cited in Bourque et al. 2005). Bourque et al. (2005) found that concentrations of larvae of *Styela clava* in the upper 1-m of the water column at a field location in Prince Edward Island, eastern Canada, peaked around noon. They pointed out, however, that timing of peak concentrations of larvae may vary among locations and over time at the same location, in response to factors such as day-length, water temperature and light intensity. Cohen 2005 and ISSG Global Invasive Species Database 2006 indicate that *Styela clava* is only able to spawn at water temperatures above 15^{o}C and salinities above 25-26 psu (no sources are given for this information).

Larvae of *Styela clava* do not usually travel more than a few centimetres by active swimming (Minchin et al. 2006). Consequently they tend to congregate close to the parent population, although they can be passively dispersed over distances covered by 1-2 tidal excursions (equivalent to the duration of the larval period). Larvae are negatively buoyant but negatively geotactic and positively phototactic, particular at higher hydrostatic pressures, and consequently tend to settle near the water surface (Davis 1997, cited in Minchin et al. 2006). Suitable conditions for establishment occur in sheltered localities with salinities of >22 psu and temperatures ≥16^{o}C for several weeks (Minchin et al. 2006). Individuals apparently reach maturity at 3-10 months (Cohen 2005).

7.8.3 Habitat and biology

Styela clava occurs in low wave-energy environments and sheltered embayments from the upper sublittoral zone to at least 25 m depth (ISSG Global Invasive Species Database 2006). It is especially abundant 10-200 cm below the sea surface (Lützen 1999), and the fact that it has been recorded up to 30 cm above the level of extreme low water of spring tides in southern England (Holmes & Coughlan 1975, cited by Lützen 1999) suggests that it is able to withstand a degree of regular exposure to air. It can apparently survive for up to 3 days out of water under cool, damp conditions (Lützen & Sørensen 1993, cited in Minchin et al. 2006). Based on a survey of the distribution of *S. clava* in harbours of the Southern Californian Bight, Lambert &

Lambert (2003) noted that the species was consistently more abundant closer to the entrances to bays, where water currents were stronger and that it differed from *S. plicata* in this respect.

Substratum type

Natural substrata for attachment of *Styela clava* include rocks, the blades of macroalgae and the shells of live and dead bivalves (Lützen 1999, NIMPIS 2002, Bourque et al. 2005). *S. clava* is also found on a range of artificial structures, including floating pontoons, tyre fenders, vessels, buoys and anchors, and diverse materials, including concrete, cement, wood, ropes and the steel or fibreglass hulls of vessel (Bourque et al. 2005, Gust et al. 2005, 2006a, ISSG Global Invasive Species Database 2006, Minchin et al. 2006). In a survey of harbours in southern California, Fay & Johnston (1971, cited in Lambert & Lambert 2003) recorded *Styela clava* only on floats and pilings and not on any natural substrata.

According to Holmes (1976), *Styela clava* colonises only those surfaces bearing a well-developed epibiota. It can attach to larger individuals of its own species and individual *S. clava* may be extensively fouled with smaller tunicates of their own or other species, algae, sponges, hydroids and bryozoans (Lützen 1999, Cohen 2005, Minchin et al. 2006).

On natural substrata, such as rocks or bivalve shells, *Styela clava* is reported to reach population densities of 50-100 m^{-2} (Lützen 1999). On artificial substrata, however, much higher densities have been reported (500-1500 m^{-2}: Holmes 1976, NIMPIS 2002).

In New Zealand *Styela clava* has been found attached to floating pontoons, wooden pier piles, suspended mooring lines and vessel hulls (Gust et al. 2006a). It has also been reported attached to dead bivalve shells on a muddy shore in the Tamaki Estuary, Auckland (Chris Hickey, NIWA, pers. comm.).

Food preferences

Styela clava is a suspension feeder, feeding on suspended, particulate matter, such as phytoplankton, zooplankton and organic detritus, filtered from water pumped through its branchial sac.

