Understanding Biosecurity in Native Oyster Restoration

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CHAPTER 1
UNDERSTANDING BIOSECURITY IN NATIVE OYSTER RESTORATION

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INTRODUCTION

Invasive Alien Species or Invasive Non-Native Species (hereafter INNS; see Box 1.1) and diseases are a significant threat to biodiversity throughout European waters. In the case of the native oyster (*Ostrea edulis*), the disease caused by *Bonamia ostreae* (see Figure 1.5), is a major driver of decline in remnant populations. Given the magnitude of the threat posed by both INNS and disease, biosecurity (see Box 1.1), is a critical aspect of work in the marine environment. In this chapter we introduce the threats posed by INNS and diseases, their possible pathways of introduction, and the legislative and policy background practitioners of native oyster restoration should familiarise themselves with. All activities involving site visits pose a possible biosecurity risk. Practitioners should therefore work with their partners to understand and mitigate these risks wherever possible. These guidelines provide an introduction, but are not a substitute for working with the relevant authorities to ensure that work on site exceeds the mandated requirements and we encourage practitioners to participate in dialogue with the relevant authorities from early on in the planning process.

The presence or introduction of INNS or diseases negatively impacts the conservation objectives for protected species and habitats, the biodiversity associated with a healthy biogenic habitat, and the livelihoods of other sea-users. INNS and diseases also threaten the success of native oyster restoration through inducing high mortality (e.g. from Bonamiosis), competition for food or space, by acting as predators/pests of native species, or even altering the local environment to a less suitable condition, as well as risking the reputation of restorative projects if they are in any way implicated. Since both INNS and translocated oysters can be vectors of disease that can have devastating impacts on the receiving areas, wild populations, and aquaculture stocks, it is important to understand and mitigate against risks of disease transmission.

BOX 1.1: KEY DEFINITIONS

Biosecurity is defined as preventive measures to reduce the risk of spread of infectious diseases, pathogens, pests, invasive non-native species, and modified organisms, and can include toxins and pollutants. The way the term is applied can depend on the context and over a range of scales, including local, national, and transnational, even global.

Cultch is any substrate, such as rock or shell, that a juvenile native oyster is attached or may attach to.

Disease is defined as a disorder of structure or function in an organism that produces specific signs that are not caused by physical injury alone.

Invasive Non-Native Species (INNS) are organisms that have been introduced deliberately or accidentally across a biogeographic boundary by humans, and which go on to have a negative ecological or economic impact in their new range. Not all non-native species become invasive. Many non-native species have little impact in their new range and may have positive economic benefits associated with them. INNS refers specifically to those species which do have a negative impact. INNS are also referred to as Invasive Alien Species (IAS) in Article 3 (2) and (3) of Regulation (EU) No 1143/2014 of the European Parliament where they are defined as: “an alien species whose introduction or spread has been found to threaten or adversely impact upon biodiversity and related ecosystem services”.

Pathogen is defined as the biological agent (especially microorganisms such as bacteria, viruses) that cause disease in their host.

Translocation is the movement of populations of native oysters, adult, juvenile or larval, from one location to another that could be considered a different body of water.

Native oyster restoration as a pathway for INNS and disease

Shellfish movements have been a major vector of INNS and disease. Attempts to replenish oyster stocks in Europe through importing both native and non-native oysters (e.g. *Crassostrea virginica*, *Crassostrea angulata*) have been documented since the 1870s. Over the decades, many non-native species and diseases were inadvertently translocated with these introductions, some with severe negative effects for marine ecosystems. A number of the species introduced with live oysters have become highly invasive and a threat to the native oyster itself, including the American tingle *Urosalpinx cinerea*, the slipper limpet, *Crepidula fornicata* and the native oyster parasite *Bonamia ostreae* (see Figure 1.3 for examples). It is as a result of these lessons learned that legislation was developed, both nationally and internationally, to prevent future spread of INNS and disease. It is therefore critically important that practitioners familiarise themselves with the relevant legislation.

Native oyster restoration often involves the movement of people, equipment, cultch material and live shellfish between sites and as such represents numerous opportunities to accidentally move species between sites as well. Whilst such movements are by no means the only vectors of INNS or disease, efforts to restore native oyster populations must adopt rigorous biosecurity protocols in order to reduce the risk that an action with an intended positive ecological benefit, results in a negative impact.

Biosecurity as an integrated part of restoration practice

It is important to note that the responsibility for ensuring the highest possible level of biosecurity lies with the projects themselves. Inadequate biosecurity measures present a very real biological, ecological, and reputational threat. It must be recognised that it is not always possible to accurately identify threats to the environment prior to damage occurring, and that there is usually a time lag between arrival and recording of INNS and diseases. Therefore, vigilance is needed, even in areas where INNS or diseases are not yet recorded. As well as adhering to the legal requirements, projects should apply a "Precautionary Approach" to prevent harm being caused by accidental or poorly considered transfers. The IUCN has published "Guidelines for applying the precautionary principle to biodiversity conservation and natural resource management", which can helpfully be referred to.

It is the responsibility of the restoration practitioners to seek advice from the relevant authorities and ensure that they meet legal requirements. Failure to do so can result in legal consequences.

Figure 1.1: Solent Oyster Restoration Project staff carrying out fieldwork in marina sites in the Solent. Photo: Blue Marine Foundation.

Stop the spread **The success and reputation of a restoration project can be negatively impacted by accidental introductions of invasive species and pathogens. Project equipment such as vans, boats and field kit can all be vectors for their transmission, which will ultimately damage the marine environment and wildlife.**

CHECK **Check** your equipment, clothing and boats after carrying out fieldwork for fouling material. Ensure that you remove anything that you find and dispose of it in the appropriate manner.

CLEAN **Clean** all fieldwork items thoroughly with freshwater as soon as possible. Ensure that you pay attention to items such as fieldwork clothing, restoration equipment, trailer wheels and areas that are damp or hard to reach.

DIS INFECT **Disinfect** – Where the risks are higher, include disinfection as part of cleaning procedures.

DRY **Dry** – Ensure that you drain water from any water remaining on fieldwork items, and equipment such as a trailer and boat. Try to dry all equipment for as long as possible before next usage.

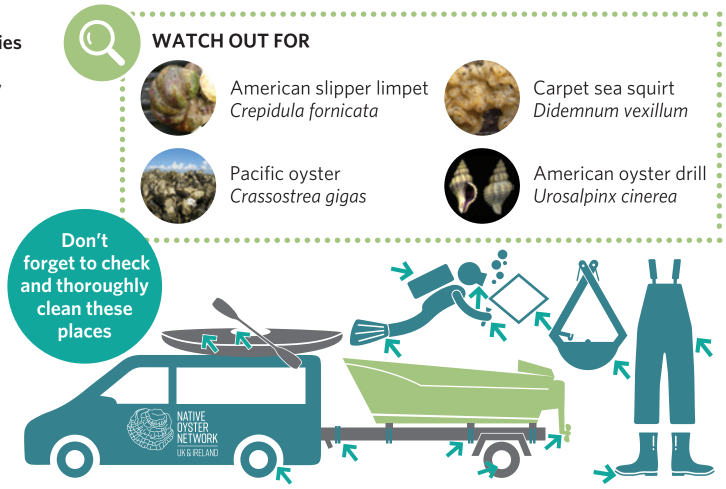


Figure 1.2: Biosecurity considerations to prevent transmission during restoration practice and fieldwork:
Areas to be vigilant with when cleaning after carrying out fieldwork for oyster restoration projects: Check – Clean – Disinfect – Dry.

INNS and diseases can be moved between sites whenever people and equipment are moved, as well as when oysters or cultch material are placed in the water. As such it is important that all people participating in oyster restoration activities, including science and monitoring, comply with both standard 'Check, Clean, Disinfect, Dry' protocols, as well as with relevant European and national legislation relating to aquatic animal health when moving between sites (see Figure 1.2).

Check before leaving a site all equipment including wetsuits, vessels, boots, buckets etc. Remove all visible hitchhikers, sediment, and debris. If this occurs away from the site, ensure that all material is at least disposed of safely, and under no circumstances near a watercourse. Under circumstances of enhanced risk, disposal should be to a specified biological waste disposal route (possibly including incineration).

Clean all equipment including the vessel and bilge tank with freshwater. Do not let water drain back into the sea, as spores and eggs can persist for some time.

