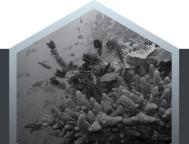




Rapid response manual for *Mytilopsis sallei* and *Perna viridis*

Version 2.0, January 2020



Nationally agreed guidance material endorsed by the Marine Pest Sectoral Committee

© Commonwealth of Australia 2020

Ownership of intellectual property rights

Unless otherwise noted, copyright (and any other intellectual property rights, if any) in this publication is owned by the Commonwealth of Australia (referred to as the Commonwealth).

Creative Commons licence

All material in this publication is licensed under a Creative <u>Commons Attribution 4.0 International Licence</u> except content supplied by third parties, logos and the Commonwealth Coat of Arms.

Inquiries about the licence and any use of this document should be emailed to copyright@agriculture.gov.au.



Cataloguing data

This publication (and any material sourced from it) should be attributed as: Marine Pest Sectoral Committee 2015, *Rapid response manual for 'Mytilopsis sallei'* and '*Perna viridis'*, Department of Agriculture, Canberra, CC BY 4.0. Document modified in December 2018 to meet accessibility requirements.

This publication is available at marinepests.gov.au/what-we-do/publications

Marine Pest Sectoral Committee Secretariat Department of Agriculture GPO 858 Canberra ACT 2601 Email <u>mpsc@agriculture.gov.au</u> Web <u>marinepests.gov.au</u>

Disclaimer

These manuals are part of a series of documents providing detailed information and guidance for emergency response to key marine pest species or groups of pest species.

The manuals are made available on the understanding that the Commonwealth of Australia is not thereby engaged in rendering professional advice. The Commonwealth does not warrant the accuracy, currency or completeness of the guidelines, or their relevance for any particular purpose. In particular, it should be noted that legislation, regulations and by-laws may vary between different jurisdictions and ports in Australia. Consequently, the guidelines do not purport to state what is necessary or sufficient to comply with laws applying in any place.

Before relying on the manuals in any important matter, users should obtain appropriate professional advice to evaluate their accuracy, currency, completeness and relevance for their purposes.

Note

Rapid response manuals are a key element of the Australian Emergency Marine Pest Plan. They provide detailed information and guidance for emergency response to a marine pest incident. The guidance is based on sound analysis and links policy, strategies, implementation, coordination and emergency management plans.

Preface

The Australian Government Department of Agriculture maintains a series of emergency response¹ documents to ensure national coordination of emergency responses to incursions by exotic pests and diseases or significant range expansions of established pests and endemic diseases. The Emergency Marine Pest Plan (EMPPlan) Rapid Response Manuals for marine pests provide detailed information and guidance for emergency response to key marine pest species or groups of pest species of national significance.

The EMPPlan is adapted from the Australian emergency plans for terrestrial and aquatic animal diseases—the Australian Veterinary Emergency Plan (AUSVETPLAN) and the Australian Aquatic Veterinary Emergency Plan (AQUAVETPLAN). The format and content have been kept as similar as possible to those documents to enable emergency response personnel trained in their use to work efficiently with these manuals in the event of a marine pest emergency.

This manual describes the principles for an emergency response to an incident caused by the suspicion or confirmation of incursion by the black striped false mussel, *Mytilopsis sallei*, or the Asian green mussel, *Perna viridis*. As both species are considered exotic marine pests of national concern in Australian waters, they are listed on the <u>Australian Priority Marine Pest List</u>. Both species are highly fecund and form dense populations on marine infrastructure in sheltered tropical coastal waters. Subtropical populations of *M. sallei* and *P. viridis* are also known to occur so these species could also become pests in more temperate waters (APMPL). These marine pests could displace native Australian species and cause serious economic consequences for the aquaculture and maritime industries.

Dr Graeme Inglis and Ms Kimberley Seaward from the National Institute of Water and Atmospheric Sciences, New Zealand, and Ms Amy Lewis from the Department of Agriculture prepared the first edition of this Rapid Response Manual. The manual was revised as part of activity 3.5 of MarinePestPlan 2018-2023 (plan and implement procedures to develop and update the EMPPLIan rapid response manuals and related guidance materials). Changes to the manual include new information on molecular surveillance methods, changes based on experience gained by the *P. viridis* response near Weipa in 2017-18 and updates to biosecurity legislation on policy (*Biosecurity Act 2015*). The Marine Pest Sectoral Committee endorsed the second edition of this manual. The manual will be reviewed at least every five years to incorporate new information and experience gained with incursion management of these or similar marine pests. Amended versions will be published on the marine pest website.

¹ Note that the term 'emergency response' as used in this document does not refer to a 'biosecurity emergency' as that term is used under the *Biosecurity Act 2015*, nor do any activities described by this document undertaken during an 'emergency response' intended to be an exercise of powers provided by Chapter 8 (Biosecurity Emergencies and Human Biosecurity Emergencies) of that Act.

Recommendations for amendments

To recommend changes to this document, forward your suggestions to:

Marine Pest Sectoral Committee Secretariat Department of Agriculture GPO 858 Canberra City ACT 2601 Email <u>mpsc@agriculture.gov.au</u>

Contents

Pref	ace		iii	
	Recom	mendations for amendments	iv	
Intro	oductio	n	1	
1	Nature	of the pest	2	
	1.1	Mytilopsis sallei	2	
	1.2	Perna viridis	9	
	1.2	Summary of life history information1	16	
2	Pest pa	athways and vectors	17	
3	Policy	and rationale for incursion response	19	
	3.1	Generic policy for incursion response to marine pests in Australian waters	۱9	
	3.2	Control and eradication strategy for <i>M. sallei</i> and <i>P. viridis</i>	23	
	3.3	Policy on decision points	23	
	3.4	Policy on funding of operations and compensation	24	
4	Princip	les for containment, control and eradication	25	
	4.1	Methods for preventing spread of the organism	25	
	4.2	Tracing an incursion	38	
5	Contro	lling, eradicating and treating established populations	41	
	5.1	Eradication	11	
	5.2	Containment and control	11	
	5.3	Guidelines for delimiting surveys	12	
	5.4	Design of a delimiting survey	12	
6	Metho	ds for treating established populations	14	
	6.1	Closed or semi-enclosed coastal environments	14	
	6.2	Open coastal environments	19	
	6.3	Monitoring and ongoing surveillance	19	
		: Guidelines for using the Biosecurity Act during an emergency response to a marine onal significance	51	
Арр	endix B	: State and territory legislative powers of intervention and enforcement	53	
Арр	endix C	: Spat collection devices for <i>M. sallei</i> and <i>P. viridis</i>	55	
Glos	Glossary57			
Refe	References			

Tables

Table 1 Taxonomy of <i>Mytilopsis sallei</i>	3
Table 2 Categories of potential impact caused by <i>Mytilopsis sallei</i>	8
Table 3 Taxonomy of Perna viridis	9
Table 4 Categories of potential impact caused by Perna viridis	15
Table 5 Mytilopsis sallei and Perna viridis life history summary	16
Table 6 Pathways and vectors for <i>Mytilopsis sallei</i> and Perna viridis	18
Table 7 Management recommendations for different types of vectors	32
Table 8 Treatments achieving total mortality (LD100) of <i>Mytilopsis sallei</i> and <i>Perna viridis</i> in laboratory conditions	37
Table B1 State and territory legislation covering emergency response arrangements	53

Figures

Figure 1 External diagnostic features of <i>Mytilopsis sallei</i>	4
Figure 2 Internal diagnostic features of <i>Mytilopsis sallei</i>	5
Figure 3 Internal diagnostic features of Perna viridis	10
Figure 4 High-risk niche areas for inspection of biofouling on vessels less than 25 metres	30
Figure 5 High-risk niche areas for inspection of biofouling on vessels greater than 25 metres	30
Figure C1 Settlement collector used to monitor fouling bivalve pests	56

Photographs

Photo 1 Adult <i>Mytilopsis sallei</i> individual	2
Photo 2 Adult <i>Mytilopsis sallei</i> infestation	3
Photo 3 Adult <i>Perna viridis</i>	9

Maps

Map 1 Global distribution of <i>Mytilopsis sallei</i>	7
Map 2 Global distribution of Perna viridis	14

Introduction

Emergency response operations are most efficient if they are based on detailed knowledge of the life history, biology, ecology and susceptibility of the pest species to eradication and control measures. Species-specific <u>rapid response manuals</u> have been prepared for several marine pests that the Marine Pest Sectoral Committee (MPSC) has identified as being of national concern.

During an emergency response, detailed technical information must be collected in the investigation phase of the response. At a minimum, information will be needed on:

- the nature of the pest, including its:
 - taxonomy
 - known distribution (global/Australian, native/non-native)
 - life history and ecology
 - environmental tolerances
 - impact potential
- pathways and vectors by which the species may be spread
- methods to prevent spread of the organism
- methods for undertaking surveys to
 - delimit established populations
 - trace an incursion
 - monitor the effectiveness of management measures
- methods to control or eradicate pest populations in different marine environments
- federal, state and territory legislation and policy relevant to emergency responses.

This information must be assembled rapidly from reliable sources. Preference should be given to using primary sources of information, such as advice from scientists, engineers or other professionals with recognised expertise on the species or likely emergency operations, and from published, peer-reviewed literature. Reputable secondary sources of information, such as internet databases and 'grey' literature may be used to supplement this advice or to prepare summary information and plans for expert review.

This document provides guidance on:

- types of information needed to determine an appropriate response to the suspicion or confirmation of incursion by *Mytilopsis sallei* or *Perna viridis*.
- types of expert advice that may need to be sought
- potential sources of information for preparing a response plan
- appropriate methods for containment, control and/or eradication of established populations.

1 Nature of the pest

Understanding the life history, ecology and biology of a marine pest is fundamental to an effective emergency response. Detailed knowledge of a species allows better evaluation of the threat it is likely to pose, the feasibility of response options and the design of efficient methods for surveillance, containment, eradication and control.

1.1 Mytilopsis sallei

The black striped false mussel, *Mytilopsis sallei* (Récluz, 1894), is a small bivalve mollusc that fouls a variety of substrata in sheltered tropical and subtropical coastal waters (Photo 1). It is capable of recruiting in very large densities, causing massive fouling on wharves and in marinas, seawater systems (pumping stations, vessel ballast and cooling systems) and marine farms (Photo 2). It can form dense mono-specific stands in natural habitats that exclude most other species, leading to a substantial reduction in biodiversity.

M.sallei is listed on the <u>Australian Priority Marine Pest List</u> as a nationally significant marine pest species.

Photo 1 Adult Mytilopsis sallei individual



Source: CSIRO

Photo 2 Adult Mytilopsis sallei infestation



Source: Northern Territory Department of Primary Industry and Resources

Table 1 Taxonomy of Mytilopsis sallei

Classification	Mytilopsis sallei
Phylum:	Mollusca
Class:	Bivalvia
Subclass:	Heterodonta
Order:	Veneroida
Super family:	Dreissenoidea
Family:	Dreissenidae
Genus:	Mytilopsis

Dreissenid bivalves have been reclassified many times, at many taxonomic levels. Until recently, *Mytilopsis* was considered a subgenus of *Congeria* and some publications may still refer to *Mytilopsis sallei* under the junior synonym *Congeria sallei*. Although up to nine 'species' of *Mytilopsis* have been identified (Marelli & Gray 1985; Therriault et al. 2004), taxonomic classification of the genus is complex, as there is large intraspecific and limited interspecific morphological variation (Morton 1981). *M. sallei* and *M. leucophaeata* are considered the only extant species in this genus (Therriault et al. 2004).

The dark false mussel, *M. leucophaeata*, also has a history of invasion associated with global shipping. It is native to the Gulf of Mexico, but extralimital populations have been recorded in the north-eastern United States and in the Upper Mississippi River; along the North Sea coasts from Germany to France; and in the River Thames estuary, England (Therriault et al. 2004). European populations occupy both freshwater and brackish estuary habitats.

Diagnostic features for identification

Mytilopsis sallei can be identified in the field and in the laboratory.

1.1.1.1 Field identification

Mytilopsis sallei is a relatively small mussel that grows to an average maximum length of 25 mm. The shell is elongated, and the average ratio of length to height to width is 2.5 to 1.3 to 1 (Morton 1981).

The exterior of the shell is varied in colour; it can appear white, cream-coloured or bluish-grey to a medium brown or black (McEnnulty et al. 2000). Some specimens may have fine concentric lines. Shells of small specimens occasionally bear brown zigzag markings (Marelli & Gray 1985). The shell is thin and easily crushed. The shell valves of *M. sallei* are slightly unequal in size with the left valve fitting inside the right one (Tan & Morton 2006) (Figure 1). *M. sallei* can settle in large densities with individuals attaching by secreted byssal threads.

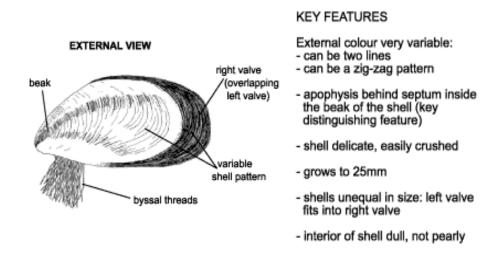


Figure 1 External diagnostic features of Mytilopsis sallei

Image: CSIRO

1.1.1.2 Laboratory identification

A key diagnostic feature for *Mytilopsis sallei* is the shape and position of the apophysis; a peg-like structure located inside the beak of the shell that is used to support the interior retractor muscles (Figure 2). In *M. sallei*, the apophysis is located lateral to the septum near the dorsal margin of the shell. The apophysis is well-developed and not closely associated with the septum. It extends laterally well into the shell cavity, and posteriorly to twice the length of the septum, generally becoming pointed postero-ventrally and appearing hook shaped; the apophysis in this species is generally closely associated with the hinge plate (Marelli & Gray 1985).

Department of Agriculture

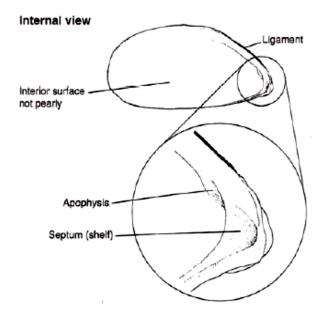


Figure 2 Internal diagnostic features of Mytilopsis sallei

Image: Stafford & Willan 2007

1.1.1.3 Sample collection and preservation for molecular diagnostics

A molecular assay for *Mytilopsis sallei* has been validated for northern Australian waters to minimise issues with cross reactivity with native species in tropical waters.

Gametes, juvenile and mature stages can be used for molecular analysis to assess the presence of target nucleic acid. Sampling methods will vary depending on the life history stage that is being targeted (see 5.4 for more information on sampling methods).

Samples for molecular analysis require specific preservation techniques to preserve DNA or RNA for analysis. The three most common methods for preservation of mussels are:

- Rapid freezing of biological material (flash freezing in liquid nitrogen) with subsequent storage at -80°C and transport in liquid nitrogen or on dry ice.
- Preservation of biological material in >70% analytical grade (not denatured) ethanol.
- <u>RNAlater</u>[®] (Sigma-Aldrich) or other nucleic acid preservation products such as DNA/RNA Shield[™] (Zymo Research) or other nucleic acid preservatives.

Contact your testing laboratory for more information regarding preservation and analysis of samples using modern diagnostics.

Life history and ecology

Understanding the ecology of *Mytilopsis sallei* involves examination of its reproduction, growth and life habit (Table 1Table 5).

1.1.1.4 Reproduction and growth

Mytilopsis sallei is a broadcast spawner. Sperm and eggs are released into the water column where external fertilisation takes place. *M. sallei* can change sex throughout its life so, at any time, a proportion of the population may be hermaphrodites (Karande & Menon 1975). In its native range, some individuals are reproductively viable throughout the year, and in Hong Kong, mass spawning

typically coincides with rapidly changing salinity levels associated with seasonal influxes of fresh water (Morton 1981). Individuals are extremely fecund, releasing tens of thousands of eggs during spawning.

Fertilised eggs develop into pelagic larvae that settle on hard substrata within a day of fertilisation. Growth is rapid and sexual maturity can occur within a month of settlement (Morton 1989). Maximum size is reached within six months of settlement and individuals live for an average of 12 to 13 months (maximum 20 months).

1.1.1.5 Life habit

Mytilopsis sallei is a suspension feeder that filters zooplankton, phytoplankton and other suspended particulate organic matter from the water column. It is an epibenthic species that attaches by a byssus thread to various substrates. It is found predominantly in sheltered intertidal and shallow subtidal habitats and is rarely recorded deeper than a few metres below mean low water springs (maximum approximately 2.5 m). Because of its high fecundity and short larval life, it is often found in dense aggregations, consisting of many hundreds or thousands of individuals per m².

