



Australian Government

MARINE
PESTS

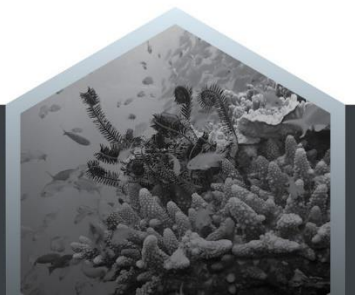
OFFICIAL

Marine pest response manual

Version 1.0, April 2024

Nationally agreed guidance material endorsed by the Marine Pest Sectoral Committee

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This publication is available at www.marinepests.gov.au/what-we-do/emergency/response-manuals

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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 marine 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.

Acknowledgement of Country

We acknowledge the Traditional Custodians of Australia and their continuing connection to land and sea, waters, environment and community. We pay our respects to the Traditional Custodians of the lands we live and work on, their culture, and their Elders past and present.

Note

Response manuals provide guidance for Australian marine pest biosecurity responses. 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, Fisheries and Forestry maintains a series of response¹ manuals 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 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 Response Manuals are 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 practical management for an emergency response to an incident caused by the suspicion or confirmation of incursion by a marine pest that is of national significance, but for which a species or taxa-specific response manual does not yet exist.

The National Institute of Water and Atmospheric Research, New Zealand, and the Department of Agriculture, Fisheries and Forestry, Australia, prepared the first edition of this Rapid Response Manual. The manual was revised as part of activity 3.4 and 3.5 of MarinePestPlan 2018-2023 (*Review the National Emergency Marine Pest Plan (EMPPPlan) framework and Plan and implement procedures to develop and update the EMPPPlan rapid response manuals and related guidance materials*, respectively). The manual has contributions from the South Australian Research and Development Institute. It has gone through an extensive process of editing and comment from the Marine Pest Sectoral Committee (MPSC) and relevant experts. The MPSC endorsed this manual on 9 April 2024.

The manual will be reviewed and updated as required 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 are 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 or corrections to this document, forward your suggestions to:

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Proposed changes will be considered by the MPSC before being incorporated into the manual.

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Introduction

Marine pests are non-native marine species introduced to areas outside their native range in which they can have negative impacts to Australia's marine environment, social amenity, or industries that use the marine environment. Preventing new introductions of marine pests is more cost effective than control (Leung et al. 2002). Where introductions occur, both short and long-term impacts and costs can be limited by a rapid and effectively managed response to the incursion (Campbell et al. 2018).

Manual purpose

Emergency response operations are most effective if they are based on detailed knowledge of the marine pest's life history, biology and ecology, ability to introduce or carry pathogens, and susceptibility to control or eradication measures. Response actions are most effective when taken immediately (or as soon as possible) after a marine pest incursion is first detected. The purpose of this document is to serve as a reference of management and technical information required to manage emergency responses to marine pests for which a taxa-specific response manual has not yet been developed.

The Marine Pest Response Manuals are a series of guidance documents that provide information on marine pest emergency response. This manual is part of the Response Manuals and is intended to be used in conjunction with other manuals to support marine pest response activities.

Taxa-specific [Marine Pest Response Manuals](#) have been prepared for several marine pests that the Marine Pest Sectoral Committee (MPSC) has identified as being of national significance:

- Response manual for invasive marine crabs
- Response manual for invasive marine bivalves
- Northern Pacific seastar (*Asterias amurensis*)
- Japanese seaweed (*Undaria pinnatifida*).

The [Biosecurity Incident Management System: Marine pest version](#) provides a uniform approach for managing responses to marine pest biosecurity incidents. It aligns with the response management approach applied to all biosecurity sectors. The manual provides guidance in contemporary practices for the management of marine pest biosecurity incident response and initial recovery operations in Australia.

The [National Introduced Marine Pest Information System](#) (NIMPIS) is a central repository of information on the biology, ecology and the distribution of over 150 marine pest species either introduced, established, or that pose a risk of future introduction to Australia. NIMPIS is a key source of information on introduced and exotic marine pest species of relevance to Australia.

Manual format

This response manual describes practical management for an emergency response to an incident caused by the suspicion or confirmation of an incursion by a marine pest of national significance, but for which a taxa-specific response manual does not yet exist. It is intended to be used in conjunction

with appropriate existing Australian Aquatic Veterinary Emergency Plan ([AQUAVETPLAN](#)) manuals, which detail the disposal, destruction, and decontamination for disease control if disease is co-introduced with a marine pest.

There are six main chapters within this manual:

1. Guidance and rationale for incursion response
2. Marine pest assessment
3. Pathways and vectors of introduction and spread
4. Preventing and monitoring spread
5. Containment, delimitation, and eradication
6. Decontamination, destruction, and disposal.

The [Australian Priority Marine Pest List](#) (APMPL) and [Exotic Environmental Pest List](#) (EEPL) have been developed to include marine pests that have national significance (Table 1). Within the appendices is taxa-specific information on 12 high-risk marine pests. The [AQUAVETPLAN](#) provides information on disease agents that could be introduced with some of these marine pests. This manual does not intend to replace AQUAVETPLAN information but provide linkages between marine pest management and aquatic disease management. 'Aquatic' for the purposes of this manual and the AQUAVETPLAN disease manuals includes marine, freshwater, estuarine and hypersaline waters.

1 Guidance and rationale for incursion response

Every biosecurity incident is unique, as is the response to the incident. Management actions taken during marine pest responses will differ based on variables such as the:

- pest-specific traits and their taxonomic or functional characteristics
- significance (environmental, economic, and social impacts)
- extent of the incursion
- value and location of the receiving environment
- likelihood of eradication.

This section discusses national policies that guide and support marine pest responses by providing a biosecurity response framework, operational guidance, and potential financial arrangements that can be tailored to meet the needs of each unique incident.

1.1 Sources of information

Information on the distribution, ecology, and effects of marine pests can be found via a variety of sources, including:

- scientists and technical experts
- primary sources of scientific literature
- online resources on marine pests.

The Marine Pest Sectoral Committee (MPSC) maintains a database of professionals and experts that can provide information on the life history, ecology, and biology of a marine pest. Contact the MPSC for more information: mpsc@aff.gov.au.

Several useful online resources contain summary information on various marine pests. These include:

- [National Introduced Marine Pest Information System](#) (NIMPIS)
- [Marine Pest Response Manuals](#)
- [National Priority Pests: Part II Ranking of Australian Marine Pests](#)
- [National Control Plans](#) are available for six species:
 - Northern Pacific seastar (*Asterias amurensis*)
 - Asian bag or date mussel (*Arcuatula [Musculista] senhousia*)
 - European green shore crab (*Carcinus maenas*)
 - Japanese seaweed or wakame (*Undaria pinnatifida*)
 - European basket shell clam (*Varicorbula gibba*)
 - European fan worm (*Sabella spallanzanii*)

- Additional distribution databases that can be used to search information on invasive species include:
 - [Atlas of Living Australia](#)
 - [World Register of Marine Species \(WoRMS\)](#)
 - [FishBase](#)
 - [Global Invasive Species Database](#)

1.2 Policies for management of marine pest responses in Australian waters

The [Biosecurity Incident Management System: Marine pest version](#) (BIMS: Marine pest version) manual provides guidance on policies and procedures for the management of biosecurity incident responses, including responses to marine pest emergencies within Australian waters.

1.2.1 Commonwealth, state, and territory authority responsibilities

Lead agencies in a response to a marine pest emergency should collaborate with and keep the Consultative Committee on Introduced Marine Pest Emergencies (CCIMPE) informed.

For incidents that are contained to a single jurisdiction, state coordination centres and local control centres may be established depending on the scale of the response. A national coordination centre is established to help manage concurrent incursions in more than one jurisdiction. National coordination operations will work in consultation with the CCIMPE representatives and relevant industry and community sector organisations. For further information on local, state and national control and coordination centres refer to the [BIMS: Marine pest version](#).

1.2.1.1 Consultative Committee on Introduced Marine Pest Emergencies (CCIMPE)

CCIMPE provides national technical coordination for managing marine pest emergencies and comprises biosecurity representatives from each Australian jurisdiction with coastal borders (the Australian Capital Territory is not represented).

CCIMPE is a national technical body that advises the National Management Group (NMG) on marine pest incidents and whether they meet the criteria for national cost-sharing under the [National Environmental Biosecurity Response Agreement 2.0](#) (NEBRA). The NMG is the peak, national biosecurity decision-making forum through which parties seek decisions in the event of an incident of a pest or disease (DAFF 2024a). See the [NEBRA](#) for the NMGs role and responsibilities.

CCIMPE provides response advice to lead agencies and assists in developing and implementing response actions such as a National Biosecurity Incident Response Plan (NBIRP). CCIMPE may also act as an information sharing forum to provide national biosecurity agencies with updates on marine pest responses that are not cost shared under the NEBRA.

1.3 Funding of operations and compensation

The [NEBRA](#) establishes national arrangements for responses to nationally significant biosecurity incidents where there are predominately environmental or public benefits. The NEBRA provides a mechanism to share responsibilities and costs for a response when eradication is considered feasible, the pest is considered to be of national significance, and the benefits of a response outweigh the costs and calculated to be cost-effective as per Schedule 3 of the [NEBRA](#). Guidance on undertaking a

benefit-cost analysis (BCA) for marine pest responses is available from Summerson, Hester & Graham (2018). Demonstrating that the benefits of a response outweigh the costs is required when seeking cost-sharing under the NEBRA.

CCIMPE may recommend to the NMG to consider a national cost-shared eradication response under the NEBRA if an incident is considered nationally significant, technically feasible to eradicate and cost beneficial to do so. Species on the [APMPL](#) and [EEPL](#) are already pre-considered to be of national significance.

Cost sharing must be agreed to by the NMG. The eligible costs of emergency eradication responses are shared as follows:

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

The 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, the 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 the NMG on a case-by-case basis where there is one or more private beneficiary and no existing arrangements.

Marine pest biosecurity incidents that do not meet the criteria for cost-sharing under the NEBRA will predominately be the responsibility of the lead agencies in the affected jurisdiction undertaking the response, however *ad hoc* resourcing (e.g. financial, human and physical) may be available through national biosecurity support programs such as the [National Biosecurity Response Team](#).

Please refer to the current version of the NEBRA or contact the NEBRA custodian nebra@aff.gov.au for more information as the NEBRA may be periodically revised.

1.4 Decision points

Decision points may include decisions to stand down eradication or control operations and determining the current status of marine pests.

Detection of any marine pest not known to occur in Australia should initiate an investigation phase. This phase will likely be run concurrently with the initial control actions if initial indications are that the infestation is limited. If the emergency investigation revealed that the incursion was potentially eradicable, then the incident manager will prepare a NBIRP and forward to CCIMPE for urgent consideration.

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

- 1) [investigation](#) and [alert phase](#)
- 2) [operational phase](#)

3) [stand-down phase](#).

Further details on decision points can be found in the [BIMS: Marine pest version](#). It is important to note that not all detections of marine pests will initiate a response involving all three phases. For example, the detection of marine pests on a vessel may involve a truncated response.

1.4.1 Calculating optimal sample numbers and when to stand down a response

Quantification of response sampling numbers and the best time to stand down a response are technical assessments. Advice from statisticians, ecologists, economists, or other relevant experts should be sought.

[EpiTools](#) offers several tools to assist in decision making for sampling numbers and is freely available and easy to use. The South Australian Research and Development Institute (SARDI) has developed a sample number calculator for surveillance using plankton samples tested with quantitative polymerase chain reaction (qPCR) assays ([Survey Sample Number Calculator](#)). Both tools require estimates of survey confidence, target abundance and test performance to calculate the number of samples required.

In many cases a decision on a surveillance program to meet the requirements of the situation may be discussed and agreed by CCIMPE. This will take into account the context of the situation and the issues around conducting a surveillance program.

1.4.2 Determining the current status of marine pests

The current status of marine pests (previously called ‘proof of freedom²’) aims to demonstrate to an agreed level of confidence that a pest, if present, is at a low enough abundance that it can be regarded as effectively eradicated. It requires a robust and intensive surveillance program during the operations phase of the response. The purpose of determining marine pest status is to inform future decisions, mainly whether a response can be stood down once the associated surveillance is complete, or whether further ongoing management is required. The outcome of surveillance for marine pest status may influence management actions such as movement restrictions, ballast water and biofouling management.

The Marine Pest Sectoral Committee (MPSC) has developed national *Policy principles for determining the current status of marine pests* ([Appendix F](#)). The policy principles provide stakeholders (governments, industry and others) with nationally agreed and flexible principles for determining the status (likelihood of presence/absence) of marine pests in defined areas within Australia.

Responses that are cost-shared under the [NEBRA](#) require a ‘proof of freedom’ phase if eradication is thought to have been achieved. The NEBRA custodian (nebra@aff.gov.au) can provide guidance on developing surveillance programs for marine pest status on request.

² The term ‘proof of freedom’ was previously used in marine pest responses. However, ‘proof of freedom’ has different connotations, especially from an agricultural disease perspective. As such, the MPSC agreed to retire usage of ‘proof of freedom’ for marine pests, and instead have adopted ‘current status of marine pests’ to describe the evidence that a specific marine pest is absent from a geographical region. ‘Proof of freedom’ may still be used interchangeably in some circumstances or in older documents.

Ultimately, surveillance for marine pest status will depend upon the context and requirement. CCIMPE can provide advice and connection to expertise to assist in developing a 'proof of freedom' surveillance plan.

Information for calculating the optimal number of surveys to conduct after freedom is assumed to have been achieved is available from Regan et al. (2006).

1.5 Health, Safety and Environment

1.5.1 Safety of response personnel

The safety of personnel involved in response activities is paramount. Handling certain aquatic animals may be dangerous. Many methods for response activities also involve divers working under water or in outdoor environments. Personnel may work extended hours to achieve control and eradication. Fatigue in personnel can compromise their safety and that of others, particularly if they are working with machinery or in dangerous environments.

1.5.2 Work health and safety during a response

All operations associated with a marine pest incursion must consider relevant Commonwealth, state and territory government work health and safety (WHS) requirements, standard operating procedures (SOPs) and safety data sheets (SDS') for response activities, including handling chemicals and samples. For example, chlorine in liquid form can cause 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 SDS, which should be available to staff working with a chemical.

1.5.3 Environmental considerations

When a response takes place there may be considerable waste generated which needs consideration prior to commencing. Certain techniques will generate large quantities of plastic wastes or involve chemical applications, some of which may have residual effects (e.g. cupric compounds). Disposal of large quantities of organic wastes needs careful consideration and appropriate disposal areas and transport corridors identified. See [Section 6](#) for more information.

Response actions may have impacts on non-target species within the response area and an environmental impacts assessment should include non-target species. This may include threatened or listed species and culturally significant species.

Response actions also need to consider the surrounding environment. Some high priority areas such as reserves, Sea Country, national parks and Ramsar wetlands will need consideration as to what methods of management are most appropriate. Effective communication of public access, including potential restrictions, to locations and when they will be completed is crucial.

2 Marine pest assessment

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 may pose, the feasibility of response options and the design of efficient methods for surveillance, containment, eradication, and control. When determining the nature of the marine pest, a manager should understand:

- does it have a demonstrable history of invasion?
- what are its life history, ecology, and environmental tolerances?
- what are its potential impacts?

To assist in an effective emergency response, this section will provide and discuss resources to identify the marine pest's history of invasion, ecology, and biology, and identify the impact(s) a marine pest may have.

2.1 Identify the pest

Determining the identity of a suspected marine pest is the first step in initiating a marine pest emergency response. This normally requires specimens recovered from a suspected incursion to be examined by a recognised taxonomic expert or diagnostic facility. Relevant research and curatorial staff within state and territory museums and research institutions should be consulted for identification as they are connected to national and international networks of taxonomic and systematic expertise.

For many organisms, identification is only possible if key diagnostic features are preserved appropriately when the specimen is collected. Guidance on appropriate techniques for collecting and preserving specimens from different marine taxa is presented in [Appendix E](#).

2.1.1 Is the species a marine pest of national significance?

The [Australian Priority Marine Pest List](#) (APMPL) includes species that have been agreed as being of national significance. Ten species are listed, with seven of these being exotic. These species meet the criteria of the [National Environmental Biosecurity Response Agreement 2.0](#) (NEBRA) as being species of national significance and would potentially be eligible for cost-sharing of a biosecurity response. An [Exotic Environmental Pest List](#) (EEPL) has also been developed and includes exotic environmental pests, weeds and diseases, including exotic marine pest species of national significance. Marine pest emergencies may be declared when a combat jurisdiction (the affected biosecurity agency, sometimes referred to as the notifying party), considers the marine pest to have real or significant impacts.

Table 1 Marine pest species listed on the Australian Priority Marine Pest List and/or the Exotic Environmental Pest List

Species	Established/Exotic	List membership
European shore crab, <i>Carcinus maenas</i>	Established	APMPL
Northern Pacific seastar, <i>Asterias amurensis</i>	Established	APMPL
Japanese kelp, <i>Undaria pinnatifida</i>	Established	APMPL
Asian paddle crab, <i>Charybdis japonica</i>	Exotic	EEPL
Chinese mitten crab, <i>Eriocheir sinensis</i>	Exotic	APMPL/EEPL
Harris mud crab, <i>Rhithropanopeus harrisi</i>	Exotic	APMPL
Black striped false mussel, <i>Mytilopsis sallei</i>	Exotic	APMPL/EEPL
New Zealand green-lipped mussel, <i>Perna canaliculus</i>	Exotic	APMPL
Brown mussel, <i>Perna perna</i>	Exotic	APMPL
Asian green mussel, <i>Perna viridis</i>	Exotic	APMPL/EEPL
Charru mussel, <i>Mytella strigata</i>	Exotic	APMPL
Carpet sea squirt, <i>Didemnum vexillum</i>	Exotic	EEPL

The Consultative Committee on Introduced Marine Pest Emergencies (CCIMPE) may consider an emergency response to marine pests not on any lists if they meet at least one of the NEBRA national significance criteria, which relate to:

- the environment
- people, including human infrastructure and social amenity
- business activity.

These species will be considered on a case-by-case basis, using as much information as possible to determine whether the species warrants activation of an emergency response and development of a National Biosecurity Incident Response Plan (NBIRP).

2.1.2 Does the species have a demonstrable history of invasion?

To demonstrate a history of invasion, the investigation must be able to show that:

- the species has previously established or has the ability to establish self-sustaining populations outside its native range as a result of intentional or accidental transport by a human-mediated vector (or vectors)
- the non-native populations have affected the economy, environment, human health, or amenity of the region in which they established.

Caution should be taken with any introduced species when there is no knowledge of the species' invasive history.

2.1.3 What is the life history, ecology, and environmental tolerances of the species?

Information on the life history, habits, ecology, and environmental tolerances of the species is needed to understand the potential range, vectors for spread, and control methods that may be used as part of a response. The type of information needed to determine the nature of the pest is summarised in Table 2.

Table 2 Life history of suspect marine pest

Feature	Further explanation
Maximum size of adult stage	n/a
Maximum age of adult stage	n/a
Maximum duration of juvenile stage	n/a
Minimum time to sexual maturity	n/a
Minimum size at sexual maturity	n/a
Type of reproduction	Sexual/asexual
Mating strategy	Internal/external fertilisation
Dispersal stage	Gametes/juveniles/adults
Potential dispersal distance (single generation)	n/a
Feeding mode	Autotrophic/herbivore/planktivore/predator/deposit feeder/suspension feeder
Depth range	n/a
Preferred habitat	n/a
Distribution within population	Gregarious/scattered/solitary
Locomotion	Motile/sedentary/sessile
Environmental tolerances	Salinity, temperature, pH, toxicant tolerance

n/a Self-explanatory.

Standardised life history categories may help estimate the potential costs and feasibility for marine pest eradication (Table 3), in combination with information within Table 1, Table 2, Table 4, Table 5, Table 6, Table 7, Table 8 and Table 9.

Table 3 Example standardised life history variables

Variable	Levels
Size of organism (include season variance)	Small: <5 cm Large: >5 cm
Appearance (camouflaged or other)	Cryptic Obvious
Habit	Solitary Grouped
Preferred habitat	Pelagic Benthic, hard substrate Benthic, soft substrate Benthic, hard and soft substrate Artificial substrate
Larval duration/incubation period	Short: hours to days Medium: days to weeks Long: weeks to months
Time to maturity	Short: <2 months Medium: 2 to 12 months Long: >1 year
Propagules per reproductive event	Low: <10,000 Moderate: 10,000 to 1,000,000 High: >1,000,000
Sexual reproductive cycles per year	Annual Biannual More frequent/continuous

Source: Crombie et al. (2007)

Multiple types of information on the life history can be useful in evaluating the likely success of an eradication attempt and in designing an appropriate response to an incursion.

2.1.3.1 Reproduction and growth

Reproductive mode

Marine organisms exhibit a wide variety of mating systems and modes of reproduction. Some are capable of reproducing both sexually and asexually. Different modes of reproduction can lead to more rapid population growth or greater dispersal potential. Identifying the reproductive mode of marine pests is highly important, as reproduction is an important factor that influences their successful establishment and further spread.

Marine species that reproduce sexually can have a range of different mating strategies. In some taxa, the sexes are separate, and fertilisation occurs internally through copulation (internal fertilisation) between separate male and female individuals (such as most decapod crustaceans and cephalopods). This requires aggregation and direct physical contact between mating individuals. Other groups release fertile gametes into the water and fertilisation occurs in the water column (such as most fish species and corals), or the female retains the eggs but males shed sperm and fertilisation can occur within the body cavity (such as some species of polychaetes).

Some marine species reproduce asexually, where fertilisation is not required to reproduce. Types of asexual reproduction can include:

- fragmentation: a new organism grows from a fragment of the parent (such as some polychaete worms, seastars, sea sponges, algae, ascidians)
- budding: small buds or tendrils are produced from the body tissues of the parent, which grow to be miniature adults and break away from the adult when they are mature (such as some hydrozoa, bryozoans, ascidians; e.g. *Didemnum vexillum*)
- vegetative growth: new individuals are formed through growth of specialised leaves, bulbs, rhizomes or stolons (such as stoloniferous algae; e.g. *Caulerpa taxifolia*)
- spore formation: some algae produce spores by mitosis that are capable of regenerating into an adult plant (such as *Polysiphonia* species)
- parthenogenesis: an unfertilised egg develops into a new individual (such as some small crustaceans; e.g. cladocerans and ostracods).

In many marine invertebrates, algae and some fish, individuals can produce both male and female sexual organs (hermaphroditism). This can occur either simultaneously or sequentially. Some hermaphroditic species can produce viable offspring through self-fertilisation ('selfing'). Species capable of selfing can establish self-sustaining populations from a single founding individual.

Life history structure

Most marine invertebrates and fish have a biphasic life history that involves morphologically distinct larval and adult stages. Often, the adult stage is demersal or benthic while the larval stages are planktonic (occurs in the water column).

Algae have a variety of complex life-history strategies. There is no typical life cycle for algae. Many algae can reproduce asexually as a result of vegetative growth or fragmentation. Others produce spores asexually which can germinate into genetically identical individuals, and still other algae have complex life cycles that involve a mixture of sexual and asexual reproductive stages.

Dispersal life stages

For many marine species, the juvenile stages (larvae or spores) are the main form of dispersal. Other species form resistant cysts that can lie dormant for long periods before releasing viable individuals. However, significant dispersal may also be achieved by movement of adults. This may occur because the adults themselves are mobile and actively move among different feeding or breeding habitats, or because sedentary individuals are transported by water currents, attached to drifting substrata, as detached adults or fragments, or via anthropogenic vectors.

Time to reproductive maturity

An eradication attempt has a greater chance of success if the incursion is discovered before the population has an opportunity to reproduce, particularly if the gametes or juvenile life stages are also important for dispersal. If the size at reproductive maturity and rate of growth from settlement to maturity are known, it may be possible to infer if reproduction has taken place from the size (or age) distribution of individuals within the population or from the presence and state of reproductive tissue.

Similarly, in some cases it may be possible to estimate how long the population has been present by using known rates of growth and the size (or age) distribution of individuals within the population to determine an estimated time of settlement or recruitment.

Fecundity

The fecundity, or reproductive capacity, of an organism is the number of potentially viable propagules or gametes (offspring) that an individual can produce during a single spawning event. Some marine organisms are extremely fecund, with a single, successful reproductive event by a few mature individuals resulting in many tens of thousands of offspring, thereby reducing the likelihood of successful eradication.