Disinfect under circumstances of increased risk, a biocide/disinfectant should also be used.

Dry all equipment thoroughly, ideally in sunlight, before moving to a new marine location.

All activities undertaken on the restoration site should be considered with regards to biosecurity. Further guidance on developing general marine biosecurity action plans, including useful worked examples were developed by Payne *et al.* 2014.

Native oyster restoration may include the translocation of cultch, spat attached to empty shells or pieces of shells (spat-on-shell), single spat (also called single seed oysters), juvenile or adult oysters. Each of these methods carries with it the risk that species and/or pathogens are also translocated. It is recommended that projects contact the relevant authority for advice on regulatory and licensing requirements early in the planning stage of any restoration project. This topic is covered in greater detail in Chapter 2.

Working with the public to understand risk

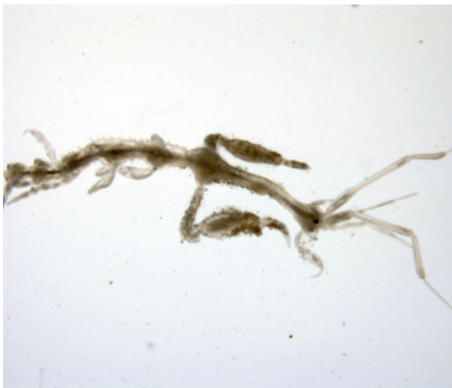
Native oyster restoration activities also present an opportunity for public engagement. Despite the many regulations in place to prevent the illegal release of oysters and the potential associated diseases and INNS into the wild, it is common practice in some coastal areas for individuals to store their live oysters in the water for a few days before consuming them, or to dump the fresh shells in the sea after a meal. Such activities may severely impact restoration and aquaculture activities and society at large, by contributing to the dispersal of pathogens and non-native species. These activities often take place both because the individuals are not aware of the risk and because they believe they may be doing something positive for the ecosystem; such an action is all the more likely if restoration efforts do not adequately communicate the risk of such activities. Restoration offers a unique opportunity to allow individuals to contribute positively to the recovery of a threatened species and a unique opportunity for the public to better understand coastal ecology and the practice of ecosystem restoration. Projects should not overlook these opportunities, in particular with regards to raising awareness of the risks associated with returning oysters or shells to the water without undertaking appropriate biosecurity measures.

Taxonomic group: Mollusca (*Bivalvia*)
Species: Pacific Oyster (*Crassostrea gigas*).
Impact: Competition, habitat change
Photo: Åsa Strand

Taxonomic group: Mollusca (*Gastropoda*)
Species: American slipper limpet (*Crepidula fornicata*)
Impact: Competition, habitat change
Photo: Zoë Holbrook

Taxonomic group: Ctenophora
Species: Warty comb jelly/Sea walnut (*Mnemiopsis leidyi*)
Impact: Predation (zooplankton), competition
Photo: Marco Faasse (CC BY-NC-SA 4.0 license)

Taxonomic group: Phaeista
Species: Japanese wireweed (*Sargassum muticum*)
Impact: Habitat change, competition
Photo: Prof. Bárbara Ignacio (CC BY-NC-SA 4.0 license)



Taxonomic group: Crustacea
Species: Japanese skeleton shrimp (*Caprella mutica*)
Impact: Competition
Photo: Joanne Preston

Taxonomic group: Tunicata
Species: Carpet sea squirt (*Didemnum vexillum*)
Impact: Competition, habitat change
Photo: Rosana Moreira da Rocha (CC BY-NC-SA 4.0 license)

Taxonomic group: Chlorophyta
Species: Green alga – "killer alga" (*Caulerpa taxifolia*)
Impact: Habitat change, competition
Photo: Boris Unger

Taxonomic group: Crustacea
Species: Acorn barnacle (*Hesperibalanus fallax*)
Impact: Competition
Photo: David Fenwick, APHOTOMARINE

Taxonomic group: Crustacea
Species: Brush-clawed/Asian shore crab (*Hemigrapsus penicillatus*)
Impact: Competition, predation
Photo: ffish.asia (CC BY 4.0 license)

Figure 1.3: A selection of high impact INNS listed as species (present and horizon) which have been selected for assessment of Good Environmental Status within GB waters, as required under the Marine Strategy Framework Directive. Please note that this list is for illustration only. Complete and current lists should be sought on national/local levels on a project by project basis. See Box 1.2 for some potential sources of information or contact the national competent authority for advice.

LEGISLATIVE OBLIGATIONS

The impacts of the introduction of shellfish diseases and INNS have long been acknowledged, and international institutions have developed legislation and relevant targets and reporting systems to address these threats. It is the responsibility of all restoration practitioners to ensure that they are aware of and adhere to relevant legislation on biosecurity to avoid falling foul of the law. Be aware that legislation and guidance functions on a variety of scales. Figure 1.4 illustrates the many levels of regulation relevant to oyster restoration. Seek the advice of the relevant authorities to ensure that the project adheres to all relevant guidance and the law.

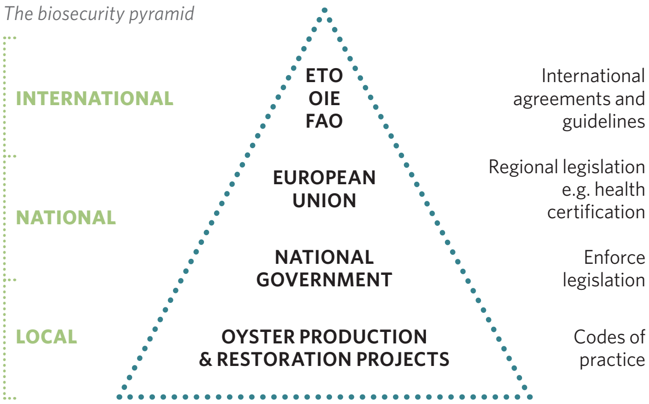


Figure 1.4: Legislation and policy regarding biosecurity function at a variety of scales, all of which projects should be aware of and seek advice on. Figure adapted from Oidtmann *et al.* (2011) Aquaculture 1-2: 22-33.

INNS and their biosecurity management

There have been few successful eradication attempts for marine non-native species or diseases in open waters. Therefore, the most effective method of control is to prevent their introduction.

A number of high-risk INNS are recognised in Europe (Figure 1.3, Box 1.2). The absence of a disease or species from the certifiable or high-risk list does not, however, mean that it is not a risk. While many non-native species have little or no impact on the receiving water bodies, it is difficult to predict which species will become problematic in an introduced range. Certain attributes related to both the life history of the species and the condition of the receiving site which can indicate the likelihood of species becoming problematic (see Geburzi and McCarthy 2018 for a good review), and invasion history from other locations can also be a useful indicator. Assessments of whether a species is likely to become invasive in a new location requires expertise. Fortunately, several databases exist for European, national, and regional records of non-native species, which projects can refer to for information. AquaNIS is an online information system for aquatic non-indigenous species introduced to marine, brackish, and coastal freshwater of Europe and neighboring regions. It includes regular updates and provides extensive species accounts, including biological traits and images to support the Marine Strategy Framework Directive (MFSD) and risk assessments for shipping and aquaculture. National platforms also provide important and up-to-date information (e.g. for Germany), or risk classification lists (e.g. the Swedish Species Information Centre (SLU)), whereas the IUCN have published a guide on INNS and their monitoring for the Mediterranean (Box 1.2). In the UK, the GB Non-Native Species Secretariat, a part of FERA/DEFRA, gives pragmatic advice and issues alerts for high impact INNS including specific marine advice and provides risk assessments for species. Further sources include: The UK Water Framework Directive Alien Species Alarm List and the EU Alien Species Regulation list of Species of Union Concern.

We recommend that projects check several platforms and ensure that the information they draw on in their risk assessment and planning is up to date. These lists and assessments can be used to identify which species are of particular concern when considering where to source oysters or cultch material from.

Every species introduced to a new area has the potential to become invasive. Therefore, while biosecurity protocols should prioritise the prevention of key identified problem species, projects should always strive to adhere to the precautionary principle and clean all materials and equipment moved, even if no INNS are believed to be present.