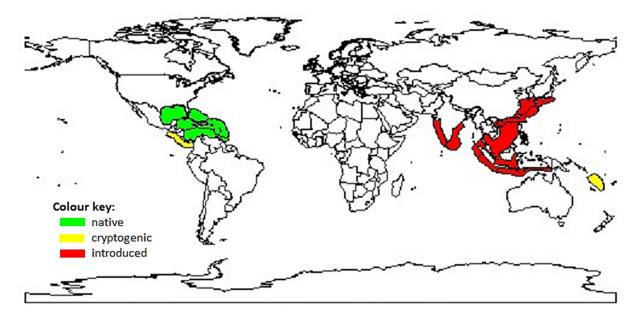
M. sallei has a broad tolerance of salinity and water temperature; it can survive temperatures of between 10°C and 35 °C and can withstand salinity levels of between zero and 80 ppt (Morton 1981). *M. sallei* is particularly suited to living in brackish waters and can withstand extended exposure (days to months) to very low salinities (0–27 ppt).

No reliable published data are available on tolerance of *M. sallei* to desiccation. Aerial exposure of zebra mussels, *Dreissena polymorpha*, a related dreissenid bivalve, causes 100% mortality after two days at 25 °C (Heimowitz & Phillips 2006). Its relatively thin-shelled valves suggest that *M. sallei* is also susceptible to prolonged periods of exposure to the air. However, unlike *D. polymorpha*, *M. sallei* occurs both intertidally and subtidally and may be regularly emersed for short periods of time (hours), so it may be more tolerant than *D. polymorpha* of exposure to the air.

1.1.2 Global distribution

Mytilopsis sallei is native to the West Indies, the Caribbean coast of Central and South America from Yucatan to Venezuela, and part of the southern peninsula of Florida in the United States. It has been introduced to Hong Kong (1980s), Taiwan (1977), Japan (1974), possibly Fiji, India, Indonesia (1967), and Singapore (Map 1). A population of *M. sallei* was discovered in Darwin, Australia, in 1999, but was successfully eradicated (Bax et al. 2002).

Subsequent detections of *M. sallei* have occurred in Darwin. Specimens have been found on the hulls of apprehended foreign fishing vessels (2000, 2005 and 2006), suspected illegal entry vessels (2010) and in the seawater systems of yachts (2006, 2007 and 2010). No populations are known to have established in Australia (Map 1).



Map 1 Global distribution of Mytilopsis sallei

Cryptogenic Unknown origin, may be native or introduced. Source: NIMPIS 2002

1.1.3 Potential impact

The high fecundity, short larval phase and rapid maturity of *Mytilopsis sallei* mean it can attain very large densities (more than 23,000 individuals per m²), rapidly becoming the dominant fouling organism in intertidal and shallow subtidal habitats (Bax et al. 2002). The dense aggregations cause heavy fouling on vessel hulls, chains, ropes, nets, mooring buoys, piles, floating pontoons, piping and other surfaces. Table 1 outlines the potential social, economic and environmental impacts caused by *M. sallei*.

Impact category	Description	Potential impact
Social amenity	Human health	No
Economy	Aquatic transport	Yes
	Water abstraction/nuisance fouling	Yes
	Loss of aquaculture/commercial/recreational harvest	Yes
	Loss of public/tourist amenity	Yes
	Damage to marine structures/archaeology	Yes
Environment	Detrimental habitat modification	Yes
	Alters trophic interactions and food-webs	Yes
	Dominates/out-competes and limits resources of native species.	Yes
	Predation of native species	No
	Introduces/facilitates new pathogens, parasites	No
	Alters bio-geochemical cycles	No
	Induces novel behavioural or eco-physical responses	No
	Genetic impacts—hybridisation and introgression	No
	Herbivory	No

Source: Hayes et al. 2005

The population of *M. sallei* discovered in Cullen Bay Marina, Darwin reached densities of more than $23,000/m^2$ in the five or six months it was present (Bax et al. 2002). In India, populations can reach a biomass of up to 100 kg/m^2 in a year (Rao et al. 1989).

Industrial structures for seawater intake, cooling intakes for vessels and aquaculture facilities are particularly susceptible to fouling from *M. sallei* (URS 2004). Dense aggregations may also smother or exclude other fouling species, altering natural biotic assemblages.

1.2 Perna viridis

The Asian green mussel, *Perna viridis* (Linneaus, 1758), is a large bivalve mollusc that fouls hard substrata in tropical coastal estuaries (Photo 3). In its natural range, it forms the basis of an important aquaculture industry and wild fishery. It is capable of recruiting in very large densities on a variety of fixed and floating hard substrata, including vessels, wharves, mariculture equipment, buoys, rocks, shells and reefs. *P. viridis* can form dense stands, precluding native habitat-forming species from the available substratum.

Photo 3 Adult Perna viridis



Source: Department of Primary Industries and Fisheries, Queensland

Table 3 Taxonomy of Perna viridis

Classification	Perna viridis	
Phylum	Mollusca	
Class	Bivalvia	
Subclass	Pteriomorphia	
Order	Mytiloida	
Super family	Mytiloidea	
Family	Mytilidae	
Genus	Perna	

Note: The genus Perna has only three species: P. viridis, P. perna and P. canaliculus.

Department of Agriculture

Diagnostic features for identification

Perna viridis can be identified in the field and in the laboratory.

1.1.3.1 Field identification

Perna viridis is a large mussel that grows to an average length of between 80 and 100 mm (maximum 165 mm). The external colour of the shell ranges from vivid green to varying dark green/brownish shades. The shell tapers to form a sharp downturned beak and the ventral margin of the shell is straight or weakly concave (NIMPIS 2002b).

1.1.3.2 Laboratory identification

The interior of the shell is pearly with shades of shiny blue-green. The ridge that supports the ligament connecting the two shell valves is finely pitted. The beak has interlocking teeth; one in the right valve and two in the left. The curved posterior end of the pallial line and the large kidney-shaped posterior adductor muscle scar are diagnostic features of this species (Figure 3). The shape of the posterior adductor muscle, the pallial line and the concave ventral margin distinguish *Perna viridis* from the two other species in this genus (*P. canaliculus* and *P. perna*). Molecular methods for the detection of *Perna spp*. are available, and have a sufficiently high sensitivity to detect *P. viridis* from larval stages or tiny settled juveniles (Blair et al. 2006).

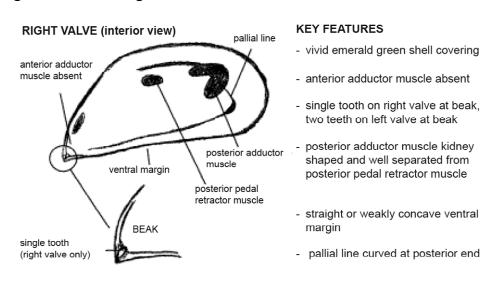


Figure 3 Internal diagnostic features of *Perna viridis*

Image: CSIRO Marine Research

1.1.3.3 Sample collection and preservation for molecular diagnostics

At the time of writing, a molecular assay for *Perna viridis* was undergoing validation for northern Australian waters to minimise issues with cross reactivity with native species in tropical waters.

Gametes, juvenile and mature stages can be used for molecular analysis to assess the presence of target nucleic acid. Sampling methods will vary depending on the life history stage that is being targeted (see 5.4 for more information on sampling methods).

Samples for molecular analysis require specific preservation techniques to preserve DNA or RNA for analysis. The three most common methods for preservation of mussels are:

- Rapid freezing of biological material (flash freezing in liquid nitrogen) with subsequent storage at -80°C and transport in liquid nitrogen or on dry ice.
- Preservation of biological material in >70% analytical grade (not denatured) ethanol.
- <u>RNAlater</u>[®] (Sigma-Aldrich) or other nucleic acid preservation products such as DNA/RNA Shield[™] (Zymo Research) or other nucleic acid preservatives.

Contact your testing laboratory for more information regarding preservation and analysis of samples using modern diagnostics.

1.1.4 Life history and ecology

Understanding the ecology of *Perna viridis* involves examination of its reproduction, growth and life habit (Table 5).

1.1.4.1 Reproduction and growth

Perna viridis is a broadcast spawner. Sperm and eggs are released into the water column where external fertilisation takes place. The sexes are separate. Spawning generally occurs twice a year between early spring and late autumn. On India's east coast the two main spawning events have been linked to changes in water temperature and food availability. Gonadal development and spawning activity showed peaks in April through June (30.9 °C) and October (31.3 °C) (Morton 1981). However, in Hong Kong spawning activity varies substantially between years, with single and double breeding periods reported in different years (Rajagopal et al. 2006).

Fertilisation occurs approximately 8 hours after spawning and metamorphosis of the planktonic larva into a juvenile mussel occurs after 8 or 10 days. Settlement and byssal thread production occurs between 10 and 20 days after fertilisation at 26 °C (Siddall 1980).

Fast growth rates are exhibited under favourable conditions. The typical minimum size for production of mature gametes is 15–30 mm, at an age of two to three months. The life span of *P. viridis* is typically two to three years. Growth rates are influenced by environmental factors such as temperature, food availability and water movement. First year growth rates vary between locations and range from 49.7 mm a year in Hong Kong to 120 mm a year in India.

1.1.4.2 Life habit

Perna viridis is a suspension feeder that filters zooplankton, phytoplankton and other suspended particulate organic matter from the water column. It is an epibenthic species that attaches by byssal threads to hard substrata. *P. viridis* is the dominant species in many rocky intertidal and shallow subtidal ecosystems, but has also been found fouling wood, concrete, metal, muddy sea bottoms, seagrass beds, and other hard substrata, including vessels, wharves, mariculture equipment and buoys.

P. viridis occurs predominantly in coastal waters (less than 10 m deep), in salinities of between 16 and 33 ppt, and at temperatures of between 11 °C and 32 °C (optimal conditions are 27–33 ppt, 26 °C to 32 °C) (NIMPIS 2002b; Power et al. 2004). It exhibits a wide tolerance for turbid conditions and pollution (Power et al. 2004). Settlement and recruitment tend to be greatest between 1.5 m and 4.0 m below mean high water spring tides (Vakily 1989), but it is capable of surviving at depths greater than 10 m. Densities of juveniles can be as high as 12,000 per m², and adults up to 4,000 per m² (Power et al. 2004).

Field and lab observations of *P. viridis* indicate that aerial exposure to ambient temperatures below 14°C (Baker et al. 2012; Firth et al., 2011) or above 25°C (McFarland et al., 2014) will result in high mortality. Experiments investigating desiccation limits of the brown mussel, *P. perna*, indicate that they have a high tolerance to aerial exposure compared with other mussel species. After seven days of continuous exposure to air, survival was found to be 78% (Branch & Steffani 2004).

1.1.5 Global and Australian distribution

Perna viridis is native to the Northern Indo-Pacific region. It occurs naturally along the Indian and South-East Asian coasts, encompassing the Persian Gulf, India, Malaysia and Indonesia. It is not native to South America, but was recorded in Trinidad in the 1990s and has subsequently spread south as far as Venezuela through natural dispersal and human-mediated pathways. It has also established non-native populations in Jamaica; Florida and Georgia in the United States; and Tokyo Bay in Japan (Map 2) (NIMPIS 2002b; Power et al. 2004; Rajagopal et al. 2006; URS 2004). During the 1970s and 1980s, *P. viridis* was introduced into French Polynesia, New Caledonia, the Cook Islands, Fiji, Samoa and Tonga as a potential species for aquaculture. It is unclear if any extant wild populations remain in the Pacific Islands (Eldredge 1994).

P. viridis has been discovered multiple times in Australian waters, attached to vessels (more than 50 times) or in the environment (indicating local recruitment). Despite numerous detections, surveillance activities have demonstrated no evidence that any populations of *P. viridis* exist in the wild in Australia.

Below are lists of *P. viridis* detections on vessels and in the environment sorted by jurisdiction:

<u>On vessels</u>

Queensland

- 2001 on a barge in Trinity Inlet, Cairns
- 2003 on a vessel in Innisfail
- 2004 on a vessel in Cairns
- 2006 on a heavy lift barge in Gladstone
- 2007 on a naval vessel in Trinity Inlet, Cairns
- 2008 in a vessel's internal seawater systems in the Port of Gladstone
- 2008 detected on a vessel in Cairns
- 2009 on a vessel in dry dock in Brisbane
- 2009 on a vessel in the Port of Bundaberg
- 2009 on a tug boat in Gladstone
- 2009 on a vessel in Brisbane
- 2011 on a vessel in the Port of Cairns
- 2012 and again in 2014 on a naval vessel at the Cairns Naval Base
- 2013 on a vessel in Hay Point
- 2019 on a cruise vessel in Brisbane.

Department of Agriculture

Northern Territory

- 1991, 2005, 2006, 2009 and 2010 specimens were collected from apprehended suspected illegal entry vessels and foreign fishing vessels in Darwin
- 1999 in the seawater systems of an Indonesian-based charter vessel in Darwin
- 2000 on a vessel in Darwin
- 2007 in the bow thrusters of a container vessel in Darwin
- 2008 on an internationally travelled sailing and motor cruiser vessel in Darwin
- 2008 on a tug boat in Darwin
- 2009 among heavy fouling on the hull and sea chest grates of a rig tender in Darwin
- 2009 on the hull and sea chests of a commercial freight vessel in Darwin
- 2009 on a naval vessel in Darwin
- 2009 and 2012 on the hull of a tug boat in Darwin
- 2012 on a naval vessel in Darwin
- 2013 on the hull of naval vessels in Darwin.

Western Australia

- 2006 on a dredge in Dampier
- 2009, 2017 and 2019 on vessels in in the Ports of Fremantle
- 2009 and 2010 on vessels in Dampier
- 2010 on a dredge in Dampier
- 2011 on a vessel in Henderson
- 2011 on multiple occasions on vessels in Rockingham
- 2013 on vessels at Barrow Island
- 2013 on a vessel in Henderson
- 2014 on a vessel in Freemantle
- 2014 on a vessel in Kwinana
- 2014 on a vessel in Dampier
- 2015 on two vessels in Freemantle
- 2015 on a tug boat in Henderson
- 2016 on a vessel in Cockburn Sound
- 2017 on a cruise vessel in Freemantle
- 2019 on two vessels in Freemantle.

NSW

• 2019 on a navy vessel docked at a naval facility in Sydney Harbour in 2019.

SA

• 2019 on a vessel following the detection and unsuccessful treatment of the vessel in WA.

In the environment

QLD

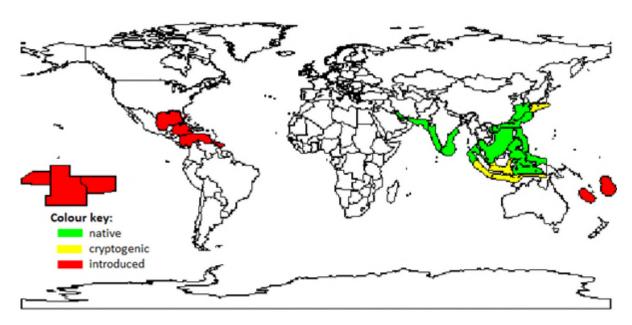
- 2001-2003 multiple detections on long stay vessels and floating infrastructure in Trinity Inlet, Cairns
- 2013 on a ghost net in Gove Harbour
- 2017 a single P. viridis was detected on a settlement array in Port Amrun, near Weipa
- 2019 six P. viridis were detected on drift wood washed up on Mornington Island
- 2019 a single *P. viridis* was detected attached to a pearl oyster in the Escape River.

Western Australia

• 2011 on a decommissioned platform in Dampier.

The other two species of the genus *Perna* (*P. perna* and *P. canaliculus*) have also been recorded outside their native range. *P perna* occurs naturally in tropical and subtropical regions of the southern Atlantic (Siddall 1980). It was introduced into the western Gulf of Mexico, presumably by shipping, in the early 1990s (Hicks & Tunnell Jnr 1993). *P. canaliculus* is endemic to New Zealand. Isolated individuals have been recorded in Tasmania (Furlani 1996), South Australia (1999, 2008) (PIRSA 1999), New South Wales (2006) and Victoria (2006), but there does not appear to be an extant population in any of those locations. Shells of *P. canaliculus* imported for human consumption are commonly found discarded in marine environments as part of marine biosecurity surveillance.

Map 2 Global distribution of Perna viridis



Cryptogenic Non-native. Source: NIMPIS 2002Potential impact

Perna viridis is a problem fouling species. It is able to attach to various surfaces, even in strong water flow, grows quickly and occurs in high population densities (Rajagopal et al. 2006). It can survive extremes of salinity (zero to 80 ppt during experimental laboratory testing; NIMPIS 2002b), and temperature (between 6 °C and 37.5 °C); it thrives in turbid coastal waters and is able to survive

prolonged exposure to biocides that kill most other fauna (Rajagopal et al. 2006). Table 2 outlines the potential social, economic and environmental impacts caused by *P. viridis*.