The fecundity of an organism can have several dimensions, including the number of:

- propagules, gametes or offspring produced during a single reproductive event
- reproductive events (or cycles) that a mature individual has in a season or a year
- seasons or years that a mature individual can continue to reproduce.

2.1.3.2 Life habit

Relevant marine environments for life stages

An effective emergency eradication response involves locating and treating all susceptible individuals or reducing the infestation to levels that cause irreversible declines in reproductive success or survival within the population (Allee effects). To determine the extent of an incursion, it is necessary to identify the range of marine environments the species can inhabit, including all life stages of the species when these occupy different environments or habitats. Table 4 summarises the range of coastal environments and habitat types that should be considered.

Table 4 Example coastal environment variables

Environmental variable	Environment type
Coastal geography	Brackish rivers and creek Lagoons and coastal lake Estuaries and coastal embayment Open coast
Water depth	Intertidal Subtidal <2 m 2–15 m >15 m
Habitat	Soft sediment (such as muds or sands) Natural hard substrata (such as rocky reef, cobbles, shell debris, encrustations) Artificial hard substrata (such as wharf piles, pontoons, jetties, buoys, ropes) Seagrass meadow Algal bed Mangrove forest Saltmarsh Coral reef Plankton/nekton

Environmental tolerances

Knowing the organism's ability to withstand short- or long-term changes in water temperature, salinity, pH, or other environmental conditions can be useful for:

- evaluating the likelihood of that species surviving and establishing self-sustaining populations within Australia
- identifying local environments in which the species may survive
- estimating the likely geographic range over which the species could survive if allowed to spread
- devising methods for treating infested vectors and marine environments.

Life-cycle models, based on temperature tolerance, have been developed for several species to predict their potential distribution range within Australia (Hayes et al. 2007) and are available on the [NIMPIS](#) website.

When published information about the range of ambient water temperatures and salinity in which the species can survive is limited, it may be possible to infer these by examining the variation in ocean temperature and salinity that occurs over the known geographic distribution of the species. Data on broadscale (1 degree of latitude and longitude) ocean climatology, including *in situ* temperature, salinity and dissolved oxygen at standard depths, are available from the [World Ocean Atlas](#).

2.2 Marine pest species impact assessment

Exotic marine species can alter the dynamics of the coastal ecosystems to which they are introduced. The type, magnitude, and extent (that is, spatial and/or temporal) of the change they cause depends on the ecology and life history of the species and the characteristics of the environment and biota into which they are introduced. Evaluating whether such changes are likely to constitute major impacts on the economy, the environment, human health, or the amenity of Australian marine resources requires consideration of the likelihood that the changes will occur and of the severity of the consequences.

The amount of information and data available to undertake such evaluations will vary with each pest, and the veracity of the evaluation will vary with the tools and expertise available. [NIMPIS](#) provides detail of known or potential impacts caused by introduced and exotic marine pest species of relevance to Australia. Quantitative or qualitative methodologies should be used to assess whether the species may cause major impacts and to estimate the uncertainty associated with the evaluation. If a qualitative assessment is undertaken, expert advice should be used to make judgements about potential impacts and their consequences.

A simple, qualitative assessment of consequences resulting from a species impact can be undertaken using these steps:

- 1) List the full range of impacts the species could have on social amenity, the economy, and the environment. A standardised list of 16 impact categories modified from Hayes et al. (2005) used to rate the potential impacts in Australia of 112 exotic or cryptogenic marine species is shown in Table 5 as a guide and is consistent with Schedule 2 of the [NEBRA](#).

- 2) Evaluate the likelihood that each impact will be realised. Likelihood values can be estimated as probabilities of occurrence or using a simple five-point scale (Table 6).
- 3) Evaluate the likely severity (consequence) of each type of impact if it were to be realised. A simple scale of consequence level can be constructed for comparative purposes (Table 6).
- 4) Calculate an overall score potential for each type of impact listed in Step 1, using the evaluations made in Step 2 and Step 3, or use a simple risk matrix table (Table 6) to identify the likely impact.

Table 5 Categories of the potential impacts of marine pests

Impact category	Impact
Social amenity	Adverse effects on human health Reduced marine environment aesthetics Reduced access for recreation activities (e.g. diving or fishing)
Economic	Adverse effects on aquatic transport Fouling of industrial pipes and canals Reduction of aquaculture, commercial or recreational fishery profitability Loss of public or tourist amenity Damage to marine structures or archaeology Management costs
Environmental	Detrimental habitat modification Adverse effects on trophic interactions and food webs Domination of or out-competes and limits resources of native species Predation of native species Introduction or facilitation of new pathogens, parasites or other non-indigenous species Alteration of bio-geochemical cycles Induction of novel behavioural or eco-physical responses Genetic impacts: hybridisation and introgression Herbivory
Cultural	Degradation of cultural assets valued by a significant segment of the community Alteration to practices and customs of a significant segment of the community Persistent and substantial negative change in national and international perception of attributes relevant to the national image

Source: Adapted from Hayes et al. (2005) and the NEBRA (DAFF 2024a).

Table 6 Example of a risk matrix used for qualitative risk analysis

Likelihood	Consequence				
	Insignificant	Minor	Moderate	Major	Significant
Rare	N	L	L	M	M
Unlikely	N	L	M	H	H
Possible	N	L	M	H	E
Likely	N	M	H	E	E
Almost certain	N	M	E	E	E

Note: Letters represent risk level for a given combination of the likelihood of an event and its consequences: **N** negligible. **L** low. **M** moderate. **H** High. **E** Extreme.

2.3 When little or no information is available on the suspected pest

When little or no information is available on a marine pest, a decision to mount an emergency response may need to be based on information of a closely related species. The closely related species should be functionally similar to the marine pest's life history (e.g. similar morphology, reproductive behaviour, or feeding modes). Information on species from similar genera or families could help determine which elements of life history and ecology are likely to be conserved across related species and may be exhibited by the marine pest.

3 Pathways and vectors of introduction and spread

Introduction pathways for marine pests can be either primary or secondary. A primary pathway moves species to new regions across biogeographic barriers, whereas a secondary pathway is the spread and dispersal of introduced species between points within or between neighbouring regions (Harrower et al. 2018). Once introduced into Australia, marine pests may subsequently spread to new locations by various vectors, which are the physical means, agent or mechanism that facilitates the transfer of organisms, or their propagules, from one place to another (NIMPIS 2023).

Details of pathways and vectors for the introduction and spread of marine pests in Australia are provided in this section. Considered to pose the most serious threat to Australian waters are:

- transport of biofouling on seagoing vessels and other maritime infrastructure
- discharge of ballast water.

Marine pests can also be introduced and spread by:

- fisheries, aquaculture, and the ornamental trade
- natural dispersal (e.g. currents)
- debris and flotsam.

Other pathways likely to be of importance for introduction and translocation of species within Australia are considered in Table 7.

[Marine Pest Response Manuals](#) have been established for several taxa, describing pathways and vectors for consideration when there is a suspected incursion. Risk assessments can also be used to identify and prioritise vectors that spread marine pests. The risk of dispersal depends on the species and range and abundance of relevant vectors operating in the invaded area. An overview of risk assessments is provided by Laeseke et al. (2020).

Species Distribution Modelling, also known as Habitat Suitability Modelling or Ecological Niche Modelling, can be used to predict distributions of aquatic species (Melo-Merino et al. 2020). There are two main families of models:

- mechanistic models – where species biology is well understood (Jofré Madariaga et al. 2014)
- correlative models – require data on species presence locations, but can be applied where species biology is not well understood (Castelar et al. 2015).

Models for predicting spread are summarised by Wonham and Lewis (2009). For more information on modelling see [Section 5](#).

Table 7 Categories of potential pest pathways and vectors into and within Australia

Pathway or vector	Description
Biofouling (vessel)	Attached fouling or free-living organisms associated with the vessel Associated with cargo With sea water systems or other deck basins
Ballast water (vessel)	With solid ballast (such as with rocks or sand) With ballast water, or other deck basins
Fisheries, aquaculture and ornamental trade	Deliberate translocation of fish or shellfish to establish or support fishery Accidental with deliberate translocation of fish or shellfish With fishery and aquaculture products, packing or substrate With or as bait, (inc. live bait wells) With 'live rock' sales or discard of aquarium life, gravel and water With aquaculture equipment
Debris and flotsam	Transport of species on marine debris (includes driftwood)
Natural dispersal	Through the movement of larvae or adults
Biological control	Deliberate translocation as a biological control agent Accidental translocation with deliberate biological control release
Navigation buoys, marine floats	As attached or free-living fouling organisms
Recreational equipment	Accidental with recreational equipment With live bait wells
Scientific research	Deliberate or accidental release with research activities
Plant introductions	Deliberate translocation of plant species (such as for erosion control) Accidental with deliberate plant translocations
Individual release	Deliberate or accidental release by individuals (e.g. aquariums, traditional belief-based live release practices)
Canals	Natural range expansion through man-made canals
Seaplanes	As attached or free-living fouling organisms

Source: Adapted from Hayes et al. (2005)

3.1 Biofouling

Biofouling can occur on all fixed or mobile structures immersed or exposed to the water. Fouling communities typically comprise of sessile and encrusting organisms such as algae, barnacles, bivalves, tubeworms, hydroids, and ascidians that have attached and are in a sessile life-stage. If the fouling layer is dense enough, it can provide shelter and support mobile species such as amphipods, crabs, seastars, and fish that may live in or among the fouling species.

Fouling communities can be found in submerged recesses and any wet surface, such as anchor wells, sea chests, bow thrusters, internal piping, and propeller shafts; collectively referred to as niche areas ([Section 4.2.1](#)). Niche areas may be more susceptible to biofouling because they are sheltered from water shear and may be free of antifouling paint. Sea chests are particularly capable of translocating abundant marine communities of both attached and free-swimming species (Coutts, Moore & Hewitt 2003).

International and domestic shipping has facilitated the spread of marine pests more than any other vector, as a result of transport in ballast water and biofouling assemblages (Brockerhoff & McLay 2011). Potential vectors include a diverse range of craft, including commercial ships, such as tankers and container ships, military vessels, fishing vessels, recreational vessels, passenger vessels, barges, dredges, and research vessels. Biofouling on the hull of vessels or in their internal seawater systems is one of two main ways that vessels can act as vectors for marine pests (the second is ballast water, see [Section 3.2](#)). Species within biofouling assemblages can be introduced by:

- spawning or fragmentation of a fouling species present on a vessel while in port followed by its successful settlement and establishment of a reproductive population
- the dislodgment or disturbance of fouling species from a vessel in port (e.g. through hull cleaning or abrasion with wharf piles)
- the sinking of a fouled vessel (MPSC 2022).

Dry-docking and in-water operation principles and recommendations are contained in the [Anti-fouling and in-water cleaning guidelines](#). The guidelines provide guidance on best-practice approaches for the application, maintenance, removal and disposal of anti-fouling coatings and the management of biofouling and invasive aquatic species on vessels and movable structures in Australia and New Zealand. The practices described in these guidelines have been aligned with international conventions intended to protect the aquatic environment from invasive aquatic species and contaminants from shipping. These include the:

- International convention on the control of harmful anti-fouling systems on ships
- 1996 protocol to the Convention on the prevention of marine pollution by dumping of wastes and other matter, 1972
- 2011 Guidelines for the control and management of ships' biofouling to minimize the transfer of invasive aquatic species.

These guidelines will be updated and released in 2024, and published on the Department of Agriculture, Fisheries and Forestry (DAFF) website; [in-water cleaning in Australia - DAFF \(agriculture.gov.au\)](#). The Australian Government *Biosecurity Act 2015* (hereafter, the *Biosecurity Act 2015*) can be used in the absence of appropriate state or territory legislative powers and may be used in circumstances that include directing conveyances.

Marine aquaculture equipment such as buoys, ropes, nets, and cages, can contribute to the spread of marine pests if they are fouled. During a marine pest emergency response, the cleaning of stock and equipment, and reduced or ceased movement of these should be appropriately managed.

Fixed marine structures such as pontoons, moorings, or piles do not represent a risk for translocation of marine pests unless they are moved while still fouled. If an emergency response to a marine pest is underway, then scheduled installation or repair of marine structures should be appropriately managed, including any support vessels or equipment used.

Biofouling management for vessels and infrastructure should be consistent with the [National Biofouling Management Guidelines](#). These are available for the following industries and operators:

- [aquaculture](#) industry
- [offshore infrastructure](#) (petroleum production and exploration industry)
- [port and marina operators](#) (marinas, slipways, boat maintenance and recreational boating facilities)
- [vessels](#):
 - [commercial fishing vessel](#)
 - [commercial vessel](#)
 - [non-trading vessel](#)
 - [recreational vessel](#).

3.2 Ballast

Ballast water is water taken on-board by vessels to maintain stability and trim. Unladen vessels arriving in a port will usually be ballasted and will need to discharge some of its ballast water in proportion to the weight increase caused by cargo loading.

Ballast water is a relatively non-selective dispersal mechanism potentially carrying species from the site where ballast water was taken up. Ships can unintentionally transport diverse assemblages of marine species in ballast water. Ballast water can contain the planktonic stages of organisms, free swimming juveniles or adults, and fouling organisms attached to the vertical walls of the ballast compartments (Carlton 2001). Sediments can be inadvertently taken up along with the ballast water and can accumulate in the ballast tank, providing habitat for benthic organisms that may be transported to other locations (Carlton 2001).

3.3 Fisheries, aquaculture and the ornamental trade

Fishing and aquaculture operations and the ornamental trade can translocate marine pests accidentally with aquaculture stock (particularly shellfish), equipment such as buoys, ropes, nets, and cages, or bait, or deliberately by illegal importation or translocation.

The risk of introduction of a marine pest into Australia via importing aquaculture stock is lower than biofouling and ballast because there are strict regulations of live animal imports (see [List of Specimens taken to be Suitable for Live Import](#) under the Environment Protection and Biodiversity Conservation Act 1999). Aquaculture can still be a significant secondary pathway for domestic spread of marine pests (MPSC 2022; Roche & Torchin 2007). Similarly, there is risk of marine pest translocation within Australia through domestic trade of live aquatic animals for socio-economic and environmental benefit (DAFF 2023a). The [National policy guidelines for the translocation of live aquatic animals](#) have been developed to guide any translocation activity of live aquatic animals.

The sale of 'live rocks' in the aquarium trade have the potential to be inhabited by marine pests. Gravel and aquarium water released into waterways may also spread any marine pests (larvae or other life stages) present. Import conditions prevent importation of live rock with viable invertebrates (see [Australian Biosecurity Import Conditions](#) (BICON)) (MPSC 2022).

3.4 Debris and flotsam

Although introductions via this vector are rare, it can be an important pathway under certain circumstances, such as following a natural disaster or shipwrecks. Debris can be carried over long distance. Debris from the 2011 Japanese earthquake and tsunami drifted by currents across the Pacific and washed-up on the west coast of North America bringing with it a diverse range of introduced species, including the Asian shore crab, *Hemigrapsus sanguineus* (Therriault et al. 2018). Debris may also be the cause of important secondary pathways within a species' introduced range.

3.5 Natural dispersal

Natural dispersal is a mechanism for the range expansion of a species through the movement of larvae or adults (including spores, tendrils, and eggs) to a new location. Natural dispersal may allow the successful settlement of recruits in a new location (NIMPIS 2023). Characteristics that enable a species to be spread via this pathway include having a planktonic dispersal phase (sexual or asexual), ability to foul floating objects or highly mobile adult life history stage. Although human-mediated dispersal is undoubtedly the most common pathway for long-distance marine pest dispersal, once a species has been introduced into an area it can disperse naturally (MPSC 2022). Control of natural dispersal from established populations is likely to be impractical or impossible, which is why response actions need to be taken before a population can establish.

4 Preventing and monitoring spread

The likelihood for eradication of a marine pest incursion increases with early detection and rapid action. Eradication is most likely to be successful in shallow and/or partially or fully enclosed waterways where the incursion can naturally be contained. In open coastal waters with moderate to high water exchange, emergency containment is likely to be limited to species with limited adult and larval dispersal or those which reproduce by vegetative growth or budding from the edges of a colony. Management to prevent or minimise further spread or reduce populations may be more appropriate where surveys indicate that an incursion is widespread.

This section covers the basis of marine pest containment or eradication from the infested area and any potentially contaminated vectors by explanation of principles for preventing and monitoring spread, including:

- management to prevent spread
- surveillance of high-risk vectors
- management of infected vectors and marine infrastructure
- tracing the incursion.

4.1 Management to prevent spread

An incident management team (IMT), established by the incident manager is appointed to confirm the identification of the suspected marine pest and to determine the likely extent of an incursion. The incident management team framework and functions can be found in the [Biosecurity Incident Management System: Marine pest version](#) (BIMS: Marine pest version).

Preventing the spread of the organism may include the following management practices, which are best implemented early in the response:

- public communication and engagement
- quarantine and movement control
- delimitation.

These management practices may also be applicable at any stage of the following response phases:

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

4.1.1 Public communication and engagement

Sometimes referred to as public relations, this is the management and communication of public information and perceptions. Communication and engagement with all stakeholders, including Commonwealth, state and territory government agencies, industry and community partners are critical to gain acceptance of management or eradication attempts, compliance with any regulations, and to encourage participation in surveillance activities and reporting.

Communication and engagement should occur early in any marine pest response and should be maintained during recovery efforts and until the end of the stand down phase.

The affected jurisdiction may establish an incident management team, which a Public Information function will be activated. The Public Information function covers the overall strategic communication approach to the incident including specific activities; call centre operation, media, social media, website content, community and stakeholder engagement, as well as the development of collateral such as flyers, signage, and similar communication materials.

The Public Information function works with the [National Biosecurity Communication and Engagement Network](#) (NBCEN) to develop nationally consistent messaging. Regardless of incident level, the NBCEN can be used to coordinate the public information response nationally (AHA 2023). The NBCEN consists of a communication representative from each jurisdiction including other relevant organisations which can provide technical expertise. A member from NBCEN (usually the Commonwealth representative) attends the Consultative Committee on Introduced Marine Pest Emergencies (CCIMPE) meetings and develops national talking points in conjunction with the combat jurisdiction to facilitate the delivery of consistent messaging that can be agreed to and used by all jurisdictions. The NBCEN is guided by the [Biosecurity Incident Public Information Manual \(BIPIM\)](#). More on the national arrangements, including NBCEN can be found on the [Outbreak website](#).

Public communication and engagement need to consider affected individuals and businesses and the economic and social (e.g. mental health) aspects of impacts of response activities. Relief and recovery support may need to be coordinated for emergency-affected individuals and communities. The [Biosecurity Incident Management System: Marine pest version](#) (BIMS: Marine pest version) provides guidance on relief and recovery roles in a biosecurity response context.

4.1.2 Quarantine and movement controls

Quarantine and movement controls can be implemented during the investigation phase, alert phase, and operations phase, and are best implemented early and refined when investigative work has provided additional information. They may end up being permanently implemented to minimise risk of spread in a long-term management program.

When a suspected marine pest is detected in an area, but a marine pest emergency has not yet been confirmed, the combat jurisdiction (notifying party) should take steps to limit the spread of the suspected pest from the investigation site or area. Limiting spread can be assisted by initiating restrictions on movements of potential vectors or release of water where this may contain propagules.

4.1.3 Delimitation

A delimiting survey establishes the geographic extent of an area considered to be infested by, or free from, a marine pest. As part of the investigation phase, delimitation informs feasibility of eradication and areas to target for eradication or control and management. Delimitation may also occur throughout the later phases of the response to inform the next steps of a response or determining the current status of marine pests. In some cases, delimitation may take over one year to capture seasonal appearance of pests.

For more information on delimiting an incursion see [Section 5.2](#).

4.1.4 Investigation and alert phase

4.1.4.1 Investigation phase

The investigation phase includes confirmation of identification of the pest and should attempt to identify all potential vectors present at their site.

Concurrent management actions need to be undertaken while identification is confirmed. If necessary, where morphological identification will take some time, molecular identification may be sufficient to act on. The CCIMPE Secretariat should be notified of the suspect incursion within 24 hours, which permits eligibility for NEBRA consideration. Once confirmation is received a Preliminary Information Data Sheet (PIDS) about the initial detection should be submitted to the CCIMPE Secretariat.

A list of potential vectors for marine pests covered in this response manual are shown in Table 7. This will involve notifying relevant parties about the investigation into a marine pest emergency – for example, port authorities, marina operators, vessel owners, and aquaculture facilities in the relevant area. Cooperation from stakeholders is important in order to stop, restrict, or inform the combat jurisdiction of the risks associated with movement of vectors from the site. Compliance with movement controls may be enhanced by communication and distribution of appropriate public awareness materials about the pest. Care needs to be taken when transporting specimens to avoid any chance of accidental escape. In this phase appropriate local authorities need to be contacted to obtain permission for relevant surveillance and sampling activities in specified areas (e.g. marine parks, conservation areas, and nature reserves) (MPSC 2022), and for dealing with species listed in relevant legislation of any state or territory waters.

As part of the investigation phase, delimitation informs feasibility of eradication and areas to target for eradication or control and management. Delimitation may also occur throughout the later phases of the response to inform the next steps of a response or to determine the current status of marine pests.

4.1.4.2 Alert phase

If the initial investigation finds that a marine pest is likely to be present, the combat jurisdiction should communicate the findings to CCIMPE for consideration of the appropriate course of action recommended by the affected jurisdiction 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 vessels, fishing gear, and aquaculture equipment into and out of suspect areas
- controlling the movement of people, such as property owners, business owners and employees, tourists, scientists, into or out of suspect sites, as appropriate. This may require police involvement
- managing water movements where possible
- awareness of methods to report sightings of the pest and access general information
- tracing potential vectors that have left the affected site
- hydrodynamic modelling to determine potential spread of larval stages

- redirecting vessels that have already left the site to appropriate sites for inspection and/or decontamination if appropriate
- informing other destination jurisdictions of movements of vessels from the -high-risk areas
- notifying relevant experts when appropriate.

If required during the alert phase and following CCIMPE endorsement, a National Biosecurity Incident Response Plan (NBIRP) may be submitted to the National Management Group (NMG) for consideration of national cost-sharing arrangements under the [National Environmental Biosecurity Response Agreement 2.0](#) (NEBRA) to help resource a national biosecurity incident response. In such instance, the NMG makes decisions that inform the national coordination of the response, while CCIMPE provides the technical advice on measures required.

4.1.5 Operations phase

The operations phase will be guided by whether eradication of the marine pest is determined to be feasible or not feasible. A technical feasibility assessment is undertaken in accordance with Schedule 3 of the NEBRA to determine the technical feasibility of eradicating the pest during a proposed national response. The feasibility of undertaking a national response is based on conclusions reached by using scientific information to evaluate the proposed response.

For more information see the Schedule 3 of the [NEBRA](#).

4.1.5.1 Eradication feasible

If an investigation reveals a potentially eradicable incursion from a marine pest, then movement restrictions implemented in the investigation phase should remain in place and amended as appropriate.

Quarantine restrictions require establishing specified areas (Figure 1):

- Infested area – all or part of a waterway in which a marine pest is known or deemed to exist
- Dangerous contact area(s) – includes 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 ‘at-risk’ area and subject to the same movement restrictions as an infested area, pending further investigation
- Restricted area – surrounds an infested area, dangerous contact area and suspect area and is subject to intensive surveillance and movement controls on potential vectors³
- Control area – surrounds the restricted area in which biosecurity conditions apply to the entry or exit of potential vectors or specific risk items.

Similar terminology is applied to potentially infested vectors within each area. For example, a vessel within a dangerous contact area would be classified as a ‘dangerous contact vessel’ and a vessel

³ The legislative ability and scope of powers to establish biosecurity restricted areas and control areas will depend on the biosecurity legislation in the relevant jurisdiction.

within an infested area would be classified as an 'infested vessel'. For more information on response area classifications, see the [BIMS: Marine pest version](#).

