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Haringvliet
Naar een dynamische delta

Recommendations for flat oyster restoration in the North Sea

Synthesis of lessons learned from the Dutch Voordelta experiments, with additional observations from flat oyster pilots in Borkum Reef and Gemini wind farm, modelling exercises and literature

April 2019

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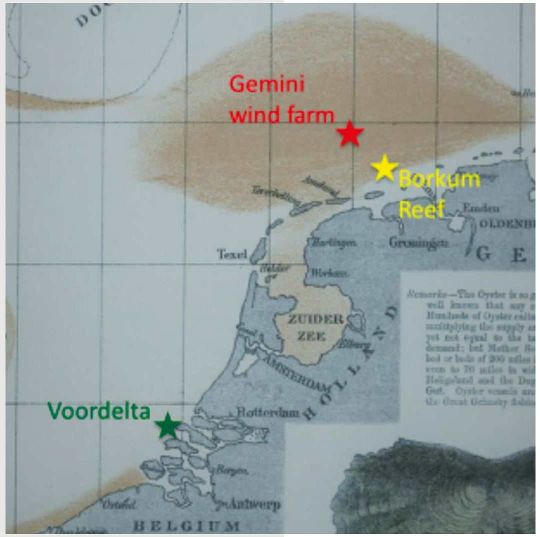


Figure S.1 Locations of the three pilots which are the basis of this report, projected onto the oyster reef areas (orange shadings) as indicated by Olsen (1883).

The main objective of this report is to provide recommendations for flat oyster restoration pilots in the North Sea. These recommendations are derived from three years of pilots and experiments with flat oyster (*Ostrea edulis*) restoration in the Voordelta area, with additional information from the Borkum Reef and Gemini wind farm restoration pilots, plus other relevant studies and model exercises. Various sponsors have supported these projects, but the most important among these are WWF Netherlands and ARK Natuurontwikkeling.

Figure S.1 shows the location of the three pilots. The underlying reports can be downloaded from www.ark.eu/schelpdierbanken.

The key objective for these pilots is to 'kick-start' autonomously growing, or at least self-sustaining, flat oyster reefs in the North Sea. Of course, we are not there yet, so pilots are still necessary. The reason for restoring flat oyster reefs is to stimulate biodiversity of the North Sea ecosystem.

The recommendations are based on our current knowledge of factors governing flat oyster restoration. An important knowledge gap is the critical surface area and density of a self-sustaining flat oyster start-population in the North Sea. This can only to be solved by more pilots and experiments at various locations, accompanied by monitoring ('learning-by-doing').

The recommendations are limited to general (i.e. non pilot-specific) factors governing flat oyster reef restoration.

The recommendations are:
For **location selection** of flat oyster restoration pilots within the Dutch part of the North Sea, in probable order of importance:

- Bottom disturbance caused by human activities, such as bottom-trawling fishery, sand or gravel extraction, and construction work should be completely absent. Possibly, even noise and/or vibrations caused by these activities are detrimental to flat oysters.
- The sediment should be firm enough to support flat oyster beds, i.e. fine or coarse sand and gravel are suitable, whereas silt is probably less suitable.

- Prolonged periods of oxygen depletion, as for example caused by thermal stratification, should be absent.
- The sea floor should be sufficiently stable and bottom shear stress should be sufficiently low since these factors will disturb oyster settlement and growth. Areas with proper conditions can be found in the northern and southernmost part of the Dutch North Sea.
- Net water current should be as low as possible to stimulate larval retention in the area. These areas are located in the southernmost and northern part of the Dutch North Sea.
- Primary production should be sufficient enough to provide food for the oysters. There is a high to low productivity gradient roughly perpendicular to the Dutch shore. However, it is not clear how far from shore this becomes limiting to the oysters.

Presence of empty shells ('shelliness') is probably important too, since this may facilitate settlement of oyster larvae. We did not identify distribution information of shelliness on the North Sea floor, but a bottom survey will easily reveal the amount of shelliness present before a pilot.

For **source selection of the flat oysters** as a seed population in a restoration pilot:

- Since the North Sea is considered to be free from the *Bonamia* disease, the source population should originate from areas free from this disease.
- The source location of flat oysters used for restoration pilots is of limited relevance to its success, as long as they are in good health at the moment of deployment.
- In principle, young oysters ('spat') produced in a hatchery should be used for the seed population, in order to avoid overharvesting and introduction of invasive alien species. However, if these are not available, employing adult oysters may be considered, but then strict protocols against overharvesting and introduction of alien invasive species must be adhered to.

It appeared that the supply of *Bonamia*-free young flat oysters in Northern Europe was too scarce to satisfy the demand for the pilots discussed in this report. Fortunately, several hatchery/nurseries are being created (among others, supported by WWF). These need to be scaled up in order to bridge the gap between supply and the rapidly growing restoration-based demand.

Oysters from a Bonamia-free area have not developed resistance against the disease, so that mortality will probably be high if the *Bonamia* infection reaches the population. This circumstance can never be completely excluded. That is why the development of a disease-free but resistant or tolerant population is crucial.

To ensure that the oysters are in **good health** when deployed:

- Due to reproduction, flat oyster condition is low between May and October. Therefore, avoid this period when harvesting as well as deploying them.
- Transport and storage before deployment should be done in cool conditions and minimised in time.
- In any case: oysters should be tested for health (e.g. by measuring the shell weight to flesh ratio of a sample) shortly before they are deployed.

If there is doubt about the appropriateness of the location, it is important to install a flat oyster **test population** of 300-600 specimen when starting the restoration pilot. The test population should be regularly checked for its survival, growth, condition, and development of reproduction organs (gonads). The oysters in this population therefore must also be measured before deployment and contained in robust racks, with meshes as wide as possible in relation to the size of the deployed oysters.

A cost-efficient method for extension of the pilot may be to employ 'spat-on-shell' around the seed population: young oysters which are allowed to settle on shell substrate. This practice is widely used in other oyster restoration projects, particularly in the USA. However, we do not know the survival rate of flat oyster spat-on-shell in North Sea waters, so this must be tested in practice.

Flat oyster settling can be stimulated by deploying clean and empty bivalve shells around the seed population, with the following provisions:

- The deployed shell substrate can consist of various types of bivalve shells (mussel, cockle, oyster).

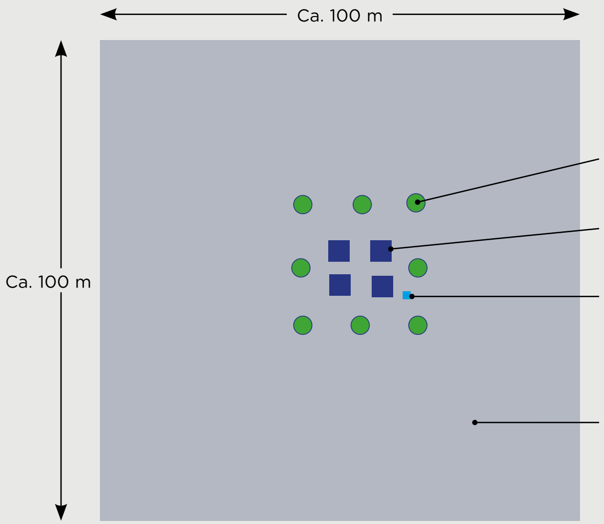


Figure S.2 Recommended basic design of a flat oyster restoration pilot.

- The shells should be clean and free of live epibionts.
- The cleaned shells should be deployed within two weeks of the first peak in larvae swarming. The latter can be determined by regular water sampling in and around the test population and subsequent laboratory analysis for larval identification and counts. This peak may also be predicted with a temperature-sum model, but this must be calibrated per location.
- If there are already many bivalve shells present on the sea floor at the pilot location it may not be necessary to deploy extra shells, but this must be tried out in practice.

Settlement can be determined by putting **'spat collectors'** into the water in or around the pilot at regular time intervals during the summer period. This is relatively costly and time-consuming, so an alternative method is to survey substrate in and around the pilot by an **ROV with camera**, by the time the young oysters have grown to at least one cm in size.

Based on these recommendations, we have developed a **standard design for a restoration pilot** (schematically depicted in Figure S.2):

- If it is not certain that the conditions at the pilot location are appropriate for flat oysters, try this out with a test population, as described above. If the conditions are known to be appropriate, the seed population can consist of oysters loosely strewn on the sea floor. A combination is possible too: loosely strewn oysters around the racks with the test population.
- If not already present, distribute empty and relatively clean shell material around the pilot, within two weeks of the first peak in larvae swarming (to be determined by larvae sampling).
- Deploy artificial reef structures in or around the pilot, in particular for enhancement of general biodiversity, protection against disturbance, and traceability of the pilot.
- Deploy and collect spat collectors for monitoring of flat oyster settlement in the first pilot years or, when newly settled oysters have grown, use an ROV with camera.