Industrial structures for seawater intake, cooling intakes for vessels and mariculture facilities are particularly susceptible to fouling from *P. viridis* (Rajagopal et al. 2006). Dense aggregations may also smother or exclude other fouling species, thereby altering natural biotic assemblages.

Because of their large size, rapid growth and abundant recruitment, species of *Perna* are a valued food item throughout their natural ranges and are intensively harvested and cultured in some countries (Rajagopal et al. 2006).

Impact category	Description	Potential impact
Social amenity	Human health	Yes
Economy	Aquatic transport	Yes
	Water abstraction/nuisance fouling	Yes
	Loss of aquaculture/commercial/recreational harvest	Yes
	Loss of public/tourist amenity	Yes
	Damage to marine structures/archaeology	Yes
Environment	Detrimental habitat modification	Yes
	Alters trophic interactions and food-webs	Yes
	Dominates/out-competes and limits resources of native species.	Yes
	Predation of native species	No
	Introduces/facilitates new pathogens, parasites	No
	Alters bio-geochemical cycles	Yes
	Induces novel behavioural or eco-physical responses	No
	Genetic impacts— hybridisation and introgression	No
	Herbivory	No

Table 4 Categories of potential impact caused by Perna viridis

Source: Hayes et al. 2005

1.2 Summary of life history information

Table 5 is a summary of the life history information for *M. sallei* and *P. viridis*, for ease of comparison.

15–30 mm

0–64 ppt

6–37.5 °C

Suspension feeder

Intertidal to (at least) 12 m

Fouling hard substrata

Gregarious settlement

Greater than 7 days

Feature	Measure Mytilopsis sallei	Measure Perna viridis
Max. size (length)	25–30 mm	100–165 mm
Max. age	1–2 years	2–3 years
Mating strategy	Separate sexes/protandric hermaphrodite	Separate sexes
Type of mating	Broadcast spawner	Broadcast spawner
Dispersal stage	Planktonic larva	Planktonic larva
Larval duration	1–3 days	10–20 days
Time to sexual maturity	Approximately 30 days	Approximately 60–90 days

unknown

Suspension feeder

Fouling hard substrata

Gregarious settlement

Up to 2 days

0–80 ppt

10–35 °C

Intertidal to approximately 2.5 m

Table 5 Mytilopsis sallei and Perna viridis life history summary

Size at sexual maturity

Feeding mode

Depth range

Distribution

Preferred habitat

Salinity tolerance

Aerial exposure limits

Temperature tolerance

Department of Agriculture

2 Pest pathways and vectors

The most likely pathway for introducing *Mytilopsis sallei* and *Perna viridis* into Australia is vessel biofouling, either on the hull or niche areas. As internal seawater systems can be difficult to clean or inspect, marine pests in these niche areas prove a potentially greater threat than those on the hull.

Both species can easily spread by movement of either the larval life stage or sedentary adults. The pelagic larval stage can spread naturally, through movement of water currents away from an infested area, or may be transported in seawater moved by humans from the infested site. The most common forms of seawater transport are uptake of ballast water or retention of water in internal spaces of vessels (such as bilge water, anchor wells and seawater piping).

Ballast water discharge is a possible pathway for introducing *P. viridis* to Australian waters, but is less likely for *M. sallei* because of the shorter larval phase of this species. However, viable *M. sallei* larvae have been reported from ballast water transported between Taiwan and Hong Kong (Chu et al. 1997), a distance of approximately 460 nautical miles. It could, therefore, be spread domestically within Australian waters over similar distances through ballast water exchange (URS 2004). Ballast water transfer is thought to be an important pathway behind the range extension of *P. viridis* in Florida waters (NIMPIS 2002b; Power et al. 2004; URS 2004). However, ballast water regulation both internationally and domestically should reduce this risk significantly.

M. sallei and *P. viridis* readily colonise hard artificial and natural substrata, and can spread as biofouling on submerged, non-permanent structures. Movement of fouled structures (including vessels, anchors, chain lockers, moorings, ropes, floats, nets) is the another pathway for introduction and spread of these species (Bax et al. 2002; Rajagopal et al. 2006; URS 2004).

Fouling on vessels is the most likely source of introduction for both species into Australian waters. Vessel biofouling includes all external wetted surfaces including niche areas such as sea chests, bilge keels, anode blocks, rudder pins, bow thrusters, propellers, shaft protectors, echo sounder transducers and log probes. It also encompasses all internal surfaces and niches that are exposed to seawater, including anchor wells, chain lockers, bilge spaces, fishing gear, bait lockers, cooling water intakes, strainer boxes and internal pipe work (AMOG 2002).

In addition to ballast water and biofouling, other pathways listed in Table 6 are likely to be of high importance for domestic translocation of species within Australian waters.

Pathway	Description	Vector for spread <i>M. sallei</i>	Vector for spread P. viridis
Biocontrol	Deliberate translocation as a biocontrol agent	No	No
	Accidental translocation with deliberate biocontrol release	No	No
Canals	Natural range expansion through man-made canals	Yes	Yes
Debris	Transport of species on marine debris (includes driftwood)	Yes	Yes
Fisheries	Deliberate translocation of fish or shellfish to establish or support fishery	No	Yes
	Accidental with deliberate translocation of fish or shellfish	Yes	Yes
	Accidental with fishery products, packing or substrate	No	No
	Accidental as bait	No	No
Individual release	Deliberate release by individuals	No	No
	Accidental release by individuals	No	No
Navigation buoys, marine floats	Accidental as attached or free-living fouling organisms	Yes	Yes
Plant introductions	Deliberate translocation of plants species (such as for erosion control)	No	No
	Accidental with deliberate plant translocations	No	No
Recreational equipment	Accidental with recreational equipment	Yes	Yes
Scientific research	Deliberate release with research activities	No	No
	Accidental release with research activities	No	No
Seaplanes	Accidental as attached or free-living fouling organisms	Yes	Yes
Vessels	Accidental as attached or free-living fouling organisms	Yes	Yes
	Accidental with solid ballast (such as with rocks or sand)	No	No
	Accidental with ballast water, sea water systems, live wells or other deck basins	Yes	Yes
	Accidental associated with cargo	No	Yes

Table 6 Pathways and vectors for Mytilopsis sallei and Perna viridis

Source: Hayes et al. 2005

3 Policy and rationale for incursion response

The policy and rationale for an incursion response is based on the generic policy for incursion response to marine pests in Australian waters, the control or eradication strategy for *Mytilopsis sallei* and *Perna viridis*, the policy on decision points and the policy on funding of operations and compensation. This chapter is an overview of marine pest emergency procedures and policy.

3.1 Generic policy for incursion response to marine pests in Australian waters

The <u>National Environmental Biosecurity Response Agreement</u> (NEBRA) establishes national arrangements for responses to nationally significant biosecurity incidents when there are predominantly public benefits. In the absence of a marine pest-specific deed, responses to marine pest incidents can fall under the NEBRA. The NEBRA provides a mechanism to share responsibilities and costs for a response when eradication is considered feasible and other criteria are met. The <u>Biosecurity Incident Management System</u> provides guidance on policies and procedures for the management of biosecurity incident responses, including responses to marine pest emergencies within Australian waters.

3.1.1 Commonwealth, state and territory authority responsibilities

Lead agencies in the response to a marine pest emergency must collaborate with CCIMPE in developing a National Biosecurity Incident Response Plan (NBIRP) as required under the NEBRA. CCIMPE will review the NBIRP and provide advice to the National Biosecurity Management Group (NMG), which will determine whether national cost-sharing arrangements should be activated. If the NBIRP and cost-sharing arrangements are approved, CCIMPE will help an affected jurisdiction implement an NBIRP. State coordination centres must be established with responsibility for strategically managing a marine pest incursion and for ensuring that community and/or industry involvement and communications are in place.

Depending on the circumstances, a local control centre with responsibility for managing field operations in a defined area may be established to enable an efficient and effective operational response. While close communication between a state coordination centre and a local control centre is imperative for effective conduct of any emergency response, it is important that strategic management (state coordination centre) and operational management (local control centre) roles be kept separate to optimise effective decision making and implementation during a national biosecurity incident response.

When a national coordination centre is established to help manage concurrent incursions in more than one jurisdiction, national coordination will be effected through consultation with CCIMPE representatives and relevant industry and community sector organisations, as appropriate.

3.1.1.1 Consultative Committee on Introduced Marine Pest Emergencies

CCIMPE provides national coordination for managing marine pest emergencies and comprises senior representatives from each Australian jurisdiction with coastal borders (the Australian Capital

Territory is not represented). CCIMPE is the national technical body that advises NBMG whether an incursion by an introduced marine pest represents a marine pest emergency (in a national context), and coordinates the national technical response. CCIMPE also makes recommendations on possible stand-down phase activities (such as monitoring).

3.1.2 Stages in an emergency response to a marine pest of national significance

Management of a marine pest emergency of national significance has four phases of activation:

- investigation phase
- alert phase
- operations phase
- stand-down phase.

The first two phases, while detailed separately in the rapid response manuals, may be run concurrently, as outlined in the <u>Biosecurity Incident Management System</u>. Progression from one stage to the next depends on the nature of the emergency and available information.

Not all detections of marine pests will initiate a response involving all four phases and certain responses (such as detection of marine pests on vessels) may involve truncated responses.

3.1.2.1 Investigation phase

The investigation phase is in effect when relevant authorities are investigating a reported detection of a marine pest. The initial report of a suspected marine pest may come from port surveys, in water vessel inspections, slipway operators, fishermen, members of the public and routine field and surveillance activities.

A notifying party must advise CCIMPE of a suspected outbreak of a marine pest within 24 hours of becoming aware of it to be eligible for cost sharing under the NEBRA. When making a preliminary assessment, the notifying party may decide that a notification is likely to trigger a marine pest emergency alert when:

- the species detected is likely to be of national significance (Schedule 2 of the NEBRA) based on available data
- the description matches a species represented on the <u>Australian Priority Marine Pest List</u> (APMPL) that is either not present in Australia or, if it is present, the detection represents a new outbreak beyond the known range of established populations of the species in Australia. All APMPL species have been assessed to be of national significance.
- the species detected has a demonstrable:
 - invasive history
 - impact in native or invaded ranges on the economy, the environment, human health or amenity
- the suspected outbreak cannot be managed through pre-existing cost-sharing arrangements
- one or more relevant translocation vectors are still operating.

If the investigation indicates that a marine pest emergency is highly likely, the notifying party will inform the reporting point and will direct implementation of the alert phase.

Given that *M. sallei* and *P viridis* are exotic to Australia and both are on the APMPL, a detection or suspected detection of either species will trigger an emergency alert. If the subsequent investigation concludes that the situation does not constitute a marine pest emergency, the notifying party will inform CCIMPE and the emergency alert will be cancelled. However, ongoing actions to limit spread of the pest may be undertaken.

3.1.2.2 Alert phase

The alert phase is in effect while confirmation and identification of a suspected marine pest is pending, and an incident management team is assessing the nature and extent of the suspected incursion. During the alert phase:

- all relevant personnel are to be notified that an emergency alert exists in the affected jurisdiction
- an incident management team is appointed to confirm the identification of the suspected pest and to determine the likely extent of an incursion
- control measures are initiated to manage the risk of pest spread from affected sites (for example, operational boundaries of restricted areas are established for potential vectors)
- the findings of an emergency investigation are communicated to CCIMPE and NMG to enable a decision to be made on whether to proceed to the operations phase.

If an emergency investigation shows there is no incursion by a marine pest of concern or there is an incursion but it is unlikely to be eradicable, the notifying party will:

- ensure interim containment measures are implemented to minimise the risk of pest translocation from any infested waterway
- provide a situation report to the CCIMPE Secretariat for the information of CCIMPE representatives and request a CCIMPE teleconference to enable consultation with all jurisdictions
- on reaching agreement from CCIMPE, request that the transition to management phase (when there is a confirmed incursion by a marine pest of concern but eradication is not considered feasible)or stand-down phase be implemented (when investigation shows there is no incursion by a marine pest of concern).
- ensure documentation relevant to the decision-making process is maintained and filed as a 'negative marine pest emergency alert' (when investigation shows there is no incursion by a marine pest of concern) or a 'non-eradicable marine pest emergency alert' (when there is a confirmed incursion by a marine pest of concern but eradication is not considered feasible).

If the emergency investigation shows there is an incursion by a marine pest of concern and it is potentially eradicable, the notifying party will:

• ensure appropriate emergency containment measures are continued to minimise the potential for pest translocation, both from and within any infested waterway

- provide a situation report and an NBIRP plan to the CCIMPE Secretariat for urgent consideration by CCIMPE representatives and request a CCIMPE teleconference to enable consultation with all jurisdictions
- following CCIMPE endorsement, submit the NBIRP to NMG for consideration of national costsharing arrangements to help resource a national biosecurity incident response.

3.1.2.3 Operations phase

The Operations phase of an emergency response commences when the marine pest emergency is confirmed by agreement through the NBMG forum and activities under a response plan are implemented. During the operations phase of a national biosecurity incident response:

- all relevant personnel and agencies should be notified that a national biosecurity incident response is being undertaken in the affected jurisdiction
- a standing committee on conservation and a local control centre should be established, if necessary
- control measures initiated in the alert phase should remain in place to manage the risk of pest spread from affected sites
- measures to eradicate the pest from infested sites should be implemented
- information from infested sites about the pest and the progress of operations should be collected, documented and analysed to enable progress of a national biosecurity incident response to be monitored
- expenditure associated with all eligible costs under cost-sharing arrangements should be documented
- regular situation reports should be communicated to the CCIMPE forum
- a decision should be made, when appropriate, on when to proceed to the stand-down phase.

3.1.2.4 Stand-down phase

The stand-down phase is in effect when, following appropriate consultation between the affected jurisdiction and CCIMPE, all agree that there is no need to progress or continue with a national biosecurity incident response. During the stand-down phase:

- a systematic approach to winding down operations must be taken to ensure operational effectiveness is not jeopardised
- all personnel, agencies and industry contacts involved in the emergency response are to be notified of the stand down.

The stand-down phase must commence once operational objectives have been achieved, or otherwise in accordance with advice provided by CCIMPE and agreed by NBMG. The advice that an emergency eradication operational response is no longer needed must be communicated to the affected jurisdiction.

3.2 Control and eradication strategy for *M. sallei* and *P. viridis*

The black striped false mussel, *Mytilopsis sallei*, and the Asian green mussel, *Perna viridis*, are listed on the <u>Australian Priority Marine Pest List</u>. Both species are highly fecund and form dense populations in intertidal and submerged marine habitats, where they may displace native Australian species were they to become established. *M. sallei* and *P. viridis* can cause nuisance fouling on marine infrastructure and can have serious economic consequences for the aquaculture and maritime industries.

M. sallei and *P. viridis* are currently considered not to be established in Australian waters. Any reports of the suspected presence of *M. sallei* or *P. viridis* in Australian waters should initiate the <u>investigation phase</u> of an emergency response.

The methods used to control an incursion of *M. sallei* or *P. viridis* in Australian waters depend on the location and size of the outbreak. If the emergency investigation revealed an incursion by *M. sallei* or *P. viridis* that was potentially eradicable, the Incident Manager would prepare an NBIRP and forward it to CCIMPE for urgent consideration.

The options for controlling an incursion by *M. sallei* or *P. viridis* in Australian waters are (see section 5 for more details):

- 1) Eradication of the pest from the infested area.
- 2) Containment, control and zoning with the aim of containing the species and slowing its further spread to other areas.

Eradication is unlikely if initial investigations show the species is widely established in open marine environments. Each control option involves a combination of strategies, such as:

- establishing declared areas to define zones where the pest is present or suspected to occur, and where emergency management operations are to be implemented
- quarantining and restricting or controlling movement of potential vectors, such as submersible equipment, vessels, marine organisms (fauna and flora) and ballast water in declared areas to prevent spread of the pest
- decontaminating potential vectors for the pest, including vessels, aquaculture stock and equipment, maritime equipment, and water that may contain larvae of the pest
- treating established populations on natural and artificial habitats in the infested area
- delimiting and tracing surveys to determine the source and extent of the incursion
- surveillance and monitoring to provide proof of freedom from the pest.

3.3 Policy on decision points

The policy on decision points includes proof of eradication and decisions to stand down eradication or control operations.

3.3.1 Proof of eradication

Proof of eradication requires a robust and intensive monitoring program during the operations phase of the response. During the operations phase, the purpose of the monitoring program is to detect new outbreaks of *Mytilopsis sallei* or *Perna viridis* for treatment and to determine the efficacy of the treatment procedure. This information can be used to refine and direct treatment.