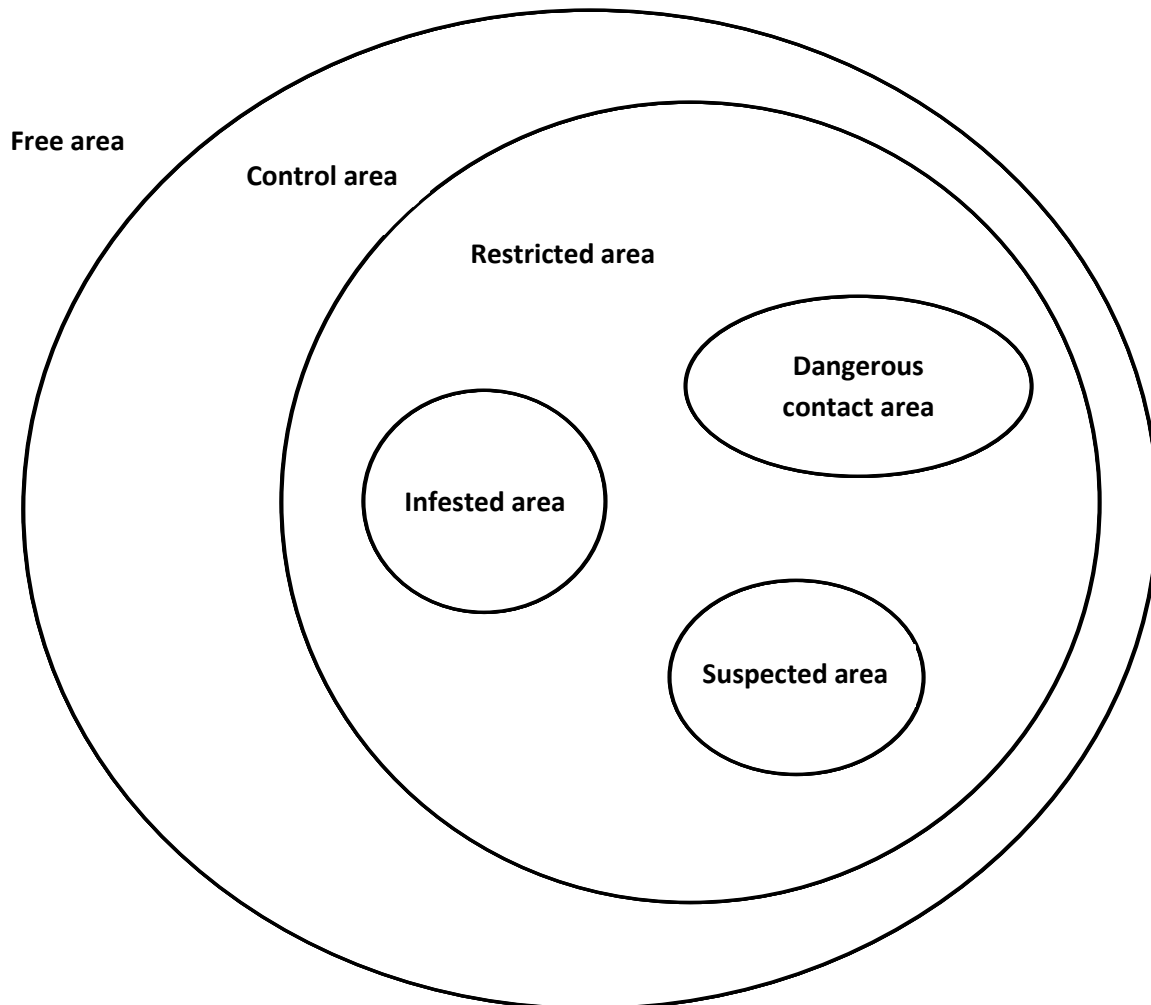


Figure 1 Areas that may be designated during a marine pest emergency

Source: Adapted from BIMS: Marine Pest Version (2020)

The extent of each specified area should be determined by:

- an initial delimiting survey of the area (see [Section 5.2](#) for guidelines on designing a delimiting survey)
- an evaluation of the length of time the species has been present and whether it is likely to have reproduced. This could be calculated by the size and distribution of the animals in the affected area, the number of cohorts apparent and, when possible, examination of the reproductive status (e.g. in crabs the evidence of berried females)
- mobility of the species
- the strength and distribution of directional or tidal currents
- expert advice.

It is important to recognise that in aquatic situations a simple radius around a detection is inadequate. Hydrodynamics and geography of the area and ecology of the target species need to be considered to determine the specified areas.

Movement restrictions may include limiting:

- the movement of vessels
- equipment exposed to the pest
- aquaculture stock or equipment
- access within certain areas
- the uptake or movement of ballast water or other water (such as influent and effluent water from land-based aquaculture or managed water bodies) within the control areas where appropriate controls are not in place.

Exposed equipment will vary depending on the target species. Above water assets will need to be considered as species such as crabs are intertidal and can remain out of the water for long periods of time (MPSC 2022).

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 circumstance, but others will be critically important for eradication or control. Effective communication and accurate information dissemination are vital to ensure compliance and acceptance of restrictions.

4.1.5.2 Restricted Area Movement Unit

The Restricted Area Movement Unit, an operations function of the Coordination Centre or Control Centre (established at the appropriate levels [national, state and/or local], to manage strategic and operational aspects of the response) 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 main duties of this unit are to:

- issue movement permits to the public
- establish and operate road and water checkpoints in the restricted area, including liaison with state transport authorities, port authorities, water authorities, police and local government
- coordinate movement and security activities across affected sites
- maintain registers of all movements, including, permits issued and people into, within and out of the restricted and affected areas.

For more information on incident management functions, see the [BIMS: Marine pest version](#).

4.1.5.3 Eradication not feasible

If an investigation reveals an incursion of a marine pest is unlikely to be eradicable, interim containment measures to prevent translocation from any infested waterway should be implemented to minimise the risk of the pest being spread from the affected area.

A stand-down phase for the NMG involvement may be entered either directly from the alert phase or from the operations phase when the CCIMPE and the NMG agree there is no need to initiate or continue a national biosecurity incident response. The stand down of the NMG does not mean that actions and consultation within the CCIMPE cease. This consultation and communication through the CCIMPE will continue as long as the affected jurisdiction(s) and/or the Chair of the CCIMPE deem it necessary. Agreement for longer term management and resourcing options should be determined. Although a stand-down phase may be entered, jurisdictions may transition from an operational phase to management.

4.1.5.4 The Australian Government *Biosecurity Act 2015*

The *Biosecurity Act 2015* can be used in the absence of appropriate state or territory legislative powers and may be used in circumstances, including directing conveyances ([Appendix A](#)):

- 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 2015*, 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). The *Biosecurity Act 2015* is only intended to be used if there is no state or territory legislation that provides appropriate powers necessary for the response, aside from ballast water which is entirely covered by the *Biosecurity Act 2015*. A provisional list of other Commonwealth, state and territory powers for intervention and detention of vessels is in [Appendix B](#).

State and territories 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 Animals](#) (DAFF 2023a). All potentially infested fishing gear, aquaculture equipment or stock should be treated and inspected before removal from the control area.

Refer to [Section 4.3.1](#) on ballast water management and [Section 4.3.2](#) on biofouling for relevant information.

For additional information on using the *Biosecurity Act 2015* during an emergency response see [Appendix A](#).

4.1.6 Stand down phase

The stand down phase is in effect when, following appropriate consultation between the affected jurisdiction and the 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
- where the pest is not eradicable, alternative ongoing management options are to be considered and the most appropriate option implemented, given the risk and required investment
- transition to management or recovery will be considered as part of stand down
- the outcomes of the response, and information on the management of the species going forward, should be communicated to stakeholders
- a comprehensive after-action review should be completed as soon as possible after the response stands down, to ensure that learnings can be captured for improvements in future responses.

4.2 Surveillance of high-risk vectors, submerged structures and habitat

In the event of an emergency marine pest response, the risk status of all potential vectors, submerged infrastructure and the environment should be determined and managed when they have been present in the restricted or control areas during the time the marine pest is suspected to have been present.

If determined to be high-risk, vessels, submerged infrastructure, the environment, and other vectors should be further assessed if they require inspection and treatment. Risk assessment may determine whether this is necessary. For example, a recently cleaned vessel, particularly with reapplied antifouling coating, will be at lower risk of picking up marine pests than one with heavily fouled niches (MPSC 2022).

All vessels, submerged infrastructure, the surrounding environment, and other vectors within the control area should be assessed and inspected for signs of the pest(s) where determined necessary. High-risk and medium-risk vectors should be assessed and required to remain within the control area until they can be inspected and declared free of the pest as determined appropriate. Likewise, submerged infrastructure and the surrounding environment should be treated according to risk status.

All high-risk and medium-risk vessels that have recently left a control area should be contacted immediately if their itinerary indicates that they present a risk for spread of the pest in Australia. If the itinerary indicates visitation to another country with biosecurity requirements on ships (e.g. New Zealand) the appropriate contact in that country should be notified. If these vessels have not entered another port or marina, they should be encouraged to remain at sea 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. A vessel that has entered another port or coastal area should be inspected immediately. If signs of the pest are discovered, then the vessel should be directed for treatment and a back tracing of the vessel's itinerary be done and surveys undertaken of the anchorages it has visited.

4.2.1 Vessel inspection

The [Australian biofouling management requirements](#) set out vessel operator obligations for the management of biofouling when operating vessels under biosecurity control within Australian territorial seas. Divers or remote operated vehicles (ROVs) should carry out in-water inspection of vessels using a standardised search protocol; see [anti-fouling and in-water cleaning guidelines](#) and [International Maritime Organization \(IMO\) biofouling guidelines](#). Divers can inspect interior spaces and crevices, such as sea chests, water intakes or outlets using endoscopes. Moist places such as anchor wells or chains will require inspection for algae, entrained organisms in sediments and semi-terrestrial air-breathing species, such as grapsid shore crabs (MPSC 2022).

Critical inspection areas for vessels less than 25 metres long (Figure 2) include:

- 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 are in water or have been recently used
- keel and keel bottom
- sounder and speed log fairings
- live bait wells and deck basins.

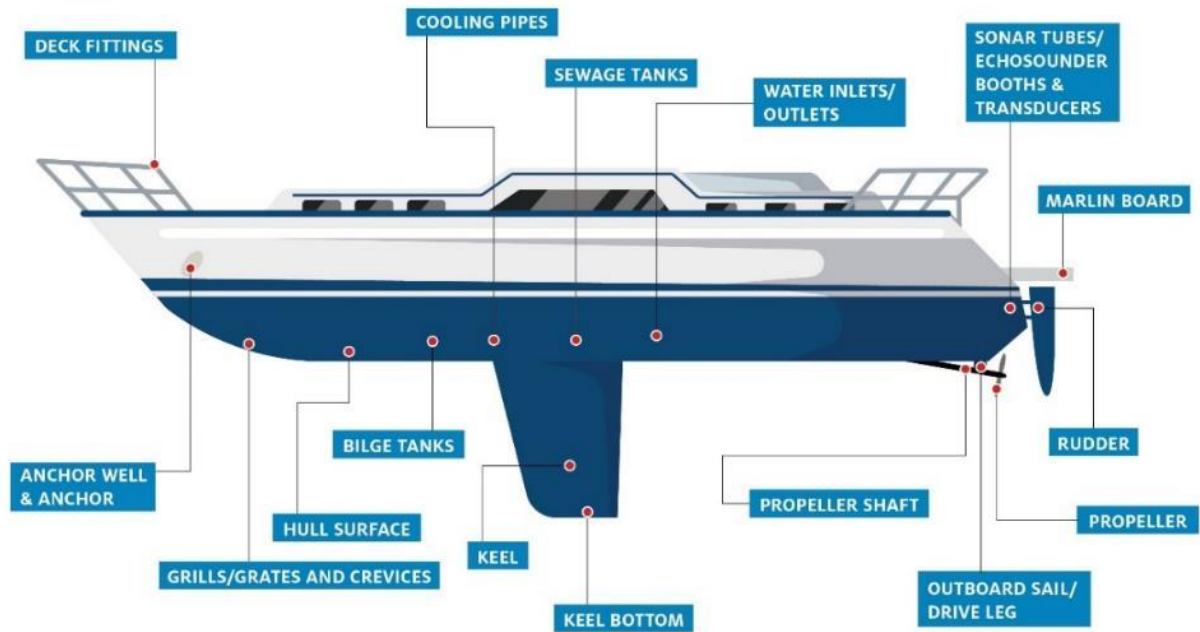


Figure 2 High-risk niche areas for inspection of biofouling on vessels less than 25 metres

Source: Floerl (2004)

Critical areas are similar for vessels longer than 25 metres (Figure 3), including additional areas:

- sea chests and gratings
- ballast tanks and internal sea-water systems
- dry-docking support strips (DDSS)
- sonar tubes
- bow and stern thrusters
- bilge keels
- other niches and cavities in the ship's wet water side.

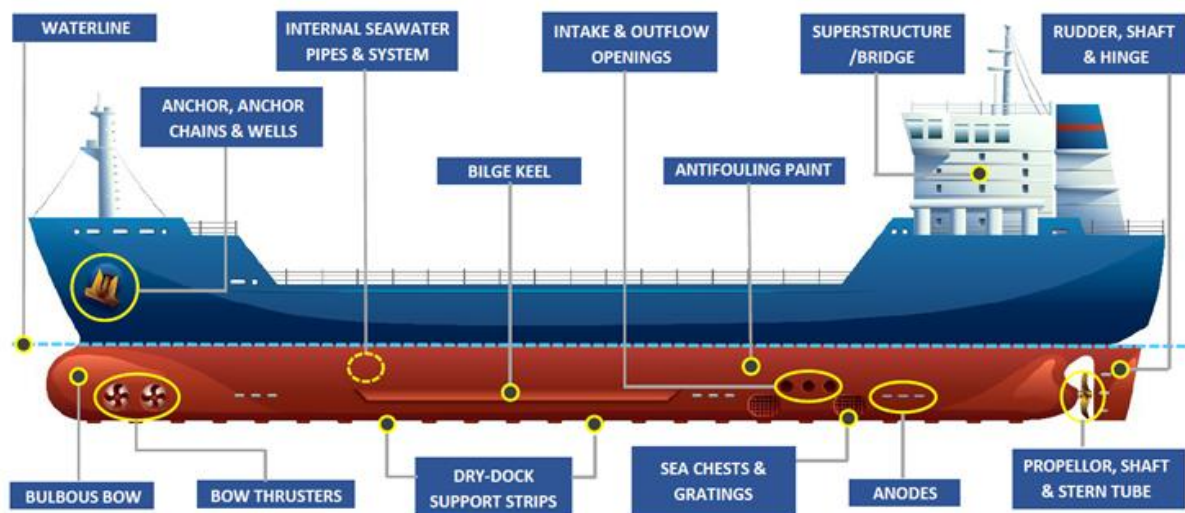


Figure 3 Schematic diagram showing the high-risk niche areas for inspection of biofouling on vessel greater than 25 metres. Vessel and its components are not to scale

Source: René Campbell – Department of Agriculture, Fisheries and Forestry

4.2.2 Submerged structure and habitat inspection

Surveillance for marine pests should be included in response measures for artificial and natural submerged structures (permanent, semi-permanent, and temporary) and habitats as they are at risk of being colonised by marine pests. For example, the infrastructure that supports vessel operations (e.g. boat harbours, marinas, slipways, recreational boating mooring areas, and fishing ports/bases) provides hotspots for the introduction and spread of marine pests from both international and domestic vessels (MPSC 2021). The environmental conditions and artificial nature of these facilities make them highly suitable for marine pests to establish new populations once they are introduced (MPSC 2021). See [Section 4.3.4](#) for management of submerged structures and habitats.

4.3 Management of infested vectors and marine infrastructure

Management of infested vectors and marine infrastructure will be different depending on the type of area where an infestation occurred. Specific details are given for the following vectors:

- ballast water
- biofouling of vessels
- aquaculture stock and equipment
- submerged marine structure.

A summary of treatments shown to cause 100% mortality (LD100) of several high-risk marine pests is provided in Appendix C. These results are largely based on laboratory trials of individual or clumped organisms and will need to be adapted to ensure complete mortality on more complex structures, such as ropes or nets, or in treatment of large quantities of equipment or stock. They may also be a useful guide for selecting appropriate efficacy trials of decontamination methods for other, similar species.

Table 8 summarises management recommendations for different types of vectors that can be applied to all marine pests.

Table 8 Management recommendations for different types of vectors

Vector	Management
International and domestic yachts <25 m, domestic fishing vessels, ferries, tugs and naval vessels	<p>Remove from water and treat and/or clean external submerged surfaces</p> <p>Contained in-water treatment with appropriate biocide</p> <p>Treat internal seawater systems</p> <p>Treat moist places (interior spaces and crevices)</p> <p>Manage ballast water</p> <p>Remove from the control area once cleaned</p> <p>Educate operators and service agents of risk</p>
Domestic merchant vessels >25 m, and international merchant vessels >25 m	<p>Inspect and treat and/or clean (if possible) external submerged surfaces</p> <p>Treat or seal internal seawater systems</p> <p>Treat moist places (interior spaces and crevices)</p> <p>Manage ballast water</p> <p>Educate operators and service agents of risk</p>
Recreational craft (e.g. jet-skis and kayaks)	<p>Remove from water and clean external submerged surfaces</p> <p>Treat and/or clean and dry internal seawater systems</p> <p>Educate users and service agents of risk</p>
Fishing gear and nets	<p>Remove from area and/or clean and dry</p> <p>Educate users and service agents of risk</p>
Fouled aquaculture stock	<p>Remove from infested area or use an effective method for decontamination</p> <p>Educate users and service agents of risk</p>
Fouled aquaculture equipment	<p>Removed from infested area</p> <p>Clean thoroughly by high-pressure water blast – for example >2,000 psi, capturing cleaned material for safe disposal</p> <p>Immerse in or apply an appropriate decontamination solution (e.g. copper sulphate solution (4 mg/L) or liquid sodium hypochlorite (200 to 400 ppm) for 48 hours)</p> <p>Rinse in seawater and air dry, preferably in sunlight</p> <p>Educate users and service agents of risk</p>
Buoys, pots and floats	<p>Restrict movement from the control area</p> <p>Treat and/or clean and dry</p> <p>Educate users and service agents of risk</p>
Water, shells and organisms for bait or aquaria	<p>Restrict movement from the control area</p> <p>Educate users and distributors of risk</p>
Flotsam and jetsam	<p>Remove from water/shoreline</p> <p>Dry prior to onshore disposal</p> <p>If possible, use barriers to prevent escape from infested area</p>
Fauna (e.g. birds)	No vector recorded†
Stormwater pipes and intakes	<p>Treat and/or clean and remove fouling</p> <p>Where possible, seal until stand down of emergency response</p> <p>Educate service agents of risk</p>

Source: Modified from Bax et al. (2002); †Fauna are recognised as vectors of disease – for example, viruses may also be carried via the gut, feathers, feet and bill of piscivorous birds (DAFF 2024b).

4.3.1 Ballast water management

The *Biosecurity Act 2015* prohibits the discharge of unmanaged ballast water within Australian seas (within 12 nautical miles of any land mass or in water less than 50 metres deep) (DAFF 2023b).

The *Biosecurity Act 2015* regulates the discharge of ballast water and ballast tank sediments in Australian waters. The Act also prohibits the discharge of ballast tank sediment within Australian seas. Vessels intending to discharge ballast water in Australia must apply for permission via the [Maritime and Aircraft Reporting System](#) and receive a valid Biosecurity Status Document prior to any discharge. Discharging untreated ballast water is now prohibited in Australia, unless granted an exemption by the Director of Biosecurity. The discharge of ballast tank sediment is an offence in Australia. Ballast water and ballast tank sediments are also managed by the [International Convention for the Control and Management of Ships' Ballast Water and Sediments](#) (International Ballast Water Management Convention) which has reduced the likelihood of this vector, however the risk is not removed. Australia is a signatory to the International Ballast Water Management Convention.

The approved methods for management of ballast water and ballast tank sediment can be found in the [Australian Ballast Water Management Requirements](#) (DAFF 2023b) and are as follow:

- use of an IMO approved [Ballast Water Management System](#) (BWMS)
- ballast water exchange conducted in an acceptable area
- use of low-risk ballast water (such as fresh potable water, high seas water or fresh water from an on-board freshwater production facility)
- retention of high-risk ballast water on board the vessel
- discharge to an approved ballast water reception facility.

Note that the International Ballast Water Management Convention requires all ships that use ballast water to comply with the regulation D-2 standard with respect to maximum amounts of viable organisms allowed to be discharged following use of an installed BWMS as of 08 September 2024. The use of ballast water exchange as a primary method of ballast water management will be phased out by the same date.

4.3.1.1 Vessels arriving in Australian waters from an international location

Vessels that are intending to discharge internationally sourced ballast water must submit a Ballast Water Report through [Maritime and Aircraft Reporting System](#) (MARS) at least 12 hours prior to arrival. To prevent the discharge of unmanaged ballast, even vessels not intending to discharge ballast water are strongly encouraged to manage their ballast water by an approved method and to submit a Ballast Water Report. The Ballast Water Report will be assessed, and a response will be issued through the Biosecurity Status Document. Following the first point of arrival, international vessels may uptake Australian sourced ballast water for discharge later in Australia ([Section 4.3.1.2](#)) or overseas ([Section 4.3.1.3](#)).

4.3.1.2 Vessels operating between Australian domestic locations

The movement of Australian sourced ballast water between Australian ports is prohibited unless it has been managed, or a low-risk exemption has been provided by the department. The approved ballast water management options are available in the [Australian Ballast Water Management Requirements](#). Low-risk exemptions are based on individual voyages with specific ballast water

uptake and discharge locations and dates; any modification to locations and/or dates or additional uptake/discharge combinations require a new application for exemption.

The domestic ballast water risk tables inform the Australian Sourced Ballast Application in the [Maritime and Aircraft Reporting System](#) (MARS) which reflects the risk status of port waters. Alterations to the domestic ballast water risk tables may be required in the event of an emergency response.

4.3.1.3 Vessels departing for international destinations

Vessels leaving a control area for destinations outside of Australia's territorial waters should be notified of the risk and be required to manage ballast water as specified by the International Maritime Organization (IMO) [International Convention for the Control and Management of Ships' Ballast Water and Sediments, 2004 \(Ballast Water Management Convention\)](#). They also need to be aware of any requirements in destination countries.

4.3.2 Vessel biofouling management

Removal of biofouling on vessels includes land-based treatment, treatment of biofouling in internal seawater systems and various in-water treatments. Refer to [Section 3.1](#) (Biofouling) for guidance on best-practice approaches for the application, maintenance, removal and disposal of anti-fouling coatings and the management of biofouling.

For vessels known to be infested with a marine pest, prevention of entry, treatment, or vessel cleaning before entry to a port are the most effective management options. Where vessel facilities are available and it is operationally practical, vessels and movable structures should be removed from the water for cleaning and maintenance, in preference to in-water operations. Australian dry dock facility information can be obtained from the [National Maritime Centre](#) (NMC). In-water cleaning in Commonwealth waters may require referral under the [Environment Protection and Biodiversity Conservation Act 1999](#) (EPBC Act). Dry-docking and in-water operation principles and recommendations are contained in the [Anti-fouling and in-water cleaning guidelines](#).

If the activity does not require referral under the EPBC Act, the activity must be self-assessed using *Appendix 1: Decision support tool for in-water cleaning* of the [Anti-fouling and in-water cleaning guidelines](#). Each state or territory jurisdiction is the primary contact for biofouling management advice. Requirements and approvals for in-water cleaning in state or Northern Territory waters differ and should be clarified with the relevant agencies as listed on the [Anti-fouling and in-water cleaning guidelines](#) webpage.

The *Biosecurity Act 2015* can be used in the absence of appropriate state or territory legislative powers and may be used in circumstances, including directing conveyances. The *Biosecurity Act 2015* defines conveyances as including vessels and floating structures ([Chapter 4](#)). The Australian Director of Biosecurity (or a delegate) can authorise state and territory officers as biosecurity officers under the *Biosecurity Act 2015*, which can enable actions in a biosecurity response. A provisional list of other Commonwealth and State powers for intervention and detention of vessels is in [Appendix B](#).

4.3.2.1 Land-based treatment

Marine pests may inhabit internal piping and water intakes that are not easily inspected or cleaned. Therefore, haul-out of vessels and other non-permanent structures, such as moorings, pontoons, chains and ropes, for inspection and treatment on land is the preferred option. This is most easily

achieved for vessels <25 metres in length and where suitable haul-out or dry-dock facilities are available near the control area. Larger vessels may need to be inspected and treated in water or suitably treated in dry-dock where possible.