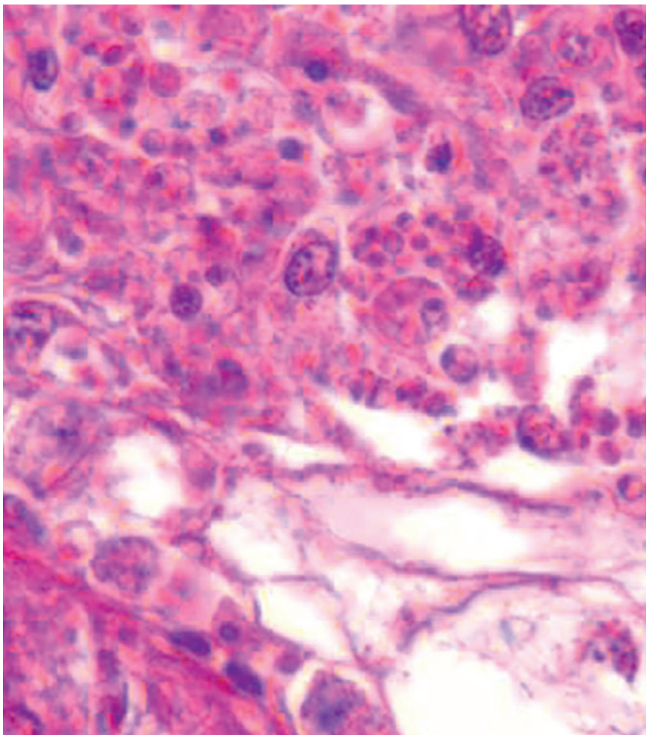


Figure 1.5: Histological slide depicting *Bonamia ostreae* cells infecting the tissue of a native oyster *Ostrea edulis*. Photo: B. Chollet/Ifremer.

BOX 1.2: EXAMPLES OF INTERNATIONAL, NATIONAL, AND SUBNATIONAL LEGISLATION AND GUIDELINES **RELATING TO INNS**

International:

Regulation (EU) 1143/2014 on invasive alien species (the IAS Regulation) entered into force on 1 January 2015, fulfilling Action 16 of Target 5 of the EU 2020 Biodiversity Strategy, as well as Aichi Target 9 of the Strategic Plan for Biodiversity 2011-2020 under the Convention of Biological Diversity.

Mediterranean – Monitoring marine invasive species in Mediterranean marine protected areas (MPAs): a strategy and practical guide for managers.

National:

France – French national strategy on invasive alien species. Reference documents for taking into account marine cultivation activities in the preservation of the marine environment (Coz & Ragot, 2020a, 2020b).

Italy – Decreto Legislativo 230/2017 – 'Disposizioni volte a prevenire e gestire l'introduzione e la diffusione delle specie esotiche invasive' (National strategies to prevent the introduction of invasive species).

Subnational:

Loch Creran Biosecurity Action Plan: A local community-level biosecurity plan set up in response to a high impact species in a shellfish growing waters.

Diseases and their biosecurity management

Native oysters are susceptible to numerous diseases which are still expanding their range in Europe and which are subject to monitoring efforts. It is critical that projects familiarise themselves with the notifiable diseases and the disease status of any locations where work is carried out. When working in shellfish growing waters consideration should also be given to the possible transmission between bivalve species. Some diseases, such as *Marteilia refringens* (including the recently proposed species *M. parafringens* sp. nov.) can be transmitted between native oysters and blue mussels (*Mytilus edulis*), and there are indications that OsHV-1 µvar can be transmitted between Pacific oysters (*Crassostrea gigas*) and native oysters.

There are several diseases which are of particular note in the context of native oyster restoration in Europe. These include the listed diseases (and the agents) of bivalves to the World Organisation for Animal Health (OIE) and/or to the European Commission (EC) (The Council Directive 2006/88/EC):

- Bonamiosis – *Bonamia ostreae* (OIE/EC – present in Europe),
- Bonamiosis – *Bonamia exitiosa* (OIE/EC – present in Europe),
- Marteiliasis – *Marteilia refringens* (OIE/EC – present in Europe),
- Denman Island Disease – *Mikrocytos mackini* (EC – not currently present in Europe),
- Herpes-like infection – Herpes virus OsHV-1-µvar (present in Europe) (notifiable in few zones in Ireland and UK only)

The law (Box 1.3) places an obligation on practitioners to report immediately any suspicion or confirmation of the presence of these listed diseases to the competent authorities. The competent authority should then investigate and decide what measures are to be taken. Measures may include an initial survey, the inclusion of a site-based risk assessment and biosecurity plan with contingency measures, as well as follow-up monitoring of the site as part of the licensing conditions of the plan or project and movement restrictions. In the case of the suspicion of the presence of a disease or non-native species, the practitioner must follow these steps:

- Report immediately to the competent authority.
- Adopt a precautionary approach – do not carry on operations that might contribute to further dispersal.
- Carry out risk assessments.
- Seek and follow advice from the relevant authorities. This may include not moving any material.
- It is important for restoration practitioners to be aware not only of the listed diseases and the requirements to follow the rules on translocations that apply internationally, but also to be mindful that there are a range of other parasites and pathogens to which the native oyster is susceptible, or may be a vector of. The following is a non-exhaustive list of known pathogens and parasites affecting the native oyster:
    - *Boccardia* (genus of),
    - *Cliona celata*,
    - *Cliona viridis*,
    - *Gyrodinium aureolum*,
    - *Haplosporidium armoricanum*,
    - *Herrmannella duggani*,
    - *Hexamita inflata*,
    - *Mytilicola intestinalis*,
    - *Nocardia crassostreae*,
    - *Ostracoblabe implexa*,
    - *Papovaviridae* (family of),
    - *Perkinsus mediterraneus*,
    - *Polydora* (genus of),
    - *Pseudoklossia* (genus of),
    - *Vibrio spp.* (e.g. *V. alginolyticus*, *V. anguillarum*, *V. coralliilyticus*, *V. neptunius*, *V. ostreicida*, *V. tubiashi*)

Haemic neoplasia may also affect oysters. In this case, no disease agent is observed, but the neoplastic cells may be infectious and cause significant mortalities.

It is the responsibility of the restoration practitioner to implement appropriate national disease prevention and management requirements and to report any unusual or unexplained mortalities, as well as any suspicion of occurrence of a listed or emerging pathogen, to the relevant authority for investigation.

Screening for diseases (see Figure 1.6) is usually carried out by national reference laboratories (European Union Reference Laboratory for Mollusc Diseases (EURL) (2020)) or other national institutions, depending upon the jurisdiction. OIE reference laboratories can be found on the World Organisation for Animal Health website (see resources section).

As with all introduced species, it is not possible to know before a disease is introduced, whether it will seriously impact in its introduced range. A disease may be subclinical in a population that has co-evolved with it, and therefore not apparent. Once transferred to a naive population it may cause high mortalities and disruption. For example, *Bonamia ostreae* was not known as a disease agent in its Californian source range but caused widespread mortalities in excess of 90% in its introduced range in Europe.

The guidance presented here is aimed at assisting native oyster restoration project managers and practitioners in ensuring that any restoration efforts in European waters are carried out responsibly. In order to ensure that all activities comply with the law, practitioners should work with the relevant authorities. Some biosecurity relevant resources from international, European, and national perspectives are given in Box 1.3.

Figure 1.6: Native oyster disease screening in University of Portsmouth lab. Photo: Luke Helmer.

Appropriate response in the event of unexpected mortality events

A critical aspect of biosecurity relating to disease management is monitoring of increased and unexplained mortality. During monitoring, restoration practitioners may notice unusual levels of mortality, changes in oyster growth, absence of larval settlement or increased or unexplained mortality. These may not have an immediate or obvious explanation and therefore require investigation. Disease is not the only cause of unexpected mortality. Pulse events, such as heavy rainfall can cause fluctuations in temperature, salinity, and turbidity, and may contribute to adult and spat mortalities, loss of planktonic larvae and cessation of reproductive activity. Storms can also increase pollution, horizontal advection, and abrasion, which can negatively impact oyster condition and possibly influence the prevalence of diseases such as infection with *Bonamia ostreae* where it is present. Because of the risk posed by disease, projects should always seek advice from the relevant authority regarding actions required in the event of an increased and unexpected mortality event.