Monitoring should also continue at sites potentially at risk of infestation. A decreasing trend in the number of new, untreated clusters of *M. sallei* or *P. viridis* detected over time in the infested area is evidence of the effectiveness of the control measures. Stand down eradication or control operations

The optimal time to stand down monitoring, eradication and control operations is a trade-off between the costs of maintaining emergency operations, including ongoing surveys (Cs), the cost of escape (including likely impacts) if eradication is declared too soon (Ce), the probability of detecting the pest species given it is present (q) and the annual probability the species remains present (p). This rule of thumb can be used to calculate the optimal number of surveys:

$$\underline{n}^* = \frac{\ln\left\{\frac{-C_s}{C_e \ln(r)}\right\}}{\ln(r)}$$

Where r = p(1-q) is the probability the pest is not detected but is still present in the survey area. See Regan et al. (2006) for guidance on calculating this decision point.

3.4 Policy on funding of operations and compensation

CCIMPE will help determine whether an incursion is likely to be eradicable and when national costshared funding under the NEBRA should be sought. Cost sharing must be agreed by NMG.

As detailed in the NEBRA, parties will share the eligible costs of emergency eradication responses as follows:

- a 50% share from the Australian Government
- a 50% share collectively from the states and the Northern Territory
 - this is calculated for each jurisdiction based on the length of coastline potentially affected by the species, and their respective human populations
 - only jurisdictions affected or potentially affected by the pest or disease are required to contribute.

NMG may commit up to \$5 million (in annual aggregate) towards the eligible costs associated with an agreed national biosecurity incident response. If this \$5 million is exceeded in any one financial year, NMG must seek ministerial approval from all parties to continue activities and/or begin new emergency responses.

Private beneficiary contributions to a response will be considered by NMG on a case-by-case basis where there is one or more private beneficiary and no existing arrangements.

4 Principles for containment, control and eradication

Successful eradication of incursions by *Mytilopsis sallei* or *Perna viridis* requires early detection and immediate action. Both species are highly fecund, reach sexual maturity quickly and have a planktonic larval phase. Eradication is most likely to be successful in shallow, partially or fully enclosed waterways. In open coastal waters with moderate to high water exchange, larvae may be dispersed over a wide area. Where surveys indicate that an infestation is widespread, eradication action is unlikely to be successful.

Characteristics of these species and the pathways by which they are spread make them difficult to eradicate. These include:

- high fecundity, with a planktonic larval stage that can be widely dispersed by water currents
- sequential (and possibly simultaneous) hermaphroditism in *M. sallei*, such that large numbers of offspring may be produced by relatively few individuals
- ability to recruit and survive as adults in confined wetted spaces, including in vessel sea chests, water intake pipes and bilge lockers, which makes them difficult to detect
- presence in estuarine environments, which can be turbid, making detection difficult
- likelihood of being detected on non-commercial vessels from infested ports or marinas, whose movements are frequent and often difficult to trace.

The basis of eradication is rapid, effective quarantine of the infested area and any potentially contaminated vectors, and elimination of the pest where it is found.

4.1 Methods for preventing spread of the organism

Methods used to prevent the spread of the organism are quarantine and movement control, and treatment for decontamination of infested vectors.

4.1.1 Quarantine and movement controls

Quarantine and movement controls include an investigation phase, an alert phase and an operations phase.

4.1.1.1 Investigation phase

When the presence of *Mytilopsis sallei* or *Perna viridis* is suspected in an area but a marine pest emergency has not yet been confirmed (see <u>section 3.1.2.1</u>), the notifying party should, when feasible, take steps to limit the spread of the suspected pest from the investigation site or area by initiating voluntary restrictions on movement of potential vectors. This may involve notifying relevant port authorities, marina operators, industry associations and vessel owners in the suspect site about the investigation into a possible marine pest emergency. Cooperation should be sought from these stakeholders to stop, restrict or inform the notifying party of movement of vectors from the site. Compliance with voluntary movement controls may be enhanced by distribution of appropriate public awareness materials about the pest. The investigation phase should attempt to identify all potential vectors present at the site and their location. Possible vectors for the spread of *M. sallei* and *P. viridis* are described in <u>chapter 2</u>.

4.1.1.2 Alert phase

If the initial investigation finds that *Mytilopsis sallei* or *Perna viridis* is highly likely to be present (see <u>section 3.1.2.2</u>), the findings should be communicated to CCIMPE for consideration of the appropriate course of action to manage the risk of spread from affected sites. The incident management team must ensure appropriate measures are implemented. These could include:

- restrictions on movement of potential vectors, such as submersible equipment, fishing gear, vessels, marine organisms (fauna and flora) and ballast water into and out of suspect sites
- controlling movement of people (such as property owners, scientists, tourists) into or out of the suspect sites, as appropriate; this may include police involvement
- a hotline phone number for reported sightings of the pests and inquiries from affected parties
- tracing potential vectors that have left the site
- redirecting vessels that have already left the site to appropriate sites for inspection and/or decontamination, if appropriate
- notifying and, where appropriate, consulting relevant experts.

4.1.1.3 Operations phase

The operations phase will be guided by whether eradication of the marine pest of national concern is feasible or not feasible.

Eradication not feasible

If investigation reveals an incursion by *Mytilopsis sallei* or *Perna viridis* that is unlikely to be eradicable, interim containment measures (to prevent translocation of a pest of concern from any infested waterway) should be implemented to minimise the risk of the pest being spread from the infested area. A stand-down phase may be entered either directly from the alert phase or from the operations phase when CCIMPE and NBMG agree there is no need to initiate a national biosecurity incident response.

Eradication feasible

If investigation reveals a potentially eradicable *M. sallei* or *P. viridis* incursion, quarantine and associated movement restrictions can be implemented.

Quarantine restrictions require establishing specified areas:

- infested area—all or part of a waterway in which a marine pest emergency is known or deemed to exist (pending confirmation of pest identification)
- dangerous contact area—an area close to an infested area in which a pest has not been detected but, due to its potential for infestation, will be subject to the same movement restrictions as an infested area
- suspect area—an area relatively close to an infested area that will be subject to the same movement restrictions as an infested area (pending further investigation)

- restricted area—a defined area around an infested area that is subject to intensive surveillance and movement controls on potential vectors²
- control area—a defined area surrounding a restricted area in which biosecurity conditions apply to the entry or exit of potential vectors or specified risk items.²

Similar terminology is applied to potentially affected vectors within each area. For example, a vessel within a dangerous contact area would be classified as a 'dangerous contact vessel'; a vessel within an infested area would be classified as an 'infested vessel'.

The extent of each specified area for *M. sallei* or *P. viridis* should be determined based on:

- an initial delimiting survey of the area (section 5.3)
- an evaluation of the length of time the species has been present and whether it has reproduced; this would be based on the size and distribution of the animals in the infested area, the number of cohorts apparent and, when possible, examination of reproductive tissue
- the strength and distribution of directional or tidal currents
- expert advice.

Movement restrictions include limiting:

- the movement of vessels, immersed equipment, aquaculture stock or equipment and other vectors for biofouling
- fishing activities within the control area
- the uptake or movement of ballast water or other water from within the control area where appropriate controls are not in place.

Implementation of restrictions will be a dynamic process, determined by the location and extent of infestation and whether the aim is to eradicate the pest or to control its spread. Some restrictions may be deemed impractical or unnecessary in a particular circumstance, but others will be critically important to eradication or control.

Restricted Area Movement and Security Unit

The Restricted Area Movement and Security Unit of the Operational Pest Control Centre is responsible for controlling movement of goods, submersible equipment, vessels, water and other vectors including people into, within and out of the restricted area as appropriate to minimise the potential for pest spread.

The unit's main duties are to:

² Note that the legislative ability and scope of powers to establish biosecurity restricted areas and control areas will depend on the biosecurity legislation that is applicable within the relevant jurisdiction.

- issue movement permits to the public
- establish and operate road and water checkpoints in the restricted area, including liaison with state transport authorities, water authorities, police and local government
- coordinate movement and security activities across infested sites
- maintain registers of all movements (in restricted and infested areas), permits issued and staff deployed.

Experience of movement controls

The emergency response to the incursion by the black striped false mussel, *M. sallei*, in Cullen Bay Marina (Darwin) in 1999, used a combination of the powers in the *Fisheries Act 1988* (NT) and the *Quarantine Act 1908 (Cwlth)* (superseded by the *Biosecurity Act 2015)* to impose sufficient quarantine measures to limit the spread of the species. The *Biosecurity Act 2015 (Cwlth)* can be used in the absence of appropriate state or territory legislative powers and may be used in circumstances, including directing conveyances³:

- into port
- to not enter a port and to obey further instruction
- to undergo a treatment action the Incident Manager deemed necessary.

The Australian Director of Biosecurity (or their delegate) can authorise State and Territory officers as biosecurity officers under the *Biosecurity Act* which will enable certain actions to be undertaken in a biosecurity response. All actions taken against a conveyance should only be taken in relation to those identified as being at risk of spreading the invasive species (Ferguson 2000). Guidelines for using the *Biosecurity Act 2015* are in <u>Appendix A</u>. The Biosecurity Act is only intended to be used if there is no appropriate State and Territory legislation that provides appropriate powers necessary for the response, aside from ballast water which is entirely covered by the Biosecurity Act. A provisional list of other Commonwealth and state powers for intervention and detention of vessels is in <u>Appendix B</u>.

Each state and territory should consider enacting relevant fisheries or other legislation to prevent or control fishing within a control area, and prevent or control translocation of stock and equipment from within it. Any requested movement of fishing gear or aquaculture stock or equipment should be subject to risk assessment consistent with procedures outlined in the National Policy Guidelines for the Translocation of Live Aquatic Organisms (Department of Agriculture 2020). All potentially infested fishing gear, aquaculture equipment or stock should be treated and inspected before removal from the control area.

³ Under the *Biosecurity Act* the definition of conveyances includes vessels and floating structures

4.1.2 Surveillance for high-risk vectors

In the event of an emergency marine pest response, movement controls on potential vectors and pathways will be easier to manage if efforts can be targeted at vectors that pose the greatest risk of spread.

All vessels and other vectors that have been within an infested area or dangerous contact area during the time the pest is known or suspected to have been present should be considered at high risk of transporting the pest. Vessels, oil rigs, barges and other moveable structures that have been present in suspect, restricted or control areas, that have marine fouling on them, should also be treated as high risk. The risk status of vessels may be changed if inspections or surveys find no sign of the pest

Vessels that have not been within the infested or dangerous contact areas, but which have been in close proximity to a high-risk vessel that have departed these areas or the control area should also be considered high risk. All high-risk vessels should be required to proceed to an approved inspection and treatment facility.

Where resources allow, all vessels and potential vectors within the control area should be inspected for signs of the pests. The preferred method for inspecting and cleaning vessels is by haul-out on a slipway. Alternatively, inspections may be carried out by divers or Remotely Operated underwater Vehicles (ROVs). All high risk vessels should also have their internal seawater systems treated (see Section 5.1.3.2). Medium-risk vectors should be required to remain within the control area until they can be inspected and declared free of the pest.

Divers and ROV operators should perform in-water inspection of vessels using a standardised search protocol. Biofouling is likely to be greatest in wetted areas of the vessel that are protected from drag when the vessel is underway and/or where the antifouling paint is worn, damaged or was not applied.

For vessels smaller than 25 m in length (Figure 4), particular attention should be given to inspecting:

- rudder, rudder stock and post
- propellers, shaft, bosses and skeg
- seawater inlets and outlets
- stern frame, stern seal and rope guard
- sacrificial anode and earthing plate
- rope storage areas and anchor chain lockers
- ropes, chains or fenders that had been left over in the water
- keel and keel bottom
- sounder and speed log fairings.

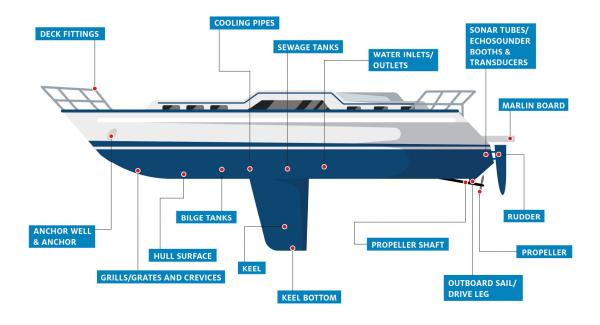
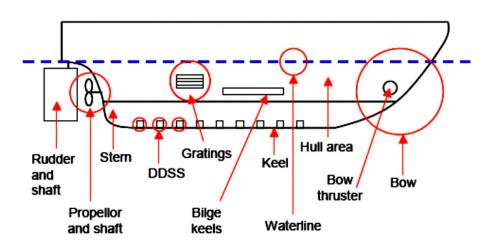


Figure 4 High-risk niche areas for inspection of biofouling on vessels less than 25 metres

For vessels larger than 25 m in length (Figure 5), additional high-risk niche areas include:

- dry docking support strips (DDSS)
- sea chests and gratings
- sonar tubes
- bow thrusters
- keel and bilge keels
- ballast tanks and internal systems.

Figure 5 High-risk niche areas for inspection of biofouling on vessels greater than 25 metres



Department of Agriculture

Image: Floerl 2004

Divers can inspect interior spaces and crevices (such as seachest, water intakes or outlets) using endoscopes.

All high-risk and medium-risk vessels that have recently left a control area should be contacted immediately. If they have not entered another port or marina they should be encouraged to remain at sea, no closer than 1.5 nautical miles to the nearest land until inspection and/or quarantine arrangements can be made. Biosecurity risks detected before or during this inspection must be dealt with before the vessel can be brought further inshore. Where the vessel has entered another port or coastal area, it should be inspected immediately and, if signs of the pest are present, the vessel should be directed for treatment, a back tracing of the vessel's itinerary be done and surveys undertaken of the anchorages it has visited.

4.1.3 Treatment methods for decontaminating infested vectors

Treatment methods differ depending on the type of area in which the infestation occurred. It could have been found in ballast water, on vessels or on equipment and marine organisms.

Table 7 summarises management recommendations for different types of vectors.

Potential vector	Suggested management	
International and domestic yachts and other	Clean external submerged surfaces	
vessels smaller than 25 m	Treat internal seawater systems	
	Manage ballast water	
	Remove from the control area once cleaned	
Domestic fishing vessels, ferries, tugs, naval	Clean external submerged surfaces	
vessels	Treat internal seawater systems	
	Manage ballast water	
Merchant vessels larger than 25 m departing for	Inspect and (where possible) clean external submerged surfaces	
other Australian destinations	Treat or seal internal seawater systems	
	Manage ballast water	
Merchant vessels larger than 25 m departing for	Inspect and (where possible) clean external submerged surfaces	
international waters	Treat or seal internal seawater systems	
	Restrict uptake of ballast water from the control area	
	Restrict ballast discharge within the Territorial Sea	
	Recommend exchange of any ballast sourced inside the control are	
	once the ship is in international waters (greater than 12 nautical miles)	
Recreational craft (such as dinghies, jet-skis,	Clean external submerged surfaces	
kayaks, outboard motors)	Clean and dry internal seawater systems	
	Educate users and service agents of risk	
Fishing gear and nets	Clean and dry on removal from area	
	Educate users of risk	
Aquaculture stock (fouled)	Remove from infested area and destroy	
Aquaculture equipment (fouled)	Remove from infested area	
	Clean thoroughly by high pressure (greater than 2,000 psi) water blasting	
	Immerse in copper sulphate solution (4 mg/l) or liquid sodium	
	hypochlorite (200–400 ppm) for 48 hours	
	Rinse in seawater and air dry	
Buoys, pots, floats	Clean and dry	
	Restrict removal from the control area	
	Educate users on risks	
Water, shells, substratum, live hard-shelled	Restrict removal from the control area	
organisms from the control area (such as aquaria, bait)	Educate users on risks	
Flotsam and jetsam	Remove from water/shoreline	
	Dry prior to onshore disposal	
	If possible, use barriers to prevent escape from infested area	
Fauna (such as birds, fouled crustacean)	Verify the importance of the vector during delimitation surveys	
Stormwater pipes, intakes	Clean	
	Where possible, seal until stand down of emergency response	

Table 7 Management recommendations for different types of vectors

Source: Bax et al. 2002

4.1.3.1 Ballast water

In the event of an emergency response, all ballast water sourced from the area would be considered high-risk to the Australian marine environment. The *Biosecurity Act*, which implements the <u>International Convention for the Control and Management of Ship's Ballast Water and Sediments</u> (Ballast Water Convention) together with the *Biosecurity (Ballast Water and Sediments) Determination 2017* (Ballast Water Determination), prohibits discharge of ballast water anywhere within Australian seas⁴, subject to certain exceptions.