Once successive analysis of the test population shows that pilot conditions are appropriate for flat oysters, large-scale extension of the pilot can be undertaken by deploying extra oysters (possibly in the form of spat-on-shell) and substrate.

Given the various uncertainties inherent to a flat oyster restoration pilot, it is essential to accompany it with monitoring. We present a full monitoring programme. Whether this is achievable depends on the available budget in combination with the specific pilot objectives.

Our recommendations for a full **monitoring programme** of flat oyster pilots are:

- Regularly inspect the installation and oyster racks (if used) for damage and remove growth of attached organisms (if necessary, clean and/or repair).
- Continuously record environmental data at the pilot location near the sea floor: temperature, food availability, oxygen level, and current speed.
- Measure survival, growth, gonad development, and condition of the test population, i.e. the flat oysters in the racks, and if possible also oysters on the sea floor.
- During the reproductive period (June-August) sample water in the pilot, near the bottom, and analyse for concentration of flat oyster larvae when planning to deploy settlement substrate.
- Deploy and collect spat collectors, and later use ROV with camera, to determine flat oyster settlement in and around the pilot.
- Survey development of biodiversity in and around the pilot, among others to investigate ecosystem services provided by the oyster reef.

These guidelines are in line with the monitoring recommendations by the Native Oyster Restoration Alliance (NORA, 2017).

Given the uncertainties about flat oyster restoration, **we recommend a pilot duration of about 10 years** (if achievable). **This period may be roughly divided into four phases, with the following recommendations per phase:**

Phase 1 **Preparation and deployment: year 1**
T_0 monitoring, prior to deployment. Deployment period: well before or after the summer period. Start with a test population if it is not known whether local conditions are suited for flat oysters (go to phase 2), otherwise start with higher amounts of oysters and substrate (go to phase 3).

Phase 2 **Test phase: years 1 to ca. 3**
Perform monitoring and maintenance of the test population. Monitoring focuses on survival, growth, reproduction, and the occurrence of recruitment (SGRR) as such, and on environmental parameters. Extra oysters and/or settlement substrate can also be deployed, but this entails risks as long as SGRR are not yet proven to be sufficient. Based on the SGRR results in this phase, a stop/go decision can be made.

Phase 3 **Extension phase: years ca. 3 to 6**
If it is decided to continue, pilot extension can be undertaken by deploying more oysters (possibly in the form of spat-on-shell) and/or more substrate. Monitoring emphasis can shift away from survival, growth, and reproduction, focussing more on recruitment and the reef's spatial distribution around the pilot, as well as the development of biodiversity.

Phase 4 **Reef development phase: years ca. 6 to 10 (or longer)**
Gradually decrease measures aimed at extension of the reef, to investigate whether the reef maintains or grows by itself. Continue environmental, recruitment, and biodiversity monitoring. Consider measures to enhance ecosystem functions and structure if keystone species are missing (especially those which are functional in maintaining the reef).

Mainly due to expensive ship time, working in offshore conditions is extremely costly. Hence we recommend combining monitoring, maintenance, and extensions visits to the pilot as much as possible.

1 Introduction

1.1 Background and history

In their feasibility study of flat oyster (*Ostrea edulis*) restoration in the Dutch North Sea, Smaal *et al*. (2015) concluded that the time was right to start flat oyster restoration in the Dutch part of the North Sea. The main reasons behind this conclusion were, and still are:

- Probably due to improved health, the flat oyster population shows signs of autonomous extension in the Dutch Delta area (in the extreme southwest of The Netherlands), where it has been kept in culture since about a century ago.
- The areas without bottom-trawling fishing activities will probably increase in the North Sea, due to the current extension plans of wind farms and Marine Protected Areas. Bottom-trawling fishery is destructive to epibenthic flora and fauna, including flat oyster reefs.

Almost simultaneously with the Dutch study, a German feasibility study (Gercken & Schmidt, 2014) was undertaken, coming to similar conclusions.

2015 also saw the start of the Haringvliet Dreamfund Project, aiming at restoring an estuary, which is part of the Delta area, in the southwest of The Netherlands. The project includes flat oyster reef restoration in the Voordelta, which is the seaward extension of the Delta. While looking for a suitable location to start restoration pilots, we discovered a mixed Pacific oyster (*Crassostrea gigas*) and flat oyster reef near the Brouwersdam that separates Lake Grevelingen from the Voordelta (Christianen *et al.*, 2018). The flat oysters are probably the offspring of aquaculture flat oysters in the Grevelingen. This discovery confirmed that flat oysters are spreading autonomously beyond their original culture area.

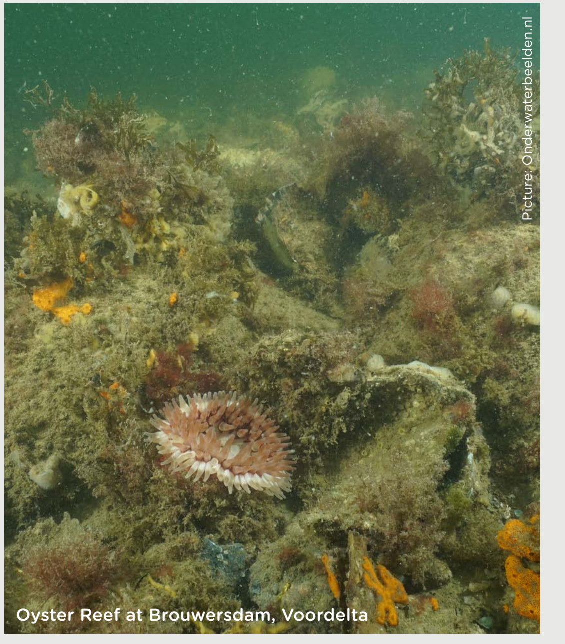
During the past three years, co-created and mainly funded by ARK Natuurontwikkeling and WWF Netherlands, we have worked on a method for extending and creating shellfish reefs in the Voordelta, with an emphasis on flat oysters. We did this by studying the structure of this reef, by performing various experiments, and by starting an additional restoration pilot in the area (Sas *et al.*, 2017, 2018a; Didderen *et al*, 2019a). This has helped to identify several of the steering factors of flat oyster reef development and restoration. In a sense, the Voordelta has become a 'field laboratory' for flat oyster reef restoration. This is an essential asset, since the factors influencing development and sustainability of wild flat oyster reefs in the marine environment are largely unknown. Moreover, the reefs in the North Sea disappeared before anyone realised that they were worth studying. Fortunately, oysters have been kept in culture, in the Delta area as well as in other inlets and estuaries around the North Sea, which gives us some basic information about the flat oyster life-cycle and the initial population for restoration attempts (in the Delta only in that very area, due to *Bonamia* infection; see Chapter 4). However, knowledge of how to maintain and extend an oyster culture in the sheltered coastal zone is something different from restoring wild oyster reefs in the open North Sea.

The Voordelta oyster reef discovery and the results of the experiments have drawn wide-spread attention and have stimulated various organisations to start surveying other areas for flat oyster presence and restoration potential (e.g. Fariñas-Franco *et al*., 2018). This resulted in the discovery of a small population in the western Wadden Sea (Van der Have *et al*., 2018a). After successful pilots nearshore, and drawing on the experience of these pilots, in spring 2018 two new and relatively large oyster reef restoration pilots were started in the deeper waters of the Dutch North Sea: in the Borkum Reef area and the Gemini wind farm (Didderen *et al*., 2019b,c). WWF is the main driver behind the Borkum Reef pilot. The Gemini pilot is an initiative of Jan Groot, WWF and the Gemini 'Buitengaats' consortium. Monitoring in these pilots yielded important additional results, which we have therefore used for this synthesis, together with results of relevant modelling exercises and scientific literature.

1.2 Objectives and scope of the report

The main objective of this report is to present recommendations for flat oyster restoration in the North Sea (offshore as well as nearshore), based on the research results over the past three years. In this, we do not distinguish between restoration 'pilots' and 'attempts'. A pilot is intended to try out certain locations and methods, whereas a true attempt is aiming at actually restoring a population at a certain location. But in both cases, the initiator aims to get insight in the critical success factors for the population to survive and proliferate, for which our recommendations are intended.

The recommendations should be considered as 'work in progress' towards guidelines for native oyster reef restoration in the North Sea, since there still are uncertainties. Firstly, the various factors behind flat oyster recruitment (i.e. spat settlement and growth to a size at which most oysters can survive and reproduce) are not all known. Recruitment is crucial to maintenance and growth of an oyster reef. Secondly, and related to this, we do not yet



Oyster Reef at Brouwersdam, Voordelta

know how extensive, in terms of oyster density per surface unit and total surface area, a reef should be in order to sustain itself and/or expand in the open North Sea. These uncertainties can only be filled in through more, well-monitored experiments and restoration attempts, at different locations, with differing areas, oyster densities, and time scales.