As a guideline in instances where there are sudden, increased and unexplained high mortalities or recruitment failure, practitioners must report any abnormal mortality event to the authority and investigate the possible involvement of an infectious agent (as outlined in article 26 of the EU Directive 2006/88/EC, and also in Article 18 of Regulation 2016/429 which replaces 2006/88 in 2021). For that purpose, oysters including moribund ones (but not dead ones) should be sampled and processed according to recommendations of the European Union Reference Laboratory (EURL) for Mollusc Diseases in order to carry out histology, bacteriology and PCR for the detection of specific pathogens. These diagnostic analyses must be carried out by a recognised or agreed laboratory.

Going beyond legislative requirements and 'owning' the risk

Most existing national policies and legislative frameworks relevant to translocations for restoration are based on risk profiles of the aquaculture industry. It is important to understand that restoration carries potentially far higher risks because oysters go permanently back into the ecosystem. Routine monitoring by the government may be infrequent and reflective of a perceived low risk. In most cases, it will therefore be necessary for restoration projects to take responsibility for the biosecurity of their operations and apply a greater stringency than may be legally required. Maintaining a high level of biosecurity in restoration work is imperative both for ecological success, and to maintain a social license for such activities.

It is important to recognise that even with the most stringent testing and biosecurity procedures, it remains possible that a disease agent or INNS may become present at the restoration site where translocations have occurred (see Figure 1.7).

This may be because there is currently no 100% accurate method of disease-screening all translocated materials in a consignment.

1. Any biosecurity for the translocation of live oysters runs the risk that not all INNS individuals will be eliminated because, inevitably, the system must allow for the survival of the oysters.
2. Third-party activities in the area may have introduced a disease or INNS at or around the time of the project translocation.
3. The disease or INNS may have already been present and undetected in other biological reservoirs.

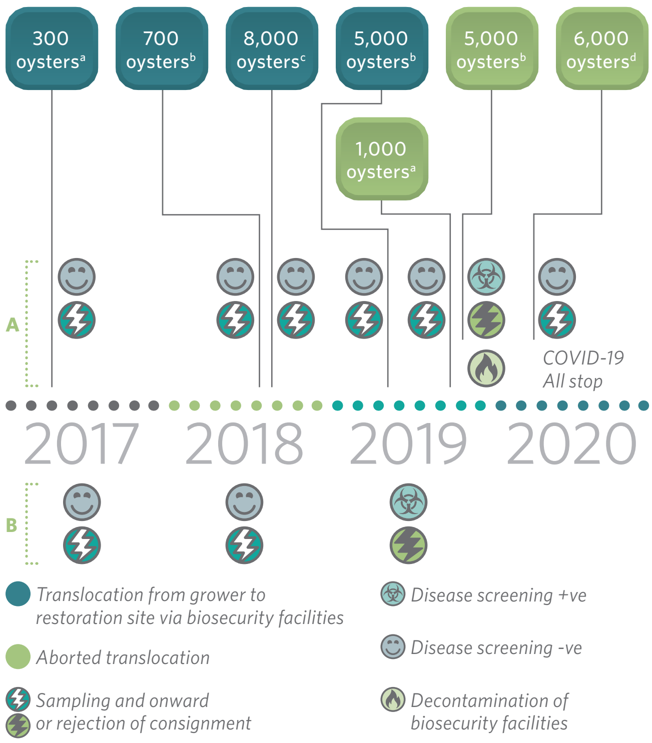


Figure 1.7: Schematic of biosecurity disease-screening activity of a restoration project based on a redacted but real case study. A = independent project-based testing of consignments translocated between oyster growers and the restoration site (via closed-circuit biosecurity holding facilities). B = project-based confirmatory testing of restoration site. All project testing (A&B) in addition to favourable (for disease) statutory government testing and accreditation of suppliers. Despite screening of all consignments, confirmatory annual screening of restoration site and rejection of consignment that tested positive for *Bonamia ostreae*, the restoration site tested positive in 2019 for said disease. Superscript letters indicate the four different suppliers.

BOX 1.3: EXAMPLES OF INTERNATIONAL, NATIONAL, AND SUBNATIONAL LEGISLATION AND GUIDELINES RELATING TO DISEASE MANAGEMENT

International:

Marine biosecurity has an international legislative framework: The European Union Member States, Council Directive 2006/88/EC (24/10/2006) sets out animal health requirements for aquaculture animals and products, and on the prevention and control of certain diseases in aquatic animals. The OIE Aquatic Animal Health Code (2019) provides standards for the improvement of aquatic animal health worldwide and the Regulation (EU) 2016/429 ('Animal Health Law') sets rules to control transmissible animal diseases and that have broad impacts on public or animal.

National:

UK – The Aquatic Animal Health (Scotland) Regulations 2009, AAH (England and Wales) Regulations 2009, and AAH (Norther) Regulations 2009 implement Council Directive 2006/88/EC (as amended) in the UK. NB: EU Directive 2006/88/EC will be replaced by Regulation 2016/429 from April 2021. Some useful advice on Marine Biosecurity Planning, INNS and marine diseases can be found at NatureScot and CEFAS.

Sweden – The translocation of organisms for aquaculture purposes (which also governs translocations of wild populations) is governed by the fisheries law (2 kap. 16 § Förordning (1994:1716) om fisket, vattenbruket och fiskerinäringen) complemented by SJVFS 2014:4 2 kap 7-10 § Statens jordbruksverks föreskrifter om djurhälsokrav för djur och produkter från vattenbruk and Fiskeriverkets föreskrifter (FIFS 2011:13) om utsättning av fisk samt flyttning av fisk i andra fall än mellan fiskodlingar. The general rules are that permission for culture and translocations cannot be granted for alien species, organisms with a contagious disease and some more specific cases.

Subnational:

On a regional level, Inshore Fisheries and Conservation Authorities or communities may produce Biosecurity Action Plans to manage shellfish (e.g. North western Inshore Fisheries and Conservation Authority Biosecurity Plan).

CHAPTER 2
BIOSECURITY GUIDELINES FOR NATIVE OYSTER AND CULTCH TRANSLOCATION

CHAPTER AUTHORS

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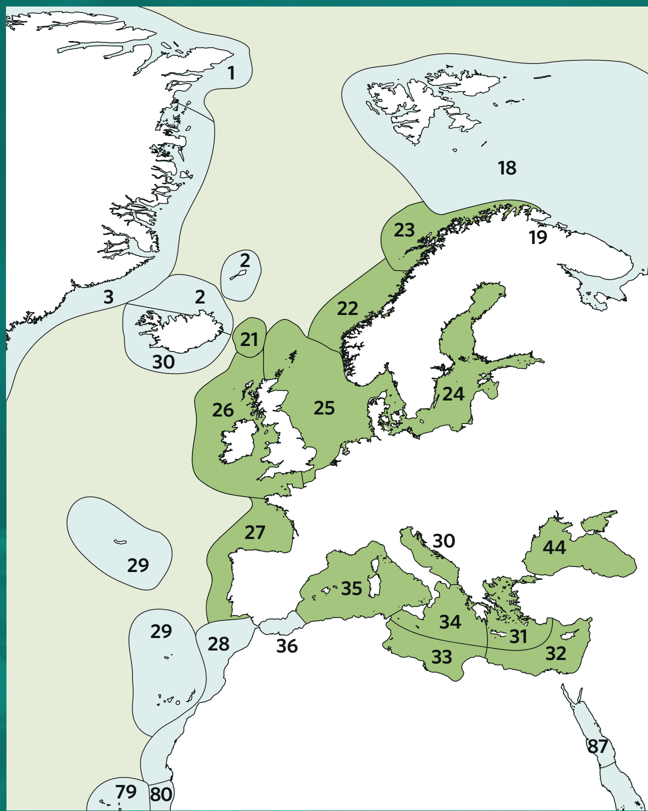

Figure 2.1: Marine ecoregions map adapted from Spalding *et al.* 2007. The known biogeographic range of *Ostrea edulis* is shaded in green. *Ostrea edulis* will only exist in areas within these ecoregions with suitable abotic and biotic habitat characteristics.

INTRODUCTION

Given that all translocations carry with them a risk of accidental introduction, it is important that **avoiding the risk by avoiding translocations be considered in project planning**. If projects decide to proceed with translocations despite the inherent risks, comprehensive protocols and actions to mitigate and reduce the risks presented should be developed on the project level. **It is critical that the relevant authorities are informed of all planned activities and projects seek advice from, and work in partnership with, the relevant authorities throughout the project**. Furthermore, projects should ensure that all required permits are obtained before the initiation of any restoration activities. Projects should seek to exceed the legally mandated standard, because the current framework was not developed with restoration (and hence the permanent placement) of native oysters in European waters in mind. And because the central aim of ecological restoration is to support ecosystem recovery and improve the environmental status of European waterbodies, which demands that a rigorous approach be applied.

