

# Handbook for restoration of eelgrass in Sweden



National guideline



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## Handbook for restoration of eelgrass in Sweden

## National guideline

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Handbook for restoration of eelgrass in Sweden

# Preface

The state of the coastal and marine environment needs to be improved. A large number of international and national commitments and decisions raise demands for measures to reduce impact and strain, as well as for restoration of the coastal and marine environment; primarily the Water Framework Directive, the Marine Strategy Framework Directive, the Habitats Directive, and the national environmental quality objective Sea in balance and the living coast and archipelago. An important prerequisite for the restoration work is a well-functioning toolbox, with scientifically based methods.

This manual provides a detailed guideline to eelgrass restoration and addresses all important steps in the restoration process, from site evaluation and selection, consultation and permitting, harvesting and planting, to monitoring and evaluation of results. The methodology is primarily developed for the conditions along the Swedish west coast, but parts can also be applicable in the Baltic Sea and other areas in northern Europe after the methods have been investigated there. The manual forms part of the action program for the Marine Strategy Framework Directive (measures 29, 30 and 31; Swedish Agency for Marine and Water Management report 2015: 30).

Target groups for the handbook are primarily environmental officers and managers of marine coastal environments on county boards and municipalities that organize and handle eelgrass matters, but also business operators whose activities may adversely affect eelgrass as well as consulting companies that may carry out the practical work with eelgrass. The handbook can also form the basis for courses at universities and colleges.

Although well-functioning methods for eelgrass restoration are now available for Swedish conditions, restoration of eelgrass is time-consuming, expensive and associated with uncertainties. Consequently, it is of the utmost importance that the management primarily focuses on protecting the remaining eel meadows, and only as a final measure allows compensation restoration as a solution in exploitation.

The work was initiated by the County Administrative Board in Västra Götaland in 2007 through an international compilation of the state of knowledge regarding eelgrass restoration (Västra Götaland's report 2009:26). Based on this, among other things, the Swedish Environmental Protection Agency in 2010 granted the County Administrative Board funding for a research project on eelgrass restoration in Swedish marine areas (NV Dnr 309-863-10 Nh, HaV Dnr 1514-12). The work has subsequently been developed through the formation of the interdisciplinary research program Zorro at the University of Gothenburg with additional research funding from the university, FORMAS (Dnr 212-2011- 758) and the Swedish Agency for Marine and Water Management (HaV Dnr 2283-14).

A big thank you to all those who contributed with information, data and opinions during the course of the work. The handbook has been produced by a research group from the University of Gothenburg. The group consists of researchers in marine ecology, environmental law and environmental economics. For the part of the Marine and Water Authority and the County Administrative Board, the project manager has been Ingemar Andersson and Ingela Isaksson.

Gothenburg May 2016,

Björn Sjöberg, Head of the Department of Marine and Water Management

As part of international knowledge sharing, the Swedish version of the report (2016:9) has been translated into English (HaV dnr 1477-20). The translation has been done in collaboration with Flora Kent at Scotland`s Nature Agency (NatureScot).

Gothenburg March 2021,

Johan Kling, Head of the Department of Water Management

# Summary

More than 60% of the eelgrass has vanished from the Swedish northwest coast since the 1980s as a result of nutrient pollution and overfishing. Although measures have significantly improved water quality in recent years, no natural recovery of eelgrass has occurred. Instead, the losses of eelgrass continue as a result of e.g. coastal exploitation. Restoration of eelgrass constitutes a potential tool to recreate historic habitats and to mitigate eelgrass meadows that are destroyed during exploitation.

This handbook provides detailed technical guidelines for eelgrass restoration in Scandinavian waters and includes all important steps in the restoration process, from site selection and permit processes to harvest and planting of eelgrass, and monitoring and evaluation of results. The described methods are based on extensive studies carried out along the northwest coast of Sweden, from 2010 to 2015, and are mainly applicable to the Skagerrak – Kattegat area including the Sound. Some of the methods may also be appropriate for the southern part of the Baltic Sea, but complementary studies will be needed before they can be recommended for this area as well.

Although functional methods for eelgrass restoration are now available for Swedish waters it is important to note that eelgrass restoration is very labor intensive, expensive and the results are many times uncertain. When an eelgrass meadow is lost, the physical and biological environment may change so much that it no longer allows eelgrass to grow in the area. It is therefore not always possible to restore a lost eelgrass bed. Hence, it is imperative that environmental managers prioritize the protection and conservation of remaining eelgrass habitats, and only as a last option use compensatory restoration as a measure to mitigate losses caused by coastal exploitation.