All vessels that contain ballast water will need to be appropriately managed according to the <u>Australian Ballast Water Management Requirements</u>. This includes via an approved method of ballast water management, or disposed of safely, such as through an approved ballast water reception facility. If *Mytilopsis sallei* and *Perna viridis* are present in an area, steps can be taken by the Department of Agriculture to ensure no low-risk exemptions to discharge ballast water would be granted under section 23 of the Ballast Water Determination.

Since the Ballast Water Convention has come into effect, certain ships are no longer allowed to manage ballast water through exchange. These vessels are required to install acceptable ballast water management systems to ensure appropriate treatment of ballast water on-board. These systems eliminate harmful pests from ballast water by using methods such as UV treatment or chlorination. Vessels that are allowed under legislation to meet ballast water management requirements through exchange (subject to certain exemptions), would be required to conduct ballast water exchange outside Australia's 12 nautical mile territorial sea limit. Additional measures may need to be investigated where vessels utilise ballast water exchange and operate exclusively within a declared Same Risk Area, detailed within the *Biosecurity (Ballast Water Same Risk Area) Instrument 2017*.

Operators may choose to retain high-risk water within a ballast water tank if there is no intention to discharge the water in Australian seas. However, carrying high-risk ballast water into Australian seas is strongly discouraged, as a vessel's itinerary may change, or discharge may be necessary in the case of safety or pollution considerations.

Vessels departing for international destinations

Vessels leaving the control area for destinations outside Australia's territorial waters should be notified of the risk and required to exchange ballast water sourced from the control area in oceanic waters, outside 200 nautical miles at depths greater than 200 m, as specified by the International Maritime Organization (IMO) <u>International Convention for the Control and Management of Ships'</u> <u>Ballast Water and Sediments, 2004</u> (Ballast Water Management Convention). Permission should not

⁴ Under the Biosecurity Act, the definition of Australian seas changes depends on the Administration (the country's flag under which the vessel is registered) of the vessel. For Australian or foreign vessels whose Administration is party to the Ballast Water Convention, Australian seas is waters within the outer limits of Australia's exclusive economic zone (EEZ) (200 nautical miles from the territorial sea baseline). For other vessels, Australian seas is the waters within the outer limits of the territorial seas of Australian (12 nautical miles from the territorial sea baseline).

be given for discharge of high-risk ballast within the 12 nautical mile limit. Options for oceanic exchange of ballast water are described in the <u>Australian Ballast Water Management Requirements</u> Version 7 (Department of Agriculture 2017) and are consistent with the IMO's Ballast Water Management Convention Guidelines for Ballast Water Exchange.

Vessels departing for Australian destinations

When possible, vessels travelling to other Australian ports should be encouraged to exchange ballast sourced from the control area in oceanic waters or treat it using an approved on-board ballast water management system. Australian law prohibits discharge of high-risk ballast water anywhere inside Australia's territorial waters (12 nautical mile limit). To avoid discharging high-risk domestic ballast water, the ship may elect to hold the ballast water on-board or transfer it from tank to tank within the ship. This is an acceptable way of managing ballast water risk. However, ships' masters should ensure that, when using this method, they have carefully considered their cargo plans to negate any need to discharge any high-risk ballast water within Australian ports.

The <u>IMO's Ballast Water Convention</u> came into effect in 2017, and ballast water management systems are now an accepted alternative to ballast water exchange. These systems eliminate harmful pests from ballast water by using methods such as filtration, UV treatment, electrolysis, active substances and cyclonic separation.

4.1.3.2 Biofouling of vessels and other possible vectors

Mechanical removal of biofouling on vessels includes land-based treatment, internal seawater systems and various in-water treatments.

Land-based treatment

Because *Mytilopsis sallei* and *Perna viridis* are able to inhabit internal piping and water intakes that are not readily inspected underwater, haul-out of vessels and other non-permanent structures (such as moorings, pontoons, ropes) for inspection and treatment on land is the preferred option for decontamination. This may only be possible for vessels smaller than 25 m in length where suitable haul-out or dry-dock facilities are available within or close to the control area. Larger vessels may need to be inspected and treated in the water.

High pressure water-blasting (of 2,000 psi or greater) of external surfaces with hot or cold water can successfully remove fouling mussels. Care should be taken to treat the niche areas identified in Figure 4 and Figure 5, and any seawater entry points. Cleaned vessels should be left to dry on the hard stand for a minimum of seven days (Ferguson 2000).

Mussels dislodged during haul-out or cleaning of a vessel may remain viable and could start a new population if returned to the sea. The Incident Manager must approve haul-out facilities used for decontamination. Such facilities should be fully contained so material removed from vessel hulls cannot move back into the marine environment by direct disposal, run-off, aerosol-drift or any other means. All macro (greater than 1 mm) particles removed from vessels cleaned out-of-water should be retained and disposed of in landfill (or as biohazard material, if appropriate). All liquid effluent (run-off) from out-of-water vessel water-blasting and cleaning should be collected for treatment in a liquid effluent treatment system.

Woods et al. (2007) provide guidance for identifying vessel cleaning facilities suitable for removal of marine pests. Approved facilities should also comply with relevant state requirements for waste containment and disposal from slipways, boat repair and maintenance facilities.

High-pressure water blasting followed by prolonged (more than seven days) aerial exposure may also be used to treat other fouled structures removed from the infested area (such as mooring blocks, pontoons, floats, fenders). However, materials such as ropes with fine interstices that may be protected from the blasting and which can retain moisture, should be treated chemically or be disposed of to landfill.

Internal seawater systems

Internal seawater systems should be cleaned to the greatest extent possible with:

- 5% (by volume) industrial detergent (Conquest or Quatsan) in water (preferably fresh) for 14 hours (Lewis & Dimas 2007)
- chlorine at a concentration of 24 mg/L for 90 hours (Bax et al. 2002)
- hot water 60 °C for 1 hour (Growcott et al. 2016)
- copper sulphate solution at a concentration of 1 mg/L for 38 hours (Bax et al. 2002).

The Incident Manager may approve other treatments. The marine descaler, Rydlyme, dissolves biofouling and is non-toxic and biodegradable. There is a linear relationship between the level of fouling and the volume of Rydlyme required to digest fouling (Lewis & Dimas 2007). Rydlyme technical application information recommends a 1 to 1 ratio of Rydlyme to water to be circulated in a closed system for at least four hours, and a freshwater flush of build-up to remove excessive scale (Rydlyme Marine 2004). At this concentration, 14 hours is the recommended application time to dissolve significant mussel growth (Lewis & Dimas 2007).

In-water cleaning

The <u>Anti-fouling and in-water cleaning guidelines (2015)</u> state that where practical, vessels and moveable structures should be removed from the water for cleaning, in preference to in-water operations. When removal is not economically or practically viable, the guidelines accept in-water cleaning as a management option for removing biofouling, provided risks are appropriately managed.

Applicants who wish to perform in-water cleaning in Australian waters should familiarise themselves with the principles and recommendations contained in the guidelines. In Commonwealth waters, applicants should first check their obligations under the Environment Protection and Biodiversity Conservation Act 1999 (EPBC Act). If the activity does not need to be referred under the EPBC Act, then applicants should self-assess their activity using the decision support tool in Appendix A of the Anti-fouling and in-water cleaning guidelines. Applicants who wish to perform in-water cleaning in state or territory waters should contact the relevant agency in each state or territory jurisdiction for advice.

Vacuum and brush cleaning

The most commonly available in-water cleaning technologies are brushing and scraping, soft cleaning tools, and water or air jet systems. These methods vary in their effectiveness in removing and containing biofouling organisms, and in their suitability for use on different anti-fouling coating

types. Further information about these cleaning methods can be found in the <u>Anti-fouling and in-</u><u>water cleaning guidelines (2015)</u>.

Rotating brush and vacuum systems have been trialled in New Zealand to remove fouling pests (Coutts 2002). Preliminary results suggest these systems remove a large proportion (more than 90%) of low-to-moderate levels of fouling and collect, on average, more than 90% of the material that is removed. Problems associated with vacuuming include dislodgement of bivalves and other fouling organisms by divers or dragging hoses, reduced efficiency with variable hull shape. When there is heavy fouling, blockages by large organisms can damage filter valves. Gametes and planktonic larvae of *M. sallei* and *P. viridis* are microscopic and filtering systems must be able to deal with their removal (Coutts 2002). Because of these problems, brush and vacuum systems should only be used when there are no other options for vessel treatment.

Wrapping and encapsulation

Wrapping and encapsulation of the submerged surfaces of vessels using impermeable barriers, such as polyethylene plastic, have been used to treat fouling on vessels of up to 113 m long (Mitchell 2007). The wrapping deprives fouling species of light and food while continued respiration and decomposition of organisms within the barrier depletes dissolved oxygen in the water, thus creating an anoxic environment that is eventually lethal to all enclosed organisms.

Polyethylene silage plastic wrap (15 by 300 m, 125µm thick) is cut to size to suit the vessel type and is deployed by divers in association with a topside support team. The plastic is passed from one side of the vessel to the other, overlapped and secured tightly using PVC tape or ropes to create a dark, anaerobic, watertight environment. Sharp objects on the hull (such as propeller blades) should be wrapped separately or covered with tubing or cloth before encapsulation to prevent tears in the plastic.

Properly deployed, the wrap should contain the pest species and its larvae; care should be taken to ensure that biofouling is not dislodged when the wrap is deployed. The wrap must remain in place for at least seven days to ensure mortality. Wrapping of vessels larger than 25 m in length is labour intensive and may take up to two days to deploy per vessel. In addition, the time needed for effective treatment (seven days) may be too slow when rapid treatment and turnaround of vessels is crucial.

This method of treatment is only suitable in relatively sheltered environments with slow current flow, since strong currents create difficulties in deploying the wrap and increase the chances of tears in the plastic.

Where very large vessels or several vessels need to be treated, the encapsulation technique will generate large amounts of plastic waste. Wrap and equipment used to deploy it must be disposed of in landfill or an approved solid waste treatment facility.

Commercial encapsulation tools are available which can be applied to a vessel arriving in port, or to a vessel at anchor, alongside a wharf or in a marina berth.

Relevant agencies in each state or territory jurisdiction should be consulted about the suitability of a wrapping and encapsulation method for a vessel or moveable structure.

Chemical treatment

Mortality can be accelerated by adding chemical agents to the encapsulated water (Coutts & Forrest 2005). For example, sodium hypochlorite (NaOCl, 12.5% w/v) can be added to the sea water enclosed in the sheath to achieve a concentration of 200 to 400 ppm. The sheath and chemical treatment remain in place for 36 to 48 hours for each vessel. Because this technique may release some chloride ions to the surrounding water, consent is required from relevant state or territory authorities to undertake the treatment.

4.1.3.3 Aquaculture stock and equipment

Various treatments have been evaluated to remove fouling pests from aquaculture operations. These evaluations have mostly considered removal of soft-bodied fouling pests, such as ascidians or macroalgae, from cultured shellfish stock or equipment. Their efficacy for removing bivalves, such as *Mytilopsis sallei* and *Perna viridis*, has not been fully tested.

Both *M. sallei* and *P. viridis* are highly tolerant of changes in salinity and chlorine concentrations (Bax et al. 2002; Rajagopal et al. 2002). Their strong byssal attachments and hard shells make them more resistant than most soft-bodied fouling organisms to mechanical damage or to short-term exposure to toxicants. Bivalves close their shell valves in response to potentially toxic chemicals in the water, which reduces the effectiveness of many chemical immersion treatments. The effectiveness of toxicants may also be affected by salinity, temperature, dissolved oxygen, pH, water flow and size and nutritional status of the bivalves.

Table 8 is a summary of treatments shown to cause 100% mortality (LD100) of *M. sallei* and *P. viridis*. These are largely based on laboratory trials and may need to be adapted to ensure complete mortality on more complex structures such as ropes or nets or for large applications.

Treatment	Duration of immersion and concentration for 100% mortality for <i>M. sallei</i>	Duration of immersion and concentration for 100% mortality for <i>P. viridis</i>
Chlorine	111 hours at 12 mg/Lª 90 hours at 24 mg/Lª	48 hours at 10–15 mg/L ^b
Chlorine and copper sulphate	48 hours chlorine at 12 mg/L, then 48 hours copper at 1 mg/L	na
Copper sulphate	38 hours at 1 mg/L	na
Water temperature	120 minutes at 40 °Cª	5 hours at 39 °C
	30 minutes at 50 °C ^a	30 minutes at 60 °C ^d
	30 minutes at 60 °Cª	

Table 8 Treatments achieving total mortality (LD100) of *Mytilopsis sallei* and *Perna viridis* in laboratory conditions

a Bax et al. 2002. b Rajagopal et al. 2003b. c Rajagopal et al. 2003a. d Azanza et al. 2005. na No information available.

Ropes and equipment

Due to uncertainty about the efficacy of the techniques for treating *M. sallei and P. viridis*, the protocols recommended for treating ropes and aquaculture equipment, such as buoys, floats, nets and traps, are:

- 3) Remove to land taking care not to dislodge mussels when removing structures from the water.
- 4) Clean thoroughly by high pressure (greater than 2,000 psi) water blasting.

- 5) Immerse in copper sulphate solution (4 mg/L) or liquid sodium hypochlorite (200–400 ppm) for 48 hours.
- 6) Rinse in seawater and air dry.

This is based on the protocols used during the 1999 incursion response to *M. sallei* in Darwin (Bax et al. 2002).

Aquaculture stock

Some cultured species with hard shells (such as molluscs and crustaceans) and macroalgae that may be fouled by *M. sallei* and *P. viridis* are potential vectors for their spread. Any decontamination of aquaculture stock depends on the relative robustness of the pest and cultured stock to the treatment. For example, adults of thick-shelled bivalves, such as oysters, may be more resistant than *M. sallei* to treatment by hot water or high-pressure water blasting, because *M. sallei* has a relatively weakly calcified shell. Spat and less calcified juvenile oysters are not as resistant to these treatments. The more robust shell and larger size of *P. viridis* means it is better able to withstand mechanical and physiological shocks for longer periods than *M. sallei*. However, small *P. viridis* are less tolerant than some oyster species to elevated water temperatures (Rajagopal et al. 2003a).

Disinfection of bivalves and other aquaculture stock for external hitchhikers is not always effective and must be weighed against potential environmental impacts of any treatment and its effect on stock. Where the treatment cannot be effective, it may be precautionary to either destroy potentially contaminated stock and dispose of it to landfill or harvest and process stock for human consumption.

4.2 Tracing an incursion

Tracing is used to discover the method and pattern of the spread of the pests and may include traceforward and trace-back. It is crucial to defining and modifying the dimensions of the specified areas and requires investigations that determine:

- the length of time the species has been present
- the initial source and location of infestation
- whether the pest has reproduced
- the possible movement of water, vessels, animals, submersible equipment and other potential vectors for the pest
- the existence and location of other potentially infested areas.

If the Local Control Centre is established, it is responsible for managing tracing and surveillance activities within the control area.

Several methods are useful for estimating how long the pest has been present. The demography of the population may be inferred from the size distribution and reproductive state of animals collected during initial investigations.

For example, specimens of *Perna viridis* that are less than 15 mm long are likely to be no more than three months old and non-reproductive. The growth rates that Rajagopal et al. presented in 1998 and 2006 for *P. viridis* in India suggest that individuals greater than 90 mm long could be up to a year or more old, and specimens greater than 120 mm long could be more than a year and a half old.

However, there is large variability in the reported growth rate of this species, which may be as slow as 5 mm/month in highly polluted waters (Rajagopal et al. 2006).

Mytilopsis sallei are reproductively mature within one month of settlement (see <u>section 1.1.3</u>). Very dense populations can build up rapidly. For example, the population in Cullen Bay Marina, Darwin, in 1999 reached densities of more than 23,000 individuals per m² within six months of the initial incursion (Bax et al. 2002).

A population of either species with individuals varying widely in size, or that contains two or more distinct size cohorts, may indicate successful local reproduction and multiple recruitment events.