Oyster habitat restoration in Europe is still in its infancy and the science to support best practice protocols has yet to be fully developed. This chapter seeks to outline the many considerations in planning a translocation activity and to provide some examples of solutions that have been used in oyster restoration to date. **While this chapter seeks to outline the considerations and provide some detail of existing practice, it is critical that project managers work with the relevant authorities to develop appropriate protocols in each case, and that validation of the efficacy of the actions undertaken are assessed on a case by case basis**. Project managers should be transparent about their protocols and share where possible, relevant validation data regarding the efficacy of the protocol. In this way the community of restoration practitioners can work together to refine approaches to ensure they are as effective and cost effective as possible.

Considerations before translocation

It is critical when considering a translocation the following questions are addressed:

1. **Is translocation necessary?** Consider why translocation is the best option. Are there local stocks that could be used? Can the project timeline be adapted to allow for the use of 'on-site' or biosecure hatchery reared stock or local spat collection? If possible, do not translocate oysters.
2. **Are there local sources?** Identify potential local sources and if possible, use oysters from local sources and environments.
3. **If translocating, use the following hierarchy in selecting donor material to minimise risk:**
   1. **Do not consider donor sites outside of the historical native range of *Ostrea edulis* (see Figure 2.1).** While there are now populations of *O. edulis* outside of its native range (e.g. on the east coast of North America), reintroducing oysters from outside its native range should be avoided at all costs. This is to avoid the potential introduction of non-native species and diseases associated with the native oyster in its introduced range. As an illustration of the risk, the European presence of more than sixty species, native to the Pacific Northwest USA, can be attributed to movements of the Pacific oyster since the 1960's alone.
   2. **Do not consider donor sites with high-risk invasive species or diseases that are not present at the receiving site.** Moving an oyster from an area where a listed disease is known to be present to a disease-free area is illegal where disease controls are applied. Similarly, releasing known INNS into the wild is a criminal offence in some countries. This aside, the ecological and societal risk of introducing either a disease or high-risk INNS into an area is unacceptable, given the possible impacts such action could result in.
   3. **Minimise the physical distance between the donor and receiving site.** To reduce the risk of unknown diseases or INNS being introduced to an area, it is best to reduce the physical distance between the donor and receiving site. This may also have the additional benefit when moving live oysters, of maintaining any local or regional genetic structure in the oyster population (see Box 2.1).
   4. **Avoid movements across latitudinal gradients.** The native oyster can be infected by a large number of pathogens (see list in Chapter 1). Within their co-evolved range and the local temperature regime, pathogens may have limited impact on their host. There is, however, a risk that pathogens may become more virulent when moved to a different environment (see Figure 2.2). As it is not possible to know which diseases may have an impact in the novel environment, and it is in any case challenging to screen for all known diseases, movement of oysters to a largely different environment is not recommended.

BOX 2.1: GENETIC IMPLICATIONS OF *OSTREA EDULIS* TRANSLOCATION

Human-mediated translocation of either wild or hatchery-produced *Ostrea edulis* has long occurred within the fisheries context but has, in the past decade, also been applied as a conservation management strategy for endangered species. The movement of live oysters from one location to another may pose an environmental risk due to not only the spread of diseases/pests, but also the genetic erosion of wild populations.

Translocation may induce an increase in genetic diversity in recipient populations, by mixing genetically divergent populations, and may reduce genetic divergence among geographically distant populations, hence possibly reducing the existence of adaptation to local conditions and consequently overall population fitness in recipient area populations. The impacts of translocation are therefore an important issue for the management of exploited or endangered species.

The selection of local sources should be prioritised, both to reduce the risks of accidentally introducing diseases/pests and to maintain genetic structure and adaptations present in the local population. Where the use of local stock is not an option, practitioners should consider genetics when selecting donor stock. The genetic structure for the native oyster at local and regional scales is currently the subject of ongoing investigation. It is therefore not included further in this report, but rather it is recommended that the current scientific literature be examined when seeking donor material.

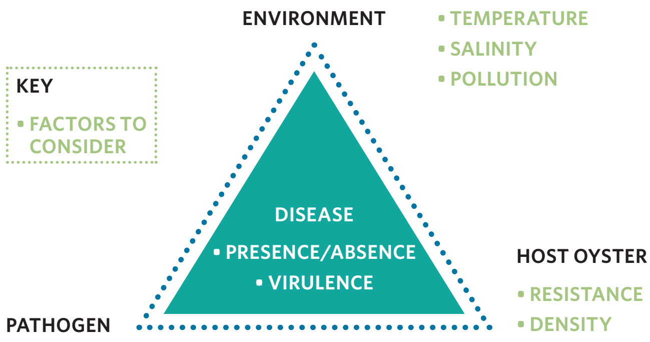

Figure 2.2: The disease triangle: The occurrence and severity of disease is a product of the host and pathogen co-occurring, and the environmental conditions being suitable for disease outbreak. Movement of oysters from one environmental regime to another therefore carries the risk that novel environmental conditions may favor the pathogen and that a previously subclinical disease in the oysters may become problematic.

Figure 2.3: Comparative survey of donor and recipient sites to establish INNS and disease transfer risks by W.G. Sanderson, DEEP Project. Photo: Richard Shucksmith.

Translocating live oysters

If translocation is necessary and potentially appropriate donor material has been identified, the next step is to undertake thorough biosecurity measures, under advice from the relevant authorities, to reduce the risk of accidental transfer of species. Initial risk assessments should be undertaken in order to understand the risk and map out the appropriate action. Assessment of risk should include consideration of ongoing activities in both the donor and receiving site.

It is critical that adequate time for completing comprehensive biosecurity measures is planned into the project. For example, undertaking field surveys to assess the suitability of donor sites should ideally take place in the summer, when species are most abundant and therefore likely to be encountered and identified (see Figure 2.3). Additionally, the time required to physically clean and screen oysters should be accounted for, as this can be a rate or scale limiting step. As an indicator, substantial epifaunal growth can mean that it takes one person one hour to clean 100 oysters. Projects should not seek to translocate a greater number of oysters than they have time to clean and check thoroughly. Translocating large numbers of oysters is an arduous and time-consuming process, and translocating more individuals increases the risk of unintended introductions. These realities should be considered when planning translocation activities.

Undertaking a risk assessment

The first steps in any risk assessment is risk identification or mapping and analysis. This may be facilitated by classifying risks according to different criteria, e.g. local/regional/global extent, continuous/instant, manageable/unmanageable, internal/external. The identified risks should then be analysed regarding likelihood and consequence. Moreover, to rank risks, they must first be comparable. This can be achieved through literature reviews and potentially expert consensus round table processes where the identified risks are prioritised and weighted. The different weights can then be combined in a structured way. High ranking risks, i.e. above the acceptance levels given by the risk evaluation, will proceed to treatment/management. As a rule of thumb, negative risks with high likelihood and low consequence should be treated with preventive measures, low likelihood and high consequence should be treated with mitigating measures and very high/catastrophic consequences should be avoided (see Cook *et al.* 2014 for further guidance). Bear in mind that risk assessment should be an ongoing, iterative, and adaptive process (see Figure 2.4).

Prior to translocating animals, it is important to consider the disease status of both the donor and recipient sites. Comprehensive existing OIE, EU and local regulation surrounding the testing, movement and monitoring of pathogens and disease should be adhered to as an absolute baseline with regards to decisions on movements. Details of legislation in EU areas is located on the European Commission website (see Box 1.2 and Box 1.3 in Chapter 1).

Recommended routes to compile relevant data for the donor and recipient site include:

- Search the confirmed designation notices for notifiable diseases and work with the competent authority to ensure that the site is a candidate donor site under animal health law and to verify compliance with the relevant aquatic animal health regulations.