A critical first step, before large-scale restoration is initiated, is to evaluate if existing environmental conditions at potential restoration sites allow eelgrass to grow. Monitoring of physical and biological conditions and test-planting of eelgrass should therefore be carried out for at least 12 months prior to selecting a restoration site. The dominant causes of why eelgrass plantings fail along the Swedish northwest coast are poor water quality resulting from site sediment resuspension, disturbance from bottom-drifting perennial algal mats and shore crabs, and shading from ephemeral algae. In general, it is recommended that eelgrass restoration should only be attempted at sites where the light availability at the planting depth is at least 25% of the surface irradiance, and where test-planted shoots show positive growth after one year.

Before any restoration work is started, it is important to contact relevant site authorities to obtain information regarding necessary permits and required communication with stakeholders. For the methods recommended in this manual, only consultation with the County Administrative Board is normally required.

For eelgrass restoration in Sweden, the single-shoot method is recommended where single, adult shoots are harvested and planted by hand, without sediment from the donor meadow, using diving. To reduce winter mortality resulting from ice-scouring or insufficient light, it is generally recommended that shoots are planted in early June, between 1.5 and 2.5 m depth. It is also recommended that shoots are planted 0.25 to 0.50 m apart (equivalent to a planting density of 4 to 16 shoots per square meter) and that the size of the planted area is at least 1000 m<sup>2</sup> to

increase the chances of positive feedback mechanisms from the restored meadow. The recommended methods for harvesting do not result in any measurable impact on the donor meadows, and the planting methods are relatively fast. Studies suggest that 4 divers could harvest and plant 40,000 shoots covering one hectare in 10 working days. During optimal conditions the shoot density can increase 10 times before the winter. Since harvesting and planting is done by hand, the method is likely to limit the size of possible restoration projects to less than 10 hectares per year, which is a very small amount compared to the 1000s of hectares that have been lost along the Swedish west coast since the 1980s.

Thus, the available restoration methods probably cannot alone recreate the historic distribution of eelgrass. However, in combination with large-scale measures that improve conditions for eelgrass growth along the Swedish west coast, restoration at strategically chosen locations may constitute an important complement that could enable and accelerate natural recovery of Swedish eelgrass habitats.

Monitoring of the restored eelgrass bed is critical to evaluate if the goals of the restoration are met, and must be part of every restoration project. This is particularly important in mitigation projects to ensure that no net loss of eelgrass occurs. This handbook recommends that the result of the restoration be primarily evaluated by comparing eelgrass shoot density, biomass and area extent of the planted bed with the same variables in a natural, reference bed over a period of 10 years.

The total cost of restoring one hectare of eelgrass using the recommended methods is estimated to vary between 1.2 and 2.5 million SEK. These values include the cost of site selection for one year and monitoring for 10 years (0.38 and 0.39 million SEK, respectively), which are independent of the size of the restoration project. The cost of harvesting and planting, on the other hand, is directly proportional to the size of the planted meadow, and the shoot density used, and varies between SEK 0.44 and 1.73 million per hectare for the recommended methods. If anchoring techniques need to be used the planting cost could double. Thus, it is important to identify optimal planting methods during evaluation of restoration sites to keep costs down.

Methods for eelgrass restoration using seeds have also been developed for Swedish conditions. However, seed methods cannot presently be recommended due to very high and variable losses of seeds, and high costs. Compared to the single-shoot method, seed methods have higher risks of failure, take two additional years to obtain a functional eelgrass meadow, and are estimated to cost two to three times more with available methods.