In this circumstance, it is important to mention that the number of flat oyster restoration pilots/attempts in the North Sea is still increasing. For instance, in autumn 2018 a pilot was started in the Luchterduinen offshore wind farm, initiated by a consortium of NGO's and companies (The North Sea Foundation, Natuur & Milieu, Eneco,

Wadden Sea
Flat oysters found in 2017
Hatchery/nursery started in 2018

PROGRAMMA NAAR EEN RIJKE WADDENZEE | Natuurmonumenten
NIOZ | WWF | Jan Groot

Wind farm Luchterduinen
Started in 2018

Eneco | Van Oord Marine ingenuity
Stichting De Noordzee | Natuur & Milieu | asn bank

Voordelta project
Started in 2016

Haringvliet Naar een dynamische delta | ARK Natuurontwikkeling | WWF
provincie Zuid Holland | Ministerie van Landbouw, Natuur en Voedselkwaliteit | Ministerie van Infrastructuur en Milieu | Port of Rotterdam | LIFE IP } Deltanatuur

Wind farm Gemini
Started in 2018

Gemini | Jan Groot | WWF

Borkum Reef
Started in 2018

WWF | ARK Natuurontwikkeling

Overarching projects:

Hatchery/nursery of Bonamia free flat oysters

Zeeland's Roem | Wageningen University & Research | Ministerie van Landbouw, Natuur en Voedselkwaliteit

EcoFriend: innovative monitoring in wind farm pilots

TKI Wind op Zee Topsector Energie | Gemini

Figure 1.1 Current flat oyster restoration projects in the Dutch North Sea area, projected onto the flat oyster reef map by Olsen (1883). The orange areas indicate the historic flat oyster reefs.

ASN Bank, and Van Oord). In addition, there are various hatchery/nursery initiatives in order to produce the growing number of oysters required.

See Figure 1.1 on page 12 for a complete overview of current pilots and other relevant flat oyster initiatives in the Dutch North Sea and coastal waters, projected onto the flat oyster reef map produced by Olsen (1883). As can be seen, most projects are within, or close to, the historical oyster reefs.

Other important flat oyster restoration examples in the North Sea area and nearby Channel are:

- The pilot near Helgoland, undertaken by the German Alfred Wegener Institute (AWI) at Bremerhaven (Pogoda, 2019), based on the positive conclusions by Gercken & Schmidt (2014).
  See: https://www.awi.de/en/science/biosciences/shelf-sea-system-ecology/main-research-focus/european-oyster.
- The Dornoch Environmental Enhancement Project (DEEP), an initiative to restore Native European oysters to the Dornoch Firth by the Glenmorangie whisky distillery, Heriot-Watt University, and the Marine Conservation Society. See: www.theglenmorangiecompany.com/about-us/deep
- The Solent Oyster Restoration Project (SORP), aiming at restoring the native oyster to the Solent, the strait that separates the Isle of Wight from mainland England, by the Blue Marine Foundation in partnership with the Universities of Portsmouth and Southampton, Ben Ainslie Land Rover BAR, and MDL Marinas. See: www.bluemarine-foundation.com/project/solent
- The Essex Native Oyster Restoration Initiative (ENORI) on the UK east coast aims at supporting self-sustaining populations of native oysters within several estuaries there. See: www.essexnativeoyster.com

The increased interest in flat oyster restoration led to the establishment (in 2017) of the Native Oyster Restoration Alliance (NORA), a European network aiming at reinforcement and restoration of the native European flat oyster. Network members are representatives of governmental agencies, science, non-governmental organizations, as well as oyster growers and other private enterprises.
See www.noraeurope.eu for general information and www.noraeurope.eu/restoration-projects for information on various restoration initiatives.
Given the growing interest in flat oyster restoration, we assume that more flat oyster restoration pilots will be initiated in the North Sea area. This synthesis and the related recommendations are intended for all those who want to give it a try, and for those who are trying. Its scope is to give guidance based on our key field work results, with emphasis on the general (i.e. not pilot-specific) factors governing flat oyster reef restoration. Such factors are mostly of a biological and ecological nature, though some information on costs and legal factors are also general and therefore included. A description of costs, permits, design, etc. specific to an offshore wind farm pilot is given in Sas *et al.* (2018b).

1.3 Brief description of contents

In **Chapter 2**, we describe the overall strategy of flat oyster restoration we are currently following. We have already set important steps, but a much larger effort is required before we attain the objective of self-sustaining flat oyster reefs in the North Sea.

Chapter 3 describes the flat oyster life cycle and the steering factors we humans can employ to achieve reef restoration.

Chapter 4 describes the pilots and experiments in Voordelta, Borkum Reef, and Gemini wind farm.

Chapter 5 provides the recommendations we have derived from these pilots and experiments, model results, and literature data.

Chapter 6 shows how a flat oyster pilot can be set up, monitored, maintained, and expanded.

2 Objectives and overall strategy of flat oyster reef restoration

Flat oysters once covered a large area of the North Sea, as depicted in Figure 1.1. The map illustrates that flat oysters can survive and reproduce in offshore areas, up to 60 metres water depth and maybe even more. The importance from the viewpoint of nature restoration is that flat oyster beds provide a natural hard-substrate habitat in a predominantly soft-bottom marine environment and influence other environmental conditions as well by filtering, providing, and recycling nutrients, providing shelter and possibly also by storing carbon.

An oyster reef is a species assemblage with higher species richness than a sandy or silty sea bed. In the Voordelta mixed oyster reef we observed that in quadrats with oysters the number of epibenthic species was 60% higher compared to adjacent sand patches (Christianen *et al.*, 2018).

For North American waters, field surveys and ecological modelling showed that fish and mobile crustaceans thrive on oyster beds (with Virginia oysters, zu Ermgassen *et al.* (2015)). We assume that this is the case for the North Sea too. Hence, North Sea marine life, including fish and mobile crustacean production, is expected to increase when flat oyster reefs are restored. Of course, this must be checked by monitoring, once a reef is established. Taking measures to enhance biodiversity should even be considered if keystone species are missing (especially those which are functional in maintaining the reef).

Other offshore shellfish reefs (e.g. horse mussels *Modiolus* modiolus) were, as far as we know, never so extensive in the southern North Sea. Flat oyster reefs are therefore considered to have the highest potential for enhancing marine biodiversity.

Given adequate conditions, shellfish reefs should be able to maintain themselves and expand, which gives them an extra characteristic that artificial hard substrate does not have.

Our key objective in flat oyster restoration is to ‘kick-start’ self-sustaining reefs in the open North Sea, with all the naturally associated species and ecosystem services. In other words, after an initial stimulation phase, it should not

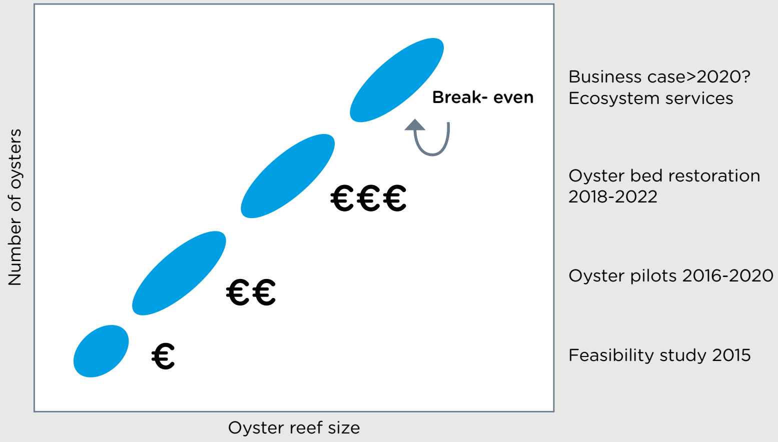


Figure 2.1 Illustration of the time-line and scaling up of oyster reef restoration in the Dutch North Sea.

be necessary to add new or other specimens and biodiversity should develop. Obviously we are not there yet, but this is the direction restoration practices should go and for which the recommendations of this report are i ntended.

The overall strategy of scaling up flat oyster restoration attempts is depicted in Figure 2.1 below. We are currently in the second phase, in which restoration attempts of the scale of 1 ha are initiated and in which we also attempt to extend the existing Voordelta oyster reef (which spreads across about 40 ha).

Intuitively, we suspect that at the single ha scale, flat oyster reefs are not yet self-sustaining. The required dimension lies probably somewhere around 1000 ha (with sufficient oyster density, i.e. at least five individuals per m2; see par. 3.3). This, in turn, will require major investments. But ultimately flat oysters create important ecosystem services, so in the long run gains probably outweigh costs. One way to approach this is to monetize ecosystem services, such as offshore oyster culture and increased stocks of fish and large mobile crustaceans (Figure 2.1).