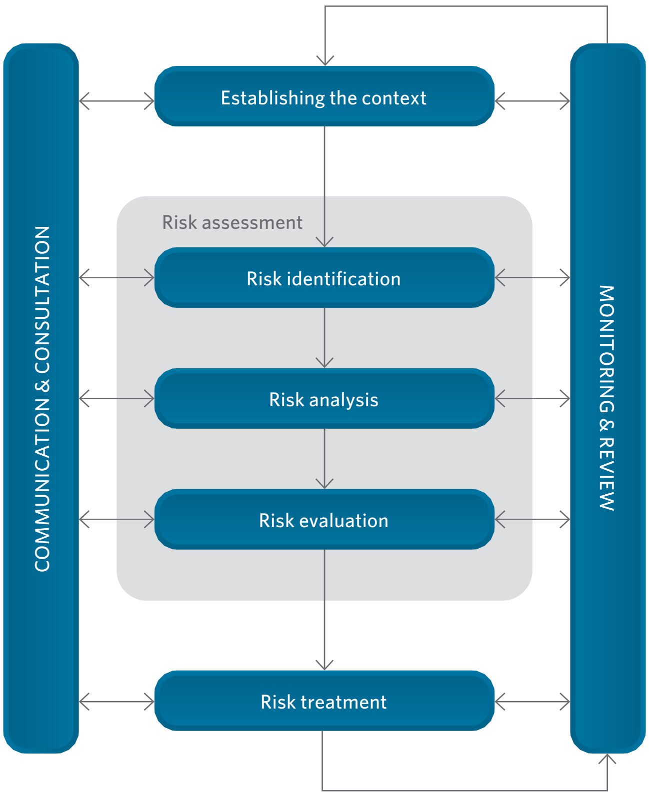

Figure 2.4: An overview of the steps required in undertaking a risk assessment. Risk assessment is an ongoing process and information derived from operations should be fed back to ensure that the assessment remains current.

BIOSECURITY MEASURES PLAN SCHEMATIC

1. Online assessment of INNS and notifiable shellfish diseases described in donor and recipient sites.

Websites to consult:
WoRMS, MarLIN, CABI, NBN atlas, NEMESIS

- INNS present at donor equal to or less than recipient.
- Disease status of donor site equal or less than recipient.

- High impact INNS/disease present at donor but not recipient.

No movement of oyster to take place.

2. Physical assessment of INNS and notifiable shellfish diseases at donor and recipient sites.

Particular attention paid to shellfish themselves.

- INNS present at donor equal to or less than recipient.
- Disease status of donor site equal or less than recipient.

- High impact INNS/disease present at donor but not recipient.

No movement of oyster to take place.

3. Plan to receive oysters from donor site.

Physical biosecurity measures put in place:

- Oysters to be monitored for INNS and signs of mass mortality.
- Oysters to be cleaned appropriately to remove epibionts.
- Water used to clean oysters either to enter the origin site or treated appropriately with chlorine or other.
- Plan for short-term contingency storage area if conditions prevent deployment.

- Epibionts removed or killed to satisfactory level so that risk of INNS transfer is removed.
- No signs of mass mortality.

- Individuals with compromised shells discarded at donor site.

- High impact INNS/disease present at donor but not recipient.

No movement of oyster to take place.

4. Receive oysters from donor site.

Transport oysters in most suitable conditions:

- Forecasted weather to be cool and unlikely to cause delays in marine vessel travel or travel by road.
- Transport to occur at a time of day as to avoid long traffic delays i.e. over night.
- Oysters to be transported in a damp and cool environment.

- No evidence of further epibiont growth.
- No signs of mass mortality.

- INNS observed.
- Large mortality event occurring during transport.

No deployment of oyster to take place.

5. Deploy oysters to seabed.

Figure 2.5: Decision tree outlining the factors to be considered when assessing suitable donor sites and whether to go ahead with relaying oysters into the recipient site. Figure modified from Blue Marine Foundation schematic.

Figure 2.6: Native oysters in hatchery in the Netherlands. Photo: Oscar Bos/Wageningen Marine Research.

- Contact the Statutory Nature Conservation Bodies or regulators directly for any non-native species abundance data from Water Framework Directive or Marine Protected Area surveys.
- Some useful data on non-native species can be found within the JNCC Marine Recorder snapshot from the National Biodiversity Network atlas, within the Federal Agency for Nature Conservation's information portal on alien and invasive species in Germany, SWAM (Swedish Agency for Marine and Water Management), or within the French National Inventory of Natural Heritage and in sources listed in Chapter 1.
- Consider which non-native species are present in areas with high connectivity to the donor areas (e.g. adjacent waterbodies, ports, or bays). Is there a high risk of these spreading into the donor site?

Survey the donor site

Once a potential donor site has been identified, the current disease status of the site should be confirmed through further testing. Legally mandated controls and disease testing, as well as biodiversity surveys, take place at insufficient frequency to ensure that the current disease/INNS status of a site is accurately reflected. Restoration practices should therefore endeavour to undertake their own field surveys and testing to ensure that the risk assessments are undertaken with the most current and relevant information.

Screening for disease

Pathogen screens should be done using recommended methods as specified in the OIE aquatic manual and as recommended by the EU legislation (Commission Implementing Decision (EU) 2015/1554 of 11 September 2015 laying down rules for the application of Directive 2006/88/EC as regards requirements for surveillance and diagnostic methods (notified under document C(2015) 6188). This should include all the notifiable diseases for the native oyster: Bonamiosis (*B. ostreae* and *B. exitiosa*) and Marteiliosis (*M. refringens*), as well as oyster herpes virus (see Chapter 1 for list of notifiable diseases). Sample sizes should follow or exceed those recommended OIE aquatic manual and EU legislation. In the aforementioned decision there are specific recommendations about the surveillance and diagnostic of *B. ostreae* and *M. refringens*. In addition to screening listed pathogens, general screening based on histology and bacteriology should be implemented (see EURL SOPs). Consideration should also be given to diseases which are not listed. Attention should therefore be paid to the general health of the oysters and the recent history of mortality at the donor site.

The OIE manual is a ready source of methods for screening, but it should be noted that this document can take considerable time to update and that there may be more appropriate, recent methods available in publication. When possible, the diagnostic analyses should be carried out by internationally recognised laboratories. In addition, it is prudent to use the appropriate national guidelines for disease monitoring to determine the number of individual samples required to 2nd para, sought from the relevant National Reference Laboratory.

Surveying for INNS

When undertaking the biodiversity survey to inform the project risk assessment, particular care should be paid to potential and high-risk INNS. As INNS include a full range of species with differing life histories, no one sampling protocol will be best suited to all potential species of interest. Project managers should therefore consider using a range of methods that cover: 1. species that are likely to have low densities and are dispersed and 2. species that are likely to have higher densities and/or be less patchily distributed. For example, grab sampling nested within drop down video may be suitable for some locations, whereas where the predominant habitats are dominated by epibiota, *in situ* surveys are more suitable, as some high risk species such as *D. vexillum* (see Figure 1.3 in Chapter 1) or *Styella clava*, are difficult to identify on video tows where there is already substantial epiphytic growth.

For detailed assessment of benthic fauna, a standard methodology (e.g. 0.1m^2 Day or Hamon grab, with samples sieved over 1mm sieve, and/or a drop-camera/towed video survey of the seabed) is commonly used by statutory bodies. Project managers can use the JNCC Marine Method Finder to identify suitable monitoring approaches for each habitat.

While biodiversity surveys, in particular in the intertidal, are a good way to engage local volunteers, it is critical that those undertaking the survey are trained to identify all the potential problem species and, equally, have enough knowledge to recognise species that 'don't-look-right' (i.e. a previously unrecorded INNS). This is because there are INNS we do not yet know about (cryptic and/or recent introductions). Furthermore, whilst positive identification for some species can be undertaken during the survey, project managers should ideally also collect samples for a full assessment of non-native species using lab-based specialist benthic taxonomists.

Once the site surveys have been undertaken, the initial risk assessment should be revisited with the updated information in mind. If an aggressive INNS such as *D. vexillum* or a notifiable shellfish disease is recorded at the donor site, then oysters should not be translocated from that site (see Figure 2.5). Should other non-native species be identified from previous data or surveys of the donor site, then a marine biosecurity plan should be written to identify measures that can reduce the risk of those non-native species being introduced. This may be required by regulators before consent is given for the translocation. Guidance on authoring such a plan can be found in Cook *et al.* 2014 (see resources).

Figure 2.7: Individual inspection of each oyster in closed biosecurity holding facilities. Oysters translocated to the restoration site after disease screening, cleaning, surface sterilisation and UV depuration. Photo: Phil Wilkinson/DEEP project.