# Terms and definitions used in this handbook

Abundance	- Number of individuals of an organism.
Apical shoot	- A fully grown head shoot (see Figure 5.4a).
Biogeochemistry	<ul> <li>The chemical, physical, geological and biological processes and reactions that control the composition of an environment.</li> </ul>
Biotope	<ul> <li>A natural type with natural boundaries where certain plant or animal communities belong</li> </ul>
Bioturbation	<ul> <li>Mixing and transport of sediment caused by activity of animals in or on top of the sediment surface.</li> </ul>
Clonal growth	<ul> <li>Vegetative, non-sexual reproduction that results in offspring (shoots) that are genetically identical to the parent plant.</li> </ul>
Compensation restoration	<ul> <li>The compensation is done through restoration of damaged habitat where the goal is to compensate for all resources and ecosystem services lost so that no net loss occurs.</li> </ul>
Conductivity	- Electrical conductivity that in water is a measure of the salinity of the water.
Connectivity	- Ability of movement by organisms between areas.
DIN	- Dissolved inorganic nitrogen.
Donor meadow	<ul> <li>A natural eelgrass meadow where plant materials (flower shoots or vegetative shoots) are harvested for use in restoration.</li> </ul>
Ecologic restoration	<ul> <li>Restoration where the goal is to restore a degraded ecosystem to its historical condition.</li> </ul>
Ecosystem engineer	<ul> <li>An organism capable of creating or modifying its physical and / or biological habitat, and affecting a variety of other organisms.</li> </ul>
Epiphyte	<ul> <li>A plant that lives on the surface of another plant (e.g. filamentous algae on the surface of the eelgrass leaf).</li> </ul>
Ecosystem service	- Features of an ecosystem that supplies humans with goods or services.
Ecosystem functions	<ul> <li>The biological, chemical and physical processes and constituents of an ecosystem.</li> </ul>
Faeces piles	- Piles formed by the excrement of animals
Fetch	- The stretch of wind has blown over open water.
Fitness	- Degree of genetic adaptation to a biological environment.
GIS	- Geographic information system.
Habitat	- An environment where a particular organism can live.
Hydrodynamics	- Study of the movement of fluid.
Internode	<ul> <li>The 'scars' that form along the rhizome of the eelgrass when some leaves are dropped.</li> </ul>
Intertidal	<ul> <li>Areas of coast that are exposed to air during low tide and are located under water at high tide.</li> </ul>
Invertebrates	- Animals without vertebral column, e.g. crustacean, molluscs, worms.

Kd	- The extinction coefficient of light in water which indicates how quickly the light is absorbed.
Limnic	- Freshwater living.
Lux	<ul> <li>Dimensions of light defined as the total luminous flux (lumens) per square meter.</li> </ul>
Macrofauna	<ul> <li>Organisms that live on and below the sediment surface and are so large that they are caught by a 0.5 or 1 mm sieve.</li> </ul>
Meristem	- A plant's growth zone.
Morphology	- Form / appearance.
Mortality	- Deadliness, fatality.
Orthophoto	- Geometrically corrected aerial image.
PAR	<ul> <li>Photosynthetically Active Radiation. Light metric for photosynthetically active light which includes the wavelengths of sunlight (400–700 nm) that plants can use for photosynthesis.</li> </ul>
Plasticity	<ul> <li>Phenotypic plasticity means that an organism has the ability to change its own appearance depending on the environment they are exposed to.</li> </ul>
Plug method	<ul> <li>Eelgrass restoration method where eelgrass shoots and accompanying sediment is harvested by means of a pipe that is pressed down into the meadow.</li> </ul>
Pore water	- The water found between the grains in a sediment.
Reference meadow	<ul> <li>Natural unaffected meadows as close to the restoration area as possible used as a reference to evaluate the results of eelgrass restoration.</li> </ul>
Regime shift	<ul> <li>Major and sustained changes in structure and function of an ecosystem that is maintained via feedback mechanisms.</li> </ul>
Remote sensing	- Measurements of properties of an area from satellite or aerial imagery.
Resuspension	<ul> <li>Occurs when sediment that has settled on the bottom again swirls in the water mass due to e.g. wave movements or currents.</li> </ul>
Rhizome	- The stem part of plants located below the sediment surface.
Single shot method	<ul> <li>Method for eelgrass restoration where individual shoots are planted by hand without sediment from the donor meadow and without anchoring.</li> </ul>
SSM	- (See single shot method.)
Staple method	<ul> <li>Eelgrass restoration method where eelgrass shoots are anchored in the sediment at the restoration site using the staples that are pressed down over the rhizome.</li> </ul>
Subtidal	- Areas of the coast that are under water both at high and low tide.
TSS	<ul> <li>Total Soluble Solids. Total content of suspended material (in the water mass).</li> </ul>
Turbidity	- A measure of how much particles are in the water, the murkiness of the water.