There is a risk that marine pests dislodged during haul-out or vessel cleaning may remain viable and could start a new population if returned to the sea. For example, some crab species are intertidal and move readily across open, dry spaces (MPSC 2022). Similarly, bivalves can withstand extreme environments and are tolerant of many treatment types. The incident manager must approve haul-out facilities used for decontamination. Such facilities should be fully contained so that material from vessel hulls cannot accidentally or intentionally be returned to the marine environment. All macro (>1 mm) particles removed from vessels cleaned out of water should be retained and disposed of in landfill (or as biohazard material in secure landfill if appropriate). All liquid effluent (runoff) from out-of-water vessel water blasting or cleaning must be filtered to 10 µm (Sherman, Jennings & Miller 2020) then collected for treatment in a liquid effluent treatment system or disposal in a secure landfill/seepage system that does not connect with waterways.

Approved vessel cleaning facilities should comply with relevant jurisdictional requirements for waste containment and disposal from slipways, boat repair and maintenance facilities. Guidance for identifying and selecting approved vessel cleaning facilities suitable for removing marine pests are given by Woods et al. (2007).

Depending on the marine pest, high-pressure water blasting followed by prolonged (>5 days) desiccation (aerial exposure, preferably to the sun) may also be used to treat other fouled structures removed from an infested area, such as mooring blocks, pontoons, floats, and fenders. Consideration needs to be given if using this method for marine pest that can survive for extended periods out of the water, particularly intertidal species (e.g. crabs or bivalves).

4.3.2.2 Internal seawater systems

Some marine pests, such as bivalves, are robust organisms capable of tolerating extreme environments. Internal seawater systems of vessels should be cleaned to the greatest extent possible with:

- [chemical treatment](#)
- [thermal treatment.](#)

Concentrations of chemical treatments will need checking at intervals to ensure they are maintained, particularly for chlorine which degrades rapidly in the presence of organic matter.

4.3.2.3 In-water cleaning

The [antifouling and in-water cleaning guidelines](#) 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 and where supported by relevant jurisdictional authorities. Applicants who wish to perform in-water cleaning in Australian waters should familiarise themselves with the principles and recommendations contained in the guidelines.

Depending on the location of the intended clean, there may be a range of legislative requirements for in-water cleaning in Australia waters. Applicants who wish to perform in-water cleaning in Commonwealth, state, or territory waters must first contact the relevant agency in each jurisdiction for approval. The relevant agencies are listed on the [Anti-fouling and in-water cleaning guidelines](#) webpage.

4.3.2.4 Sea chests and other niche areas

Sea chests and internal seawater systems of vessels can accumulate biofouling and are structurally complex, making access for inspection and treatment difficult. Both mobile and sedentary species are found in these areas (Coutts, Moore & Hewitt 2003). Fouling communities that include dense patches of bivalve shellfish can be attractive habitats for other marine pests. Biofouling of sea chests, internal pipework and other niche areas can be independent to biofouling on the hull, and a clean hull does not imply clean niche areas.

Treatments of these areas for marine pests include chemical and non-chemical methods.

There are considerations for effective in-water treatment. For instance, a key element of in-water treatment of sea chests is being able to seal off the confined spaces so that the treatment can be administered effectively. This can be achieved by sealing off external gratings using commercially available [magnetic tarpaulins](#) or bespoke sealing units. Sealing off confined spaces can also assist in preventing mobile marine pest species from avoiding the treatment.

There are published reports demonstrating that acetic acid and commercial descaler formulations can be effective against intact fouling assemblages within 48 hours (Cahill et al. 2019; Cahill et al. 2021). These preparations effectively clean attached molluscs and would be expected to attack calcareous shells of crustaceans. An important consideration for chemical treatments is its risk to the environment and operator against its efficacy. Acetic acid and chlorine are considered safe to use within the marine environment; however, their efficacy needs to be determined prior to use. Maintaining active concentrations of these chemicals requires careful monitoring. Local authorities should be contacted for requirements around use of chemicals in natural waterbodies. Some non-acidic chemical treatments (e.g. quaternary ammonium compounds) can be effective in as little as ten hours.

Thermal stress can feasibly be applied to pipework and niche areas by use of heated water between 50 to 60 °C which can render taxa non-viable in under two hours (Cahill et al. 2019; Growcott, Kluza & Georgiades 2016). The application of thermal stress does need to be considered against which marine pest species is being targeted as it may be more effective on temperate species than tropical species. Physical removal of a pest from niche areas is not always possible or feasible. There is risk of inadvertently releasing the biofouling organisms into the environment without significant measures to ensure that no viable material can escape. Deoxygenation and osmotic shock could take many days to several weeks to kill more resilient marine pests, such as crabs (Cahill et al. 2019), meaning they are unsuitable for response actions for sea chests and other niche areas. Acceleration of deoxygenation by use of oxygen scavenging chemicals may increase efficiency in killing marine pests but efficacy should be monitored (Cahill et al. 2021).

4.3.3 Management of aquaculture stock and equipment

Marine pests may be transported either on equipment used to culture marine species (such as ropes, nets, cages, buoys, harvesting vessels) or on the stock itself. Movement of aquaculture stock or equipment from the control area during a marine pest emergency response should be permitted only if it can be demonstrated that steps taken to decontaminate the equipment and stock are able to effectively remove all life stages of the pest (that is, 100% mortality). This should require efficacy trials of the decontamination methods and approval of the protocol by the incident manager.

Different marine pests vary in their susceptibility to physical removal or exposure to toxicants. Species such as bivalves or barnacles, which have strong basal attachments and/or hard exoskeletons that allow them to withstand short periods of exposure to toxicants, are likely to be more resistant to decontamination methods than soft-bodied pests, such as ascidians or macroalgae. The effectiveness of any treatments may be affected by the conditions in which they are applied, including the ambient salinity, temperature, dissolved oxygen, pH, water flow, and the size and nutritional status of the treated species.

For all aquaculture stock and equipment treatment methods which are land-based, there is a risk that marine pests dislodged during haul-out and may remain viable and could start a new population if returned to the sea. Containment and treatment of the waste, including influent and effluent water, may be necessary and similar precautions should be applied as per land-based treatment in [Section 4.3.2.1](#).

4.3.3.1 Aquaculture stock

The translocation of aquaculture stock is a probable secondary vector for spread of marine pests in Australia. Species such as oysters or seaweed can provide habitats that support the accidental co-transfer of other species like crabs (MPSC 2022). Similarly, invasive bivalves can settle on the shells of other bivalves and on equipment used to culture bivalves.

Aquaculture stock can be treated by:

- manual removal/destruction
- detergents
- osmotic treatment.

The utility of treatment methods used to decontaminate aquaculture stock relies on the therapeutic ratio. A therapeutic ratio is the highest exposure to an effective treatment that results in no stock loss or reduced viability of stock because of the treatment (Cahill et al. 2021). Trials should be carried out to determine rates of mortality of the treatment on aquaculture stock and on the target marine pest (Cahill et al. 2021). 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.

Import of aquaculture stock is strongly regulated and most jurisdictions have conditions on movements of aquaculture stock to manage biosecurity and other risks.

4.3.3.2 Aquaculture equipment

The protocols recommended for treatment of ropes and aquaculture equipment, such as buoys, floats, nets, and traps are:

- 1) Remove to land taking care not to dislodge motile species when removing structures from the water
- 2) Clean thoroughly by high-pressure water blasting (>2000 psi at distance of 100 mm)
- 3) Immerse in 2% liquid sodium hypochlorite (200 to 400 ppm) for >4 hours, or 2% detergent (e.g. DECON 90) solution for >8 hours, or hot water (>50° C) for >1 hour
- 4) Rinse in seawater or freshwater and air dry for >48 hours.

4.3.4 Submerged structures and habitat

All infrastructure submerged or exposed to the marine environment is at risk of being colonised by marine pests. This includes permanent, semi-permanent and temporary infrastructure. For fouling organisms, submerged structures, both artificial and natural, that cannot be removed from the water are to be considered high priority. These include, but are not limited to, structures such as:

- aquaculture infrastructure and facilities
- petroleum production and exploration industry infrastructure and facilities
- marinas, slipways, boat maintenance and recreational boating facilities
- projecting piles
- breakwaters
- groynes
- rock walls
- rip-rap
- wrecks
- hulks
- hulls
- steel facings
- ropes and buoys
- moorings and mooring dolphins
- natural seabeds and reefs.

Biofouling management for infrastructure should be consistent with the National Biofouling Management Guidelines. These are available for the following industries and operators:

- [aquaculture](#) industry
- [offshore infrastructure](#) (petroleum production and exploration industry)

- [port and marina operators](#) (marinas, slipways, boat maintenance and recreational boating facilities).

4.4 Tracing an incursion

Tracing is used to discover the method and pattern of the spread of the pests and may include trace-forward and trace-back. Tracing is used to discover where an incursion may have originated from and identify potential additional sites of outbreak within Australia. The first location to have the detection of a marine pest may not be the original site of introduction. Tracing is crucial to defining and modifying the dimensions of the specified areas.

For more information on tracing an incursion see [Section 5.2](#).

5 Containment, delimitation, and eradication

Containment is a component of control which aims to restrict an incursion of an invasive species to a limited geographical range. Delimitation establishes the geographic range of an area considered to be infested by, or free from, a pest. Eradication aims to eliminate a marine pest from the infested area. Delimitation informs the feasibility of controlling or eradicating a marine pest. The feasibility will depend on the nature and location of the incursion and the management option adopted.

Management options include:

- containment of the marine pest to the infested areas and prevention of further spread; ongoing costs and efforts, or,
- eradication of the marine pest from an infested area; highest initial control measure and cost.

This section provides information on:

- containment and control
- delimiting surveys
- eradication.

For methods suitable for containment, delimitation and eradication see [Section 5.4](#).

5.1 Containment and control

Containment aims to prevent secondary spread and assists to maintain the possibility of eradication. If a decision is made to implement containment and control, then the incident manager will (in consultation with stakeholders) recommend that interim containment measures be implemented to minimise the risk of the marine pest translocation from the infested waterway. This may include movement controls on potential vectors, public information campaigns, policies and practices for vessel and equipment sanitation and surveillance, and control of secondary infestations outside the infested waterway.

5.2 Delimitation of an incursion

After the detection of a marine pest, a delimiting survey should be conducted quickly to establish if the area considered to be infested is localised or widespread. This information will assist in determining which response option, containment, eradication, or ongoing management is most feasible (van Havre & Whittle 2015). Until the response option is known, containment measures around all suspected infected area(s) should be implemented to reduce the potential spread of the marine pest. An incursion can generally be declared delimited when no new infested area has been discovered for a period of time, given that surveys into new areas are performed to indicate spread has not occurred (van Havre & Whittle 2015).

The below section outlines considerations when planning a delimiting survey and some survey methods that may assist in delimitation, including:

- tracing an incursion (trace-back and trace-forward)
- perpendicular and margin transects
- adaptive sampling
- approach, decline, delimit.

We provide an overview of the different sampling methods for marine pests that could be used during delimiting surveys in [Section 5.4](#). In some cases, a sampling method is not necessarily consistent across life stages, for instance a method that is effective for trapping juvenile stages may be ineffective at trapping adult life stages.

5.2.1 Planning a delimiting survey

When planning a delimitation survey strategy, a manager should consider:

- the allocation and management of available resources to delimit an incursion most effectively, including:
 - funding of the operation (see [Section 1.3](#) for more information)
 - personnel and equipment (including personnel training)
 - SOPs for consistency of sample collection, preservation and record keeping
 - ability to obtain identification confirmation from a recognised taxonomic expert or diagnostic facility.
- the location where the pest was initially detected:
 - how long the pest has been present at the site before it was detected
 - the dispersal characteristics of the pest, including:
 - 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.
- pest biology, such as survival reproductive rate and current stages of reproductive development
- pest habitat, such as distribution and suitability of potential habitats around restricted areas and control areas
- survey design sensitivity (factoring detection method sensitivity, including pest biology), sampling logistics, and operator safety.

Local knowledge and site inspections as well as satellite imagery, habitat suitability maps or risk maps, hydrographic charts, and online databases such as [Seamap Australia](#) can be useful for identifying areas that may contain habitat suitable for the pest. Where they exist, habitat suitability maps and hydrodynamic models such as [Connie3](#) (accessed on request from CSIRO) may also be useful to simulate the likely directions of current flow. This information can provide possible rate and extent of spread of planktonic larvae from the known area of infestation (Inglis et al. 2006). Graphical summaries that plot the areal extent of new detections relative to the area searched can be used to evaluate the progress of delimitation and control of the pest (Panetta & Lawes 2005).

Knowledge of habitat requirements may assist in targeting surveillance to habitats likely to harbour the invasive species. Habitat suitability models and particle dispersion models may also assist to identify survey locations (Inglis et al. 2006). For example, shore crabs tend to inhabit areas with rocks and temperate mangroves in intertidal zones rather than bare, sandy areas, whereas many swimming crabs prefer open habitats at subtidal depth ranges (MPSC 2022). Further examples include clams that are infaunal, inhabiting soft benthic environments such as mudflats and sandy shores, rather than hard rocky intertidal substrates, whereas mussels and other epifaunal bivalves typically prefer settling on hard structures, such as wharves, pontoons, and vessel hulls.

5.2.1.1 Tracing an incursion

Usually conducted at the same time, trace-back and trace-forward information is used to determine how and where a marine pest first entered a site and where it may have spread to (van Havre & Whittle 2015).

Tracing an incursion requires investigation into:

- the length of time the pest has been present
- the initial source and location of infestation
- whether the pest is likely to have reproduced
- the possible movement to and from the site of water, vessels, animals, submersible equipment, and other potential vectors for the pest
- the existence and location of other potentially infested areas, particularly areas of suitable habitat.

Trace-back

Trace-back information can be used to determine the possible extent of an incursion, particularly for a primary incursion where a single size or age 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 estimate the source of a spawning event. This source information can be used to determine where else in the area the prevailing currents could have spread the larvae (Burgman et al. 2013; Hauser et al. 2016). The use of DNA-based methods can help identify both source and connected populations and areas of provenance (Roux et al. 2020).

Elements of demography of the marine pest populations may be inferred from the size or age distribution within the population and reproductive state of animals collected during investigations. A population that contains individuals that vary widely in size, are reproductively active, or contain two or more distinct size cohorts could be indicative of successful local reproduction and multiple recruitment events.

Trace-forward

Trace-forward information can be used to identify locations outside the infested area that may have been exposed to the pests by vectors that have departed the known infested area (van Havre & Whittle 2015). Areas near detections can be surveyed in more detail on pest distribution or abundance if needed for assessment of eradication feasibility. Surveillance of areas of potential secondary spread can then be prioritised based on risk, informed by vectors, modelling and habitat suitability (Brown et al. 2013).

Data sources for tracing vectors

Vessels

Tracing the movements of vessels to and from an incursion is important to know where a marine pest may have originated or be translocated within Australian waters. Some useful data sources on movements of large, registered commercial vessels are:

- [Australian Government Department of Agriculture, Fisheries and Forestry](#)
- [Lloyd's List Intelligence](#)
- [MarineTraffic](#)
- [Australian Fisheries Management Authority](#)
- [Bureau of Infrastructure, Transport and Regional Economics](#)
- [Australian Border Force](#)
- [Australian Maritime Safety Authority](#)
- local port authorities keep records of all vessel movements at their port berths and associated anchorage points.

Specific industries operating in marine environments may have information on movement of vessels and equipment such as aquaculture, natural resource extractors, maritime transport and logistics industries. 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 been used in their facilities. Local industry groups, such as fishing groups, may provide point-of-contact for vessels and the movements of their respective industry sectors. Logged vessel trip reports held by the Australian Volunteer Coast Guard may also provide some data on vessel movements.

Some states and territories have developed vessel-tracking systems for a range of vessel types. For example, during the operational period of *Mytilopsis sallei* incursion in Darwin, an access database was developed that contained vessel names, contacts, current location, history of individual vessel movements and the risk status of the vessel.

Ocean current modelling

Ocean current modelling may be an effective forward and back tracing method for estimating the source and locations as part of a marine pest response. Some tools that can assist with modelling current movements include:

- [Connie3](#) (accessed on request from CSIRO)
- [Regional Ocean Modelling System](#)
- [Marine Invader Tracking and Information System](#)
- [International Comprehensive Ocean-Atmosphere Data Set](#)
- [Global Marine Environment Datasets](#)
- [National Oceanic and Atmospheric Administration](#).

5.2.1.2 Perpendicular and margin transects

Allocating surveys along perpendicular and margin transects can rapidly lead surveyors to the outer reaches of an invasion, particularly at times when infestations are dense at the point of introduction and decline with distance (Hauser et al. 2016). Alternatively, survey effort could be made at the margins of the known infestation.

5.2.1.3 Adaptive sampling

Using probability-based sampling, adaptive sampling designs use sample points located on systematic grids or gradients away from the site of known infestation (Thompson 2004; Brown et al. 2013). This is most useful to ensure the greatest possible area is covered, while providing the best chance of detecting established and founding populations. The general approach is to sample at predetermined locations (often across a grid), and when the target is found, to sample more intensively near the detection (Thompson 2004). Adaptive sampling can be effective for detection of rare species, but has the disadvantages that the final sample size and survey cost are unknown prior to the survey, and field implementation may be complicated (Thompson 2004).

5.2.1.4 Approach, decline, delimit (ADD)

Approach-decline-delimit (ADD) can estimate an incursion area of a spreading marine pest in situations where the extent of spread is difficult to measure, such as when time has lapsed since initial detection or pest density is low (van Havre & Whittle 2015). Approach-decline-delimit delimits an incursion assuming very little prior information (e.g. site of first detection) by measuring the decline in density of occurrence (Leung et al. 2010). See Leung et al. (2010) for detail on ADD application.

5.3 Eradication

Eradication programs will be more successful if initiated early and are well designed and resourced. Eradication is more likely to be successful or feasible if initial investigations determine that the species is not widespread, can be contained, is not difficult to detect, or is present or potentially present in closed/semi-closed environments.

Eradication is the preferred option when:

- the pest can be determined to be technically feasible to eradicate
- discounted benefit-cost analysis favours eradication over control
- the socio-political environment supports using eradication methods.

For example, the early detection and removal of a small population of *Perna canaliculus* in South Australia resulted in eradication with minimal resource expenditure (McEnulty et al. 2001).

The [National Environmental Biosecurity Response Agreement 2.0](#) (NEBRA) establishes national arrangements for responses to nationally significant biosecurity incidents when there are predominately environmental or public benefit. The NEBRA provides a mechanism to share responsibilities and costs for a response when eradication is considered feasible, the pest is considered to be of national significance, and the response calculated to be cost-effective.

A marine pest's biology and reproductive strategy will influence the effectiveness of an eradication program. For example, marine pests with high fecundity and long planktonic larval durations can

spread over large distances by tides, ocean currents, and anthropogenic pathways. Due to spread by tides and currents, eradication may not be possible in open coastal waters where there is high movement of water. Eradication is most likely to be feasible when:

- the area inhabited is small, that is, <1,000 m²
- the infestation occurs within an area of minimal flushing or exchange of water
- the available habitat occurs in relative shallow water, such as <15 m
- the population is relatively aggregated and has not yet reached reproductive maturity (Crombie et al. 2007)
- the infestation is detected and controlled before spawning can occur.

Tracking the success of eradication to ensure effectiveness of response management can inform the next steps of the response. Expert modelling can give a measure of progress during an eradication program. For example, an eradograph, uses the specific characteristics of the marine pest and the incident managers eradication objective to generate the temporal trajectories of delimitation. It can imply the reallocation between search and control activities or to discontinue, maintain or increase an eradication program. However, any applications or suggestion of changes in an eradication program must be evaluated against a benefit-cost analysis (Burgman et al. 2013).

In planning an emergency eradication response, it is important to obtain good descriptions of the nature of the incursion, including the environment in which it is located and the distribution and abundance of the pest. As much as possible, these descriptions should be spatially explicit (that is, geo-referenced) to guide application of treatment methods.

Table 9 summarises the variables that may be used to describe the nature of a marine pest incursion and help define likelihood of eradication.

Table 9 Variables to describe distribution of marine pest incursion

Variable	Distribution level
Area currently infested	Very small (<100 m ²) Small (100–1 000 m ²) Medium (1 000–10 000 m ²) Large (1–10 ha) Very large (>10 ha)
Abundance	Low Moderate High
Pattern	Continuous Fewer than 5 patches 5 or more patches
Use of suitable habitat	Low (<10%) Moderate (10–50%) High (>50%)
Maturity of organisms found	Juveniles Sub-adults

Variable	Distribution level
	Adults
Maximum depth of infestation	Shallow (<2 m) Moderate (2–15 m) Deep (>15 m)
Maximum depth of available habitat	Shallow (<2 m) Moderate (2–15 m) Deep (>15 m)
Turbidity	Clear (visibility >5 m) Moderate (visibility 1–5 m) High (visibility <1 m)
Water exchange in incursion area	Minimal Low High

Source: Modified from Crombie et al. (2007)

5.4 Methods for containment, delimitation, and eradication

Methods that have been trialled for containment, delimitation and eradication of established populations are listed below. The methods used to treat marine pests will vary in efficacy according to the size and location of the incursion, whether they are in open, closed, or semi-enclosed coastal environments. The methods can be used at any phase of a response for which they are determined most appropriate for containment and control, delimitation or eradication. More details on the efficacy of these treatments can be found in summaries by Aquenal (2007), McEnnulty et al. (2001) or in the primary references cited in [Appendix D](#).

This section provides understanding on:

- open coastal environments
- closed and semi-closed coastal environments
- monitoring and ongoing surveillance.

The methods used to treat marine pest incursions, which can be divided into three generic types:

- [physical treatment](#)
- [chemical control](#)
- [biological and ecological control](#).

A summary of these treatments in both artificial and natural substrates is provided in [Appendix D](#).

Taxa-specific [Marine Pest Response Manuals](#) have been prepared for several marine pests and include methods for containment and control, eradication and guidelines and designs for delimiting surveys:

- invasive marine crabs
- invasive marine bivalves

- Northern Pacific seastar (*Asterias amurensis*)
- Japanese seaweed (*Undaria pinnatifida*).

[National Control Plans](#) (NCP) have also been developed for several marine pests that are already established in Australia and are having significant impacts on the marine environment or marine industries:

- Northern Pacific seastar (*Asterias amurensis*)
- Asian bag or date mussel (*Arcuatula [Musculista] senhousia*)
- European green shore crab (*Carcinus maenas*)
- Japanese seaweed or wakame (*Undaria pinnatifida*)
- European or basket shell clam (*Varicorbula gibba*)
- European fan worm (*Sabella spallanzanii*).

5.5 Open coastal environments

There are limited emergency eradication response options available for marine pests in open coast environments, particularly on high energy coastlines or water >10 metres deep. Many treatment options described in the following sections and [Section 4](#) may be applied to small-scale incursions in the open ocean environment, but the primary difficulties are containing the wide dispersal of larvae if reproduction is occurring and maintaining treatment conditions at a lethal level for enough time to be effective. For instance, the application of chemicals will require development of support structures or technologies account for current and wave action effects. Most chemical treatments also cause impacts on non-target species and may have significant environmental effects which requires consideration.

Successful eradication of small incursions may be possible using methods, such as physical removal, smothering, small-scale containment, and chemical treatment if the incursion is detected early or where site- and species-specific conditions allow removal or containment. Successful eradication usually combines a range of methods, some of which may be selected on factors such as population distribution and density (Green & Grosholz 2020).

5.6 Closed or semi-enclosed coastal environments, aquaculture stock and equipment

Eradication is most achievable in closed or semi-enclosed coastal environments, such as marinas and coastal lakes, or from aquaculture stock and equipment because the marine 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 freshwater, application of chemical biocides, physical removal, and ecological control. Timeliness is essential, because if the marine pest has spawned and the larvae have settled then control will be far more difficult.

If the infestation is confined to a relatively small, enclosed, or semi-enclosed waterway, it may be possible to treat the entire water body and all aquatic habitats within it (Willan et al. 2000). Similarly, if the infestation is confined to specific aquaculture equipment or stock then it is possible

to treat the equipment or dispose of the infested stock. If this is not possible then the management success will depend more heavily on the ability of monitoring and delimitation surveys to locate and treat all clusters of the population.