Costs aside, we consider oyster reef restoration as a part of good stewardship of the North Sea.

3 Flat oyster life cycle and reef restoration steering factors

3.1 Flat oyster life cycle

Flat oysters have a complex life cycle shared by all members of the genus Ostrea (Figure 3.1). After a short pelagic period (6-10 days for flat oysters), oyster larvae settle permanently by cementing themselves to hard substrate. In the six-month period after settlement they are called 'spat' (Walne 1974).

In the first three years they function as males (protandrous life cycle) and in subsequent years they can function alternately as females or males. This sex change depends on environmental conditions (temperature, food). The males produce sperm clumps (spermatozeugmata; Ó Foighil *et al.*, 2000) which after spawning are inhaled by the females. The females are larviparous, that is, the eggs are present in the cavity and after fertilization the larvae are brooded by the female, 8-10 days in flat oyster. After brooding, the larvae swarm into the water column in June-August.

The critical success factors during the life cycle are survival rate (larvae, spat and mature oysters), growth rate, sex organ (gonad) development, larvae production, and recruitment of new oysters into the population by successful settlement of larvae as spat, as already indicated in Figure 3.1.

These success factors are summarized as the Survival, Growth, Reproduction, and Recruitment (SGRR) conceptual model: a self-sustaining or growing oyster reef will develop if there is sufficient survival, growth, and reproduction of mature oysters and recruitment of young oysters into the local population.

3.2 Steering factors in oyster reef restoration

Restoration can be defined as: "The process of establishing or re-establishing a habitat that, in time, can come to closely resemble a natural condition in terms of structure and function" (Baggett *et al.*, 2014). Shellfish (flat oyster) reefs can be defined as intertidal or subtidal biogenic structures formed by shellfish (flat oysters) living at high densities and building a habitat with significant surface complexity.

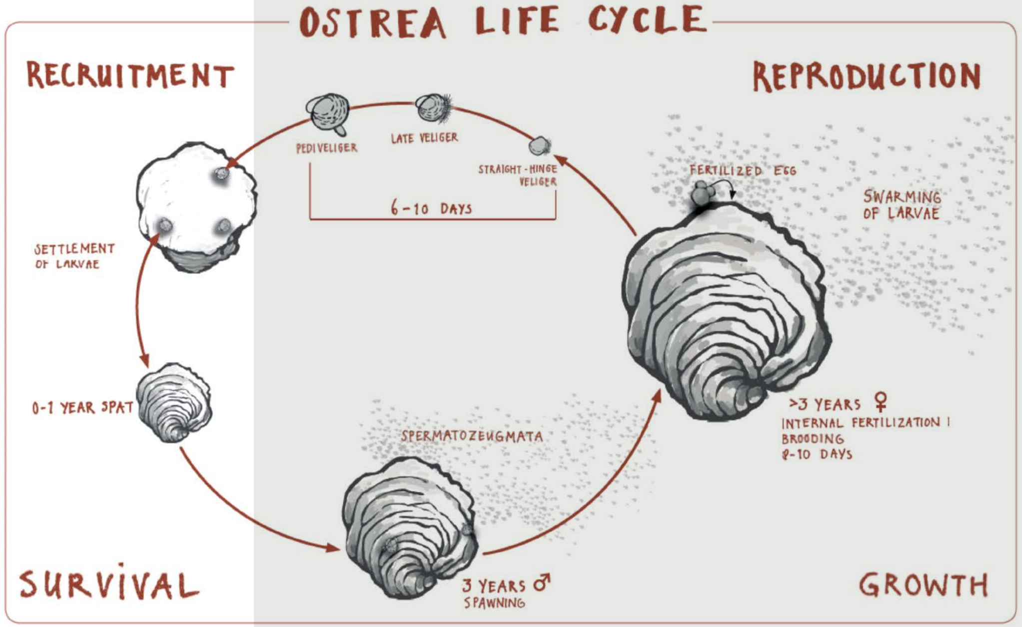


Figure 3.1 The general life cycle of an Ostrea oyster.

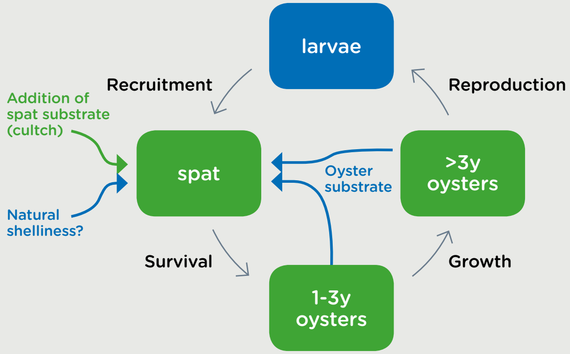


Figure 3.2 Important steering factors of flat oyster bed development (in green): deployment of live oysters (of both sexes and different ages) to produce larvae and deployment of substrate for settling of these larvae. Blue arrows indicate that oysters (1-3 y and >3y) and natural shelliness also provide settlement substrate.

Several experts insist on using the term oyster 'bed' instead of 'reef'. We stick to the term 'reef' however, to underline that flat oysters create a three-dimensional structure on the sea bed.

Shellfish reef restoration is successful if high densities with substantial structural complexity occur and maintain themselves. In the case of flat oysters, such a reef should have a minimal density of five individuals per m^2 (OSPAR, 2000).

This can be accomplished, in principle, by influencing key steering factors in the oyster life cycle (see Figure 3.2). These include (Beck *et al.*, 2009):

- Providing or adding a 'seed population', i.e. a mix of oysters of both sexes to 'kick-start' a population, by producing larvae, if the population is recruitment limited (which is the case in the North Sea).
- Providing settlement substrate for spat if the population is substrate limited (which may not be the case in some areas of the North Sea).
- Both if oysters and suitable substrate are absent.

Choice of a presumed optimal location is an additional steering factor.

In Chapter 5 we will elucidate more specific recommendations for these steering factors, based on the Voordelta, Borkum Reef, and Gemini pilots, as well as additional sources.

4 Pilots: locations and design

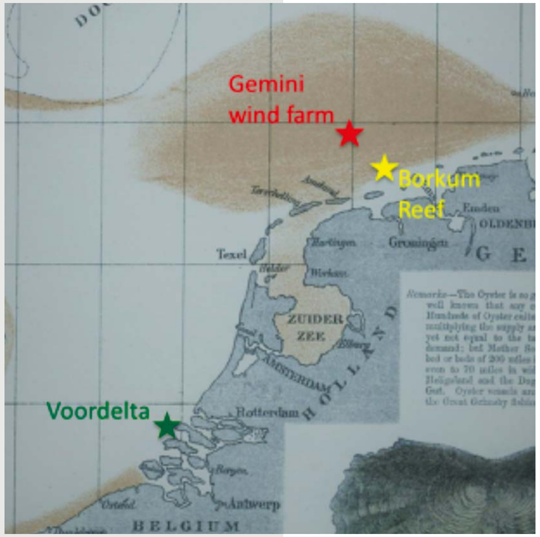


Figure 4.1 Locations of the three pilots which form the basis of this report, projected onto the Olsen (1883) map

4.1 Introduction

This synthesis is based on the results of experiments and pilots in the Voordelta (2016-18), with additional information from the Borkum Reef and Gemini oyster restoration pilots (2018). The locations of these pilots are presented in Figure 4.1.

Full information about these pilots and experiments is presented in the annual reports. These are:

- Voordelta:
  - Sas H., Kamermans P., Have T. van der, Lengkeek W. & Smaal A. (2017) Shellfish reef restoration pilots Voordelta: Annual report 2016.
  - Sas H., Kamermans P., Have T. Van Der & Christianen M. (2018a) Shellfish reef restoration pilots Voordelta: Annual report 2017.
  - Didderen, K., P. Kamermans, A. van den Brink, T.M. van der Have, J.H. Bergsma, H. van der Jagt, W. Lengkeek, M. Maathuis, H. Sas (2019a) Shellfish bed restoration pilots Voordelta, Netherlands, Annual report 2018.
- Borkum Reef: Didderen, K., P. Kamermans, W. Lengkeek (2019b) Borkum Reef oyster pilot, results 2018.
- Gemini wind farm: Didderen, K., P. Kamermans, W. Lengkeek (2019c) Gemini wind farm oyster pilot, results 2018, BuWa report 19-012.

These reports can be downloaded from https://www.ark.eu/natuurontwikkeling/dieren/schelpdierbanken

4.2 Voordelta experiments and pilots

The Voordelta is a nature conservation area and part of the EU Natura 2000 network. A map of the designated Natura 2000 area is presented in Figure 4.2.