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# 1 Introduction

## 1.1 Background – eelgrass restoration

Unlike terrestrial environments and wetlands where habitat restoration has been successfully used for over 100 years to recreate or improve damaged environments, seagrass restoration is a relatively young science that still has knowledge gaps and challenges (Paling et al. 2009). Although eelgrass transplantation (*Zostera marina, L.*) has been used as a method to restore damaged or lost eelgrass habitat in the United States since the 1940s, overall survival of planted stocks has been lower than 50% globally (Fonseca et al. 1998, van Katwijk et al. 2009, 2015). However, the relatively poor success is largely due to the use of inappropriate sites or incorrect methods and there are also many examples where eelgrass restoration has been very successful, especially in the USA (Fonseca et al. 1998, Orth et al. 2012, NOAA 2014).

In Europe, the experience is more limited and known eelgrass restoration attempts have only been carried out in the Netherlands and Denmark, but so far no successful large-scale restoration attempts have been carried out (Paling et al. 2009). In the Netherlands, more large-scale projects (> 20,000 shoots) with restoration of eelgrass stocks in the tidal zone have been implemented since the 1980s, but they have generally not been successful. However, dwarf grass (*Z. noltii*) has been restored more successfully in some areas of the Dutch coast and in Spain (van Katwijk et al. 2009, 2015). In the Limfjord in Denmark, small-scale attempts were made to restore eelgrass in the 1990s after the nutrient load in the area decreased. These studies showed that planted eelgrass shoots can have high survival and growth in areas with good water quality, while plantings with seeds largely failed (Christensen et al. 1995). No large-scale restoration efforts have yet been carried out in Denmark, but studies are underway to develop methods where seeds are used (see www.NOVAGRASS.dk). In Sweden, small-scale studies to develop methods for eelgrass restoration with shoots and seeds have been carried out in Bohuslän in NW Sweden since 2010 (plantings of the size of 1–100 m<sup>2</sup>; see below), but no attempts to carry out large-scale restoration (here defined as  $\geq 0.1$  ha) has yet been performed.

## Need for eelgrass restoration in Sweden

Eelgrass meadows are very important and rich habitats on shallow soft sediment bottoms in Sweden that provide nature and humans with several important ecosystem functions and services (Cole & Moksnes 2016). In Bohuslän, more than 60% of all eelgrass has disappeared since the 1980s, which corresponds to a loss of approximately 12,500 ha (Moksnes et al. 2016, Appendix 1). Similar losses have not been indicated in other parts of Sweden where the eelgrass occur naturally. In the Skagerrak and Kattegat area of the North Sea, overfertilization in combination with overfishing has led to increased growth of fast growing epiphytic algae and mats of macroalgae that covers eelgrass meadows. This is considered to be a main reason for the eelgrass's reduced distribution. Although measures have been put in place to reduce eutrophication and the water quality has improved in coastal water in western Sweden for the past 10 years, no recovery of eelgrass has been seen. Instead, a slow loss of the remaining eelgrass meadows continues in Bohuslän as a result of increasing exploitation pressure on shallow coastal areas. In recent years, therefore, interest from managers has increased to use eelgrass restoration as a measure to reduce habitat loss, both by trying to recreate historic habitat and as a compensatory measure when eelgrass is damaged and at risk of disappearing during exploitation or accidents. For example, restoration of eelgrass in the North Sea is one of the new measures in the Swedish Agency for Marine and Water Management's action program for the marine environment in accordance with the Marine Strategy Framework Directive (Swedish Agency for Marine and Water Management 2015). In the fall of 2015, for the first time in Sweden, a court decision was also made where restoration of eelgrass was required as compensation for the losses of eelgrass caused by the Port of Gothenburg during its planned expansion (see Moksnes et al. 2016, section 8.2 for details).

This handbook is a result of studies conducted in Bohuslän 2010–2015 by researchers at the University of Gothenburg in collaboration with the County Administrative Board of Västra Götaland and the Swedish Agency for Marine and Water Management with the aim of developing methods for restoration of eelgrass in Swedish water. Although extensive studies are the basis of this manual, there are still gaps in knowledge regarding methods for large-scale restoration of eelgrass in Scandinavian waters. Part of the recommendations given in the manual may therefore be revised as new knowledge becomes available. For this reason, it is **important that all attempts to restore eelgrass in Swedish water are closely monitored and evaluated scientifically so that the knowledge and methods for eelgrass restoration in Scandinavian areas can be improved.** 