The wide range of physical tolerances of marine pests present challenges for their control. Control will require continued coordination and communication between affected parties. When resources allow, all habitat potentially suitable for the pest should be surveyed and treated where required. When this is not possible, habitats should be prioritised based on suitability for the pest and delimitation survey results.

5.7 Monitoring, delimiting, and ongoing surveillance

Monitoring, delimiting, and ongoing surveillance are used to detect marine pest populations, to inform control programs or to support that eradication has been successful.

Active surveillance for any marine pest 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, then it will be necessary to have targeted active surveillance for the species outside the restricted and control areas to support declaration of zones free from the marine pest under surveillance.

The [Australian marine pest monitoring manual and guidelines](#) can be used to help determine quality assurance and control, and appropriate sampling intensity for ongoing surveillance.

Several surveillance types include:

- systematic and targeted searches of suitable or treated subtidal habitat within the restricted area or at sites at risk of infestation
- systematic and targeted searches of suitable or treated intertidal habitat within the restricted area or at sites at risk of infestation
- targeted searches and inspection of vessels and other vectors departing, or which have left, the control area
- regular monitoring of recruitment within the restricted area or at sites at risk of infestation, including use of molecular detection techniques.

5.7.1 Molecular delimitation and surveillance

Molecular detection techniques can be rapid and cost-effective tools for marine pest surveillance. These techniques are highly sensitive and can assist in detecting target species, even at low abundances. Molecular methods can also be used to confirm identification of sample specimens when morphological identification is difficult or unresolved. A range of tools and resources exist to support molecular surveillance and are referenced throughout this section. For molecular techniques to effectively support marine pest management, issues such as assay validation, sampling procedures, marker/DNA probe selection and interpretation of molecular surveillance results should be considered.

Molecular delimitation surveillance involves identifying the spatial population boundaries of a target species. This in turn, can assist in the prioritisation of other methods for containment, eradication,

and control of established populations. The species may be present at low population densities and have a heterogeneous distribution, which can increase the time and resources required to undertake comprehensive delimitation (Bott et al. 2010; Darling & Mahon 2011; Darling et al. 2017; Darling & Frederick 2018; Goldberg et al. 2016; Hauser et al. 2016; Trebitz et al. 2017; Zaiko et al. 2018). In aquatic environments, detection probability is influenced by the decay rate of genetic material and passive dispersal from the source under local hydrodynamic conditions (Ellis et al. 2022; Darling & Frederick 2018). The high sensitivity and low costs of molecular techniques make them an effective tool for delimitation surveillance (Goldberg et al. 2016) and the ability to test historic environmental samples can improve temporal surveillance resolution and assist in trace-back activities. Other benefits of molecular surveillance include the ability to detect life stages that lack diagnostic morphological characteristics such as eggs and larvae, cryptic or morphologically ambiguous taxa, and viable but non-culturable microorganisms (Darling & Frederick 2018).

Molecular methods for detection of marine pest species have been developed using primarily either a polymerase chain reaction (PCR) approach generally targeting specific species, or a high throughput sequencing (HTS) approach that attempts to identify sequences to the lowest taxonomic level in a community but may lack the specificity to identify sequences to a species level. In delimitation surveillance, usually one species or taxon will be targeted, therefore the PCR or quantitative PCR (qPCR) approaches are recommended. Targeted species approaches aim to determine the presence or absence of a species in a sample, the abundance of the target species in a sample, and whether the sample complies with a standard. For community-based approaches, HTS metabarcoding or next-generation sequencing may be used to identify multiple species in a complex sample to infer species richness and biodiversity (Darling & Frederick 2018).

Validated assays should be used where possible to maximise detection probability and so assay performance (sensitivity and specificity) can be quantified. The Department of Agriculture, Fisheries and Forestry developed [Guidelines for development and validation of assays for marine pests](#) that advise on consistent and comparable validation processes to develop assays. The CSIRO have also developed the [Environmental DNA test validation guidelines](#) and [Environmental DNA protocol development guide for biomonitoring guidelines](#) which provide quality control and minimum standard considerations for developing, validating and using eDNA/eRNA assays for single- and multi-species detection. The [Compendium of introduced marine pest molecular studies relevant to Australia](#) contain species-specific information including validated assays. PCR assays developed overseas should still be validated for Australian conditions because of the potential for cross-reaction with native species that could affect test performance.

Sensitivity levels of PCR tests are high, allowing detection even where target DNA is present at very low concentrations. However, where the target organism is rare, DNA may not be present in every sample. Sample quality and DNA quantity, inhibition, false positive or negative errors and seasonal variability in DNA presence in the water can influence results (Goldberg et al. 2016). Use of validated assays enables calculation of the optimal sample number as part of surveillance program design. The South Australian Research and Development Institute (SARDI) have developed a [sample number calculator](#) for surveys using plankton tow samples and qPCR assays.

Molecular surveillance results should be interpreted in conjunction with other surveillance tools, methods, and considerations to best inform management. Positive molecular detections of target

DNA do not guarantee target organisms are present at the location and may be due to false positive results (DNA probe specificity or sample contamination) or translocation of target DNA (e.g. through ballast water or hydrodynamic dispersal). Positive detections using molecular methods should be confirmed using traditional surveillance methods where possible.

5.7.2 Divers and remote operated vehicles

Divers and ROVs may be used for both surveillance activities and delivering treatment methods for which they are determined most appropriate.

Divers can be particularly effective at detecting marine pests that tend to aggregate around complex structures. However, the ability to observe a marine pest while diving relies heavily on water visibility, identification training, and search techniques. Divers can use touch very effectively to detect some marine pests in inaccessible niches. On several occasions, mussels have been detected by divers using touch. Cost of professional divers needs to be considered by managers. If visibility is low, then visual surveys will be compromised. These same visibility limitations apply to ROVs. However, ROVs can be used in place of divers, particularly when hazards are present (e.g. crocodiles, sharks, stinging cnidarians), but their full use in surveillance or pest detection is still being optimised and few data are available on their effectiveness.

Divers are required for the application of several treatments, as well as subtidal surveys, around wharf piles, vessels, floating pontoons, and other artificial structures in port and marine environments, and on intertidal and shallow subtidal reefs:

- [underwater vacuum, suction and filtering systems](#)
- [wrapping and encapsulation](#)
- [smothering](#)
- [osmotic treatment](#)
- [direct chemical injection.](#)

5.7.3 Settlement arrays

Settlement arrays are commonly used to study recruitment of sessile marine organisms from planktonic life stages, such as larvae, into a benthic juvenile or adult phase.

Settlement arrays are likely to be an effective sampling method and surveillance tool for fouling species like epifaunal bivalves (i.e., mussels and oysters), ascidians, bryozoans, macroalgae, some tubeworm polychaetes (i.e., *Sabella spallanzanii*), sea sponges, some corals, and barnacles. They are unlikely to be an effective sampling method for infaunal species (i.e., infaunal clams such as *Mya arenaria*) or motile species (i.e., crabs, seastars).

Settlement arrays have many advantages including:

- being cost-effective to make
- simple to use and easy to deploy by non-specialists
- can sample species continuously over a long period of time (temporal scales)

- can be deployed in different areas and depths of the water column (spatial scales)
- can sample species inaccessible to divers or other sampling methods because of organism size or seasonality
- fouling organisms growing on plates can be used for both taxonomic identification and molecular diagnostics.

A disadvantage of settlement arrays is the relationship between the presence and abundance of the target species within the environment and its detection on the settlement surface is complex and difficult to quantify, which is similar to other methods of passive sampling. For fouling species of marine pests, this means that:

- Uncommon or rare biofouling species, including those that are at an early stage of establishment, will be under-sampled
- Absence from an array does not necessarily mean the absence of an established population because of species-specific variation in settlement preferences.

Important factors influencing settlement array results include: timing, duration, and depth of deployment, orientation (and shading) of the surfaces, surface rugosity and material, water currents and tidal movements, predation, and the presence of antifouling coatings (Tait et al. 2016). The number of settlement arrays or surface area of settlement substrata must be relatively high, and the settlement area must be attractive for settlement of the target fouling species to detect it during sampling (Floerl et al. 2012).

Various designs of settlement arrays have been proposed (Floerl et al. 2012; Sutton & Hewitt 2002). Generally, settlement arrays consist of a collection of plates of varying materials and surface features that act as settlement substrata for larval phases of sessile marine species (Figure 4). Plates are usually placed about 2 metres below water level at low tide and attached to a fixed structure in the environment such as a wharf piling or pontoon.

Settlement plates are typically deployed for a minimum of three months to allow biofouling to reach a size and maturity to enable effective taxonomic identification. Where tidal amplitudes are large, a suspended settlement array is essential to maintain a constant depth.

Different orientations, water depths, and numbers of settlement plates deployed can be used (Tait et al. 2016). Settlement arrays can also be deployed in a staggered manner to enable continuous sampling over the reproductive period of the target fouling species, while minimising overgrowth of biofouling organisms. For example, two months after deploying a settlement array, a second settlement array could be deployed. After four months of deployment the original settlement surfaces are retrieved while the second set of surfaces is retrieved two months later. This allows for two overlapping deployments each of four months duration.

Plastics (PVC, Perspex), wood, cement/rock, metal (steel aluminium) and fibreglass have successfully captured sessile species such as marine bivalves (Tait et al. 2016; Vekhova 2006). The likelihood of a sessile species to settle will differ between settlement surfaces and this needs to be considered. For example, natural rope or other filamentous fibres are used to promote *Perna canaliculus* settlement in aquaculture settings in New Zealand.



Figure 4 Example of a standard settlement array showing square plates attached to a frame

Source: Leigh Tait, NIWA

5.7.4 Shore searches

Shore searches may be used for surveillance activities and as a treatment method.

Shore searches can be an effective way to observe and catch live intertidal marine pests. Once searchers are familiar with the identity of the target marine pest then many searchers can be deployed, covering large areas. A standard shore search may involve 10-minute timed searches along a transect or be based on the number of rocks/boulders overturned. For example, in Port Phillip Bay, Victoria it was reported that *H. sanguineus* were found to occur in very discrete patches roughly 5 m² where up to 15 individuals could be found in 2 minutes. Between patches, up to 100 rocks could be turned over before another patch was found (MPSC 2022).

Often shore searches are used to augment other sampling regimes, such as baited or unbaited traps. Shore searches can also aid with surveillance activities. Shore searches are less effective at sampling species that are subtidal or cryptogenic species because they are hard to identify. Complex or inaccessible habitats such as mangroves, steep limestone cliffs/rocks and areas with high boat traffic or swell can also impede shore searches.

5.8 Physical treatment

Physical treatments include a range of methods that rely on the ability to detect and either remove marine pests or kill them *in situ*. Physical treatments are generally the most socially and environmentally acceptable way of removing marine pests from a system. Physical treatments can be difficult to achieve in complex habitats, such as oyster reefs or mangrove forests, making operations challenging or environmentally destructive. Consequently, physical treatments are mostly effective in

small and accessible areas, such as on a relatively flat seabeds or artificial structures, such as a hull surface in a contained marina.

Manual removal

Manual removal typically refers to collection and removal of the pest organism by hand or by using handheld implements. Manual removal has been used as a rapid response and long-term control option for some introduced macroalgae, molluscs, seastars and crabs (McEnnulty et al. 2001). Hand collection and containment of bivalves can be achieved by divers, in the intertidal zone, or removal from man-made structures on land (e.g. vessels and aquaculture equipment).

The advantages of manual removal are selectivity for the target pest and limited damage to non-target species. However, as it requires visual detection of the pest it cannot be applied effectively in turbid environments where such detection is impaired. Manual removal is of greatest utility when incursions are small and spatially confined or when they are in sensitive environments (such as marine reserves or areas of high biodiversity value). Oyster farmers in New South Wales have used hammers to smash Pacific oyster, *Magallana gigas*, to control feral populations and the oyster virus ostreid herpesvirus-1 microvariant (OsHV-1 μ var) (Cavanagh 2014). Similar methods for destruction of wild *M. gigas* were used in South Australia (Keen 2010; Sierp 2019).

Mechanical removal

Mechanical removal entails use of machinery to directly remove the target species and may involve techniques such as mowing, dredging, trawling, or mopping. Care must be taken during mechanical removal as fragile species, such as many polychaetes and colonial organisms, may be capable of regenerating from fragments. *Perna perna* was successfully eradicated by dredging in central New Zealand following accidental introduction following de-fouling of a drilling rig (Hopkins et al. 2011).

The efficacy and environmental impact of tools should be considered when implemented as part of a management response. Some of these practices can cause considerable bycatch or ecological damage, either through direct disturbance of the assemblages or through modification of habitat (e.g. removal of habitat-forming species, increased turbidity, release of toxic chemicals from the seabed).

5.8.1 Harvesting

Harvesting can reduce numbers of some marine pests with community assistance programs, recreation and commercial harvest incentive schemes and fishing methods, including netting and trapping. Harvesting has moderately high selectivity applicable to fish, crabs, seastars and jellyfish. Harvesting is more suitable to control strategies or for local depletion than for eradication (Pasko and Goldberg 2014). It can represent an opportunity to support ecosystem and natural resource management, but it can also incentivise intentional spread of the marine pest.

5.8.1.1 Recreation and commercial harvest incentive schemes

Any consideration of recreational or commercial harvest must bear in mind that often harvesters may aim to maintain stocks rather than reduce them to non-viable levels, which may not be consistent with management aims. Additionally, transfer of valued species to new areas is common and difficult to manage so this must be considered. Marine pests that are recreationally harvested for food may become socially important which has the potential to impair eradication aims.

Acceptability as a food source may be high so implications for food safety may also need to be considered.

Incentive schemes may be offered in several ways (Pasko and Goldberg 2014):

- contract operation (commercial):
 - payment to a service provider for the removal or harvest of the marine pest
- commercial market (commercial):
 - effort undertaken, usually privately, to harvest and sell the marine pest when a perceived market exists
- recreational harvest (recreational):
 - encouragement of recreational fishing of the marine pest.

Incentive schemes do not necessarily rely on the marine pest being marketable, although more attractive species require less additional incentive for capture. Many pests have existing markets, and these species may be viewed as a potential resource in their introduced range (Andreakis and Schaffelke 2012). Where incentives are offered, the value of these may need to increase as the pest population decreases to reflect the additional effort required to capture rare individuals (Pasko and Goldberg 2014). To determine if commercial harvest of an introduced species is viable, data are needed on catchability, cost of fishing methods, and product value (St-Hilaire et al. 2016). Production of by products such as compost or fertiliser may be viable options for large quantities of product with fewer concerns about degradation.

5.8.1.2 Community assistance programs

Community assistance in removal of highly abundant marine pest species can increase awareness but generally reduces numbers in the short term. There is also potential for 'by-catch' of misidentified non-target species, and sustained pressure needs to be maintained at appropriate times.

5.8.2 Epibenthic sled

Benthic sled tows effectively sample epibenthic assemblages over large areas (Figure 5). Dredge and sled catch efficiency can be affected by operational factors such as speed of towing, fullness of catch, depth, substrata and motile species' ability to escape. It may be necessary to determine dredge and sled efficiency to help inform survey design (Hopkins et al. 2011). Benthic sampling will be unsuitable for reef habitats or other complex structures. Epibenthic sleds could be used to augment other sampling regimes but are not recommended as a single survey method or single method for containment, eradication or control.



Figure 5 Epibenthic sled

Source: Chris Woods, NIWA

5.8.3 Netting

Nets, including seines, midwater trawl nets, gill nets, fyke nets and dipnets can be used to catch active species. The appropriate net mesh size to marine pest species size needs to be considered. For example, netting can be effective at capturing larger numbers of adult crabs but may be ineffective at capturing juveniles and small crab species if they can escape through the mesh (MPSC 2022). An environmental and logistical drawback of netting is the amount of bycatch. For example, fyke nets caught significantly more *Carcinus maenas* than baited box traps but also caught significantly more bycatch than the baited traps (Poirier et al. 2017). Nets can be difficult to operate in complex habitats such as coral and rock areas and seagrass meadows. Also, nets are usually prohibited in port areas where surveillance operations are commonly undertaken, or in sanctuary zones and some marine park areas.

5.8.4 Trapping

A variety of trap types (baited and non-baited) can be used to target mobile marine species such as decapod crustaceans (crabs, lobsters), fish and seastars (Aqueal 2007). Trapping programs are simple, quick to initiate and require a minimal level of training and familiarity with equipment. Traps can be deployed in a variety of locations such as near intertidal rocky shores, wharf pilings, break walls and other habitat with complex physical structure, such as seagrass meadows, temperate mangrove channels, saltmarshes, and shellfish beds.

Trapping has limited collateral impact on the environment and is consequently viewed as a socially acceptable form of pest control. However, trapping has the potential to capture bycatch and such impacts should be considered by incident managers (Clark 2011). Trapping efficacy will depend on the target species, availability of alternative, natural food sources, and is most effective in areas with high populations (Andrews, Whayman & Edgar 1996). Although trapping can remove large numbers of (usually) adults from a population, most trapping techniques tend to be highly selective and are therefore effective for only some life stages and sizes or one gender of a population. Because of this, trapping is not an effective eradication tool on its own but is best used as part of an integrated pest management program along with other treatment methods.

Traps may be destroyed or dislodged when deployed in areas of crocodile habitat, high swells, or large tidal movements. Traps placed in areas with public access such as popular beaches can be at risk from human theft and tampering.

5.8.4.1 Baited traps

Baited box traps (Figure 6) are logistically convenient because they are relatively small, lightweight and collapsible, meaning they can be carried in large numbers on board small boats, whereas larger commercial crab pots may require specialist boats to deploy. Baited traps can usually be deployed over a relatively short duration (24-hour soak time) enabling a large area to be sampled (Mabin et al. 2020). Baited traps are attractive to aggressive or active predators and scavengers but can miss herbivorous and some omnivorous species. Some species of crabs that are egg-bearing may also forage less (MPSC 2022). Smaller species are less likely to be attracted inside baited traps or may not be detected as cannibalism and predation inside traps is common.

Bait choice is of vital importance as this is the lure for the target species (Favaro et al. 2020). Bait fish such as sardines are commonly used and typically considered highly effective (Dittmann et al. 2017; Favaro et al. 2020). Sardines and pilchards are commercially available and can be purchased in bulk, making them a cost-effective bait choice. Squid and cod are not as effective, whilst mussels are completely ineffective at attracting some species such as crabs (Favaro et al. 2020).

In some jurisdictions the use of traps with specific designs is regulated and permission may need to be sought prior to deployment. It is also important to note that when using baiting methods relevant animal welfare legislation should be considered as part of the trapping activities (MPSC 2022).



Figure 6 Baited box traps

Source: Chris Woods, NIWA

5.8.4.2 Unbaited traps

Unbaited traps include pitfall traps, plastic crates filled with bivalve shells, and crab condos (Figure 7). These types of traps are engineered to attract species by providing shelter. Experimentation has shown that some crab species select habitat based on structure rather than food availability (Riipinen et al. 2017). These types of traps are much better at catching and attracting marine pests such as crabs at different life-stages, from new settling megalopae to reproducing adults (Fowler et al. 2013).

Refer to [Response manual for invasive marine crabs](#) for specific detail on crab condos and plastic crates filled with bivalve shells.

Pitfall traps can be constructed in intertidal soft sediments by sinking buckets (~20 L) filled with seawater into the substrate so the rim is flush with the sediment surface. Foraging species fall into the buckets and are unable to escape. Unbaited traps that extend across stream and river channels have effectively been used to trap one million *Eriocheir sinensis* over two years in Belgium (Schoelynck et al. 2020). Use of barrier systems like this will need to account for the other wildlife, particularly dugongs and turtles, as well as consideration of freshwater species, such as platypus, within water bodies with a brackish boundary (MPSC 2022).



Figure 7 Crab condos

Source: Chris Woods, NIWA

5.8.5 Barriers

Physical barriers deployed during migration of species such as *E. sinensis* can lower population densities, however, they may not be effective in large waterbodies with high degree of connectivity between tributaries (Schoelynck et al. 2020). Chemical barriers have been considered, although there are significant legal and public health issues associated with handling or deploying poisons in the marine environment, and they are not sufficient on their own to effect eradication.

5.8.6 Underwater vacuum, suction and filtering systems

Underwater vacuum systems are flexible suction hoses attached to small dredges to suck the target organism from marine sediments or from fouling surfaces. Care must be taken to properly filter the water and capture all material to prevent the spread by fragments or release of any larvae (Coutts 2002). Vacuum removal of *D. vexillum* colonies has been implemented on wharf piles and vessels. In New Zealand this was effective on vessels when filtering to 50 µm to minimise release of larvae (Coutts & Forrest 2007). Underwater vacuum is best suited to infrastructure or sites where substrates are primarily sandy.

Use of this method is not suitable for seabeds as poor visibility can be caused by the diver's contact with the seabed, the dragging of the vacuum pipe and the reverse flushing action used to clear blockages. When used in fine, muddy sediment or where there is a large quantity of biofouling, vacuum filters are easily clogged. Due to the labour-intensive nature and thus high cost of the procedure, diver assisted underwater vacuum is most effective against small infestations.

5.8.7 High-pressure water blasting

High-pressure water blasting is a cost-effective and an environmentally acceptable method of treating biofouling on infrastructure and should remove all mobile biofouling species (Inglis et al. 2013). High-pressure (>2000 psi for 2 seconds at 100 mm distance) may be required to dislodge biofouling from fissures and crevices. Water blasting has been used to remove established

populations of mussels, macrophytes and tunicates from vessel hulls or other hard substrata, as well as from infected aquaculture equipment. High-pressure water blasting can clean a wide variety of structures. Water blasting could promote release of gametes, so high-pressure cleaning may best be combined with additional treatments such as chemical treatment, heat or desiccation.

High-pressure water sprays typically require treated areas to be either intertidal or removed from the water. *In situ* cleaning by underwater blasting should not be considered for an incursion response unless all viable biological material can be collected.

5.8.8 Shading and light attenuation

Light attenuation deprives autotrophic fouling species of light and ability to photosynthesise. Screens, covers and dyes can change or reduce the amount of light to which the plant is exposed, causing it to die. For plants with large carbon reserves (in rhizomes, tubers or other vegetative structures) shading can take a long time to be effective and can be difficult to maintain. Shading may also not effectively treat all life stages of the plants that have different light requirements. Use of this technique is likely to be limited to enclosed water bodies such as coastal lagoons, closed estuaries or enclosed breakwater harbours, where structures or dyes used to shade plants can be kept intact for long periods.

5.8.9 UV light treatment

The application of ultraviolet light (UVC; 100–280 nm) can prevent recruitment on ship hull coatings and reduce biofouling settlement on reverse osmosis membranes (Hunsucker et al. 2019; Rho et al. 2022). UVC is the most germicidal wavelength in the UV spectrum and it breaks chemical bonds between DNA and RNA polymers in microorganisms (Braga et al. 2020). This treatment has the potential to cover small and large areas depending on lamp size and transmission intensity. Effectiveness of treatment will be dependent on the light's power, exposure time, frequency of treatment, distance from treatment area and water quality for light penetration (Hunsucker et al. 2019). Ship hull construction material and anti-fouling coatings need to be considered as long-term exposure with UVC light has been shown to damage copper coatings (Hunsucker et al. 2019).

5.8.10 Thermal treatments

Thermal treatment has low selectivity, but impacts are localised and leaves no residues. It is most suitable as a management tool against biofouling, microscopic life-stages, soft-bodied organisms and species with thin shells such as dreissenid species (Cvetkovic et al. 2015). Complex topographies, heavy fouling, or taxa with thicker shells such as corbiculid species may require higher temperatures and/or longer exposure times (Inglis et al. 2013).

Thermal treatment may be applied as elevated temperature via:

- hot water
- steam
- created by:
 - electrical elements
 - hydrodynamic cavitation
 - heat torches.

Thermal treatment may also be applied as reduced temperature via cold or ambient water to materials and equipment in containment.

The efficacy of any thermal treatment is dependent on the susceptibility of the target organisms life stages and the ability to maintain the required temperature to achieve mortality. The mass of fouling and exposure time will need to be considered when planning a treatment. Generally, heat treatment is a favourable treatment option because of its efficacy and low risk to environment and operations. Organisms such as crabs and bivalves with hard shells require hotter treatments (50 to 70 °C) than soft-shelled organisms. The use of heated water between 50 to 60 °C can render taxa non-viable in under two hours (Cahill et al. 2019; Growcott, Kluza & Georgiades 2016).