7.8.4 Physiological tolerances (range and preferences)

Temperature

Styela clava is reportedly able to tolerate temperatures ranging from –2 to 23°C (Minchin et al. 2006). Holmes (1969, cited in Holmes 1976) described a population living in southern England, where water temperature raged from 2-23°C, and breeding in all but the coldest 2-3 months of the year. On the Pacific coast of North America it has been found at water temperatures ranging from 11-27°C (Cohen 2005). Larvae are able to survive temperatures from 10 - 30°C (Boothroyd et al. 2003).

Parker et al. (1999) reported no evidence of gametogenesis in individuals sampled in early February in southern Ireland, when the water temperature was 3-4°C, but small numbers of ripe gametes in individuals sampled in the middle of the same month, when the temperature had risen to 8°C. There was evidence that gonad maturation occurred at temperatures below 8°C. Gametogenesis and spawning peaked in August-October, when water temperatures ranged from 14-18°C.

Depth

The reported depth range for *Styela clava* ranges from just above the level of extreme low water of spring tides (in southern England: Holmes & Coughlan 1975, cited by Lützen 1999) to at least 25 m (NIMPIS 2002). Lützen (1999) described *S. clava* as a "predominantly littoral species, which is especially abundant 10-200 cm below the sea surface in areas without tides or when attached to floating objects….The species may extend to depths of 15-25 m…but a record of 40 m depth…is probably exceptional".

Salinity

Styela clava appears to avoid areas with estuarine conditions (Lützen 1999). Sims (1984, cited in Lützen 1999) found that Californian specimens showed poor vital functions after 3-d immersion in 26.5 psu seawater. This corresponds with Lambert & Lambert's (2003) observation of die-offs of *S. clava* on floating structures in southern California after heavy rain (followed by rapid recolonisation). They also cited an earlier study (MacGinitie 1939) in the same area that found complete mortality of *S. clava* below a sharp halocline that formed at a depth of ca 2.2 m following heavy rain. Below this depth there was no evidence of any mortality. Individuals can, however, survive shorter periods of salinity as low as 8 psu, presumably by closing their siphons (Sims 1984, cited in Lützen 1999).

Other populations of *Styela clava* may be more tolerant of lower salinities than those studied in California. In the eastern Limfjord (Denmark), populations exist in salinities averaging 26-28 psu, with decreases to <20 psu for periods of several days (Lützen 1999). Individuals experimentally exposed to stepwise decreases in salinity from 31-18 psu showed >50% survival for 40 d (at 12°C) and 50% survival when the salinity was further reduced to 16 psu (Lützen & Sørensen 1993, cited in Lützen 1999). Lützen (1999) cited a report that larvae of *S. clava* from the Sea of Japan were able to complete metamorphosis at salinities of 20-32 psu, but that <18 psu was "deleterious" (no definition given). Cohen (2005) stated that adult *S. clava* die in salinities <10 psu, but did not give a source for this information.

In summary, salinity tolerance of adults and larvae appears to extend as low as 18 psu for extended periods (and much lower for short periods), but may be dependent on the salinity regime to which the population has previously been exposed.

7.8.5 Route of introduction

Styela clava may have reached the Pacific coast of North America as fouling on ships' hulls, but it may also have been introduced as fouling on imported live oysters (Cohen 2005). It is known to occur on oysters (*Crassostrea gigas*) in Japanese oyster farms, and oysters from Japanese farms were transplanted to Elkhorn Slough (California) in 1929-1934, roughly coincident with its date of first detection in California (1932). From Elkhorn Slough it could have been transported to other parts of California as fouling on coastal shipping or via further transfer of oyster stock (including its recent appearance in Humboldt Bay: Cohen 2005).

The introduction of *Styela clava* to southern England is commonly ascribed to fouling on naval vessels returning from the Korean War in 1952 (Minchin & Duggan 1988, cited in Minchin et al. 2006), having acquired fouling in the Yellow Sea. It is likely to have spread from the original site of introduction to other parts of the United Kingdom and continental Europe on coastal shipping or, locally, by dispersal of eggs and larvae (Lützen 1999). It has also been suggested that *S. clava* reached the Danish coast, where it was first recorded on an oyster bed in the Limfjord, attached to oysters imported from the English Channel and re-laid in the Limfjord (Lützen 1999). Oyster spat imported from Japan in the 1970s, or transplanted within the English Channel region, may have contributed to the establishment of Dutch and French populations (Lützen 1999).