PREPARING ADULT OYSTERS FOR TRANSLOCATION

Physical cleaning of oysters

If the origin and donor sites have been deemed suitable by the preceding steps, the oysters obtained for translocation should be first inspected and then physically cleaned to ensure no visible epibiota persists (see Figure 2.7). This process should be completed at the donor site pre-transport to ensure no epibiota is transferred elsewhere. It may be necessary to require suppliers to cost for this activity, which may be different to their normal aquaculture practices and restoration practitioners should therefore expect such oysters to be comparatively more expensive. Wastewater from the cleaning can be disposed of at the origin site rather than being transferred elsewhere. It is also recommended that treatment and transport of oysters takes place in the late autumn to late winter to minimise epibiotic growth.

As part of the visual inspection, a record of species present and in what number on the oyster shells should be recorded pre- and post-treatment. This is not only helpful as an audit trail to demonstrate statutory compliance but could contribute to the evidence base for best practice of future restoration work. Note that oysters with associated heavy infestations of boring sponges (e.g. *C. celata*, see Figure 2.8) will have holes which can be difficult to clean. Heavily undermined shells with many crevices should be discarded along with other oysters with physically compromised shells. These should be discarded responsibly at the donor site. If further fouling is found at a later stage, or if cleaning must occur remotely, material should be disposed of responsibly. Under circumstances of enhanced risk, disposal should be to a specified biological waste disposal route (possibly including incineration). During cleaning, care should be taken to ensure that there are no small bivalves hidden in the hinge-line of the oysters such as spat *Mytilus* (that could be the INNS *Mytilus trossulus* in some scenarios).

Physical cleaning can be done by hand (scrape/scrub off) and/or mechanical methods, such as cement mixers or shellfish cleaning machines. If using mechanical methods, large oysters can be tumbled in batches and so it is a more time-effective procedure for a large number. This treatment may not be suitable for smaller oysters.

Figure 2.8: The exterior (left) and interior (right) of a native oyster infested by a boring sponge (*Cliona celata*). Photos: Luke Helmer.

While cement mixers have been shown to be successful at removing epibiota in existing projects, they also found some or parts of organisms, such as holdfasts, may persist. Repeat treatment may be required. It is critical that, if mechanical treatment (as opposed to cleaning by hand) is undertaken, a large sample size of the treated oysters be examined by hand in order to determine that the epifauna have been effectively removed.

Following physical cleaning, oysters should be left to recover in running filtered seawater for a minimum of three days before undergoing chemical treatment. Subjecting them to immediate chemical treatment would put oysters with chipped shells at risk of unnecessary exposure and may result in increased oyster mortality. During this time, oysters also have the opportunity to "depurate" some of their internal microbiota. Disposal of water used in this phase should therefore be subject to biosecurity and chemical pollutant risk assessment and where necessary, treatment before disposal.

CHEMICAL TREATMENT OF OYSTERS

The purpose of chemical treatment is to kill any shell epibiota that may have survived the physical cleaning of the oysters and therefore reduce the risk of INNS transfer (see Figure 2.9). Remaining epibiota might include scraps of clonal organisms such as sponges, sea squirts or certain types of seaweed, as well as hardy spores and resting/reproductive stages of other organisms. As well as the oysters themselves, some organisms such as keel worms, barnacles and other bivalves can clamp-shut to avoid ingress of fluids: they are therefore theoretically able to survive the chemical treatment just as well as the oysters. Care should therefore be taken in the physical cleaning stage to make sure that the tubes of keel worms are removed or broken open, that barnacles are removed or broken open.

Various chemicals have been used for the surface sterilisation of oysters and they range in their expense and availability, including hypochlorite, formaldehyde, and commercial fish-farm treatments such as Virkon. There is not a clear evaluation of the relative effectiveness of different treatments, but the obvious abiding principle is that it should be toxic to the epibiota in the concentration and exposure time used. Exposure-times can vary, and bulk dunking methods have been used, akin to the use of a chip-fryer-basket. Dunking methods may be preferable and efficient with younger oysters (e.g. 10g) because the shells appear to seal-shut well. Sponging oysters with the chemical treatment (whilst using appropriate Personal Protective Equipment) might be deemed more appropriate in larger adult oysters where the gape of the shell may be worn or damaged and therefore less likely to seal well if fully submerged in a chemical bath.

Turrell *et al.* 2018 undertook a thorough review of the literature regarding chemical treatments of oysters in order to develop recommendations for moving *C. gigas* from an aquaculture site with a high risk INNS (*D. vexillum*). A complete review of the tested options and the resulting impacts on the target INNS and the shellfish are provided in their report. The method recommended for field tests as a result of the review was immersion in freshwater (salinity < 2ppt) for at least 24 hours.

Quarantine

Once oysters have undergone both physical and chemical external cleaning, it is important that the efficacy of the biosecurity protocol is quantitatively assessed and that internal contaminants are given an opportunity to be expelled. A quarantine period should therefore be imposed, during which the oysters are given time to recover from the treatment under controlled conditions, given time to be depurated of internal microorganisms, and can be monitored to assess the efficacy of treatment thus far. Given that it is not yet known whether the oysters have been successfully cleaned, water used in this period should be handled as potentially high-risk waste and should be disposed of accordingly. At this stage, oysters may be kept in closed circulation or flow through systems. Filtered water from the receiving site may be used for this stage. Ideally moderate numbers of oysters should be kept in each tank, and tanks should not share water circulation. In circumstances of high risk, it may also be desirable to use artificial seawater and u/v recirculation systems to 'flush' in-shell water and

Figure 2.9: Intermediate storage of scraped and rinsed oyster broodstock before their chlorination bath. Photo: Bérenger Colsoul.

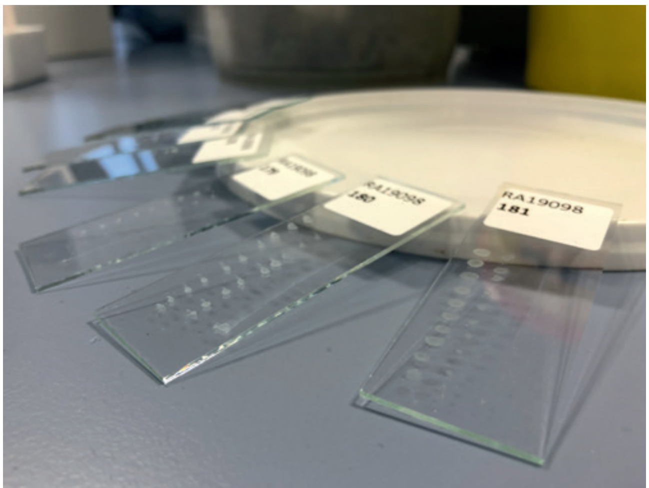

Figure 2.10: Smears of native oyster's heart for *Bonamia* screening. Photo: Chantelle Hooper.

decontaminate the oysters from waterborne organisms (**note:** "decontamination" will not "flush out" diseases). During this time individual batches of oysters can be screened and assessed independently (see Figure 2.10). The bottom of the tanks should be checked daily for evidence of recently dead organisms that may have crawled out of crevices or from within the shells. Anecdotally, it seems that slightly reduced salinities may encourage this process. Should any evidence of living associated biota be found, oysters should be subjected to further treatment. It should be noted that currently there is insufficient evidence regarding the effective duration of a quarantine period, but project-experience suggests that remnant live epibiota may take 3 days to emerge under these conditions, therefore a shorter period is not recommended.

Preparing spat for translocation

In the case of translocation of spat (settled larvae > =2mm or juvenile oysters < 10mm), it should be considered that the sensitivity of the young oysters may mean that many biosecurity treatments (both physical and chemical) are inappropriate and locally sourced spat from sites with the same (or better) disease and INNS status may be the only appropriate option. The relevant authorities should be informed throughout the process.

Spat on shell (or other substrate) is likely to have less well-established biofouling relative to adult oysters, having spent less time in the water. However, as young oysters are more sensitive to both physical and chemical cleaning methods, and the shell or substrate can be a vector of unwanted organisms, the potential risks associated with translocation of spat on shell are great. Therefore, as dictated by the precautionary principle, **spat which have been in contact with open water prior to translocation, should only be translocated locally**. Nevertheless, it is critical that the following ground rules be followed for all translocations: Without exception, animals must only be moved to recipient sites from donor sites with equal or higher health status.
For example, moving native oysters from an OsHV-1 µvar positive to negative site, or moving animals from an area with close proximity to a Bonamiosis positive zone, should not take place.