We performed our main pilots and experiments near the Blokkendam location (see Figure. 4.3). The main characteristics of this part of the Voordelta are:

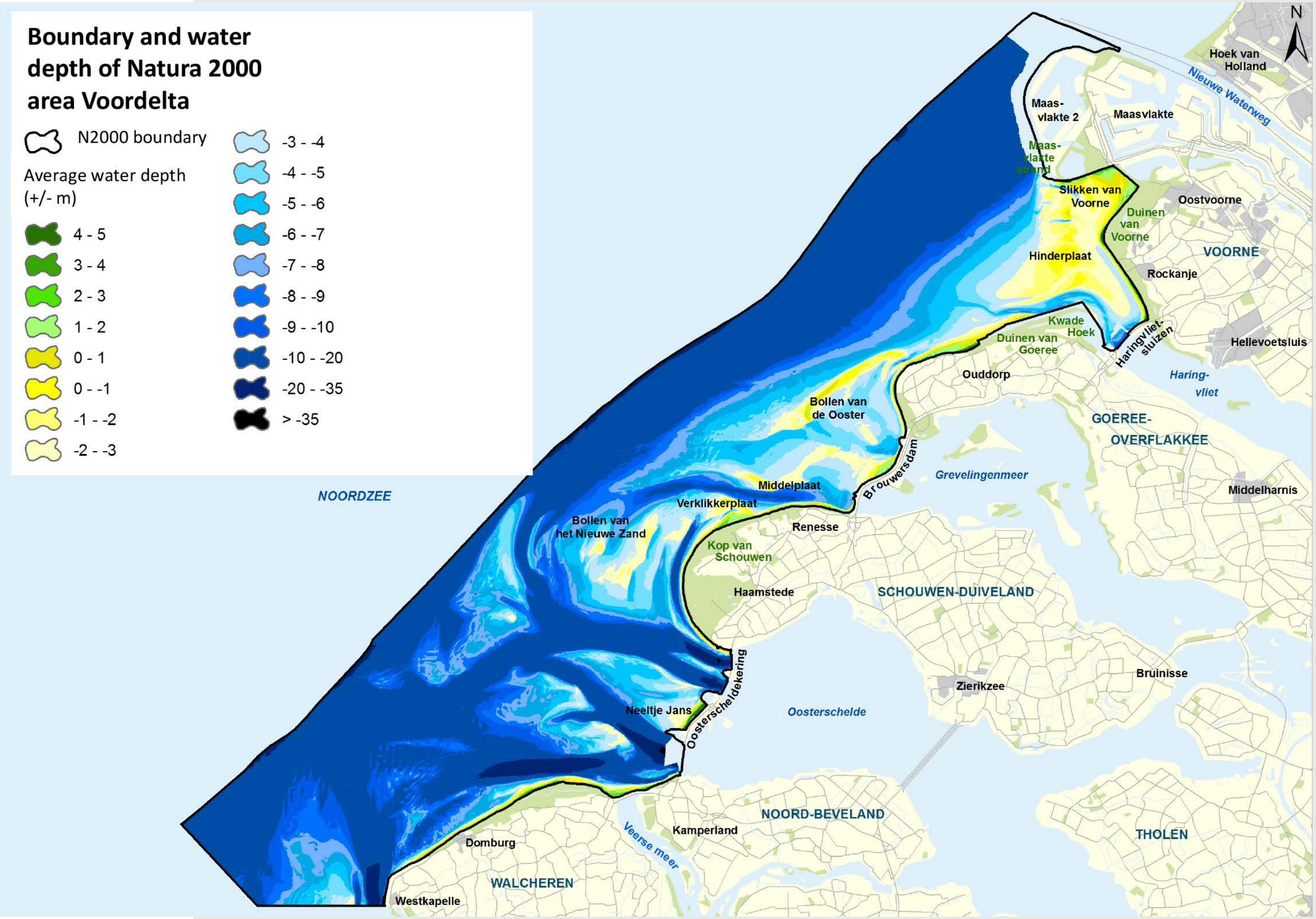


Figure 4.2 Map of the Voordelta Natura-2000 area (Rijkswaterstaat & Royal Haskoning/DHV).

- The area is shallow, with water depth varying from 1 to 6 metres.
- There is a mixed flat oyster and Pacific oyster (*Crassostrea gigas*) reef present near Blokkendam location. Shrimp fishery is the only type of bottom-trawling fishery in the Voordelta area, but it is even absent near the Blokkendam, due to the shallow water depth and the presence of large stones.
- The oyster density varies strongly within the reef. Its area is at least 40 ha. Most probably, the flat oyster larvae originally came from nearby Lake Grevelingen (Figure. 4.3), a saline lake with limited tidal range and in open connection via the Grevelingen outlet to the Voordelta since 1998. In this lake, flat oysters occur naturally and on culture plots. As a result, in early summer, flat oyster larvae can be detected swarming in the Voordelta area.
- Other shellfish species also occur in the area, such as razor clam (*Ensis sp.*), surf clam (*Spisula sp.*), soft shell clam (*Mya arenaria*), and blue mussel (*Mytilus edulis*).

Figure 4.3 Map with relevant pilot and experiment locations in the Voordelta area.

As stated in Chapter 1, the main objective of the overall Voordelta project (2016-2018) is to develop a method for extending and creating shellfish reefs. In order to attain this objective, secondary objectives are:

- Understand the critical factors for formation and development of these reefs.
- Identify suitable locations for shellfish reefs in the Voordelta.

Specific objectives and related activities for the experiments are (see Sas *et al.*, 2017 and 2018a; Didderen *et al.*, 2019a):

- To measure survival, growth and reproduction of flat oysters. This was done by deploying racks with a defined population at two different locations: Blokkendam and Hinderplaat. Conditions for flat oyster survival near Hinderplaat appeared to be unsuitable, probably due to large and sudden fresh water inflows from Haringvliet. Therefore, it was decided to stop monitoring at this location.
- To unravel the dynamics of spat settlement as a basis for recruitment of flat oysters, since this constitutes the ‘Achilles heel’ of the establishment of flat oyster reefs. The main question here is: How can flat oyster spat settlement be stimulated and how can this knowledge be used to create new flat oyster reefs? A related question is: Which substrate is most suitable for flat oyster spat settle-

ment? These questions are addressed by monitoring the occurrence of flat oyster larvae in the water column and deploying substrate when flat oyster larvae appear to be present, as well as by mapping the substrate used by flat oysters in the Blokkendam reef.

- Based on the results of the above experiments:
  - To attempt to extend the reef, by timely deploying substrate at locations considered suitable.
  - To develop a new pilot, on the lee (east) side of the tidal flat Bollen van de Ooster. This location was chosen because shellfish surveys conducted earlier showed the presence of living flat oysters and because fishery or other bottom-disturbing activities are not allowed there. The pilot was started in 2018.
- To map the biodiversity associated with the oyster reef, in order to demonstrate its value for marine nature development.

The oysters employed for the Voordelta pilots usually came from the region itself: Lake Grevelingen, Eastern Scheldt, and sometimes from the reef in the Voordelta. These are infected with *Bonamia*. So, we tested for *Bonamia* in the Blokkendam reef and it is indeed present in that population (Sas *et al.*, 2018). Therefore, in this case, oysters infected by *Bonamia* can be used for reef restoration. A Dutch regulation is being prepared limiting the deployment of Bonamia infected oysters to this part of the Voordelta only.

We also performed a survival and condition test with Norwegian oysters, from Hafrsfjord (see below for source description).

4.3 Borkum Reef pilot

The Borkum Reef pilot, which was started in spring 2018, is a larger-scale restoration attempt. It consists of:

- A small number of oysters in racks, to allow monitoring of growth, health, and development of gonads (reproduction organs) of this subset.
- Artificial sandstone reefs with and without oysters.
- About 80,000 oysters loosely distributed on the sea floor, within an area of approximately 1 ha.

See Didderen *et al.* (2019b) for full description of the pilot. It is performed with Norwegian oysters. The same regulation as mentioned in par. 4.2 prohibits the deployment of *Bonamia*-infected oysters in the open North Sea because this is considered as a *Bonamia*-free zone. The Norwegian oysters are free from the Bonamia disease. They originate from wild oyster reefs, located in relatively shallow water (up to 5-metre depth) and are harvested by divers. A sustainable harvest protocol was set up and maintained by WWF and the oyster company.

The oyster population contains a naturally occurring variety in age classes. The use of different age classes is important, since such a population is productive. Young oysters (up to three years of age) are all males, while older oysters can both be male and female (see par. 3.1).

Water depth at the pilot location is about 25 metres. The pilot is located about 20 km from shore. The artificial reefs have various functions: protection and references points for the racks, settlement substrate and elevation form the seabed for oysters, substrate and/or shelter for other species. Local fishermen have agreed to avoid the area with bottom trawling equipment and work together with WWF and ARK regarding monitoring and maintenance.