4.2.1 Data sources for tracing vectors

4.2.1.1 Vessels

Tracing the movements of vessels to and from an incursion is made difficult by lack of a consolidated system for reporting or managing data on vessel movements in Australian waters. Some potentially useful data sources on movements of large, registered commercial vessels are:

- The <u>Lloyd's List Intelligence</u> maintains real-time and archived data on movements of more than 120,000 commercial vessels worldwide. It contains arrival and departure details of all vessels larger than 99 gross tonnes from all major Australian and international ports. The database contains a searchable archive that includes movement histories of boats since December 1997. Searches can be purchased for specific ports, vessels or sequences of vessel movements.
- <u>MarineTraffic</u> provides real-time data on the movements of more than 550,000 vessels. It maintains archived data going back to 2009. Searches can be purchased for specific ports, vessels, areas or periods of time.
- Local port authorities keep records of all vessel movements at their port berths and associated anchorage points.
- The <u>Australian Fisheries Management Authority</u> manages data on the locations of all fishing vessels that have Commonwealth fishing concessions. All Commonwealth fishing concession holders must have installed and be operating an integrated computer vessel monitoring system. The system is also required for some fisheries managed by state and territory fisheries management agencies (such as the Queensland East Coast Trawl Fishery).
- The <u>Bureau of Infrastructure, Transport and Regional Economics</u> maintains statistics on maritime trade, markets, shipping lanes, key trade routes, traded commodities and passenger services throughout Australia.
- The <u>Department of Agriculture</u> and the <u>Australian Border Force</u> maintain data on all vessels arriving in Australian waters from overseas. These data are for proclaimed first ports of entry into Australia.
- The <u>Australian Maritime Safety Authority</u> deals with maritime safety, protection of the marine environment and maritime and aviation search and rescue services. It also coordinates a vessel tracking program, which works as an umbrella for managing related vessel information from the Modernised Australian Ship Tracking and Reporting System (MASTREP) the Great Barrier Reef and Torres Strait Vessel Traffic Service, the Automatic Identification System, the Long Range Information and Tracking system and the Australian Maritime Identification System.

• The aquaculture industry deals with equipment, stock and boat movements between aquaculture sites.

There are no consolidated data on domestic movements of smaller coastal vessels within Australian waters. Ports and some marina operators keep records of vessels that have used their facilities. Local industry groups (such as fishing, petroleum exploration) may provide points of contact for vessels from individual industry sectors that have visited the infested area. Some data may also be available from sources such as the Australian Volunteer Coast Guard, in the form of logged vessel trip reports.

Some states and territories have developed vessel-tracking systems for a range of vessel types. During the operational period of the *Mytilopsis sallei* incursion in Darwin, the Northern Territory Police and the Australian Government Department of Agriculture, with support and input from the Darwin Port Authority, Australian Border Force, the Northern Territory Fisheries Division Licensing Branch, the Australian Fisheries Management Authority and Coastwatch, developed an access database that contained vessel names and contacts, current location, history of individual vessel movements and the risk status of the vessel.

4.2.1.2 Ocean current modelling

Ocean current modelling may be an effective forward and backward tracing method for estimating the source and sink locations as part of marine pest incursions. There are a number of tools that can assist with modelling of current movements:

<u>Connie3</u> uses archived currents from oceanographic models and particle tracking techniques to estimate connectivity statistics from user-specified source or sink regions. A range of physical and biological behaviours can be specified including vertical migration, horizontal propulsion, swimming, flotation or surface slick formation.

<u>Regional Ocean Modelling System (ROMS)</u> is an ocean model used for a diverse range of applications. ROMS has pre and post-processing software for data preparation, analysis, plotting and visualisation.

5 Controlling, eradicating and treating established populations

The feasibility of controlling a *Mytilopsis sallei* or *Perna viridis* infestation in Australian waters depends on the nature and location of the incursion and the management strategy adopted. Two control options are available:

• eradication or complete elimination of *M. sallei* or *P. viridis* from the infested area (highest level of control measure and cost)

or

 containment and control by limiting the species to the infested area, preventing further spread and protecting uninfected areas (has ongoing costs and implementation so may have higher cost in the long term).

5.1 Eradication

Eradication of *Mytilopsis sallei* or *Perna viridis* requires complete removal from the infested area or destruction. Eradication is unlikely to be successful or feasible if initial investigations determine that the species is widespread, cannot be contained, is difficult to detect, or is present or potentially present in open coastal environments.

Because the planktonic larvae of *M. sallei* and *P. viridis* can be spread rapidly over large distances by movement of tidal and coastal currents, eradication may be impossible in open coastal waters where there is high exchange of water. Eradication is most likely to be feasible when:

- the area inhabited by *M. sallei* or *P. viridis* is small (less than 1,000 m²)
- the infestation occurs within an area of minimal flushing or exchange of water
- the available habitat occurs in relatively shallow waters (less than 5 m)
- the population is relatively aggregated.

See section 6 for treatment options.

5.2 Containment and control

If the decision is made not to attempt eradication but to implement containment and control, the Incident Manager will recommend that interim containment measures be implemented to minimise the risk of pest translocation from the infested waterway. This may include movement controls on potential vectors, public awareness campaigns, policies and practices (in consultation with stakeholders) for vessel and equipment sanitation and surveillance, and control of secondary infestations outside the infested waterway.

5.3 Guidelines for delimiting surveys

A delimiting survey establishes the boundary of an area considered to be infested by or free from a pest. The survey should be conducted to establish the area considered to be infested by the pest during the emergency response and to decide if eradication is feasible. The State or Local Control Centre will plan a survey strategy with reference to appropriate confidence limits based on:

- the location where the pest was initially detected
- pest biology—survival, reproductive rate, spread, dispersal and influence of environmental factors
- pest habitat—distribution and suitability of potential habitats around restricted areas and control areas
- survey design—should take into account the sensitivity of the methods to detect the pest species and the ease with which a sample may be obtained, as well as operator safety
- sampling methods—should take into account the area of expected occurrence
- a predictive analysis of areas where the pest is likely to occur
- expected prevalence of the pest if unrestricted
- statistical methods to specify the different confidence limits for targeted and general surveillance.

When possible, the survey should be consistent with national standards and contain estimates of confidence based on best available information.

5.4 Design of a delimiting survey

The location at which the pest was first detected is a useful starting point for a delimiting survey, but it is important to recognise that it is not necessarily the initial site of the infestation. When designing a delimiting survey, it can be useful to work backward, to try to trace the initial source of the incursion (trace-back) and also to try to predict where the pest has, or could, spread to (trace-forward).

The geographic extent of an incursion will be determined by:

- how long the pest has been present at the site before it was detected
- the frequency and quantity of reproductive output from the population since the initial incursion
- the effects of environmental and human factors on the spread of dispersal stages.

Local knowledge and site inspections as well as satellite imagery, hydrographic charts and online databases such as <u>Seamap Australia</u> can be useful for identifying areas that may contain habitat suitable for the pest. Where they exist, hydrodynamic models (for example, CSIRO's <u>Connie3</u>) may also be useful for simulating the likely directions of current flow and the possible rate and extent of spread of planktonic larvae from the known area of infestation. Trace-forward techniques should be used to identify locations outside the infested area that may have been exposed to the pests by vectors that have departed the area known to be infested.

Trace back information can also be used to determine the possible extent of an incursion (particularly a primary incursion where a single size class is present). Working backwards from the estimated age of the specimens and the known settlement biology and larval lifecycle of the species, ocean current modelling can predict the source of a spawning event. This source information can then be used to determine where else in the area the prevailing currents could have spread the larvae.

The greatest survey effort should be made at the margins of the known infestation. Adaptive sampling designs with sample points located on systematic grids or gradients away from the site of known infestation (Eberhardt & Thomas 1991; Gust & Inglis 2006) are most useful to ensure the greatest possible area is covered, while providing the best chance of detecting established and founding populations.

The type of sampling method chosen should be based specifically on the species being targeted, the habitat being searched and the conditions at the site. *Mytilopsis sallei* and *Perna viridis* both occur predominantly on hard natural and artificial substrata, although both may also foul shelled organisms (such as epifaunal bivalves) that occur in soft-sediment environments. In shallow subtidal waters (less than 10 m), where *M. sallei* and *P. viridis* are most abundant, visual surveys by divers or snorkelers are likely to be the most efficient, as a large area can be searched relatively quickly, and complex artificial structures, such as wharf pilings, pontoons and niche areas of vessels can be inspected. ROVs can be used to do this where diving is not an option for safety reasons.

The ability of divers or ROV operators to detect *P. viridis* and *M. sallei* depends on sufficient training in identification and search techniques, water clarity at the site and abundance and degree of aggregation of the population. Large *P. viridis* may also be heavily fouled making visual detection difficult. Where underwater visibility is less than 1 m, visual surveys may be severely compromised.

Artificial structures, such as projecting piles, steel facings, ropes and mooring dolphins associated with wharf structures, are to be considered a high priority during surveying as they are highly susceptible to fouling by invasive marine species. Other surfaces with potential for colonisation include breakwaters, groynes, rockwalls, wrecks, hulks, moorings, navigational markers, hulls, aquaculture facilities and natural rocky reefs.

For soft-sediment habitats, visual surveys may be replaced by benthic sled tows, modified scallop dredge, beam trawls or similar techniques that will effectively sample epibenthic assemblages over large areas. *M. sallei* and *P. viridis* also inhabit intertidal areas, which should be surveyed visually (from shore or sea) during low tide for infestation.

Samples from plankton tows and settlement plates can be analysed for environmental DNA (eDNA) to detect the presence of DNA from *M. sallei* and *P. viridis*. Settlement plates can also be used for visual detection of fouling organisms such as *M. sallei* and *P. viridis*. Time of larval settlement should be considered in relation to the settlement plate deployment period. See the <u>Australian marine pest</u> monitoring guidelines, version 2 (NSPMMPI 2010) for additional information that can be adapted for delimiting surveys.

6 Methods for treating established populations

Methods used to treat established populations of *Mytilopsis sallei* and *Perna viridis* will vary in efficacy according to the size and location of the incursion. This chapter summarises treatment options for closed or semi-enclosed coastal environments and for open coastal environments.

6.1 Closed or semi-enclosed coastal environments

Eradication is most achievable in closed or semi-enclosed coastal environments (such as locked marinas and coastal lakes) because the pest can be more easily contained and it is possible to maintain conditions necessary to achieve mortality for longer. Various treatment options are possible in these circumstances, including draining, de-oxygenation and/or flushing of the waterway with fresh water, application of chemical biocides, physical removal and ecological control (Aquenal 2007).

If the infestation is confined to relatively small, enclosed or semi-enclosed waterways, it may be possible to treat the entire water body and all marine habitats within it. If this is not possible, the success of management will depend more heavily on the ability of monitoring and delimitation surveys to locate and treat all clusters of the population. Where resources allow, all habitat potentially suitable for *Mytilopsis sallei* and *Perna viridis* should be treated. Where this is not possible, habitats should be based on suitability for the pest and delimitation survey results.

6.1.1 Chemical treatments

Major constraints for chemical treatment of water bodies are the volume of water that needs to be treated (a function of the area, depth and degree of flushing of the waterway), the presence and susceptibility of valued non-target organisms that may also be affected, residual effects of any toxicants on the surrounding environment and human health and safety management when handling large volumes of chemicals. Legal issues can also influence the ability to administer chemicals as a rapid response, due to the large number of chemical products available and different legislative requirements between Australian states and territories (Aquenal 2007). Consideration should be given as to whether a permit for the use of chemicals is required from the relevant state or Northern Territory environment agency or the Australian Pesticides and Veterinary Medicine Authority.

6.1.1.1 Chemical options

Many chemicals have been examined as possible treatments for removing fouling mussels. These comprise oxidising biocides and non-oxidising biocides. Oxidising biocides include chlorine (gas, or sodium or calcium hypochlorite), bromine, active halogen compounds, ozone, hydrogen peroxide and chlorine dioxide. Non-oxidising biocides include aldehydes, amines and quaternary ammonium compounds, organobromines and organometals (Jenner et al. 1998).

Chlorine

Chlorination is the most common form of chemical control used in enclosed water systems because of its economy, availability and wide-spectrum efficacy. Chlorine breaks down naturally and has minimal long-term effects on the environment.

However, chlorination does have some inherent problems associated with its use:

- hazards of handling chlorine gas cylinders
- difficulty in maintaining chlorination plants in the operational area
- non-uniform distribution of residual chlorine (Rajagopal et al. 2006)
- impacts on non-target organisms.

Chlorine is unstable in water. Exposure to light, elevated temperatures and reaction with organic compounds in the water accelerates the reduction in chlorine concentration so it can be difficult to maintain desired levels. For this reason, it is important to monitor levels of 'free available chlorine' in the treated area.

Chlorine in liquid form is capable of causing severe burns and is highly toxic if swallowed or inhaled. Operational staff should be appropriately trained in the safe handling and application of dangerous chemicals. Further information on the hazards, safe handling, emergency procedures and disposal of chemicals is available on the material safety data sheet, which should be available to staff working with a chemical.

Liquid sodium hypochlorite was used as the initial biocide in attempts to eradicate *M. sallei* in the Cullen Bay Marina, Darwin. Chlorine concentrations in the marina water were raised to 10 mg/L using several hundred tonnes of sodium hypochlorite (Bax et al. 2002). To disperse the chlorine and reduce stratification of fresh and salt water, the propellers of a large cruising sports fishing vessel, moored to a pontoon, were run to 'mix' the 12 hectare marina and large pumps mixed the marina's 'dead-ends'. However, chlorine treatment was not as effective as was hoped; death rates were not as high as expected. Also, obtaining sufficient chlorine to maintain an effective concentration in the marina became logistically difficult.

Following a trial of copper sulphate (Cu 1.5 mg/L) in another infected Darwin marina that resulted in 100% mortality, copper sulphate was used in combination with chlorine in the Cullen Bay Marina to raise the mortality rate. Copper sulphate powder was dissolved in a road construction watering truck tank and hosed over the water surface of the 'mixed' marina.

Compared with other mussel species, *P. viridis* is very resistant to chlorine (Rajagopal et al. 2003c). Chlorine concentrations are most effective and cause 100% mortality in all size classes when residual levels are kept between 8 and 15 mg/L. Sublethal responses of *P. viridis* (filtration rate, foot activity index and byssus production) are negatively correlated with chlorine concentration (Rajagopal et al. 2003c). Because of the ability of *P. viridis* to tolerate chlorine, continuous low-dose concentrations are not as effective at removing this species as they are with other species. Power stations generally employ low-dose chlorination to prevent larvae from settling rather than killing already established populations (Rajagopal et al. 2003c).

Other chemicals

M. sallei and *P. viridis* are also susceptible to copper toxicity (Chan 1988). Copper persistence in the environment should be considered when weighing up treatment options. Other chemicals have also been examined for their lethal effects on *P. viridis* but their use is not common because of difficulties with large-scale application and general safety.

These other chemicals include:

- silver nitrate—mean concentration = $30 \mu l/L$, LC50 = 96 hours (Mathew & Menon 1983)
- nitric acid—mean concentration = 4520 μm/L, LC50 = 168 hours (Chan 1988)
- pyrene—mean concentration = $32 \mu l/L$, LC50 = 10 hours (Peachey & Crosby 1996)
- selenium dioxide—mean concentration = 1,000 μ /L, LC50 = 96 hours (Patel et al. 1988)
- zinc sulphate—mean concentration = 300 μl/L, LC50 = 96 hours (Chan 1988)
- tributyltin oxide—mean concentration = $48 \mu l/L$, LC50 = 96 hours (Karande et al. 1993)

Use of the African endod berry has also received attention because of its ability as a molluscicide and the rapid degradation of its toxicity (Lemma et al. 1991). It has been used to control zebra mussels (at a mean concentration of 100,000 μ g/L, LC50 = 72 hours) and could be a cheap, biodegradable and effective control tool (Lemma et al. 1991). Products such as <u>BioBullets</u> can tailor the size of the toxin delivery system, thus improving target specificity and reducing impacts on non-target molluscs, however it may not be commercially available in Australia at the time of writing.

Descaling materials act by dissolving calcium components of the animals involved and can be effective means of controlling molluscs in small areas. Rydlyme is a commercial application of hydrochloric acid which has been used to treat *Perna viridis* infestations in niches of boats, its application in larger areas would require investigation.

6.1.1.2 Salinity manipulation

Salinity reduction through redirection of stormwater or other freshwater sources into the infested waterway (and/or by removal of seawater), has been used to control the algal pest *Caulerpa taxifolia* in enclosed waterways in South Australia. Both *Mytilopsis sallei and Perna viridis* can tolerate prolonged exposure to a wide range of salinities, from almost freshwater to seawater, with no obvious adverse effects. To attempt eradication of these species, salinity within the infested area would be need to be reduced to very low levels (less than 5 ppt) for a month or more.

Extreme salt concentrations have been shown to affect the ionic balance between the body fluids and the external environment in *M. sallei*. Salinity concentrations of more than 60 ppt reduce activity and spawning, and rapidly increase mortality (Ramachandra Raju et al. 1975).

Desiccation and oxidative stress are also effective ways of reducing mussel populations, but this requires control of water levels. This type of control option will be most applicable to fouling species in confined areas such as power plants, raw water systems, reservoirs, locked marinas and impoundments where water level is most easily manipulated (Almeida et al. 2005).

6.1.2 Physical treatments

Several physical treatments have been trialled for removing invasive biofouling bivalves. These include manual or mechanical collection, filters and sieves in water piping, temperature treatment (thermal shock, high heat or freezing), desiccation, and changes in water flow velocities (McEnnulty et al. 2000). Use of these treatments depends on the type of habitat in which the pest is located and the potential effects on the surrounding environment.