Effective and safe deployment is likely to be limited to depths no greater than 30 m, but only small areas can be treated without the use of multiple dive teams. Its acceptability is high and requires specialised equipment, operator training and may require jurisdictional approval.

Substrates that can be removed from the water can be immersed in hot water, or heated water can be applied to contained areas such as niche spaces and piping (Forrest and Blakemore 2006). Heat produced by vessel engines or hydrodynamic cavitation can be used to treat ballast water or vessel internal niches (Quilez-Badia et al. 2008; Leach 2011).

Underwater flame torches cause rapid (<30 s) mortality in clams but substrates need to be considered as clams buried in mud required heating for up to 5 minutes (Coughlan 2019). Flame torches have been used to destroy intertidal *M. gigas* in South Australia and were deemed suitable for small-scale destruction with added benefit of killing OsHV-1 μ var in oyster tissues. Powerful flame torches may be deemed unsafe and risk damage to infrastructure and handlers.

5.8.11 Desiccation and water level manipulation

Lowering water levels in a water body can cause mortality of submerged organisms through desiccation. Desiccation also involves the removal of equipment and attached fouling communities from the water and drying them. These tools can be effective control options for marine pests, although the practicalities associated with manipulating water bodies or removing infested structures from the water will need to be considered. Application of these techniques may be restricted to structures that can be removed from the water, or to contained areas where draining of water (drawdown) is feasible.

Where applicable, the recommended length of time required for equipment to be fully dried ensuring that all biofouling is killed will be ~21 days (Hilliard, Polglaze & LeProvost 2006). However, this may be longer for some marine pests that have a range of tolerances to aerial exposure. For instance, intertidal crabs and thicker-shelled bivalves, such as *M. gigas*, typically have a high tolerance for air exposure (MPSC 2022).

For this reason, material being desiccated needs to be well spread out. Weather conditions need to be considered as the air temperature and/or relative humidity may prolong the period required for desiccation. For example, *P. perna* can tolerate air exposure for around 18 days at 15°C and high relative humidity, compared with only around 1 day when exposed at 25°C and low relative humidity (Hicks & McMahon 2003).

Sunlight in combination with desiccation is extremely effective as a general disinfectant.

5.8.12 Wrapping and encapsulating

Wrapping deprives fouling species of light and food, with continued respiration and decomposition of organisms within the barrier depleting oxygen to lethal levels.

Effectiveness of this method:

- relies on smothering material to be continuously applied without gaps or breaks to avoid escape of fragments or larvae or ingress of clean water
- is greatest in sheltered environments with low currents as strong currents can make deploying the wrap difficult and increase the risk of tearing
- is improved through the addition of biocides.

Polyethylene silage plastic wrap (125 µm thick) can be cut to size to suit the vessel type and is deployed by divers in association with a topside support team (Mitchell 2007). 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, watertight environment. Application has also been applied to wharf piles by dive teams using rolls of black polyethylene (1 m wide × 50 µm thick) by wrapping around the piles in a circular motion overlapping each successive wrap by ~400 mm (Figure 8; Coutts and Forrest 2007). Sharp objects on the hull or pylon, such as propeller blades, oysters, or fixings, should be wrapped separately or covered with tubing or cloth before encapsulation to prevent tears in the plastic. 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. Commercially available floating boat docks up to 30 m have been shown to be useful for emergency treatment of biofouling on small vessels.

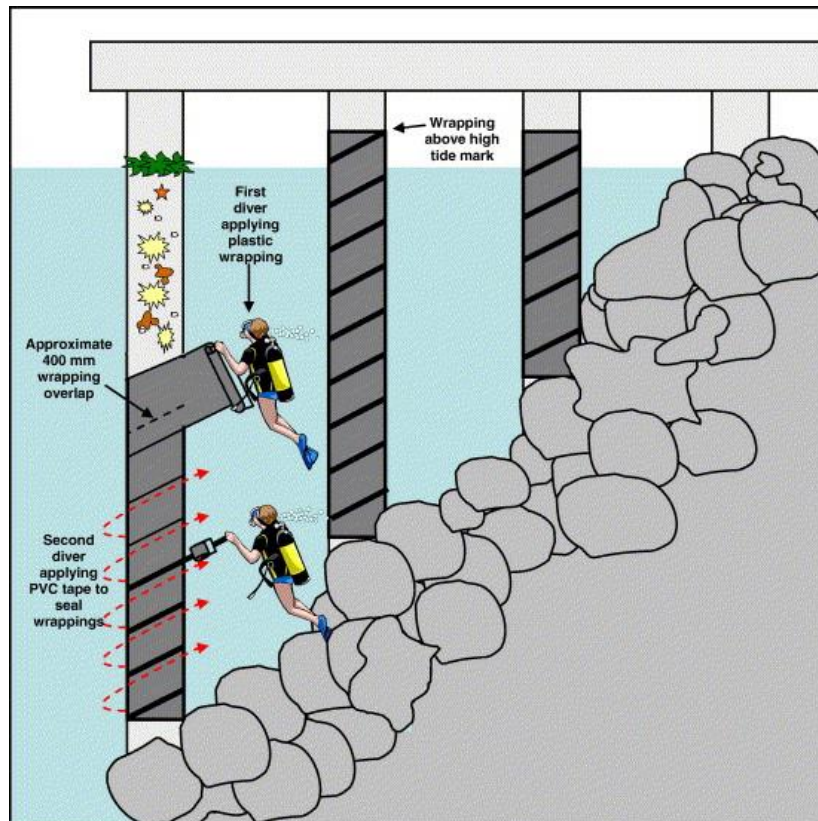


Figure 8 A a schematic of the polyethylene wrapping method used to treat wharf piles

Source: Coutts and Forrest (2007)

If properly deployed, the wrap should contain the pest species and its larvae. Extreme 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 7 days if no biocide is used to achieve the desired effect (Inglis, Floerl & Woods 2012). Wrapping of vessels >25 metres is labour intensive and may take up to two days to deploy. The time needed for effective treatment is around 7 days, which may be too long when rapid treatment and vessel turnaround time is crucial.

With any wrapping method it must be noted that some marine pests, such as crabs (*E. sinensis* or *H. sanguineus*) can leave the water to respire and feed. Wrapping techniques should ensure that this cannot happen for mobile species capable of survival out of water. Wrapping also produces large amounts of plastic waste. This waste must be disposed of in landfill or an approved solid waste treatment facility. For more information on disposal see [Section 6](#).

5.8.13 Smothering

Like wrapping and encapsulation, smothering benthic habitats by covering them with plastic, geotextile fabric or burial with sediment (such as dredge spoil) can effectively treat relatively localised infestations. Smothering has:

- low selectivity, but impacts are localised
- leaves no residues
- applicable to sessile or sedentary species (on surfaces that can be covered)

- relatively affordable.

The material used to smother the surface must be continuous, without gaps or breaks in material to avoid escape of fragments or larvae. Tolerances to burial by sediment is variable between marine pests and some can tolerate prolonged periods (>2 weeks) of burial (Glasby, Creese & Gibson 2005). Control programs must consider if the physical burial of marine pests such as macroalgae with sediments could potentially cause environmental damage or improve the habitat for settlement and expansion of other introduced species.

The application of smother material to be used is dependent on the seabed or substrate. For example, smothering of flat or gently sloping soft-sediment seabeds with uncontaminated dredge spoil was successful in eliminating *D. vexillum* in New Zealand (Coutts 2006; Coutts and Forrest 2007). Divers carried down polypropylene woven bags (840 x 460 mm) filled with the dredge spoil and emptied them across the infected seabed with a coverage of 100 mm (depth) (Coutts 2006). Geotextile fabric sheets proved to be more suitable for steep gradient rip-rap seabeds as they are unable to hold dredge spoil.

5.8.14 Containment combined with chemical treatment

Speed of effectiveness of wrapping, encapsulation and smothering can be improved through the addition of biocides such as chlorine or acetic acid (Ammon et al. 2019). This may be particularly effective for marine pests, such as bivalves, that need to have their tolerance of anoxic environments taken into consideration. This treatment method is most suitable for sheltered environments. In high-energy conditions, deploying and maintaining containment structures is problematic, as is handling and deploying chemicals. The addition of chlorine (sodium dichloroisocyanurate, 'dichlor') at an initial concentration of 200 mg/l to a vessel in a floating dock rendered 90% of the invasive polychaete *Sabella spallanzanii* non-viable within 4 hours of exposure (Morrisey et al. 2016). In addition, other fouling organisms were killed within 6 days; *M. gigas* oysters attached to the hull were gaping and empty and macroalgae had been bleached (Morrisey et al. 2016).

5.9 Chemical treatment

The dynamic nature of marine environments means that any biocides or chemical agents, such as chlorine, salt, herbicides, or pesticides released into them are rapidly diluted and dispersed. This is problematic when the agent must be above a threshold level to be lethal. Very large concentrations may need to be released or the area may need to be enclosed for the treatment to be effective (Ferguson 2000; Anderson 2005). Conversely, where the agent is effective at very low concentrations, rapid dispersion by water may achieve broad dispersal.

The major considerations for the use of chemical treatments in water bodies, including the:

- volume of water that needs to be treated (a function of the area, depth, and degree of flushing of the waterway)
- presence, susceptibility, and value of non-target organisms that may also be affected
- water quality (e.g. organic matter may consume chemical)
- residual effects of any toxicants on the surrounding environment and human health
- safety management when handling large volumes of chemicals.

Incident managers should consider the use of chemical control in aquaculture and the potential for negative effects on future marketability of a product or useability of the infrastructure, e.g. copper compounds may inhibit phytoplankton production in ponds.

The [Australian Pesticides and Veterinary Medicines Authority](#) (APVMA) is the Commonwealth authority responsible for assessment and the registration of all agricultural and veterinary chemical products in the Australian marketplace. The primary legislative acts the APVMA operates under are the *Agricultural and Veterinary Chemicals (Administration) Act 1992* and the *Agricultural and Veterinary Chemicals Code Act 1994*. The APVMA maintains a list of all approved chemical products that are available in Australia, the list can be found at the [APVMA PubCRIS database search](#). Any variations required to be made to registered and approved use of these chemicals must be approved by APVMA.

In most states and territories, registered chemical products must only be used for the purposes specified on the label. Any use of chemicals for the control of marine pests is likely to differ from that specified on the label. In these cases, permits need to be sought from APVMA to use chemicals in a different way. APVMA can also consider applications for permits allowing limited use of an unregistered chemical product.

In addition to seeking APVMA approval for use of chemicals to control marine pests, there will often be other stakeholders that need to be consulted and consent sought for their use, such as port authorities, local governments, and national park managers.

An extensive range of chemicals have been trialled in the laboratory for their efficacy against marine pests (McEnulty et al. 2001). Several effective chemicals are presented below in more detail and a summary of chemical applications in [Appendix D](#):

Chemicals that have been evaluated for their efficacy against marine organisms comprise two forms:

- oxidising biocides:
 - chlorine (gas, or sodium or calcium hypochlorite)
 - bromine
 - active halogen compounds
 - ozone
 - hydrogen peroxide
 - chlorine dioxide
- non-oxidising biocides (Jenner et al. 1998):
 - aldehydes
 - amines
 - quaternary ammonium compounds
 - organobromines
 - organometals
 - mild acids (such as acetic acid)
 - brine or lime.

5.9.1 Reproductive inhibitors

The endocrine-disrupting compound, asterosaponins, has been suggested as a species-specific method of reproductive inhibitor for reducing *A. amurensis* populations. The difficulties of depositing large amounts of endocrine-disrupting compounds into seriously affected areas have been suggested as a limitation to the potential use of these types of chemicals (Goggin 1998). The use of pheromones as aggregators, disruptors, attractants, or behaviour modifiers is under investigation.

5.9.2 Industrial detergent, disinfectant or de-scaler

Commercial marine detergents, disinfectants, and de-scalers, such as Conquest[®], Quatsan[®] or Rydlyme[®], respectively, deteriorate and/or dissolves biofouling and are biodegradable. Conquest[®] is a highly effective detergent and Quatsan[®] a highly effective disinfectants that cause 100% mortality of fouling mussels within 14 hours at concentrations of 1% and above (Lewis & Dimas 2007). Rydlyme[®] at 25% concentration for 14 hours is the recommended application time to dissolve significant mussel growth (Lewis & Dimas 2007). A linear relationship between the level of fouling and the volume of Rydlyme[®] required to digest fouling has been developed for this treatment (Lewis & Dimas 2007). Rydlyme[®] may dissolve growth in this period, whereas other preparations may weaken it but not dissolve it.

Consultation with vessel or infrastructure owners needs to be considered as some of these preparations have been associated with damage to internal seawater systems. Toxicity of detergents, disinfectants, and de-scalers need to be considered prior to use.

5.9.3 Osmotic treatment

Manipulation of salinity levels (osmotic treatment) has been used in several marine pest incursions. Depending on the marine pest's tolerance, exposure to hyposaline (via addition of freshwater) or hypersaline (via addition of sodium chloride) conditions can disrupt the osmotic balance resulting in death.

It can take the form of immersion of infested structures or equipment in fresh water, manipulation of salinity in enclosed water bodies through re-diversion of fresh or salt water, or through application of large quantities of salt in close proximity to the target organism.

Salt is inexpensive, easy to obtain, safe to handle and can be applied on a large scale with the appropriate resources such as a barge, backhoe, hopper, or diver guidance. This technique becomes less efficient as the area being treated increases or when applied to steep slopes and high-relief habitats (such as rocky reef). Salt treatment is not suitable for application in high-energy environments since salt would be rapidly dispersed by ocean-generated swell. The efficacy depends on absolute salinity change and the rate of change in salinity. The rate of salinity change is likely to be slow for large treatment volumes, so treatments are likely to be most effective for small, enclosed areas. Whilst application of salt can be effective, it can also be detrimental to other species and should be considered when planning response activities.

Manipulation of salinity is an effective technique for treating aquaculture equipment and seed stock and for *in situ* treatment of pests where the incursion and treatment can be contained. Freshwater *in situ* treatment may be restricted to habitats within enclosed environments or structures that can be removed from the water for treatment. It is a non-species-specific technique and is likely to have lethal effects on non-target biota. It is effective for treating hull infestations of attached species, by

mooring affected vessels in areas with low salinity or fresh water but may not be effective on some euryhaline species.

The salinity tolerance of a species can vary according to life-stages and may also be affected by other factors such as temperature, nutrient, or oxygen levels. The efficacy of salinity manipulation for marine pests will depend on their ability to withstand prolonged exposure to an altered regime, for example:

- catadromous mitten crabs, *Eriocheir* sp., are unlikely to be affected by salinity manipulation as they inhabit both freshwater and marine environments (MPSC 2022)
- kelp species, *U. pinnatifida*, gametophytes survive in freshwater immersion for 1–2 days, but plantlet mortality occurs within 10 minutes. *Undaria pinnatifida* survival in hot water across the 35–55 °C range was tens of minutes to a few seconds (Forrest and Blakemore 2006)
- ascidian, *D. vexillum*, has shown resilience against simple freshwater immersion (Denny 2008). However, *D. vexillum* coverage has been reduced with brine (40, 50 and 70 ppt) for up to 10 minutes (Roldheiser et al. (2012) cited by Muñoz & McDonald 2014).

5.9.4 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. Exposure to light, elevated temperatures, and reaction with organic compounds in the water accelerates the reduction in chlorine concentration. For this reason, it is important to monitor levels of ‘free available chlorine’ in the treated area, as often as every fifteen to thirty minutes initially.

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

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

5.9.5 Lime treatment

Active lime is an economical form of chemical treatment because it is produced in large quantities for commercial purposes, is relatively inexpensive, and only small quantities of active forms of lime are needed to treat benthic organisms. Environmental concerns are associated with broadscale application of lime, and its effects on marine species and the physical environment remain poorly understood. Deployment of quicklime (calcium oxide) either directly or using porous bags has successfully controlled seastars in Korea, the United States and Canada. Toxic effects of lime have been demonstrated against echinoderms and crustaceans, but molluscs and macroalgae are generally resistant. Direct contact with lime is required for a significant effect to be achieved. This is usually done by either covering a marine pest with quicklime or having an even distribution of the lime present on the seafloor for species such as *A. amurensis* to crawl over. Appropriate PPE is required for dispersal due to the caustic effects on active lime compounds.

5.9.6 Acetic acid

Acetic acid has low selectivity and is suitable for immersion and enhancing the effect of desiccation, wrapping and encapsulating. Immersion at 4% acetic acid (in sea water) for 1 minute removes soft-bodied fouling organisms from shellfish seed stock (Forrest, Hopkins & Gardner 2007). Effectiveness of acetic acid is dependent on concentration and immersion time. Low concentrations of acetic acid (4%) are equivalent to domestic vinegar and do not represent significant environmental or occupational risks if handled appropriately (Forrest, Hopkins & Gardner 2007). When treating aquaculture stock, it is important to understand the minimum time required to remove the marine pest and minimise stock mortality.

5.9.7 Copper sulphate

This treatment will be most suited to closed water ways, internal water systems and aquaculture equipment removed from the water. A trial of copper sulphate (Cu 1.5 mg/L) used in combination with chlorine in the infested Cullen Bay Marina, Darwin, resulted in 100% mortality of *M. sallei*. Copper sulphate powder was dissolved in a road construction watering truck tank and hosed over the water surface of the 'mixed' marina (McEnulty, Jones & Bax 2001). Copper sulphate's low specificity and persistence in the environment should be considered when weighing up treatment options. Copper sulphate can have environmental impacts and may be regulated by legislation or by the waterway managers. Copper may remain in the system and be reactivated when conditions permit, and even low concentrations can affect phytoplankton.

5.9.8 Direct chemical injection

Direct chemical injection involves injecting individual organisms with a biocide using a pole spear or standard agricultural gun. The method is taxon-specific, being effective against soft-bodied, sessile, or sedentary species and relies on divers to visually locate and treat individual organisms. It is only suitable for small outbreaks because of the relatively slow injection rate (100 to 140 injections per hour). Applying toxicants through a pole-spear has been effective in controlling the crown of thorns seastar, *Acanthaster planci*. Trials using drones and image recognition software have shown promise in automating this control method, increasing efficiency.

Chemicals identified as suitable for direct injection include:

- sodium bisulphate, breaks down in sea water and is inexpensive and safe to handle
- copper sulphate, recommended as a safe and easy toxicant with close to 100% kill rates for *A. planci*
- formalin (at different concentrations)
- hydrochloric acid
- ammonia.

5.10 Biological and ecological control

Biological and ecological control occurs by the manipulation of environmental conditions to create an adverse habitat for a species survival and reproduction. They may include the use of natural predators, competitors, parasites, or pathogens to suppress population growth. Biological and ecological control are not a rapid response operation as an extensive and lengthy review process

must occur before a biological control agent can be released into the environment. The introduction of non-native species or exotic disease to effect control is not advised due to the potential issues posed by these additional introductions, especially given that impacts are likely to be irreversible (Giakoumi et al. 2019). Promoting predation or herbivory by native species, or utilising endemic diseases, are more acceptable approaches but could still produce undesirable impacts, and their efficacy is unclear (Smith 2016).

6 Decontamination, destruction, and disposal

This section contains material summarised or adapted from the Aquatic Veterinary Emergency Plan ([AQUAVETPLAN](#)) manuals because of similarities in decontamination, destruction and disposal methods suitable for marine pests. This section is intended to be used in conjunction with the AQUAVETPLAN manuals which detail methods of disease control:

- decontamination ([AQUAVETPLAN – Operational Procedures Manual – Decontamination](#))
- destruction ([AQUAVETPLAN – Operational Procedures Manual – Destruction](#))
- disposal ([AQUAVETPLAN – Operational Procedures Manual – Disposal](#)).

See [Section 5](#) for methods that can assist with decontamination and destruction in addition to the above manuals.

6.1 Decontamination

Decontamination is the cleaning or treatment of material used to remove marine pests or render marine pests non-viable, including their propagules and any parasite and pathogen that can be associated with the marine pest species (Young et al. 2017). Some decontamination occurs *in situ* and no separate disposal activities occur. Other methods, such as most physical removal, require removal and capture of both sessile and motile marine pests and it is vital that destruction and disposal occur. Appropriate decontamination procedures are required to allow personnel, machinery, and equipment to move safely between locations during response operations.

The decontamination process comprises several stages (DAFF 2023c):

- planning:
 - identification and assessment of risks
 - design of efficient and effective procedures
 - training of personnel
- implementation:
 - cleaning
 - disinfection
 - waste treatment and disposal.

If decontamination is required, a plan should be developed considering the following information:

- the nature of the pest and how is it most effectively removed
- type of environment, material or equipment requiring decontamination
- water supply quality and quantity:
 - organic matter rapidly inactivates a number of chemical disinfectants

- available options for disinfection:
 - including disinfectant chemical compatibility if multiple agents are in use
- risks to the safety of personnel and equipment:
 - disinfectants can be corrosive, and most are irritants to people
- environmental pollution risks:
 - most disinfectants are toxic to aquatic life, although some are degraded quickly
- relevant legislation or regulations that must be complied with.

Effective cleaning is responsible for more than 90% of the success of decontamination. However, accumulations of soil, dirt, organic matter, or biofouling provide an effective barrier which may protect propagules or pathogens from disinfecting agents. Wash water may still contain viable propagules associated with the marine pest and must be disposed of appropriately. Effectiveness of cleaning compounds and disinfectants will depend on:

- water quality (such as suspended matter) and hardness
- concentration and contact time
- temperature and pH.

6.2 Destruction

Destruction occurs to aid in disposal of a captured marine pest or to control the spread of disease (in case of disease management) via methods employed during containment, eradication, or control of established population efforts. For example, destruction may be required after the collection of a vessel fouling material, aquaculture, stock or equipment. However, destruction of stock or equipment may not always be required since removal from water will ultimately result in death for the marine pest. The marine pest and aquaculture stock tolerance to desiccation should be considered prior to removal from the water. Treatment for closely related marine pest and stock species may result in the death of the stock. For example, a marine pest bivalve and cultured bivalve stock may have similar desiccation tolerance and be destroyed at the same time when removed from the water.

Marine pests may be destroyed *in situ*, but in some instances, organisms may be removed and destroyed elsewhere. The time for death to result is variable, and exposure to air will result in stress to the organism. The time between removal of the marine pest from the water and destruction should be as short as practically possible. This will minimise the organism's stress and the risk of escapes.

Where marine pests are removed and destroyed elsewhere, the site used for destruction should be contained to prevent release of the marine pest, viable propagules or pathogens and parasites. Ideally the destruction site should be close to the area from which pests are being removed, and/or to the disposal site. Appropriate disposal sites and methods should be identified prior to commencing destruction activities. Due to the volumes of fluid associated with destroying marine pests, surface or groundwater contamination and seepage back into marine environments must be managed carefully.

Destruction plans should consider (DAFF 2023d):

- if destruction will occur *in situ* or at another location
- the volume and type of marine pest to be destroyed
- how the marine pest will be contained until destruction
- any pathogens or parasites carried by the marine pest that will also need to be destroyed
- facilities and equipment available for destruction method
- appropriate destruction methods (see [Section 5](#) and [AQUAVETPLAN – Operational Procedures Manual – Destruction](#))
- potential environmental and human health impacts and any relevant legislation (e.g. chemical use and dead biomass)
- any required decontamination and disposal method
- any permits required by authorities for dealing with species listed in legislation.

Information pertaining to ethical concerns, which will depend partly on legislative and legal requirements of the jurisdictions involved, can be found in the following resources:

- [Australian Animal Welfare Strategy \(AAWS\) and National Implementation Plan 2010-14](#)
- [Australian code for the care and use of animals for scientific purposes](#).

6.3 Disposal

The primary reasons for disposing of a marine pest, their products, materials, and waste is to remove or deactivate the marine pest's reproductive, regenerative or disease transmission potential. Disposal should be completed as soon as possible after capture or destruction. Disposal has social, environmental, and aesthetic impacts that need to be considered.

Several considerations for a disposal plan for marine pest waste are summarised below (DAFF 2023e):

- selection of disposal site and transport to the disposal site
- method of disposal
- items that may require special consideration (e.g. liquid waste, control of scavengers)
- media and community communication.

Appropriate arrangements are required for the disposal of marine pest waste. A decision-making framework developed for identifying appropriate disposal has been developed ([AQUAVETPLAN – Operational Procedures Manual – Disposal](#)). In summary, an incident manager should consider the following:

- is the method consistent with international agreements and standards?
- are acceptable transport methods available?