4.4 Gemini wind farm pilot

The pilot in the Gemini windfarm (see Didderen, 2019c), which was also initiated in spring 2018, is of smaller scale than the one in Borkum Reef. It is performed with about 15,000 Norwegian oysters from the same batch as used for the Borkum Reef project. A small part of these was also put in racks, while the majority was loosely strewn in and around wind farm monopile scour protection stones. Water depth varies between 28 and 36 metres. The wind farm is 85 km from shore.

5 How to restore a flat oyster reef: lessons from the Voordelta, Borkum Reef, Gemini wind farm pilots and other sources

5.1 Habitat requirements for restoration pilots

Smaal *et al.*, (2015) argued that abiotic conditions (including bottom shear stress and stability, current velocity, salinity, sediment composition, temperature, oxygen availability, water depth, and absence of bottom disturbance) and biotic conditions (diseases, food availability, predators) should allow flat oyster growth and reproduction in large parts of the North Sea nearshore and offshore.

A primary condition for oyster reef development is the total absence of seabed disturbance in the designated reef area, including trawling fishery, construction works, and gravel/sand extraction. Possibly, even noise and/or vibrations caused by these activities are detrimental to flat oysters (Tamis *et al.*, 2019).

In model studies, Smaal *et al.* (2017) and Kamermans *et al*. (2018a,b) investigated other relevant conditions for flat oyster restoration pilots in the Dutch part of the North Sea. We recapitulate these below, with additional information from the pilots in Borkum Reef, Gemini and some German experiments, to provide recommendations for optimal conditions and locations for restoration attempts/pilots.

The relevant conditions are:

- Sediment composition without too fine fragments: the sediment should be firm enough to support flat oyster beds, i.e. fine or coarse sand and gravel are suitable, whereas silt is probably less suitable. This is the case in most parts of the Dutch North Sea. Possibly with the exception of (large parts of) the Oyster Grounds. Explanations for this could be that the organic part of this silt was produced by the oyster reefs that were once present there, and/or that currently sedimenting algae are no longer consumed by oysters.
- Presence of empty shells ('shelliness') or other hard substrate probably facilitates settlement of oyster larvae. The type of shell is probably of little importance (see par. 5.4). We did not find information on spatial distribution of such substrates on the North Sea floor, but a bottom survey will easily

Oyster reef at Brouwersdam

reveal the amount of shells or other hard substrate present before a pilot.

- A sufficiently stable sea floor: a mobile bottom will disturb oyster settlement and growth. The model studies mentioned above showed that, in the northern and southernmost part of the Dutch North Sea areas, a sufficiently stable bottom can be found.
- A sufficiently low sea floor shear stress: a high shear stress can damage oyster reefs. The model studies showed that this should not pose a problem outside the nearshore area. Borkum Reef is located relatively near to shore, but current velocity was measured and not deemed a limiting factor (Reuchlin, 2018).
- A low net sea water current, in any direction: current will cause dispersion of flat oyster larvae. Hence, they will tend to drift away from the parent reef, leaving it insufficiently rejuvenated. At present the magnitude of net current, which causes larvae to drift away too far, is based on model predictions. In addition, the settling rate of flat oyster larvae and size of the reef will also play a role (see par. 5.3). But if there is a choice, the areas with lowest possible net current should be selected for restoration pilots. A tidal current, moving to and from the pilot (hence: without a net current) will be less problematic and occurs everywhere in the North Sea anyway. Such locations do exist in the Dutch part of the North Sea, as the model studies show. In particular, there is a very low net current in the Gemini wind farm. At Borkum Reef, modelling shows a moderate net current to the east.
- Absence of prolonged periods of oxygen depletion, as for example caused by thermal stratification. Lack of oxygen is lethal to all epibenthic flora and fauna, although flat oysters appear to be relatively resistant (Smaal *et al.*, 2015). Choosing a location without prolonged stratification periods is therefore preferred. The model studies show that it may occur in a zone just to the north of the Dutch Wadden Sea. There are indeed indications that temporarily stratification occurs during spring and summer in the Gemini wind farm. Fresh water stratification occasionally occurs in a narrow band along the Dutch coast, hence that phenomenon is less relevant to the open North Sea.
- Sufficient primary production, to provide food to the oysters: there is a high to low productivity gradient roughly perpendicular to the Dutch shore. The mentioned model studies indicate that food limitation could occur far from shore, though not just how far. In illustration, the condition of the oysters in Borkum Reef (close to shore) appears to be slightly better than those in the Gemini wind

Deploying substrate at Voordelta

farm (85 km from shore). Pogoda *et al*. (2011) also report a moderate oyster condition near Helgoland, which is about 55 km from the German mainland, while oysters at stations more inshore showed a better condition.
A factor which may, in time, cause problems for flat oyster reef development in wind farms is the high number of mussels attached to monopiles. These compete with the oysters for phytoplankton in the water column. With the planned growth of the wind farm area, this could possibly lead to food limitation for the oysters (Kamermans *et al*., 2018), but that is a hypothesis that remains to be tested.

- Minimal suspended sediment concentration: a high concentration of suspended inorganic particles (according to current estimates above ca. 50 mg/l) can interfere negatively with oyster filter feeding. The mentioned model studies did not yield Dutch North Sea areas with too high suspended sediment concentrations for flat oysters.
- Sea water temperature near the bottom between 3 and 30 °C: flat oysters require a winter temperature not below 3 °C and summer temperature not above 30 °C. This is probably not exceeded anywhere in the Dutch part of the North Sea.
- Sedimentation rate not too high: a high rate tends to bury the oysters. There are no North Sea data or model studies of (the influence of) this factor.
- Low predation pressure, in particular of starfish and crabs: predation may pose a severe threat, specially to young oysters. There are no indications that parts of the Dutch North Sea are particularly infested with these predators, though they may of course be attracted to flat oyster reefs and their associated organisms. So, the influence of this factor must be determined in practice.

The Voordelta pilots, with the Hinderplaat location in particular, showed that a high freshwater flow is lethal to flat oysters, but this is obviously only relevant at locations very near to shore and at main river outlets.

In summary, we recommend the following qualitative criteria for location selection of flat oyster restoration attempts/pilots, in order of probable importance:

- Bottom disturbance by human activities (such as trawling fishery, sand/gravel extraction, and construction works) should be completely absent.

- The sediment should be firm enough to support flat oyster beds (i.e. fine or coarse sand and gravel are suitable, whereas silt is probably less suitable).
- Prolonged periods of oxygen depletion should be absent (as for example caused by thermal stratification).
- The sea floor should be sufficiently stable and bottom shear stress should be sufficiently low. Such areas can be found in the northern and southernmost part of the Dutch North Sea.
- Net water current should be as low as possible. These areas are again located in the southernmost part and in the northern part of the Dutch North Sea.
- Primary production should be sufficient; in far offshore areas it may become limiting to flat oyster growth.

The sea floor at the chosen location should be surveyed before deployment, in order to ascertain that its basic physical conditions are met (as summarised in the above recommendations).

5.2 Oyster sources and treatment prior to deployment

***Bonamia* free**

Currently, a Dutch regulation is being prepared which specifies rules for the oysters to be deployed as a start population. It specifies that:

- In the open North Sea (since this area is considered to be disease-free) the oysters should originate from areas that are free from *Bonamia* and *Marteilia*.
- In a small section of the Voordelta (since *Bonamia* already occurs there) the oysters can originate from an area which is infected by *Bonamia*.

In the North Sea area, *Bonamia* is considered to be a higher threat than *Marteilia*; therefore our practical recommendations concentrate on the former.

Oysters from a *Bonamia*-free area have not developed resistance against the disease, so mortality will probably be high if the *Bonamia* infection reaches the population. This circumstance can never be completely excluded. That is why the development of a disease-free but resistant or tolerant population is crucial.

Wild versus hatchery oysters

The oysters deployed in the Borkum Reef and Gemini wind farm are harvested from Hafrsfjord in Norway. This is a wild population, of various age classes. The principal reason for ARK/WWF employing these oysters is that the required amounts of flat oysters were not available from a hatchery at the time the pilot was starting. Secondary reasons are that only a population containing various age classes is immediately fertile (see par. 3.1) and that oysters from a wild population are genetically more diverse than from a hatchery.

It is less desirable to harvest and deploy wild flat oysters for restoration purposes: firstly, because wild oyster reefs are already rare, secondly because alien invasive species can be attached to them. That is why NORA strongly advises against harvesting and deploying wild oysters for restoration purposes (see www.noraeurope.eu/wp-content/uploads/2018/Berlin-Oyster-Recommendation-Part-1).