6.1.2.1 Removable structures

Ropes, mooring lines, buoys, floating pontoons and other structures within the infested area that can be removed from the water should be removed and treated on land. Procedures for treating these structures are described in <u>section 4.1.3.3</u> and could include:

- disposal to landfill
- air-drying for a minimum of seven days
- high-pressure water blasting
- immersion in chemical or fresh water baths.

6.1.2.2 Hard substrata and structures that cannot be removed from the water Hard substrata and structures that cannot be removed from the water include intertidal and submerged habitats.

Intertidal habitats

Hard intertidal substrata, such as wharf piles, exposed jetties and rocky shorelines may be treated when they are exposed at low tide.

Manual or mechanical collection can reduce populations of mussels, but both are relatively labour intensive. Localised patches of mussels may be removed by hand or scraped from affected surfaces and the removed animals disposed of to landfill. However, manual collection is costly, time consuming and inefficient when the incursion is large. As *P. viridis* and *M. sallei* generally inhabit hard or rocky substrates, mechanical collection is difficult over large areas.

Backpack heat guns or flame throwers have been suggested for removal of mussels above the low tide mark (National Taskforce on Black Striped Mussels 1999). This method was used in Port Adelaide River in 2018 to destroy *Crassostrea gigas* potentially infected with Oyster Herpesvirus type 1 in an attempt to reduce the viral load entering into the environment.

Submerged habitats

Physical removal by divers may be effective only in small areas and shallow water depths (less than 12 m). For example, in 1996 divers successfully eradicated a small, introduced population of the New Zealand greenshell mussel, *P. canaliculus* in the Gulf of St Vincent (South Australia) by manual collection (McEnnulty et al. 2000). Success with this technique requires intensive monitoring and repetition of removal as new individuals or clusters are located. If the source of the population is not contained or detection probabilities are low, this technique can rapidly become cost prohibitive. Destruction of bivalves through smashing of their shells can be less labour intensive than physical removal, however considerations such as the disease risk of the organisms or release of gametes should be considered.

Wrapping and encapsulating submerged substrata using impermeable barriers, such as polyethylene plastic, have been used successfully to treat fouling on structures such as wharf piles, jetties, pontoons, vessel moorings, small reefs and aquaculture facilities that cannot be removed from the water (Aquenal 2007). Black polyethylene bale wrap (1 m wide and 50µm thick) is wrapped over the structures, with an overlap of about 0.4 m on each successive layer of wrap, and secured using PVC tape to achieve a watertight seal. Aquenal provides details on the procedures for deploying the wrap on different structures and the costs involved with this treatment technique (Aquenal 2007). The wrappings can remain in place for extended periods (up to 12 months), thus providing some protection from reinfection. Should the outside of the wrappings become reinfested, their removal provides a second treatment option provided the animals are able to be retained when the wrap is removed. Parts of the structures that cannot be wrapped effectively may be treated by commercial divers using a hammer or an underwater flame torch.

Encapsulation techniques are most suited to treating small to medium-sized incursions (less than 10,000 m²) in relatively sheltered waters. The procedure is labour intensive and hazardous for divers. The wrap is susceptible to puncture and tearing by shipping, strong water currents and sharp oysters or tubeworms, which reduces its effectiveness. The technique is non-selective; all organisms contained within the wrapping will be killed.

Encapsulation or other containment techniques may also be used in combination with chemical treatment to achieve faster kill rates. Chemicals are injected into the covered area to maintain elevated concentrations of the biocide in close proximity to the fouled surface (Aquenal 2007).

6.1.2.3 Soft sediment habitats

In soft muddy sediments, large *Perna viridis* and clumps of *Mytilopsis sallei* may be removed by trawling or dredging. The type of dredge used will determine the effectiveness of the technique. Dredges normally used in bivalve fisheries (such as scallop dredges) are most efficient on soft, flat, muddy sediments, but are highly size-selective, with the size of mesh used on the dredge determining the size range of animals captured. The small size of *M. sallei* (less than 25 mm) means this form of dredge is unlikely to efficiently retain most animals.

In shipping ports and marinas, it may be possible to use the large, cutter-suction dredges used for capital works to remove upper layers of sediment (including any mussels) from throughout the infested area. Screening and disposal of sediment removed from the infested area must ensure no viable bivalves are returned to the water. To be effective, dredging must be accompanied by monitoring to ensure it treats as much of the infested area as possible.

An alternative to sediment removal is smothering by deposition of uncontaminated dredge spoil (Aquenal 2007). Technical advice should be sought on the source, type and quantity of sediment needed to ensure mortality of bivalves in treated areas. The efficacy of dredge spoil as a treatment option is also influenced by conditions at the site. It is most likely to be a viable option in sheltered areas where the seabed topography is relatively simple, to maximise persistence of capping. Deposited sediment will be dispersed rapidly in high energy, or highly complex habitats (such as rocky reef). The availability of a sufficient volume of uncontaminated dredge spoil should also be considered, along with any permits or government requirements (Aquenal 2007).

6.2 Open coastal environments

Limited emergency eradication response options are available to deal with marine pest incursions in open coastal environments, particularly on high energy coastlines or in deep water (more than 10 m). Many treatment options described in <u>section 6.1</u> may be applied to small-scale incursions in these environments, but the main difficulties occur in containing the bivalve larvae and in maintaining treatment conditions in a lethal state for sufficient time. The latter requires deployment of structures or application technologies that allow delivery of chemicals or encapsulation techniques over large areas and which are robust to water movement.

Successful eradication of small incursions may be possible using simple methods (such as manual removal, smothering, small-scale containment, or chemical treatment) if the incursion is detected early or where site-specific conditions allow containment and treatment. Trials of steam sterilisation on subtidal rocky reefs have shown some effectiveness for treating relatively small areas, but the efficacy of this technique is compromised in complex topographical environments, such as rocky reefs.

6.3 Monitoring and ongoing surveillance

Monitoring and surveillance are used to detect new populations or clusters of mussels and to inform eradication and control programs. Active surveillance for the presence of *Mytilopsis sallei* or *Perna viridis* or in restricted and control areas should continue until the incursion is declared eradicated or until the emergency response is stood down. If a zoning program is implemented, it will be necessary to implement targeted active surveillance for the species outside the restricted and control areas to support declaration of zones free from *P. viridis* or *M. sallei*. The Australian Monitoring Design Package (Version 1c), including the <u>Australian marine pest monitoring manual and guidelines</u>, can be used to help determine appropriate sampling intensity for ongoing surveillance.

Several methods may be appropriate for surveillance:

- systematic, targeted searches by divers or ROVs of suitable or treated subtidal habitat within the restricted area or at sites at risk of infection
- systematic, targeted searches by shoreline observers of suitable or treated intertidal habitat within the restricted area or at sites at risk of infection
- targeted searches and inspection of vessels and other vectors, including aquaculture stock and equipment, departing, or which have left, the control area
- regular monitoring of settlement arrays within the restricted area or at sites at risk of infection
- regular sample collection and analysis using molecular detections methods such as polymerase chain reaction (PCR).

A description of a spat collection device designed by the CSIRO for monitoring settlement of *M. sallei* is in <u>Appendix C</u>. This device is likely to be suitable for monitoring spatfall of *P. viridis*. Alternatively, 'hairy' ropes of different configurations can be used to monitor spatfall. These are commercially available in large quantities and are widely used to harvest wild spat of *Perna* and other mussel species for aquaculture.

PCR tests should be validated to quantify the sensitivity and specificity of assays. Work is underway to validate assays for both *P. viridis* and *M. sallei* in Australian environments. Your testing laboratory should be consulted as part of planning molecular surveillance activities.

Appendix A: Guidelines for using the Biosecurity Act during an emergency response to a marine pest of national significance

The following is an interim process for using the Biosecurity Act for action on vessels to treat contaminations by a marine pest of national significance. The Biosecurity Act may be used in certain circumstances, including where a biosecurity officer suspects on reasonable grounds, that the level of biosecurity risk associated with the vessel is unacceptable. Under these circumstances, a biosecurity officer may, in relation to a vessel that is under biosecurity control direct:

- the person in charge or operator of a vessel not to move, interfere with or deal with the vessel
- the person in charge or operator of a vessel to move the vessel to a specified place, including a place outside of Australian territory
- a vessel to undergo treatment action deemed necessary by the biosecurity officer
- that other biosecurity measures which may be prescribed by regulations be undertaken.

In addition, biosecurity officers may exercise certain powers, such as taking samples of ballast water from vessels, for the purpose of monitoring compliance with provisions for the management of ballast water at a port or offshore terminal within the outer limits of the EEZ of Australia. Where the Director of Biosecurity (or delegate) is satisfied that a sample of the vessel's ballast water indicates that the vessel poses an unacceptable level of biosecurity risk, then the Director may give a direction to the vessel not to discharge ballast water until conditions specified in the direction are met.

The conditions of using the Biosecurity Act are:

- The Australian Government Department of Agriculture is to be contacted before taking the proposed action to determine the appropriate provisions of the Biosecurity Act that apply.
- Directions to take action under the Biosecurity Act are to be given by a biosecurity officer. Officers of a state or territory government must be authorised as biosecurity officers under the Biosecurity Act to be able to give directions under the Act.
- Actions under the Biosecurity Act should only be taken for vessels currently identified as at risk of spreading a marine pest of national significance.

Responsibility for directing and approving action under the Biosecurity Act rests with the biosecurity officer, but the actual vessel control and treatment actions are handled by the Local or State Control Centre. As a matter of policy, the following information should be provided to the Australian Government Department of Agriculture to help determine appropriate application of the Biosecurity Act:

• the proposed course of action

- the location of proposed action
- details to identify the vessel involved in the proposed action
- contact details of local management agencies that will be managing the vessel control and treatment.

Appendix B: State and territory legislative powers of intervention and enforcement

The Intergovernmental Agreement on Biosecurity (IGAB), is an agreement between the Australian, state and territory governments. It came into effect in January 2019 and replaced the previous IGAB which started in 2012. The agreement was developed to improve the national biosecurity system by identifying the roles and responsibilities of governments and outlining the priority areas for collaboration to minimise the impact of pests and disease on Australia's economy, environment and community. The <u>National Environmental Biosecurity Response Agreement</u> was the first deliverable of the IGAB and sets out emergency response arrangements, including cost-sharing arrangements, for responding to biosecurity incidents primarily affecting the environment and/or social amenity and when the response is for the public good. In combination with the IGAB, Commonwealth, state and territory governments are responsible under their principle fisheries management legislation to respond consistently and cost-effectively to a marine pest incursion.

Jurisdiction	Agency	Principle acts covering emergency response arrangements	Marine pest contact website
Commonwealth	Department of Agriculture and Water Resources Department of Agriculture	Fisheries Management Act 1991 Biosecurity Act 2015	agriculture.gov.au/fisheries
New South Wales	NSW Department of Primary Industries	Fisheries Management Biosecurity Act 1995 Fisheries Management (General)Biosecurity Regulation 2017 Fisheries Management (Aquaculture) Regulation 2012 Ports and Maritime Administration Act 1995 Marine Parks Regulation 1997 Marine Safety Act 1998	<u>dpi.nsw.gov.au/fishing/pests-diseases</u>
Victoria	Victorian Fisheries Authority; Department of Jobs, Precincts and Regions (Agriculture Victoria)	Fisheries Act 1995 (protection of fisheries) Environment Protection Act 1970 (management of ballast water) Marine and Coastal Act 2018	https://vfa.vic.gov.au/operational-policy/pests-and- diseases/noxious-aquatic-species-in-victoria/aquatic-pests

Table B1 Commonwealth, state and territory legislation covering emergency response arrangements

Jurisdiction	Agency	Principle acts covering emergency response arrangements	Marine pest contact website
		<i>Marine Safety Act 2010</i> (power of Harbour Masters to direct vessels and duty of harbour masters to minimise adverse impacts on environment)	
		<i>Port Management Act 1995</i> (where no harbour master appointed, powers to direct vessels and act to minimise adverse effects on the environment)	
Queensland	ueensland Department of	Fisheries Act 1994	daff.qld.gov.au/fisheries/
Agriculture and Fisheri	Agriculture and Fisheries	Biosecurity Act 2014	www.qld.gov.au/environment/coasts-waterways/marine-pests
South Australia	Primary Industries and Regions SA	Fisheries Management Act 2007	pir.sa.gov.au/biosecurity/aquatics
Western Australia	Department of Fisheries	Fish Resources Management Act 1994 (under review)	fish.wa.gov.au/Sustainability-and-Environment/Aquatic- Biosecurity/Pages/default.aspx
Tasmania	Department of Primary Industries, Parks, Water and Environment	Living Marine Resources Management Act 1995	dpipwe.tas.gov.au/biosecurity-tasmania/aquatic-pests-and- diseases
Northern Territory	NT Department of Primary Industry and Resources	Fisheries Act 1988	nt.gov.au/marine/for-all-harbour-and-boat- users/biosecurity/aquatic-pests-marine-and-freshwater
			nt.gov.au/d/Fisheries/index.cfm?header=Aquatic%20Biosecurity

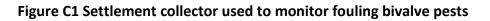
Appendix C: Spat collection devices for *M. sallei* and *P. viridis*

The spat collector consists of a rope backbone supporting horizontal pipe arms, to which artificial settlement surfaces are attached: PVC plates (120 mm by 120 mm) and rope mops (Figure C1). The arms of the collector are two 600 mm lengths of 20 mm poly pipe (plumbing risers), with holes drilled where settlement plates are attached with cable ties. A 600 mm (top piece) and a 300 mm (bottom piece) length of poly pipe are attached vertically to the cross piece and a 10 mm rope is threaded through both vertical pipes. A suitable weight is attached to the bottom of the rope. The top end of the rope is secured to a float or an appropriate structure, such as a mooring buoy or pontoon, allowing the device to move vertically with the tides while keeping the settlement surfaces at a depth of about 2 m. Cable ties are attached to one arm of the collector to identify each side as either 'tagged' or 'untagged'.

For the first three months, only the tagged side of the settlement collector is fitted with three settlement plates and a rope 'mop'. Inspections of the plates and rope mop are carried out monthly for marine pests. Inspections can also include recording a brief description of fouling organisms growing on the plates and within the rope mop, and photographing the plates (front and back) and rope mop.

After three months, three new settlement plates and a rope mop are fixed to the untagged side and similar monthly inspections occurs. To establish a quarterly collection regimen, collect the settlement surfaces from the tagged side only, three months after deployment.

Samples that cannot be inspected immediately should be immersed in ethanol or methanol, and the containers shipped to the nearest appropriate state agency or expert for identification. Removed plates and rope mops are to be replaced with new ones.



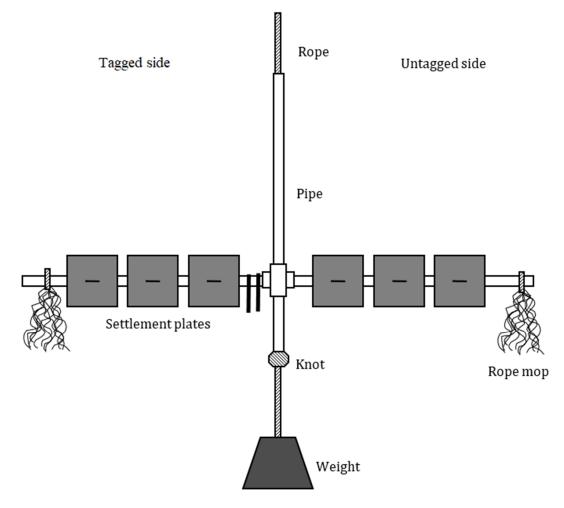


Image: Ferguson 2000 (in Maher et al. 2011)

Glossary

Term	Definition
CCIMPE	Consultative Committee on Introduced Marine Pest Emergencies
EMPPlan	Emergency Marine Pest Plan
IGAB	Intergovernmental Agreement on Biosecurity
IMO	International Maritime Organization
NBIRP	National biosecurity incident response plan
NEBRA	National Environmental Biosecurity Response Agreement
NIMPIS	National Introduced Marine Pest Information System
RRM	Rapid response manuals

Department of Agriculture

References

Almeida, E, Bainy, A, Dafre, A, Gomes, O, Medeiros, M & Di Mascio, P 2005, 'Oxidative stress in digestive gland and gill of the brown mussel (*Perna perna*) exposed to air and re-submersed', *Journal of Experimental Marine Biology and Ecology*, vol. 318, no. 1, pp. 21–30.

AMOG 2002, Hull fouling as a vector for transferring marine organisms: phase 1 study—hull fouling research, report by AMOG Consulting and MSE to the Commonwealth Department of Agriculture, Fisheries and Forestry, Canberra.