Finally, it should be noted that cultch used in wild settings for translocation should be subject to the cultch biosecurity protocol below before being placed in the spatting pond or other structure.

DEPLOYING OYSTERS

Once all the steps have been taken to identify possible biosecurity risks and to address identified risks, it is critical that the effectiveness of the measures is assessed prior to oysters being relayed into the receiving site. Only when the restoration practitioner and that relevant authorities are confident that the associated risk is acceptable, should the translocation be completed (see Figure 2.11). Figure 2.5 provides an overview of the steps at which risks should be assessed.

Practice due diligence

There is currently no method that, when applied, renders living oysters completely biosecure for translocations. Although general protocols for cleaning aquatic organisms exist (see resources for some examples), protocols suitable for relaying of live oysters for ecological restoration have yet to be tested and confirmed effective. There are outstanding knowledge gaps regarding the efficacy of possible treatments, in particular if efforts involving translocations are scaled up. **It is therefore critical that each translocation attempt validates the efficacy of the biosecurity measures undertaken with a thorough screening of the treated oysters. The sample size should be large enough to ensure a high degree of confidence that the consignment of oysters has met the desired biosecurity standard.** Screening for epifauna should, as a minimum, involve visible examination of the shell and hinge. Screening for associated biota should as a minimum involve examination of the base of quarantine tanks for signs of recently emergent and dead individuals for several days.

Figure 2.11: Transport and relaying of oysters during restoration project. Photos: Åsa Strand.

While disease screening is one of the first steps undertaken in determining whether the stock is suitable for translocation from an approved donor site, a further and final screening for diseases may be undertaken before the stock are released into the wild. The rationale for this further final testing is that it is possible that oysters start to express the disease when under stress (e.g. having undergone treatment). Therefore, tests taken towards the end of the quarantine period may pick up disease presence overlooked in the initial stages.

Translocating cultch

When considering cultch translocation, it is critical that the following questions are posed.

1. **Is translocation necessary or are there local sources?** If possible, use cultch from local sources and environments to reduce the risk of introducing novel diseases or species.
2. **If translocating, consider the following factors when determining where to source cultch:**
   i. Do not accept any cultch from donor sites with high-risk invasive species or diseases are not present at the receiving site.
   ii. Use appropriately 'weathered' land-based or cooked/heat treated sources where possible (see Figure 2.12 and Figure 2.13).
   iii. Ensure that all cultch material is safe with regards to heavy metal content and other toxins.
   iv. Minimise the physical distance between the donor and receiving site.
3. **What is the waste designation and associated legislation for selected cultch?** If the cultch material is shell, cooked or otherwise, it will be necessary to weather the shell to ensure that all residual biology is rendered inert. There may be legislation pertaining to the storage of shell and the use of "animal waste". Check with the relevant authorities regarding waste management regulations.

Risk assessment

The first step in assessing the appropriateness of the identified source of cultch, is to undertake a risk assessment. If sourcing cultch from a known marine location, a survey of the cultch donor-site should be undertaken, following the same protocol as outlined for donor sites of live oysters (see above). For example, cultch material from a location with a known notifiable disease or high impact invasive species should only be considered for use in a site with a similar or lower status.

Clutch can also be purchased from aggregate suppliers. In such cases either the exact origin of the cultch may be unknown, or impractical to survey. In these cases, the supplier should be asked to provide information regarding whether the cultch material has been heat treated or weathered for any length of time or if any kinds of contaminants (heavy metals, organic chemicals, etc.) are present. Where this information is unknown, the material should be treated as though it were freshly extracted, and appropriate treatments applied.

The case study from the Essex Native Oyster Restoration Initiative (see Box 2.3) provides further details project managers may want to consider.

Treatment of cultch

Non-local and marine derived cultch material (shells or stones) must be treated in order to ensure that living marine organisms, spores or resting stages are not unwanted contaminants of the material. What is deemed suitable treatment should be agreed with the relevant authorities. One cost effective means is to weather (expose to the elements) the material for a minimum of 12 months, turning the shells every two months where material is deposited < 15m height, and twice monthly if deposited more deeply.

Assessment of effectiveness

As with all other stages of biosecurity practice, it is the responsibility of the project to ensure that the treatment has been effective in removing any unwanted organisms and spores. This may include visual examination of the material. As a general guideline, material should be weathered until there is no evidence of residual biology remains, dried or otherwise. An effective method of assessment and the appropriate sample size for assessing the status of the clutch material should be agreed with the relevant authority, whereas the methods outlined above have been shown to be effective under some circumstances, there is a limited scientific basis for establishing exact and reliable guidelines. There is therefore an emphasis throughout this document on practitioners assessing each step of the process. In order to optimise treatments moving forward, and promote cost effectiveness in restoration, further research is needed.

BOX 2.2: CONTRIBUTE TO IMPROVED BIOSECURITY GUIDELINES

Rendering living oysters' low risk for translocation from a biosecurity perspective is costly both in terms of time and money. The efficacy of actions and the investment required, in particular with regards to time and manpower, is not well documented. We therefore urge projects to submit their experiences to the NORA Secretariat or the Native Oyster Network – UK & Ireland.

Figure 2.12: Scallop shell weathered for a year and with no residual biology. Photo: Bill Sanderson, DEEP.

BOX 2.3: CASE STUDY: ESSEX NATIVE OYSTER RESTORATION INITIATIVE, U.K.

The Essex Native Oyster Restoration Initiative (ENORI) is a collaboration working towards the Essex estuaries having self-sustaining populations of native oysters. The restoration location is a designated Natura 2000 site and is a nationally important Marine Conservation Zone (MCZ). The MCZ is both substrate and recruitment limited for oysters. The project is unusual in benefitting from an adjacent oyster fishery that provided a locally-adapted broodstock, so translocation from further afield was not necessary. It was, however, necessary to translocate cultch from outside the waterbody to the restoration site. *B. ostreae* is present at this site.

Aggregate gravels were used to provide elevation off the seabed with shell cultch added as a top layer (see Figure 2.14). As it was not possible to confirm the source of marine gravels, a land-source aggregate was chosen from River Terrace Deposits quarried locally to the restoration site. For a local source of shell, a shell recycling initiative was set up to return the shell of the local fishery oysters. This was considered the lowest biosecurity risk for cultch translocation as the shell was originally removed from the same waterbody as the restoration site. However, there is a limited supply of local oyster shell and so other options were considered to achieve the volume of cultch required. Scallop, cockle, and blue mussel shell were sourced from national and European suppliers. Dialogue with suppliers was critical to confirm (where possible) the exact source of the shell and to understand risk. When shell is bought from an aggregate company, it should be noted that it is often not possible to know with certainty the geographical source as they are the 'middle-man' of the supply chain. Information such as how the shell had already been treated (if by heat for commercial shellfish processing, at what temperature and for how long) and how it was stored (location and duration) was also ascertained from suppliers.

Although this information was gathered where possible, it was agreed by ENORI that any shell (regardless of source and heat treatment) should be weathered outdoors, exposed to the elements, for a period of 12 months to ensure as far as possible that hitch-hikers and pathogens would not persist. Samples of the shell were visually inspected for living matter.

It is advised to secure storage sites as early as possible to avoid delays to deployment or multiple cultch handling and transport costs. The volume of cultch required can be substantial and to store it in a relatively thin layer and turn bi-weekly or monthly is a considerable undertaking. The resources (space, contractors, vehicles and potentially volunteers) that are required to deliver this should be considered as early in the planning process as possible and, importantly, built into project budgets.

Throughout the planning of the pilot restoration works, ENORI sought advice from shellfish health and INNS experts to ensure that the most appropriate risk management approach was adopted. On the advice of Cefas, a project record was set up that listed all the aggregate and shell used, the source, the treatment prior to delivery, the duration and location of storage, rotations and any risks associated with source and the restoration site. This was a valuable exercise for ensuring the appropriate steps were being taken by the project to minimise biosecurity risk and for any audits that may take place in the future.

Figure 2.13: Cultch for substrate enhancement, being weathered outside awaiting deployment. Photo: ENORI/ZSL.

Figure 2.14: Deployment of gravel cultch by Essex Native Oyster Restoration Initiative, UK. Photo: ENORI/ZSL.