Nonetheless, ARK/WWF are convinced that the Norwegian oysters sourced from Hafrsfjord for the purpose of the Borkum Stones and Gemini projects pose no risks of overharvesting or introduction of invasive alien species. That is because:

- The Norwegian government, IMR research institute, and the company selling the oysters state there is no overharvesting and that the harvest method (by hand, with divers) does not damage the population. A sustainable harvest protocol was developed and is maintained.
- WWF and ARK have commissioned development of a treatment protocol for wild adult oysters. This includes mechanical and chemical (bleach) treatment of the oysters before transport, as well as a quarantine period (Van der Have *et al.* 2018b). The protocol was applied to all oysters deployed in the Borkum Reef and Gemini pilots. The Dutch government has granted an oyster import permit based on the adoption and application of the protocol (Didderen, 2019b).

Test cases for Norwegian oysters

Before deploying the Norwegian oysters, it was unclear whether they would survive and grow in the open North Sea, since they were transferred from a relatively shallow and quiet fjord to much deeper water, with different current speed, temperature, light regime and probably a different algae population too. In addition, the transport process from Norway to the Netherlands was deemed to be stressful. However, monitoring two months after deployment revealed that survival rate in Borkum Reef pilot was high and surviving oysters grew well, showed gonad development and a good condition. Several female oysters had eggs and larvae were present in several oysters, which proves that spermatozeugmata were released into the water column and eggs within females were successfully fertilized. In addition, flat oyster larvae were detected in the water column just above the oysters. To our knowledge, there is no other flat oyster population nearby. The AWI pilot in particular is located north of Helgoland (cf. www.awi.de), more than 100 km away. Hence, it is almost certain that these larvae are the offspring of the flat oysters deployed in Borkum Reef pilot.

As an extra test, the Norwegian oysters were deployed in the Voordelta. It appeared they survived rather well there too and surviving specimen showed an equal or even better condition than oysters from the region itself (Eastern Scheldt and Grevelingen).

In summary, the experiments show that the deployed flat oysters rapidly adapted to a very different environment. Pogoda (2017) confirmed this finding: young French hatchery oysters, introduced in the German offshore part of the North Sea, also did well there.

The recommendations we derive from this section are:

- In the open North Sea, Bonamia-free oysters must be employed.
- The source location of oysters to be deployed for restoration attempts/pilots is probably of limited relevance to its success. As long as they are healthy when deployed (see par. 5.3).
- In principle, young oysters (‘spat’) produced in a hatchery should be used for the seed population, in order to avoid overharvesting and introduction of invasive alien species. However, if these are not available, it may be considered to employ adult oysters, but then strict protocols against overharvesting and introduction of alien invasive species must be adhered to.

In the near future, the sourcing problem of *Bonamia* free oysters may be solved by the following developments:

- Wageningen Marine Research and hatchery company Roem van Yerseke have developed a method to grow *Bonamia*-free flat oysters from a *Bonamia*-infected population (Kamermans *et al*., in prep). As indicated above, this is a very important development.
- WWF, ARK and Roem van Yerseke are setting up a hatchery with *Bonamia* free oysters.
- The Royal Netherlands Institute for Sea Research (NIOZ) at Texel, supported by WWF/ARK, Jan Groot and the Programme towards a Rich Wadden Sea, is also setting up a hatchery, with Wadden Sea flat oysters. It is yet to be determined whether these oysters are *Bonamia* free; a preliminary test indicates that they may indeed be free from the infection.
- AWI is setting up a hatchery with *Bonamia*-free oysters at its Bremerhaven laboratory.

We do not know yet which attempt will yield sufficient amounts of *Bonamia*-free/*Bonamia*-tolerant oysters in the near future.

5.3 Seed population: age structure, condition, test population and extension

‘Seed population’
As explained in par. 3.2, we call the initial population which is deployed to ‘kick-start’ an oyster reef the ‘seed population’.

Age structure
As set out in par. 3.1, the age structure of the seed population is important, since oysters less than 3 years old are males only. After three years they can become females. Probably the latter produce more eggs when they grow larger. If the restoration effort aims at immediately starting a reproducing population, it must contain a sufficient number of female oysters. We do not know the optimal sex ratio, but we assume that a significant amount (estimated at about 25%) should be female. Since adult oysters can be male as well as female, the seed population should contain about 50% adult oysters to obtain a about 25% female population. Of course, the guidelines on

harvest and treatment as set out in par. 5.2 must be adhered when using adult oysters.

Condition
The seed population should be in good condition. Condition should be estimated (e.g. by measuring the shell weight to flesh ratio of a representative sample) shortly before they are deployed. A good condition ensures sufficient reserves for the oysters to develop gonads and produce gametes, in particular eggs, which are more energy demanding than sperm.

Important condition influence factors are:
- The moment of collection (if from the wild) and deployment: during the reproductive period and a few months afterwards, i.e. from May to October, flat oysters are more susceptible to stress. So, harvest as well as deployment should be avoided in that period.
- Transport and storage before deployment: this should be done under the proper conditions, i.e. packed without water, but with moist paper around the shells, stored in cooled containers at 70C.

Young oysters (less than approximately 3 years old) are particularly prone to stress and therefore show less survival when they are not properly handled and stored prior to deployment (Pogoda, 2011).

Test population
If it is not certain that the conditions at the pilot location are appropriate for flat oysters, a pilot should contain a test population, which is well-defined (i.e. oysters are individually measured and weighted) and retraceable. In order to achieve this, they should be contained, for example in robust racks, with meshes which are as wide as possible in relation to the deployed oysters. These meshes allow water, containing oxygen and algae, to enter and excrements to leave the contained population.


Brouwersdam from above

Picture: Onderwaterbeelden.nl

The racks should be sufficiently sturdy to withstand North Sea conditions and remain stable on the sea floor and they should be retrievable. At least three racks should be deployed per pilot, in order to have enough replicates and to account for damage. We recommend each rack to contain 100-200 oysters.

The racks should be hauled up, at least annually, to be cleaned and to measure survival, growth, development of gonads and overall condition of the oysters.

Extension of the population
We do not know the critical size (e.g. surface area) and oyster density of a self-sustaining population in the open North Sea. This is related to net current speed (with respect to


Picture: Ester van den Doel/Duiken in Beeld

Flat oyster

tide), in combination with flat oyster settlement rate. Settlement rate for flat oysters in the open North Sea is, unfortunately, unknown. This is an important knowledge gap, only to be solved by learning-by-doing, coupled to proper monitoring.

Rodriguez-Perez *et al.* (2019) have done laboratory experiments with flat oyster recruitment success, and observed that the presence of adult oysters (and spat) stimulates settlement. So, it could be that here is a speeding-up mechanism in flat oyster restoration: the more adult oysters and spat are present, the higher the settlement success becomes. But this has to be tested in the open North Sea.

A cost-efficient method for extension of the flat oyster population may be to employ 'spat-on-shell' around the seed population: young oysters, which are made to settle on shell substrate. This practice is widely used in other oyster restoration projects, such as in Chesapeake Bay (see www.chesapeakebay.noaa.gov). Yet, we do not know the survival rate of flat oyster spat-on-shell in open North Sea waters, so this has to be tested in practice. Besides, many practicalities (large-scale production and transport, total costing, etc.) still have to be worked out.

Recommendations derived for starting an oyster population at a new location:

- Age structure: If one wants a productive population immediately after deployment of the pilot it should contain oysters of different age classes (ca. 50% adult). Of course, the guidelines on harvest and treatment as set out in par. 5.2 must be strictly adhered to when using adult oysters.
- The seed population should be in good health, which implies that harvest as well as deployment of the oysters should be avoided in the period May to mid-October. In addition, transport and maintenance before deployment should be done under the proper conditions (packed without water, but with moist paper around the shells, stored in cooled containers 70C).
- If it is not certain that the conditions at the pilot location are appropriate for flat oysters, a pilot must contain a test population, which is well defined and retraceable. A representative number (150 specimen or more) must therefore be contained. The best containment method is racks, with meshes just smaller than the size of the deployed oysters.
- A possible extension method of a restoration attempt/pilot is deploying spat-on-shell around the seed population.

5.4 Stimulation of settlement

Substrate
Clean and empty bivalve shells serve as settlement substrate and therefore stimulate recruitment of oysters. Adding such shells is common practice in oyster reef restoration projects and oyster culture. This principle can therefore also be employed to stimulate settlement in a restoration attempt/pilot.

If the shells originate from another area, epibionts should be killed, for example by boiling the shells in water for at least five minutes.

The experiments in the Voordelta have yielded two important observations (see Sas *et al.*, 2018 and Didderen *et al.*, 2019a):

- The type of bivalve shell is of little importance: flat oysters can settle on mussel, cockle and pacific oyster shells, possibly with a slight preference for the latter two shell types (result of substrate experiments in Voordelta pilot). This was also confirmed by observations on the small Wadden Sea population (Van der Have *et al.*, 2018a).
- The shells should be clean and deployed within two weeks after the first peak of flat oyster larvae in the water.