Aquenal 2007, *Pre-developing technology for marine pest emergency eradication response: review of current technology*, draft report prepared for the Department of Agriculture, Fisheries and Forestry, Canberra.

Azanza, MPV, Azanza, RV & Ventura, SR 2005, 'Heat shocking of Philippine green mussels, *Perna viridis*', *International Journal of Food Science and Technology*, vol. 40, no. 7, pp. 689–94.

Baker, P, Fajans, JS, Arnold, WS, Ingrao, DA, Marelli, DC & Baker, SM 2007, 'Range and dispersal of a tropical marine invader, the Asian green mussel *Perna viridis*, in subtropical waters of the southeastern United States', Journal of Shellfish Research, vol. 26, no. 2, pp. 345–55.

Baker, P, Fajans, JS, Baker, SM 2012, 'Habitat dominance of a nonindigenous tropical bivalve, *Perna viridis* (Linnaeus, 1758), in a subtropical estuary in the Gulf of Mexico', Journal of Molluscan Studies, vol 78, no 1, pp 28-33.

Bax, N, Hayes, KR, Marshall, A, Parry, D & Thresher, R 2002, 'Man-made marinas as sheltered islands for alien marine organisms: establishment and eradication of an alien invasive marine species', in CR Veitch & MN Clout (eds), *Turning the tide: the eradication of invasive species*, IUCN SSC Invasive Species Specialist Group, International Union for Conservation of Nature and Natural Resources, Gland, Switzerland and Cambridge, UK.

Blair, D, Waycott, M, Byrne, L, Dunshea, G, Smith-Keune, C & Neil, KM 2006, 'Molecular discrimination of Perna (Mollusca: Bivalvia) species using the polymerase chain reaction species-specific mitochondrial primers', *Marine Biotechnology*, vol. 8, pp. 380–5.

Branch, GM & Steffani, NC 2004, 'Can we predict the effects of alien species? A case history of the invasion of South Africa by *Mytilus galloprovincialis* (Lamarck)', *Journal of Experimental Marine Biology and Ecology*, vol. 300, no. 1–2, pp. 189–215.

Carver, CE, Chisholm, A & Mallet, AL 2003, 'Strategies to mitigate the impact of *Ciona intestinalis (L)* biofouling on shellfish production', *Journal of Shellfish Research*, vol. 22, no. 3, pp. 621–31.

Chan, HM 1988, 'Accumulation and tolerance to cadmium, copper, lead and zinc and by the green mussel *Perna viridis*', *Marine Ecology Progress Series*, vol. 48, pp. 295–303.

Chu, KH, Tam, PF, Fung, CH & Chen, QC 1997, 'A biological survey of ballast water in container ships entering Hong Kong', *Hydrobiologia*, vol. 352, pp. 201–6.

Coutts, A 2002, *The development of incursion response tools- underwater vacuum and filter system trials*, Cawthron report no. 755, Cawthron Institute, New Zealand.

Coutts, A & Forrest, B 2005, *Evaluation of eradication tools for the clubbed tunicate, 'Styela clava'*, Cawthron report no. 1110, prepared for Biosecurity New Zealand, Cawthron Institute, Nelson, New Zealand.

Department of Agriculture 2017, <u>Australian ballast water management requirements version 7</u>, Department of Agriculture, Canberra.

DPIF 2008, *Good news for users of Trinity Inlet*, media release, Queensland Department of Primary Industries and Fisheries, 2 May 2008.

DPIF 2007, *Suspect marine pest found in northern waters*, media release, Queensland Department of Primary Industries and Fisheries, 6 December 2007.

Eberhardt, LL & Thomas, JM 1991, 'Designing environmental field studies', *Ecological Monographs*, vol. 61, no. 1, pp. 53–73.

Eldredge, LG 1994, Introductions of commercially significant aquatic organisms to the Pacific Islands, South Pacific Commission, Noumea.

Ferguson, R 2000, <u>The effectiveness of Australia's response to the black striped false mussel</u> <u>incursion in Darwin, Australia</u>, a report of the marine pest incursion management workshop 27–28 August 1999, Department of Environment and Energy, Canberra.

Firth, LB 2011, Air temperature and winter mortality: Implications for the persistence of the invasive mussel, *Perna viridis* in the intertidal zone of the south-eastern United States' Journal of Experimental Marine Biology and Ecology, vol. 400 no. 1-2 pp 250-256.

Floerl, O 2004, <u>Protocol to quantify the level of fouling (LoF) on vessel hulls using an ordinal rank</u> <u>scale</u>, unpublished report for New Zealand Ministry of Fisheries project ZBS2004-03, Wellington, New Zealand.

Forrest, B & Blakemore, KA 2006, 'Evaluation of treatments to reduce the spread of a marine plant pest with aquaculture transfers', *Aquaculture*, vol. 257, pp. 333–45.

Furlani, DM 1996, <u>A guide to introduced marine species in Australian waters</u>, CSIRO technical report no. 5, Centre for Research on Introduced Marine Pests, CSIRO Division of Fisheries, Hobart.

Growcott, A, Kluza, D, Georgiades, E 2016, <u>Literature review: In-water systems to remove or treat</u> <u>biofouling in vessel sea chests and internal pipework</u>, MPI Technical Paper No 2016-16

Hayes, K, Sliwa, C, Migus, S, McEnnulty, F & Dunstan, P 2005, <u>National priority pests—part II: ranking</u> <u>of Australian marine pests</u>, report to the Department of Environment and Energy, CSIRO Marine Research, Melbourne, p. 106.

Heimowitz, P & Phillips, S 2006, *Rapid response plan for zebra mussels in the Columbia River Basin: a comprehensive multi-agency strategy to expeditiously guide rapid response activities*. Draft report, September 1, 2006.

Hicks, DW & Tunnell Jnr, JW 1993, 'Invasion of the south Texas coast by the edible brown mussel *Perna perna* (Linnaeus, 1758)', *Veliger*, vol. 36, no. 1, pp. 92–4.

Jenner, HA, Whitehouse, JW, Taylor, CJL & Khalanski, M 1998, 'Cooling water management in European power stations: Biology and control of fouling', *Hydroecologie Appliquee*, vol. 10, no. 1–2, pp. 1–225.

Karande, AA, Ganti, SS & Udhayakumar, M 1993, 'Toxicity of tributyltin to some bivalve species', *Indian Journal of Marine Science*, vol. 22, no. 2, pp. 153–4.

Karande, AA & Menon, NR 1975, '*Mytilopsis sallei*, a fresh immigrant in Indian harbours', *Bulletin of the Department of Marine Sciences*, University of Cochin, vol. 7, no. 2, pp. 455–66.

LeBlanc, N, Davidson, J, Tremblay, R, McNiven, M & Landry, T 2007, 'The effect of anti-fouling treatments for the clubbed tunicate on the blue mussel, *Mytilus edulis*', *Aquaculture*, vol. 264, pp. 205–13.

Lemma, A, Wolde-Yohannes, L, Fraliegh, PC, Klerks, PL & Lee, HH 1991, 'Endod is lethal to zebra mussels and inhibits their attachment', *Journal of Shellfish Research*, vol. 10, no. 2, pp. 361–5.

Lewis, JA & Dimas, J 2007, <u>Treatment of biofouling in internal seawater systems—phase 2</u>, Maritime Platforms Division, Defence Science and Technology Organisation, Fishermans Bend, Victoria.

McFarland, K, Baker, SM, Baker, P, Rybovich, M, Temperature, salinity, and aerial exposure tolerance of the invasive mussle, *Perna viridis,* in esturine habitats: implications for spread and competition with native oysters, *Crossostrea virginica,* Estuaries and Coasts, Vol. 38, no 5

McEnnulty, FR, Bax, N, Schaffelke, B & Campbell, ML 2001, <u>A review of rapid response options for the</u> <u>control of ABWMAC listed introduced marine pest species and related taxa in Australian waters</u>, technical report no. 23, CSIRO Marine Research, Hobart.

Maher, J, Beatty, A & Cribb, H 2011, *Monitoring for marine pests, Darwin Harbour, 2010–11 report*, Department of Resources, Northern Territory Government, Darwin.

Marelli, DC & Gray, S 1985, 'Comments on the status of recent members of the genus *Mytilopsis* (Bivalvia: Dreissenidae)', *Malacological Review*, vol. 18, pp. 117–22.

Mathew, R & Menon, NR 1983, 'Effects of heavy metals on byssogenesis in *Perna virids* (Linn.)', *Indian Journal of Marine Science*, vol. 12, no. 2, pp. 125–7.

Ministerial Council on Forestry Fisheries and Aquaculture 1999, *National Policy for the Translocation of Live Aquatic Organisms—Issues, Principles and Guidelines for Implementation*. Bureau of Rural Sciences, Canberra, ACT.

Mitchell, GK 2007, Hull encapsulation as an incursion response tool for marine fouling organisms: a method trial on a large vessel, Golder Kingett Mitchell Ltd, Auckland, New Zealand.

Morton, B 1981, 'The biology and functional morphology of Mytilopsis sallei (Recluz) (Bivalvia: Dreissenacea) fouling Visakhaptnam harbour, Andhra Pradesh, India', *Journal of Molluscan Studies*, vol. 47, pp. 25–42.

——1989, 'Life history characteristics and sexual strategy of *Mytilopsis sallei* (Bivalvia: Dreissenacea), introduced into Hong Kong', Journal of Zoology, vol. 219, no. 3, pp. 469–85.

NSPMMPI 2010, <u>Australian marine pest monitoring manual</u>, version 2, National System for the Prevention and Management of Marine Pest Incursions, Department of Agriculture, Canberra.

National Taskforce on Black Striped Mussels 1999, *National protocols for treatment of vessels and anchorages between Fremantle and Sydney potentially infected with the black striped mussel,* Department of Agriculture, Fisheries and Forestry, Canberra.

Neil, KM, Hilliard, R, Clark, P & Russell, BC 2005, *Situation and gaps analysis of IMS, vectors, nodes and management arrangements for planning the Northern Planning Area*, report by CRC Reef, URS Perth and the Museum and Art Gallery of the Northern Territory, prepared for the National Oceans Office Branch of the Department of the Environment and Heritage, Canberra.

Nel, R, Coetzee, PS & Vanniekerk, G 1996, 'The evaluation of two treatments to reduce mud worm (*Polydora hoplura Claparède*) infestation in commercially reared oysters (*Crassostrea gigas Thundberg*)', Aquaculture, vol. 141, pp. 31–9.

NIMPIS 2002a, <u>'Mytilopsis sallei'</u> (black striped mussel) species summary, National Introduced Marine Pest Information System.

NIMPIS 2002b, <u>'Perna viridis'</u> (Asian green mussel) species summary, National Introduced Marine Pest Information System.

NTG 2012, <u>Fishery status reports 2011 (PDF)</u>, fishery report no. 111, Northern Territory Government Department of Primary Industry and Fisheries, Darwin.

Patel, B, Chandy, JP & Patel, S 1988, 'Do selenium and glutathione inhibit the toxic effects of mercury in marine lamellibranchs?' *The Science of the Total Environment*, vol. 76, no. 2–3, pp. 147–65.

Peachey, RL & Crosby, DG 1996, 'Phototoxicity in tropical reef animals', *Journal of Marine Environmental Research*, vol. 42, pp. 1–4.

PIRSA 1999, *Green lipped mussels in South Australian waters information pamphlet: coast and clean seas*, Primary Industries and Resources South Australia, Adelaide.

Power, AJ, Walker, RI, Payne, K & Hurley, D 2004, 'First occurrence of the non-indigenous green mussel, *Perna viridis* (Linnaeus, 1758) in coastal Georgia, United States', *Journal of Shellfish Research*, vol. 23, no. 3, pp. 741–4.

Rajagopal, S, van der Gaag, M, Van der Velde, G & Jenner, HA 2002, 'Control of brackish water fouling mussel, *Mytilopsis leucophaeata* (Conrad), with sodium', *Archives of Environmental Contamination and Toxicology*, vol. 43, no. 3, pp. 296–300.

Rajagopal, S, Venugopalan, V, Nair, KVK, van der Velde, G, Jenner, HA & den Hartog, C 1998, 'Reproduction, growth rate and culture potential of the green mussel, *Perna viridis* (L.) in Ediayur backwaters, east coast of India', *Aquaculture*, vol. 162, pp. 187–202. Rajagopal, S, Venugopalan, V, Van der Velde & G, Jenner, HA 2003a, 'Comparative chlorine and temperature tolerance of the oyster *Crassostrea madrasensis*: implications for cooling system fouling', *Biofouling*, vol. 19, pp. 115–24.

Rajagopal, S, Venugopalan, V, Van der Velde, G & Jenner, HA 2003b, 'Response of fouling brown mussel, *Perna perna* (L.), to chlorine', *Archives of Environmental Contamination and Toxicology*, vol. 44, pp. 369–76.

Rajagopal, S, Venugopalan, V, Van der Velde, G & Jenner, HA 2003c, 'Tolerance of five species of tropical marine mussels to continuous chlorination', *Marine Environmental Research*, vol. 55, pp. 277–91.

Rajagopal, S, Venugopalan, V, Van der Velde, G & Jenner, HA 2006, 'Greening of the coasts: a review of the *Perna viridis* success story', *Aquatic Ecology*, vol. 40, pp. 273–97.

Ramachandra Raju, P, Mangapathi Rao, K, Ganti, SS & Kalyanasundaram, N 1975, 'Effect of extreme salinity conditions on the survival of *Mytilopsis sallei Recluz* (Pelecypoda)', *Hydrobiologia*, vol. 46, no. 2–3, pp. 199–206.

Rao, KS, Srinivasan, VV & Balaji, M 1989, 'Success and spread of the exotic fouling bivalve *Mytiliopsis* salei (recluz) in Indian waters', in M Mohan Joseph (ed.), *Exotic aquatic species in India: proceedings* of the Workshop on Exotic Aquatic Species in India, 25–26 April 1988, Asian Fisheries Society (Indian Branch), Mangalore, India.

Regan, TJ, McCarthy, MA, Baxter, PWJ, Dane Panetta, F & Possingham, HP 2006, 'Optimal eradication: when to stop looking for an invasive plant', *Ecology Letters*, vol. 9, no. 7, pp. 759–66.

Russell, BC & Hewitt, CL 2000, *Baseline survey of the Port of Darwin for introduced marine pest species*, report to the Northern Territory Department of Transport and Works, Northern Territory Government, Darwin.

Rydlyme Marine 2004, Cleaning engine raw water cooling water system, Rydlyme Marine, Illinois.

Siddall, SE 1980, 'A clarification of the genus *Perna* (Mytilidae)', *Bulletin of Marine Science*, vol. 30, no. 4, pp. 858–70.

Stafford, H & Willan, RC 2007, *Is it a pest? Introduced and naturalised marine animal species of Torres Strait Northern Australia*, Queensland Department of Primary Industries and Fisheries, Cairns.

Stafford, H, Willan, RC & Neil, KM 2007, 'The invasive Asian green mussel, *Perna viridis* (Linnaeus, 1758) (Bivalvia: Mytilidae), breeds in Trinity Inlet, tropical northern Australia', Molluscan Research, vol. 27, no. 2, pp. 105–09.

Tan, KS & Morton, B 2006, 'The invasive Caribbean bivalve *Mytilopsis sallei* (Dreissenidae) introduced to Singapore and Johor Bahru, Malaysia', *Raffles Bulletin of Zoology*, vol. 54, no. 2, pp. 429–34.

Therriault, TW, Docker, MF, Orlova, MI, Heath, DD & MacIsaac, HJ 2004, 'Molecular resolution of the family Dreissenidae (Mollusca: Bivalvia) with emphasis on Ponto-Caspian species, including first report of *Mytilopsis leucophaeata* in the Black Sea basin', *Molecular Phylogenetics and Evolution*, vol. 30, pp. 479–89.

URS Australia 2004, <u>Best practice for the management of introduced marine pests: a review</u> (pdf 1.92 mb), Global Invasive Species Programme.

Vakily, JM 1989, *The biology and culture of mussels of the genus 'Perna'*, International Center for Living Aquatic Resources Management and Deutsche Gesellshaft fur Technische Zusammenarbeit, Manilla, Philippines and Eschborn, Federal Republic of Germany.

Wells, F. 2018 A low number of invasive marine species in the tropics: A case study from Pilbara (Western Australia). *Management of Biological Invasions* 9:227-237 DOI: <u>10.3391/mbi.2018.9.3.05</u>

Woods, C, Floerl, O, Fitridge, I, Johnston, O, Robinson, K, Rupp, D, Davey, N, Rush, N & Smith, M 2007, *Efficacy of hull cleaning operations in containing biological material: II. Seasonal variability*, MAF Biosecurity New Zealand Technical Paper Series 08/11, prepared for BNZ Pre-clearance Directorate by National Institute of Water and Atmospheric Research Ltd.