- does the method meet legislative requirements, and can the necessary regulatory approvals be obtained?
- is the method consistent with industry standards and agreements?
- is the method cost-effective?
- how quickly will the method resolve the disposal problem?

Appendix A: Using the *Biosecurity Act 2015* during an emergency response

The following is an interim process for using the *Biosecurity Act 2015* for action on vessels to treat contaminations by a marine pest of national significance. The *Biosecurity Act 2015* 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 exclusive economic zone 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 2015* are:

- the Australian Government Department of Agriculture, Fisheries and Forestry is to be contacted before taking the proposed action to determine the appropriate provisions of the *Biosecurity Act 2015* that apply
- directions to take action under the *Biosecurity Act 2015* 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 2015* to be able to give directions under the Act
- actions under the *Biosecurity Act 2015* 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 2015* 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, Fisheries and Forestry to help determine appropriate application of the *Biosecurity Act 2015*:

- 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: Commonwealth, 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 [National Environmental Biosecurity Response Agreement 2.0](#) 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 principal fisheries management legislation to respond consistently and cost-effectively to a marine pest incursion.

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

Jurisdiction	Agency	Principle fisheries management acts covering emergency response arrangements	Marine pest contact website
Commonwealth	Department of Agriculture, Fisheries and Forestry	<i>Biosecurity Act 2015</i> <i>Fisheries Management Act 1991</i>	marinepests.gov.au agriculture.gov.au/biosecurity-trade/pests-diseases-weeds/marine-pests
New South Wales	Department of Primary Industries	<i>NSW Biosecurity Order (Permitted Activities) 2019</i> <i>NSW Biosecurity Regulation 2017</i> <i>NSW Biosecurity Act 2015</i> <i>Fisheries Management (General) Biosecurity Regulation 2017</i> <i>Fisheries Management (Aquaculture) Regulation 2012</i> <i>Fisheries Management Act 1995</i> <i>Marine Safety Act 1998</i> <i>Marine Parks Regulation 1997</i> <i>Ports and Maritime Administration Act 1995</i>	dpi.nsw.gov.au/fishing/pests-diseases
Victoria	Department of Energy, Environment and Climate Action	<i>Marine and Coastal Act 2018</i> <i>Marine Safety Act 2010</i> <i>Fisheries Act 1995</i> <i>Port Management Act 1995</i> <i>Environment Protection Act 1970</i>	vic.gov.au/marine-pests
Queensland	Department of Agriculture and Fisheries	<i>Biosecurity Act 2014</i> <i>Fisheries Act 1994</i>	daff.qld.gov.au/fisheries/ qld.gov.au/environment/coasts-waterways/marine-pests

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Jurisdiction	Agency	Principle fisheries management acts covering emergency response arrangements	Marine pest contact website
South Australia	Department of Primary Industries and Regions	<i>Fisheries Management Act 2007</i>	pir.sa.gov.au/biosecurity/aquatics
Western Australia	Department of Industry and Regional Development	<i>Biosecurity and Agriculture Management Act 2007</i> <i>Fish Resources Management Act 1994</i>	fish.wa.gov.au/Sustainability-and-Environment/Aquatic-Biosecurity/Pages/default.aspx
Tasmania	Department of Natural Resources and Environment Tasmania	<i>Biosecurity Act 2019</i> <i>Living Marine Resources Management Act 1995</i>	nre.tas.gov.au/biosecurity-tasmania/aquatic-pests-and-diseases
Northern Territory	Department of Industry, Tourism and Trade	<i>Fisheries Act 1988</i>	nt.gov.au/marine/for-all-harbour-and-boat-users/biosecurity/aquatic-pests-marine-and-freshwater

Appendix C: Total mortality treatments for marine pests of national significance

Table C1 Treatments that achieved 100 per cent mortality of marine pests in laboratory conditions

Organism	Species	Treatment	Conditions to achieve LD100	Reference
Macroalgae: sporophytes and gametophytes	<i>Undaria pinnatifida</i>	Freshwater immersion	8 hours at 18 °C	(Forrest & Blakemore 2006)
			10 mins at 35 °C	(Gunthorpe et al. 2001)
			45 secs at 45 °C	
			05 secs at 55 °C	
		Acetic acid (4%)	1 min. at 4% in fresh water	(Forrest & Blakemore 2006) (Forrest, Hopkins & Gardner 2007)
		Desiccation	3 days at 10 °C (55 – 85% humidity)	(Forrest & Blakemore 2006)
			1 day at 20 °C (55 – 85% humidity)	
		8 weeks at 10 °C (over 95% humidity)		
		6 weeks at 20 °C (over 95% humidity)		
		Bleach solution ^a	1 hour at 2% concentration	(Gunthorpe et al. 2001)
		Detergent (DECON 90) ^b	>30 mins at 2% concentration, over 18 °C	(Gunthorpe et al. 2001)
Crabs and other decapod crustaceans	<i>Carcinus maenas</i>	Bleach solution ^a	4 hours at 2% concentration	(Gunthorpe et al. 2001)
			Detergent (DECON 90) ^b	greater than 8 hours at 2% solution, over 18 °C
	<i>Charybdis japonica</i>	-	refer NIMPIS ^d for updated information	nimpis.marinepests.gov.au/species/species/108
	<i>Eriocheir sinensis</i>	Ammonia	48 hours at 3.12 ± 0.06 mg/L (zoea-I) 24 hours at 3.47 ± 0.36 mg/L (zoea-II) 24 hours at 6.92 ± 0.32 mg/L (juvenile)	(Zhao et al. 1997)

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Organism	Species	Treatment	Conditions to achieve LD100	Reference
	<i>Rhithropanopeus harrisi</i>	Diflubezuron (active chemical in the pesticide Dimilin)	~11 days at 10 ppb (hatch to megalopa)	(Christiansen, Costlow & Monroe 1978)
Bivalve molluscs	<i>Mytilopsis sallei</i>	Water temperature	120 mins at 40, 30 mins at 50 °C 30 mins at 60 °C	(Bax et al. 2002)
		Copper sulphate	38 hours at 1 mg/L	(Bax et al. 2002)
		Chlorine	111 hours at 12 mg/L chlorine 90 hours at 24 mg/L chlorine	(Bax et al. 2002)
		Chlorine/copper sulphate solution	48 hours at 12 mg/L chlorine, followed by 48 hours at 1 mg/L copper	(Bax et al. 2002)
	<i>Perna viridis</i>	Water temperature	5 hours at 39 °C 30 mins at 60 °C	(Azanza, Azanza & Ventura 2005) (Rajagopal et al. 2003c)
		Chlorine	48 hours at 10 – 15 mg/L chlorine	(Rajagopal et al. 2003a)
	<i>Perna canaliculus</i>	-	refer NIMPIS ^d for updated information	Species (marinepests.gov.au)
	<i>Perna perna</i>	Desiccation	for relative humidity <5%, 33%, 53%, 75% and >95%: 8.50 – 18.50 days (range) at 15 °C 4.75 – 7.25 days (range) at 25 °C 20 – 48 hours (range) at 35 °C	(Hicks & McMahon 2003)
		Chlorine	3.5 days at 5 mg/L concentration (9.6 ± 0.3 mm shell length) 4.5 days at 5 mg/L concentration (25.4 ± 0.9 mm shell length) 5 days at 5 mg/L concentration (34.1 ± 1.8 mm shell length)	(Rajagopal et al. 2003b)
	<i>Mytella strigata</i>	Chlorine	24 hours at 3 mg/L concentration (D-larvae) 24 hours 15 mg/L concentration (pediveliger larvae)	(Lim et al. 2020) (Lim et al. 2020)
			refer NIMPIS ^d for updated information	nimpis.marinepests.gov.au/species/species/146
Seastars		Bleach solution ^a	1 hour at 2% concentration	(Gunthorpe et al. 2001)

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Organism	Species	Treatment	Conditions to achieve LD100	Reference
	<i>Asterias amurensis</i>	Detergent (DECON 90) ^b	more than 2 hours at over 18 °C	(Gunthorpe et al. 2001)
		Quicklime	2 weeks	(Goggin 1998)
		Freshwater immersion	greater than 2 hours immersion at over 18 °C	(Gunthorpe et al. 2001)
Ascidians	<i>Didemnum vexillum</i>	Chlorine (sodium hypochlorite)	30 secs at 0.5% 2 mins at 0.25%	(Denny 2008)
		Caustic soda (sodium hydroxide)	20 secs at 6%	(Denny 2008)
		Desiccation	48 hours at ambient temperature after water blasting	(Coutts and Forrest 2007)

a Active ingredient 3% sodium hypochlorite. **b** Active ingredient potassium hydroxide at less than 3%. **c** Total mortality treatment applied in field. **d** [National Introduced Marine Pest Information System](#) (NIMPIS).

LD100: lethal dose for 100% mortality of all treated individuals.

Appendix D: Marine pest management options

Table D1 Physical removal options for marine pest eradication and control in and on natural and artificial substrates

Method	Efficacy and environmental circumstances										Target taxa (researched using treatment method)	Comments
	Subtidal reef	Subtidal soft sediment	Intertidal reef	Intertidal soft sediment	Vessel hulls and niche areas	Wharf piles, pontoons	Aquaculture equipment	In situ removal	Wave energy limitations	Depth restrictions		
Diving, ROVs	L	U	L	L	L	L	L	Yes	None – low energy	<30 m	Various, including echinoderms, crustaceans, molluscs, macroalgae ^a	Successful in reducing abundance; unlikely to achieve eradication exc. for small incursions
Dredging, trawling, mopping	U	L	U	L	U	U	U	Yes	None	No	Various, including echinoderms, molluscs ^a	Successful in reducing abundance; unlikely to achieve eradication
Trapping (baited)	L	L	L	L	-	L	-	Yes	None – low energy	No	Crustaceans ^b , echinoderms ^c	May be successful in reducing abundance; unlikely to achieve eradication
Trapping (unbaited, pitfall, crab condos)	L	L	L	L	-	L	-	Yes	None – low energy	No	Crustaceans ^{d,e}	May be successful in reducing abundance; unlikely to achieve eradication; suitable for catching different life-stages
Vacuum/suction (diver dredge)	L	L	U	U	P	L	L	Yes	Low energy	<30 m	<i>Didemnum vexillum</i> ^{f,m} , <i>Caulerpa taxifolia</i> ^g	Successful in reducing abundance; efficacy reduced on natural substrates
Vacuum/suction with cutting head	U	NS	U	U	-	-	-	Yes	Low energy	<30 m	<i>D. vexillum</i> ^f	Cutting head was not successful
Vacuum/suction with rotational brush (confidential report)	U	U	U	U	L	L	L	Yes	Low energy	<30 m	Various fouling taxa ^h	Preliminary results suggest fouling abundance reduced; eradication not likely

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Dredge spoil*	U	P	U	P	–	–	–	Yes	Low energy	No	<i>D. vexillum</i> ^a	Successful method; successful for gently sloping and soft sediment seabeds
High-pressure water blasting	U	U	U	U	P	L	P	Possible	Low energy	<30 m	<i>D. vexillum</i> ^k , <i>Undaria pinnatifida</i> ^l , various fouling taxa ^l	Successful for structures removed from water; unlikely to be successful for <i>in situ</i> operations
UV light treatment	–	–	–	–	P	L	L	Yes	Low energy	Yes	Macroalgae, biofouling settlement ^{l,j}	Pilot scale success on osmosis membranes and vessel hulls; more testing in a marine pest context required
Hot water baths/sterilisation	–	–	–	–	P	–	P	No	None – low energy	–	<i>U. pinnatifida</i> ^k , crustaceans ⁿ , various fouling taxa ^o	Successful method; includes internal seawater systems
Hot water box	–	–	–	–	P	L	–	Yes	No	<30 m	<i>U. pinnatifida</i> ^p , bivalves ^o , various fouling taxa ^o	Successful method for vessel hulls; requires development for natural substrates
Steam sterilisation	–	–	–	–	P	L	–	Yes	No	<30 m	<i>U. pinnatifida</i> ^q , various fouling taxa ^o	Partially effective; can only treat very small areas; includes internal seawater systems
Wrapping/encapsulation (inc. PVC, matting)	–	–	–	–	P	P	P	Yes	Low energy	<30 m	<i>Styela clava</i> ^m , <i>D. vexillum</i> ^{h,r} , <i>C. taxifolia</i> ^s , <i>Mytilopsis sallei</i> ^t , <i>Spartina anglica</i> ^u , various fouling taxa ^{h,m,r}	Successful method: mortality of pest species can be accelerated via chemical application; successful if integrity of smothering structure maintained
Containment/chemical treatment	–	–	–	–	P	P	–	Yes	Low energy		<i>C. taxifolia</i> ^v	Successful method
Wrapping plus chemicals	–	–	–	–	P	P	–	Yes	Low energy		<i>D. vexillum</i> ^f , <i>S. clava</i> ^m various fouling taxa ^m	Successful method

Letters relate to the efficacy of the treatment methods: **P** proven. **L** likely. **U** unlikely. **NS** not successful. **a** McEnnulty, Jones & Bax (2001). **b** Mabin et al. (2020). **c** Browne & Jones (2006a, b). **d** Riipinen et al. (2017). **e** Fowler et al. (2013). **f** Coutts (2002). **g** J Gilliland, PIRSA, pers. comm. (2007). **h** Hopkins (2006). **i** Hunsucker et al. (2019). **j** Braga et al. 2020. **k** Coutts (2006). **l** Forrest & Blakemore (2006). **m** Coutts & Forrest (2007). **n** Growcott, Kluza & Georgiades (2016). **o** Cahill et al. (2021). **p** Wotton et al. (2004). **q** Stuart (2004). **r** Pannell & Coutts (2007). **s** Glasby, Creese & Gibson (2005). **t** Creese, Davis & Glasby (2004). **u** Hammond & Cooper (2001). **v** Anderson (2005).

Note: For details about suitability of methodologies against a broader range of taxa, see McEnnulty et al. (2001). *Dredge spoil is classified under biological and ecological control methods but listed under physical removal options for managers focused on natural substrates.
Adapted from Aquenal 2007.

Table D2 Chemical control options for marine pest eradication and control in and on natural and artificial substrates

Method	Efficacy and environmental circumstances					<i>In situ</i> removal	Wave energy limitations	Depth restrictions	Target taxa (researched using treatment method)	Comments
	Vessel hulls and niche areas	Wharf piles, pontoons	Enclosed systems	Aquaculture equipment						
Desiccation/water level manipulation	P	L	L	P	No	–	–	Various, including <i>Styela clava</i> ^b , <i>Didemnum vexillum</i> ^c and <i>Undaria pinnatifida</i> ⁱ	Successful method for removable structures; drawdown restricted to enclosed systems	
Freshwater	P	U	P	P	Yes	Low energy	No	<i>Caulerpa taxifolia</i> ^k , various fouling taxa ^g	Successful provided organisms can be contained	
Freshwater baths	–	–	P	–	No	–	–	<i>U. pinnatifida</i> ⁱ , <i>Polydora</i> spp. ^h , <i>Ciona intestinalis</i> ^h	Successful method	
Salt treatment	U	U	L	U	Yes	Low energy	<30 m	<i>C. taxifolia</i> ^{l,m} , crustaceans ^g , <i>Asterias</i> spp., boring sponges, ascidians, hydroids and slipper limpets ^g	Suitable for control; unlikely to be successful for eradication	
Lime	U	U	L	–	Yes	Low energy	Yes	Echinoderms ^a	Successful if applied as a blanket on the substrate	
Chlorine	L	U	P	L	Yes	Low energy	Yes	Soft-bodied fouling organisms ^{f,g} including, <i>D. vexillum</i> and <i>S. clava</i> <i>U. pinnatifida</i> , <i>Sabella spallanzanii</i> ^{g,j,h} and <i>Mytilopsis sallei</i> ⁱ	Success increases with application to another method, e.g. wrapping or out of water bath	
Acetic acid	L	U	L	P	Yes	Low energy	Yes	Soft-bodied fouling organisms, dinoflagellates ^h , <i>C. taxifolia</i> ^h , settlement growth ^{g,h} , <i>D. vexillum</i> ^h , <i>S. clava</i> ^h ,	Successful for dipping of aquaculture stock; Success increases with application to another method, e.g. wrapping	
De-oxygenation (nitrogen gas)	L	L	L	L	No	–	–	Various taxa ^e	Variable success: depending on species and life history stage concerned	
Injection	U	U	–	–	Yes	No	Yes	<i>Acanthaster planci</i> ^d	Successful for small outbreaks	

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Letters relate to the efficacy of the treatment methods: **P** proven. **L** likely. **U** unlikely. **NS** not successful. **a** Woods et al. (2007). **b** Coutts & Forrest (2005). **c** Pannell & Coutts (2007). **d** Fisk & Power (1999). **e** Tamburri et al. (2002). **f** Forrest, Hopkins & Gardner (2007). **g** [National Introduced Marine Pest Information System](#) (NIMPIS), **h** Cahill et al. (2021). **i** Bax et al. (2002). **j** Coutts (2006). **k** Neverauskas & Jordan (2004). **l** Glasby, Creese & Gibson. (2005). **m** Wiltshire & Deveney (2017).

Note: For details about suitability of methodologies against a broader range of taxa, see McEnnulty et al. (2001).

Adapted from Aquenal 2007.

Table D3 Other options for marine pest eradication and control in and on natural and artificial substrates

Method	Efficacy and environmental circumstances							Target taxa (researched using treatment method)	Comments
	Vessel hulls and niche areas	Wharf piles, pontoons	Enclosed systems	Aquaculture equipment	<i>In situ</i> removal	Wave energy limitations	Depth restrictions		
Pulsed electric fields	U	U	U	U	No	–	–	Various invertebrate larvae ^{a,h}	Prevented settlement of invertebrate larvae; unlikely to be suitable for eradication
Electrolysis (chlorine production)	U	U	L	L	No	–	–	Various invertebrate larvae ^{b,h}	Prevents biofouling of artificial structures
Ozone treatment	U	U	L	L	No	–	–	Planktonic organisms ^c <i>Dreissena polymorpha</i> larvae ^d	Successful in laboratory trials against a range of planktonic organisms
Acoustic methods	U	U	U	U	No	–	–	Various invertebrate larvae ^e	These methods have shown some promise for preventing settlement of invertebrate larvae, but unlikely to be useful for eradication purposes
Electromagnetic control (including visible light, radio waves, microwaves)	U	U	U	U	No	–	–	Various invertebrate larvae ^{f,h}	
Magnetic control	U	U	U	U	No	–	–	<i>Dreissena polymorpha</i> larvae ^g	

Letters relate to the efficacy of the treatment methods: **P** Proven. **L** Likely. **U** Unlikely. **NS** Not successful. **a** Schoenbach et al. (2002). **b** Black & Veatch Corporation (2010). **c** Perrins et al. (2006). **d** Boelman et al. (1997). **e** Brizzolara et al. (2003). **f** Morgan et al. (1999). **g** Smythe et al. (1996), **h** [National Introduced Marine Pest Information System \(NIMPIS\)](#)

Note: For details about suitability of methodologies against a broader range of taxa, see McEnnulty et al. (2001).

Adapted from Aquenal 2007.

Appendix E: Specimen preservation and handling

This appendix provides general and taxa-specific specimen-handling techniques. Table E1 is a summary of the preferred and optional narcotising (relaxing) and fixing agents. Information is sourced from protocols for specimen preservation detailed by Hewitt & Martin (2001).

This appendix compliments the [Australian marine pest monitoring manual](#), which contains field guides for sampling techniques and processes for sample collection. The manual has sample handling and preferred narcotising, fixation and preservation techniques for major marine taxonomic groups. It also gives advice on appropriate levels of experience required for sample processing.

Absolute ethanol is the preferred 'all-purpose' preservative for molecular testing. Absolute ethanol must be used, denatured ethanol is not suitable. Formalin may be the preferred fixative for specimens requiring histology for identification, however it can alter nucleic acid and protein integrity making it unsuitable as a preservative for molecular testing.

Only adequately trained staff should handle samples that contain dangerous goods or hazardous substances. Dangerous goods and hazardous substances must be packed by appropriately trained and accredited personnel, or by approved couriers. The Australian Government is responsible for regulating the transport of dangerous goods by air through the [Civil Aviation Safety Authority](#) and by sea through the [Australian Maritime Safety Authority](#). The Australian states and territories are responsible for both road and rail transport of dangerous goods (DITRDCA, 2024). A list of relevant competent authorities for dangerous goods is located on the [Competent authorities for dangerous goods](#) website.

General techniques

A waterproof label containing collection details should be placed inside the collection container(s) as soon as the specimen is collected. In some cases, printing may fall off waterproof paper used for labels; pencil is preferred as some inks are soluble in fixatives. In most climates, biological and sediment samples should be placed on ice or transported to a laboratory for sorting and preservation. In all instances, material should be narcotised and preserved, as soon as practically possible, but within eight hours of collection. Narcotising is essential for some invertebrates as they must be relaxed before fixation. Narcotising and preservation agents are frequently carcinogenic, so a Safety Data Sheet (previously called a Material Safety Data Sheet) should be made available to everyone participating in specimen narcotising and preservation.

General guidelines for specimen handling include:

- any material that may be needed for DNA analysis must be either frozen or fixed in 100% ethanol. Collect both sample types if necessary
- when freezing to relax or store specimens, do not thaw and re-freeze them. Defrost once, photograph if necessary, and then fix in preservative

- all references to formalin in these guidelines mean formalin stock diluted 1:9 with sea water. Formalin stock is formalin with propylene glycol (propane-1-2-diol) mixed 1:1
- mix ethanol with deionised water to avoid precipitates
- the volume of the specimen must be included as part of, not additional to, the water volume when making up solutions. This is particularly important for large specimens or those with large water content (such as ascidians, cnidarians and sponges). Failure to include specimen volume will result in the solution being too weak
- always completely submerge specimens in preservative and make sure the specimen is not too big for the jar. If squashed into jars, specimens will distort and, more importantly, will probably not fix properly and may start to decompose
- preserving solutions (both formalin and ethanol) used to fix material rapidly become very acidic. If material cannot be processed promptly on return from the field, it is advisable to change the preserving solutions to avoid acidity problems. No material should remain in its initial fixing solution for more than one month
- sort specimens and group them according to fixing requirements. Do not mix hard and soft animals; some fragile specimens may be damaged or destroyed
- sort soft-bodied animals or unique specimens directly into individual specimen jars
- put labels inside a small plastic bag inside the sample bag or jar. The small plastic bag protects the label from chafing, discolouration, consumption or destruction by live animals or other physical damage from specimens during transport and storage. If an outside label is needed, it must be additional to that inside the jar. With very large specimens, attach the label directly to the specimen as well as attaching one on the outside of the bag
- it is important to cross-reference any photographs to the actual specimen photographed. Make sure field labels record this. It is usually best for the person who took the photos to collect the specimens and do the sorting, both in the field and in the laboratory
- material fixed properly in formalin or ethanol can be transported damp, without liquid, if it is in a sealed container. This can greatly reduce weight for transport. Preservative should be replaced as soon as practicable. Delicate specimens and ethanol specimens must have some liquid around them when transported, but the volume can be reduced. As a contingency for transport delay, ethanol specimens should have some liquid with them, otherwise they may dry out quickly, even in a sealed container.

Preservation techniques for specific taxa

Many soft-bodied animals such as ascidians and anemones require narcotising before preservation. Narcotising effectively relaxes the animal, preventing the innate defensive mechanisms induced by the shock of placing the animal in preservative.

While formalin may be the preferred fixative for specimens requiring histology for identification, where possible some additional material should be fixed in 100% ethanol to allow for planned or future identification using molecular techniques. Consult a taxonomist or laboratory expertise on requirements as histology is now only used on some taxa; ethanol allows use for taxonomic purposes

of many marine pests and is essential for molecular techniques which are sometimes the key differentiator.

Menthol crystals will not dissolve rapidly but can be floated on seawater until the animal does not respond to touch. Overnight refrigeration at 4°C can assist relaxation in some taxa.

Anemones

Photograph and relax live specimen before fixing if possible. Put in jar with enough seawater to allow the specimen to fully expand, then freeze or add menthol or magnesium chloride and leave overnight. Fix in formalin by adding the correct amount of stock formalin to the frozen specimen, making sure it mixes as it defrosts. Store in formalin.