In the Wadden Sea we observed flat oyster recruitment on various types of naturally occurring empty shells. Hence, if there are many empty bivalve shells present on the sea floor at the pilot location (high 'shelliness'), it may not be necessary to deploy extra shell substrate. But this is not certain, so we recommend deploying extra shell material (at the right time) in and around the pilot in any case.

Timing of substrate supply
We were able to construct a prediction model for the first larval peak in the Voordelta, by employing the 'temperature sum' (also known as growing degree-days, heat units or thermal time). This is the sum of the (average) water temperature per day, if the temperature is higher than a threshold temperature. With a threshold temperature of 7 °C for flat oyster (derived from Mann, 1979), the first results suggest that a temperature sum of 593 degree-days predicts the first Voordelta larval peak with an accuracy of ±7 days (Maathuis, 2018).

The temperature sum is location dependent, since environmental factors such as food availability vary between locations, so this value must be determined per pilot location. The larval concentration can be determined by regular water sampling in and around the seed population, with subsequent filtering with a plankton net and laboratory analysis. Temperature measurement can best be performed with a continuous logger. The temperature sum, which roughly predicts the first larval peak at a specific location, can then be determined easily by combining these data.

Settlement monitoring
Whether settlement actually takes place or not can be determined by deploying 'spat collectors' in the water near the pilot. These collectors are bags with clean, empty shells. They have to be hoisted up after the settlement season, to inspect them for spat. This is relatively costly and time-consuming. An alternative is to wait for several months, to allow the spat to grow to one or more cm in size, and to use a camera coupled to a ROV's to detect young oysters in and around the seed population. Since camera image quality is improving quickly, this is probably the most promising technique.

Recommendations derived from this paragraph are:

- Empty bivalve shells can be deployed to stimulate settlement and subsequent recruitment.
- If shell substrate is deployed, it can consist of all types of bivalve shells. But these should be clean, free of live epibionts and deployed within two weeks after the first peak in larvae swarming. Regular larvae sampling in the seed population and subsequent laboratory analysis can determine the correct moment. It may also be roughly predicted with the temperature-sum model and the average temperatures in spring, but this must be calibrated per location.s
- Whether settlement takes place can be determined by putting 'spat collectors' into the water in or around near the pilot and probably also by surveying in and around the seed population with an ROV equipped with camera, once the spat has grown to more than one cm size.

6 Standard restoration pilot design and monitoring

6.1 Introduction

Based on the recommendations presented in Chapter 5, we have developed a schematic standard design for a new restoration pilot (par. 6.2) and the monitoring programme required to learn from the pilot and improve its results (par. 6.3).

6.2 Standard restoration pilot design

Our standard design for a restoration pilot, based on the recommendations developed in Chapter 5, is as follows:

- If it is not certain that the conditions at the pilot location are appropriate for flat oysters, a pilot should contain a test population, contained in retrievable racks with sufficiently wide meshes, solidly placed on the sea floor. If the conditions are known to be appropriate, the seed population can consist of oysters loosely strewn on the sea floor. A combination is possible too: loosely strewn oysters round the racks.
- If not already present, empty and relatively clean shell material should be distributed around the pilot. This must be done within two weeks after the first peak in larvae swarming (to be determined by larvae sampling).
- Spat collectors can be employed for monitoring spat settlement in the first pilot years. An ROV with camera can be employed for the same purpose, but only after the spat has grown to one cm size.
- Once successive analysis of the test population shows that pilot conditions are appropriate for flat oysters, large-scale extension of the pilot can be undertaken. Probably the most cost-effective way to do this is by distributing collected spat onto empty shells elsewhere ('spat-on-shell') around the pilot, but for flat oysters in the North Sea this method still has to be proven in practice.
- Artificial reefs can be deployed in or around the pilot, in particular for enhancement of general biodiversity, protection against disturbance, and traceability of the pilot.

This basic design is depicted in Figure 6.1.

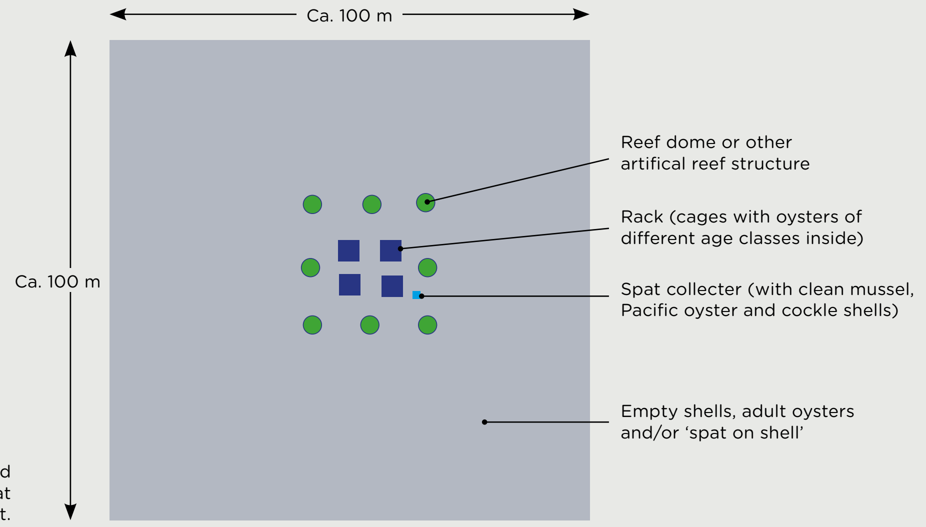


Figure 6.1 Recommended basic design of a flat oyster restoration pilot.

6.3 Monitoring

Given the various uncertainties inherent in a flat oyster restoration pilot, it is essential to accompany it with monitoring. Otherwise, it will be impossible to learn from the pilot experiences and consequently there will be no options for improvement.

For completeness sake, we present the full programme. Whether this is achievable depends on the available budget in combination with the specific pilot objectives.

Our recommendations for a full monitoring programme are:

- Survey the 'before' situation: determination of water depth, suitability of site, and biodiversity prior to deployment.
- Regularly inspect oyster racks (if used) for damage and remove growth of attached organisms (if necessary clean and/or repair).
- Continuously record environmental data at the pilot location near the sea floor: temperature, food availability, oxygen level, and current speed.
- Measure survival, growth, gonad development, and condition of the test population (i.e. the flat oysters in the racks) and, if possible, also a sample of the oysters on the sea floor.
- Sample water in the pilot, near the bottom, and analyse for concentration of flat oyster larvae.
- Deploy and collect spat collectors and/or use ROV with camera to determine settlement, in and around the pilot.
- Survey development of biodiversity in and around the pilot, for its own sake and to investigate ecosystem services provided by the oyster reef.

These guidelines are in line with the monitoring recommendations of the Native Oyster Restoration Alliance (NORA, 2017).

Sea slug, *Thecacera pennigera*

6.4 Recommended activities in subsequent pilot phases

Given the uncertainties about flat oyster restoration, we recommend a pilot duration of about 10 years. However, when the pilot is successful, extension to a longer period is desirable since this provides the (still) unique opportunity of studying stimulation and development of a flat oyster reef.

This period may be roughly divided into four phases, with the following recommendations per phase:

1. Preparation and deployment: year 1. Perform T_0 monitoring, prior to deployment. Deployment period: well before or after the summer period, i.e. before May 15 or from October onwards. Start with a test population if it is not known whether local conditions are suited for flat oysters.
2. Test phase: years 1 to ca. 3
   Monitoring and maintenance of the test population. Monitoring focuses on survival, growth, reproduction, and the occurrence of recruitment (SGRR) as such, and on environmental parameters. Extra oysters and/or settlement substrate can also be deployed, but this carries a risk as long as SGRR are not yet proven to be sufficient. Based on the SGRR results in this phase, a stop/go decision can be taken.
3. Extension phase: years ca. 3 to 6
   If it is decided to continue, a pilot extension can be undertaken by deploying more oysters (possibly in the form of spat-on-shell) and/or more substrate. Monitoring emphasis can shift away from survival, growth, and reproduction to focus more on recruitment and its spatial distribution around the pilot and the development of biodiversity.
4. Reef development phase: years ca. 6 to 10 (or longer)
   Gradually decrease extension methods to investigate whether the reef maintains or grows by itself. Continue environmental, recruitment, and biodiversity monitoring. Consider measures to enhance biodiversity if keystone species (which should naturally occur on hard substrate) are missing.

Mainly due to expensive ship time, working in offshore conditions is extremely costly, so we recommend combining monitoring, maintenance, and extensions visits to the pilot as much as possible.

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