Given the distances involved, the introduction of *Styela clava* to Australia and New Zealand is likely to have occurred via fouling on ships' hulls, either from its native

range or from introduced populations in Europe or North America. In view of the disjunct distribution of *S. clava* in New Zealand's North and South Islands, several inoculation events may have occurred (Gust et al. 2006a). Research is currently underway to determine the genetic relationships among populations of *S. clava* in New Zealand.

Minchin et al. (2006) noted that *S. clava* tend to be stripped from ships' hulls at speeds above ca 5 kt, unless they occur in more protected habitats such as sea-chests, thruster tubes, or in the lee of stabilisers and other structures on the hull. Lützen (1999) also described *S. clava* as rheophobic (i.e. avoiding strong currents), reducing the likelihood of individuals surviving as fouling on exposed parts of the hulls of rapid vessels in continuous service. Attachment to drifting macroalgae provides another potential means of dispersal. Lützen (1999) stated that fronds of *Sargassum muticum* (a macroalga introduced to Europe from Asia in the early 1970s) with *Styela clava* attached are often washed up on shores in the Limfjord. Fronds become detached from their holdfasts towards the end of the growth cycle and can float for "considerable distances".

Davis & Davis (2004) suggested that a combination of transport mechanisms, including translocation on oyster shell, dispersal on flotsam such as drift macroalgae, fouling on vessel hulls, transport of eggs and larvae in ballast water, and fouling of sea-chests are probably required to explain the present distribution of *S. clava*. Davis (2005) suggested that sea-chests were potentially of greatest importance because they offer a means of transport for established colonies of individuals, and translocated colonies are more likely to establish new populations than a single inoculum of larvae.

Slow-moving and towed vessels are particularly likely mechanisms of introduction, because of the reduced likelihood of individuals being removed from the hull by water currents during transit. Such vessels may also spend longer periods moored in ports of origin and destination than vessels in continuous service. Specimens of *S. clava* found on vessels in New Zealand have been on a tug (Lyttelton), recreational launches and yachts (Auckland, including one that subsequently travelled to Waikawa Marina, Picton, where it was found to harbour a single individual) and fishing vessels (Nelson) that had been berthed for long periods of time (possibly months in one case, years in another). Of these, recreational vessels are perhaps the most likely to have been the vector of inoculation in the ports where they were found, as the other types of vessel tend to spend most of their time in their home port.

7.8.6 Methods of sampling

- Lambert & Lambert (2003) sampled harbours by examining the sides and bottom edges of pontoons and vessels in marinas, manually removing clumps of fouling organisms to arms' depth, and recovering 5-m long ropes deployed 4 years previously.

- Minchin et al. (2006) sampled floating pontoons, supporting piles and quay walls by feeling for specimens by hand, or by scraping adhered biota from the surfaces.

- Gust et al. (2005, 2006a,b) employed above-water searches from shore or boat to detect *S. clava* on pontoons, pilings, breakwalls, buoys, heavily-fouled vessels and mooring lines. Submerged ropes were pulled up and examined. Selection of vessels to search was based on a risk-profiling approach based on empirical relationships between level of fouling and probability that the fouling assemblage includes solitary ascidians.

- Gust et al. (2005, 2006a,b) also used in-water diver searches of the undersides of pontoons, wharf piles and breakwalls. For safety reasons, and because previous studies had shown that 70% of all *S. clava* detected were found within this depth, searches were confined to the upper 5 m of the water column.

- The probability of *S. clava* being detected by searchers when it is present can be estimated for each type of substratum in a given harbour. These estimates require information on the proportion of the total area of the substrate searched and the sensitivity of the search method under prevailing environmental conditions, particularly water clarity (Gust et al. 2006a,b). Sensitivity, the ability of the searchers to detect *S. clava* when present, can be determined by searches for experimentally-deployed mimics of the organism. Details of the methods are given in Gust et al. (2006a,b).

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