Aplacophora

Best if relaxed first, usually with menthol, magnesium chloride or iced water, then fix in formalin, rinse in water and store in 70% ethanol. Do not leave in formalin for more than a few days, or the spicules will start to dissolve.

Asteroids

Photograph alive if possible. Place live into a dish of sufficient concentrated formalin (mix stock 1:5 with seawater) to cover the seastar and leave overnight. Make sure that seastars are not distorted before they are put into the fixative. Remove specimens from fixative, place on paper towel and dry in shade. Ensure specimens do not stick to the paper by moving them around regularly (keep their labels with them). When specimens start to change to a pale cream/yellow/orange, put them in a plastic bag with their label. In the laboratory, dry specimens in a microwave oven on high for 30 seconds to 1 minute; cool for a while then repeat until no more moisture is released. Beware of putting seastars with too much moisture in the microwave as they can explode.

‘Cooking’ seastars in the microwave will cause them to give off vaporised formalin. Only do this in a well-ventilated area, and in a microwave oven that is not used for food preparation. Store dry. Alternatively, fix in formalin for 24–48 hours and transfer to 70% ethanol for long-term storage. The latter method will preserve the colour in most specimens.

Bivalves

For species with valves that seal tightly, place a matchstick or similar object between valves before fixation to ensure the fixative can reach and penetrate internal tissues. To get bivalves to gape, either warm until they relax enough, or freeze them. Samples that are intended to be sorted visually should be preserved in 4% buffered formaldehyde (formalin) immediately after collection and should be stored in formalin. However, species with very thin shells should be stored in 70% ethanol.

Samples that will be analysed using the molecular probe should not be put into formalin, as formalin vapor can deteriorate the DNA. Instead, these samples should be rinsed into sample jars with SET-buffered, reagent-grade ethanol (usually 70% or 90%), ensuring that the ratio of biomass to SET buffered ethanol is no more than 1 to 3 (MPSC 2024).

Brachiopods

Fix and store in formalin for histology or fix in 70% ethanol and preserve in 70% ethanol. To allow best penetration of the fixative it helps to wedge open the valves with a matchstick or similar object.

Many species will clamp shut so tightly that this becomes impossible. Attempt similar relaxation technique to bivalves.

Cephalopods

Photograph alive, showing different colour patterns if possible. If live-caught, animals must first be anaesthetised as part of a two-step euthanasia process. Immersion in magnesium chloride ($MgCl_2$) to achieve an anaesthetic overdose, followed by immersion in 10% buffered formalin (or 70% ethanol) to ensure physical destruction of the brain, is currently considered the most humane method for euthanising cephalopods. Use a solution of 75 g of $MgCl_2$ in 1 L of sea water. For tissue preservation, place sample in 70% ethanol or 80% ethanol. Replace with a fresh 70% solution after a day or so, to minimise dilution from tissue water. If possible after collection, pour ethanol off and freeze the sample at $-80\text{ }^\circ\text{C}$ until required for analyses. Fix in formalin, arranging the arms and tentacles so they are straight, and the specimen is not distorted. It may be necessary to use weights or pins to hold the specimen in place. Enough fixative, preferably 10 times the volume of the specimen, should be used to cover the specimens completely. Specimens should be stored in 5–10% buffered formalin (one-part concentrated formalin and nine parts seawater) for at least three days. Rinse specimens in tap water and store the specimen in 70% ethanol. For cuttlefish, carefully remove the bone before fixation and store with the specimen after photographing intact animal (it is much simpler to remove the bone without breakage before fixation).

Cnidarian medusa

Photograph alive and relaxed specimens before fixing if possible. Put in a jar with enough seawater to allow the specimen to expand fully, then freeze or add menthol or magnesium chloride and leave overnight. Fix in formalin; do not freeze; store in formalin.

Crinoids

Photograph alive if possible. Fix in formalin, but not for more than two or three days. Store in 70% ethanol. Few species do not fall apart when preserved. Try to keep all fragments together and be aware that crinoids usually carry commensal organisms.

Crustaceans

Photograph specimens alive, if possible, particularly shrimps. For commensal species, it is important to also record and, if possible, collect the host. Do not freeze crustaceans unless there is no other option, as they do not fix as well after they have been frozen. Specimens are best fixed alive. Remove hermit crabs from their shells and tube-dwelling species from their tubes before fixing (keep any tubes or shells). Commensal organisms are often associated with hermit crabs and tube-dwelling species; these may need to be fixed differently to their hosts. If hermit crabs have anemones on their shells, remove the crabs and treat the anemones as detailed above. Avoid putting specimens with chelipeds in with other animals, as they may grab and damage more fragile species. It is sometimes preferable to kill large crabs individually and put them into a communal container to fix. Fix and store in 70% ethanol. Use formalin fixation if the specimen is required for histological examination. Very large specimens may need to be injected with formalin to ensure sufficient fixative reaches internal tissues. Alternatively, the carapace may be lifted to permit entry of different fixatives into the body. Some glycerine added to the fixative may help with flexibility of alcohol fixed specimens. Relaxation by refrigeration, then placing the relaxed individuals into cold fixative reduces shock and reaction to fixative.

Ctenophores (comb jellies)

Most species are virtually impossible to preserve. It is essential that good, detailed photographs and video (if possible) are taken of all specimens. A few of the more solid species, such as *Beroe* spp., and all benthic ctenophores, can be fixed in formalin, and stored in formalin or 70% ethanol. To fix benthic ctenophores flat, the methods used for platyhelminths can be successful. No matter what fixative or narcotising agent is used, most species of ctenophores simply disintegrate within minutes of being preserved, but research suggests that when preserved in 2% acidic Lugol's solution, samples of the invasive ctenophore, *Mnemiopsis leidyi*, stayed intact and were quite stable even after preservation for 105 days (Engell-Sørensen et al. 2009).

Echinoids

Treat large specimens and species with large spines as for asteroids. Place live specimens in a dish and pour preservative over them until the spines stop moving (all spines should be erect). When specimens are removed for drying, puncture the membrane surrounding the teeth with a needle to allow liquid within the test (shell) to drain out. Beware of putting echinoids in the microwave as they can explode. Fix and store in 70% ethanol. Fix smaller specimens in formalin and store in 70% ethanol.

Echiuran worms

Relax and preserve specimens as for sipunculan worms. Do not freeze, as specimens will disintegrate. In some species, the proboscis is deciduous, and usually breaks off entirely or partially; make sure it is retained. To facilitate later dissection, it can be advantageous to keep echiurans alive in clean sea water for some hours before fixing, to allow them to void sand in the gut. Echiurans exude a chemical toxic to most other animals; beware of this if putting them in containers with other invertebrates when collecting. Fix in formalin and store in formalin or 70% ethanol.

Ectoprocts

Photograph alive, if possible, as living colours can be useful identification features. Fix hard species in formalin if possible (not essential) then dry; store dried. Fix soft and lightly calcified species in formalin but do not leave for more than a few days (4 to 12 hours is best). Store in 70% ethanol. In the field, either fix specimens in formalin overnight and transfer to ethanol in the morning or fix directly in ethanol.

Holothurians

Photograph alive if possible. Always isolate large specimens when collecting, as they often eject their guts when disturbed; tubules tend to adhere to everything they come in contact with. Fix in formalin overnight, then rinse thoroughly in water or fix in 100% ethanol. Store in 70% ethanol. It is important that holothurians are not left in formalin too long, and are thoroughly rinsed when removed from it, or their skeletal plates will dissolve. These plates are essential for subsequent identification.

Leeches

Specimens must be relaxed before fixing. Use either menthol in sea water or iced sea water overnight, but do not freeze. Shark and ray leeches can be relaxed by submerging specimens in fresh water for a few hours. Transfer to fixative as soon as they stop moving or they will start to rot. Fix in formalin and preserve in 70% ethanol.

Molluscs (general)

Most molluscs can be put straight into formalin to fix and are usually preserved in ethanol. Fix and store in ethanol for molecular testing.

Nemertean worms

Photograph alive, if possible, as the colour patterns are distinctive, then relax and preserve as for sipunculan worms. Freezing does not work particularly well with these worms. Nemerteans will often break into pieces when fixed but can still be identified, so all fragments should be kept. Like echiurans, some species of nemerteans exude a toxic chemical and are best kept separate during collecting. Fix in formalin and store in formalin or 70% ethanol.

Oligochaete worms

Relax and preserve as for sipunculan worms. Photographs of live specimens can be used for subsequent identification. Fix in formalin and store in formalin or 70% ethanol.

Ophiuroids

Photograph alive if possible. Large and solid specimens should be treated as for asteroids. Fix all other specimens in formalin and store in 70% ethanol. Be aware that most species will drop arms. Specimens left in formalin for too long become fragile.

Opisthobranchs (and other reduced-shell gastropods)

Specimens must be photographed alive, as form and colour pattern are very important diagnostic features. If possible, record substrate collected from as this may be an indication of food source. Specimens must be relaxed before fixing. The best method for relaxing is to put specimen in a jar with enough seawater for it to move around with rhinophores and gills fully extended, then freeze overnight. Add enough stock formalin to frozen jar to make up solution of appropriate strength, and make sure it is mixed as the seawater thaws. If freezing (usually the most effective method) is impractical, use menthol. Magnesium chloride in seawater or iced seawater, overnight will relax specimens. Fix in formalin. Do not leave specimens in formalin for more than one or two weeks, and if possible, only for about 12 hours. Prolonged exposure to formalin will dissolve the mantle spicules or vestigial shell. Store in 70% ethanol.

Platyhelminths

If possible, specimens should be photographed alive. It is important that they are preserved as flat as possible. They can be relaxed using menthol or magnesium chloride overnight, but this is not always successful, and specimens often disintegrate. The best method is to freeze a small amount of formalin stock in a jar, then place the specimen on top. It will freeze onto the surface of the formalin, die flat and be fixed at the same time. Add the appropriate amount of seawater to make up the solution. If no other option is available, fix directly in formalin on ice and can be preserved in 70% ethanol.

Polychaete worms

Specimens can usually be fixed directly in formalin; some larger species may need to be relaxed using menthol or magnesium chloride before fixing. Try to remove tube-dwelling species from their tube to allow proper fixation, but always retain the tubes. This is particularly important with serpulid worms. Many species will fragment when fixed; all fragments should be retained. Fix in formalin and store in

formalin or 70% ethanol. In the case of species with calcareous tubes, transfer from formalin to 70% ethanol within 24 hours of fixing.

Polyplacophora

Specimens curl up when removed from their substrate. Specimens should be put onto a flat surface (such as a glass slide or wooden board) and tied flat using cotton tape. Fix in formalin, then untie and store in formalin. Store very small and deep-sea species in 70% ethanol.

Sipunculan worms

If possible, relax specimens before fixing so the proboscis is everted. This is best done with menthol or magnesium chloride in seawater overnight. Freezing does not work particularly well for sipunculans. Fix in formalin and store in formalin. Dead gastropod shells often contain sipunculans; check contents before discarding any shells.

Soft corals (octocorals)

If possible, photograph live and relax specimens before fixing. Put in a jar with enough seawater to allow the specimen to expand fully, then freeze or add menthol or magnesium chloride. Leave until relaxed, fix in formalin for up to 12 hours (two to four hours is best). Rinse thoroughly in water, then store in 70% ethanol. If any formalin remains, or the animal is left in formalin too long, the spicules will start to dissolve, and the specimen will become almost impossible to identify. Fix delicate species directly in 100% ethanol; store in 70% ethanol.

Sponges

Photograph live specimens *in situ*, if possible, to record colours and form. Some species will disintegrate when handled. In the field, freeze specimens, if possible, then fix in the laboratory. If this is not possible, use these procedures to preserve specimen, but do not leave material in formalin for more than 24 hours (8 to 12 hours is best). Fix in either 100% ethanol or in well buffered formalin overnight. Formalin is a better fixative but sponges must be thoroughly rinsed in water to remove formalin before being stored in 70% ethanol. If any formalin remains, or the sponge is left in formalin too long the spicules will start to dissolve and the specimen will become almost impossible to identify. For small or very delicate sponges, fix in 100% ethanol if possible. If formalin is used, do not leave them in formalin for more than two to three hours and rinse in water very thoroughly; store in 70% ethanol.

Tunicates (Urochordates)

Compound, colonial and other gelatinous ascidians must be photographed alive as form and colour patterns are very important diagnostic features. Photograph any other ascidians alive if possible. All ascidians should be relaxed before fixing; menthol or magnesium chloride in sea water overnight is usually effective; prod the ascidian to watch for any reaction to ensure it is relaxed before fixation. They may also need to have preservative injected into them to ensure adequate fixation. Fix in formalin or 100% ethanol. Store in formalin or 100% ethanol. Keep cool on ice or refrigerated before fixation. Formalin fixed material is essential for morphological identification, ethanol fixed material for molecular testing.

Table E1 Summary of recommended narcotising and fixation techniques

Phylum	Taxa	Photos needed	Narcotising agents							Fixatives		Notes
			None	Fresh water	Chill or freeze	Menthol	Naphthalene	MgCl ₂	70% ethanol	7–10% formalin	Formalin to ethanol	
Annelida	Leeches	No	–	Pref.	Alt.	Alt.	Alt.	Alt.	–	Pref.	–	Relax before fixing
	Polychaetes and oligochaetes	Yes	–	–	–	Alt.	Alt.	Pref.	Alt.	–	Pref.	–
Arthropoda	All	No	Pref.	–	–	–	–	–	Pref.	–	–	Do not freeze
	Barnacles	No	–	Pref.	–	–	–	–	–	Alt.	Pref.	–
	Pycnogonids	No	Pref.	–	–	–	–	–	Pref.	Alt.	–	–
Brachiopoda	All	No	Pref.	Alt.	Alt.	–	–	–	Pref.	Alt.	Alt.	Wedge valves open to allow formalin entry
Chordata	Pisces	Yes	Alt.	–	–	–	–	–	–	Pref.	–	Inject fixative into body cavity of larger specimens
	Urochordates	Yes	Alt.	–	–	Alt.	–	Alt.	–	Alt.	Pref.	Inject fixative into body cavity of larger specimens
Cnidaria	Alcyonaria	No	–	–	Pref.	–	–	Alt.	Pref.	–	–	Must be narcotised, do not use formalin
	Anthozoa: corals	No	–	–	Pref.	Alt.	Alt.	Alt.	–	Alt.	Pref.	Air dry a portion of skeleton
	Anthozoa: anemones	No	–	–	Pref.	Alt.	Alt.	Alt.	–	Pref.	–	–
	Hydroida	No	Pref.	–	Alt.	–	–	Alt.	–	Pref.	Alt.	–
	Scyphozoa and hydromedusae	Yes	Pref.	–	–	Alt.	–	Alt.	–	Pref.	–	Large volumes of fixative
Ctenophores	All	Yes	Pref.	–	–	–	–	Alt.	–	Pref.	–	Large volume of fixatives; most are ineffective

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Phylum	Taxa	Photos needed	Narcotising agents							Fixatives		Notes
			None	Fresh water	Chill or freeze	Menthol	Naphthalene	MgCl ₂	70% ethanol	7–10% formalin	Formalin to ethanol	
Echinodermata	Asteroids and echinoids	No	Pref.	Alt.	–	Alt.	Alt.	Alt.	–	Pref.	Alt.	Fix in formalin then air dry; ensure seastars are flat
	Crinoids	No	–	–	–	Pref.	–	Pref.	–	–	Pref.	–
	Holothuroids	No	–	Alt.	–	Pref.	Alt.	Alt.	Pref.	–	–	Do not use formalin
	Ophiuroids	No	–	–	Pref.	Alt.	–	Alt.	–	–	Pref.	–
Echiura	All	No	–	Pref.	–	Alt.	–	Alt.	–	Pref.	–	Must be narcotised before fixation
Ectoprocts	Cheilostomes and cyclostomes	No	Pref.	–	–	–	–	–	–	–	Pref.	Short time in formalin; can also air dry
	Ctenostomes	No	Pref.	–	–	–	–	Alt.	–	Pref.	Alt.	–
	All	No	Pref.	–	–	Alt.	–	Alt.	–	Pref.	Alt.	–
Mollusca	Bivalves	No	Pref.	Alt.	Alt.	–	–	–	Pref.	–	Alt.	Air dry valves or wedge valves open to allow formalin entry
	Aplocophora	Yes	–	–	Alt.	Pref.	–	Pref.	–	–	Pref.	–
	Cephalopods	Yes	–	–	Pref.	–	–	Alt.	Alt.	–	Pref.	–
	Gastropods: opisthobranchs	Yes	–	–	Pref.	Alt.	–	Alt.	–	–	Pref.	Air dry after microwaving
	Polyplacophora	No	Pref.	–	–	–	–	–	–	Pref.	–	Tie flat
Nemertea	All	Yes	–	–	–	Pref.	–	Alt.	Alt.	Pref.	–	Must be narcotised (see detailed methods)
Phoronids	All	No	Alt.	–	Alt.	Pref.	–	Alt.	–	Pref.	–	–
Platyhelminthes	All	Yes	–	–	Alt.	Alt.	–	Pref.	–	Pref.	–	–
Porifera	All	Yes	Pref.	–	–	–	–	–	Pref.	–	–	–

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Phylum	Taxa	Photos needed	Narcotising agents						Fixatives		Notes	
			None	Fresh water	Chill or freeze	Menthol	Naphthalene	MgCl ₂	70% ethanol	7-10% formalin		Formalin to ethanol
Sipuncula	All	No	-	-	-	Pref.	-	Alt.	Alt.	Pref.	-	-

Pref. Preferred technique. **Alt.** Alternative technique. - Not applicable.

Appendix F: Policy principles for determining the current status of marine pests

The policy principles provide a flexible approach to determining current pest status of marine pests and in the absence of agreed surveillance approaches (currently under development), general policy principles should be applied, rather than adopting a prescriptive policy. General policy principles that have been identified include:

- For incidents where the Consultative Committee on Introduced Marine Pest Emergencies (CCIMPE) convenes and provides advice to the National Management Group (NMG), the CCIMPE will recommend processes to determine pest status (e.g. likely present, likely absent, or unknown) and propose a pest status confidence level on a case-by-case basis.
- For incidents that are not referred to the NMG, the combat jurisdiction decides on processes to determine pest status. The CCIMPE may still provide non-binding advice as part of this decision.
- In scenarios not related to specific pest incursions (e.g. aquaculture site selection), jurisdictions will make the determination of presence (including range)/absence of a pest within their jurisdictional waters.
- It should be noted that pest status is valid as of the time of most recent determination but subject to change due to on-going introduction risk over time. Pest status determinations may therefore need to be repeated, with frequency dependent on the risk of introduction.
- Surveillance methods used in determining pest status should be recorded and shared upon request.
- Both quantitative and qualitative determinations of pest status can be used, as appropriate for the marine pest, location, and conditions.
- Methods used should be appropriate for the target species; pest biology should be considered with respect to surveillance duration, timing and sampling method.
- For quantitative determinations, quality assurance data are required for method accuracy as applied to the relevant situation (target species, habitat type etc.). Specifically, quantifying current pest status requires, amongst other things, knowledge of the likelihood of false negatives (failure to detect pests when present) and of false positives (apparent detection of species that are not present).
 - For eDNA approaches, the performance of both sampling collection methods and of the molecular tests applied need to be understood to provide quantitative determinations. Effects of sample timing on likelihood of detection should also be considered.
 - Qualitative determination can be made where the method has been appropriately demonstrated but its performance has yet to be quantified, e.g. eDNA methods that have

demonstrated detections in appropriate sample types but where the specific likelihood of false negatives and false positives is unknown.

- Methods that allow quantitative determination should be applied in preference where feasible.
- Pest status cannot be determined with any confidence if methods have not been validated or are inappropriate for the circumstance.
- Management implications should be considered, and caution applied when making pest status determinations because the level of confidence in presence or absence will depend on the extent and effectiveness of surveillance methods used in determining pest status.

Glossary

Term	Definition
Aquatic species	Any organism which spends all or significant parts of its lifecycle in fresh, brackish or marine waters.
Ballast water	Water with its suspended matter taken on board a vessel to control trim, list, draught, stability or stresses of the vessel.
Biofouling	Biofouling is the attachment or accumulation of aquatic organisms such as microorganisms, plants and animals, to any part of a vessel, on surfaces and structures immersed in or exposed to the aquatic environment. Biofouling is also known as hull fouling.
Biological control	Control of pests and weeds by another organism (e.g. insect, bacteria, virus etc), by a biological product (hormone), or by genetic or sterility manipulations.
Containment	Prevention of spread of an introduced species.
Control	Actions to limit spread or impacts of an introduced species, often involving partial eradication or other actions to limit population size and/or reproductive potential.
Cost benefit analysis	A comparative analysis of all costs and benefits of undertaking different options, to help decide which actions provide the best value or most suitable outcome (may include the 'do nothing' option).
Decontamination	The process of removing or destroying propagules of an introduced species, including fragments of species that can reproduce vegetatively.
Destruction	The process of killing aquatic organisms for eradication or control purposes.
Endemic species	A species with a native distribution restricted to the bioregion(s) of interest.
Established marine pest	A self-sustaining pest that occurs in Australia and is not regarded as eradicable. An established pest may be distributed widely across Australia, or be only regionally distributed. A regionally-distributed established pest may be the subject of containment measures to mitigate further spread. Native or indigenous plants and animals are not characterised as established marine pest (even if having negative impacts).
Fouling organism	Any plant or animal that attaches to natural and artificial substrates such as piers, navigation buoys, pilings or hulls. Includes crawling and nestling forms as well as seaweeds, hydroids, barnacles, mussels, bryozoans etc.
Hazard	A situation/activity that under certain conditions will cause harm. The likelihood of these conditions and magnitude of the harm produce a level of Risk.
Incursion	Occurrence of an introduced species in a region or country where it is not already established. See Interception.
Infaunal	Organisms living within substrate (e.g. burrowing).
Infestation/infested area	Population, or area with a population, of the introduced species.
Interception	Detection of a non-native organism at a pre-border or border inspection point, quarantine facility or other type of biosecurity control location.
Management	Actions taken in response to an introduced species including monitoring, control, containment, destruction etc.
Marine pest	Non-native marine plants or animals that harm Australia's marine environment, social amenity or industries that use the marine environment, or have the potential to do so if they were to be introduced, established (i.e. forming self-sustaining populations) or spread in Australia's marine environment. Many terms are used, sometimes interchangeably, to describe plants and animals that have been moved beyond their native range by humans, including alien, exotic, introduced, invasive, non-indigenous, non-native and nuisance species.

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Term	Definition
Marine species	Any aquatic species that does not spend its entire life-cycle in fresh water.
Motile	An organism capable of active movement.
Pathway	The geographic route taken by one or more vectors from point A to point B.
Pesticide	Any substance or preparation used for destroying a pest (typically associated with insects and rodents, with herbicides used for weed killers).
Plankton/planktonic	Small or microscopic organisms that drift or swim weakly in a body of water, including bacteria, diatoms, jellyfish, and various larvae.
Primary invasion	Initial establishment of an invasive marine species in a disjunct region (e.g. located beyond a land, ocean or temperature/salinity barrier).
Propagules	Dispersal agents of organisms, including spores, zygotes, cysts, seeds, larvae and self-regenerative tissue fragments.
Route	A geographic track or corridor followed by one or more vectors (see Pathway).
Regulation	A rule or order, as for conduct, prescribed by authority; a governing direction or law.
Secondary invasion	Subsequent spread within a new region due to reproduction or translocation of the initial founder population (see Primary invasion).
Sedentary	An organism that may be capable of limited movement but typically remains in one place or moves little (e.g. infaunal bivalves). See also sessile.
Sessile	An organism that is immobile and typically attached.
Surveillance	Systematic investigation over time, of a population or area to collect data and information about the presence, incidence, prevalence or geographical extent of a pest or disease; includes active and passive approaches
Targeted surveillance	Means surveillance targeted at a specific pest or life form.
Translocate/translocation	Any deliberate or unintentional transfer of an organism or its propagules between disjunct sites.
Vector	Anything capable of introducing or spreading a marine pest including a route or pathway (e.g. biofouling) or a physical or mechanical carrier (e.g. equipment or vessel)
Vessel	Any ship, boat or other craft used in marine environments; includes ships, floating platforms, boats and barges (e.g. structures that can float and be steered or moved by their own means or by other means, e.g. if towed). Also specifically includes smaller craft including recreational boats and other craft.

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