## **1.2 Purpose and delimitations**

The purpose of this manual is to provide a detailed guideline for all steps in restoring eelgrass in Swedish waters. The recommendations given can both apply to cases where eelgrass restoration is being considered as a measure to facilitate the recovery of damaged or lost eelgrass meadows, or as a compensation measure after e.g. court decision that an operator should replace the meadow that has been damaged or will be destroyed, by for example, exploitation. Although the manual is intended for the restoration of eelgrass meadows, and especially for eelgrass in the Skagerrak-Kattegat area of the North Sea, the general advice and recommendations given in the manual may also be useful in restoring other shallow habitats along the coast of Sweden. Especially for other angiosperms, but also for e.g. perennial macroalgae, mussel banks and oyster reefs.

Target groups for the handbook are primarily environmental officers and managers of marine coastal environments at authorities, county administrative boards and municipalities that organize and handle eelgrass matters, but also practitioners who may negatively affect eelgrass as well as consulting companies that may carry out the practical work with eelgrass restoration and monitoring. The handbook can also form the basis for courses at universities and colleges.

The guide is intended for **large-scale restoration of eelgrass** where the goal is to achieve a long-term recovery of a larger eelgrass habitat and its ecosystem services within a coastal area. There is no clear definition in the literature on where the boundary goes for a large-scale restoration project and in this guideline we define it subjectively for projects that aim to restore **at least 1000 m**<sup>2</sup> (0.1 ha) per area. According to the literature, it is at this size that restoration projects are beginning to become more successful, possibly because the restored meadow has a positive impact on environmental conditions. Furthermore, the scope of work on this scale is

becoming so large that it needs to be planned more industrially and requires greater work teams to be executed.

#### Areas where the manual is applicable

The manual's recommendations are most applicable for restoration of eelgrass in western Sweden, especially in Bohuslän where all studies have been carried out. Since similar environmental conditions are found throughout the Skagerrak and in large parts of the Kattegat, the methods are probably also applicable along the entire Swedish west coast down to Öresund, as well as along the Danish coast in the Kattegatt and along the Norwegian Skagerrak coast. In the more exposed parts of the southern Kattegat, waves and strong currents probably need to be taken into account when choosing sites and planting methods.

On the other hand, it is unclear how well the methods work for restoring eelgrass in the Baltic Sea, where eelgrass generally grows more exposed and deeper, or in mixed stocks with limnic flowering plants, and where seed production is very low, possibly due to stress from low salinity or short growing season (Boström et al. 2014). In the Baltic, therefore, restoration must probably be carried out with vegetative shoots. Experimental short-term studies in Kalmarsund and in the Finnish archipelago have shown that planted vegetative shoots have good survival and growth over a 5-week period, but that eelgrass leaf growth is about 50% lower in the Baltic Sea than in Bohuslän (Baden et al. 2010). This means that the establishment and spread of planted meadows would take longer, and that planting with higher shoot density may be required in the Baltic Sea. Although much of the advice given for the west coast is likely to be useful even in the Baltic Sea, supplementary studies need to be conducted in the Baltic Sea environments before applicable methods can also be recommended for this area. In 2016, the County Administrative Board in Kalmar County, in collaboration with Linnaeus University, will start such a project with funding from the Swedish Agency for Marine and Water Management (County Administrative Board in Kalmar County 2016). The aim of the project is to apply and develop the handbook's methods for restoring eel meadows in the Baltic Sea as well (Swedish Agency for Marine and Water Managment2016). Over time, therefore, this guide can be supplemented with method descriptions also from the Baltic Sea area.

## 1.3 Reading instructions

This manual consists of seven chapters and three appendices that provide a detailed technical guide for the restoration of eelgrass in the North Sea. The manual covers all important steps in the restoration process, from evaluation and selection of sites, consultation and permits, harvesting and planting, to monitoring and evaluation of the results. In most chapters, fact boxes are presented that summarize important information about methods or that describe different work steps step-by-step.

The manual recommends restoration methods in which adult eelgrass shoots are transplanted, and it is mainly these methods that are described in the text.

Detailed description of how restoration can be carried out with eelgrass seeds can be found in **Appendix 1**. As studies have shown that restoration is very difficult in areas where the environment changes as a result of the disappearance of large eelgrass meadows, a description

of new, yet untested methods that modify the physical or biological environment to enable restoration is also presented in **Appendix 2**.

**Chapter 2** describes methods for evaluating and selecting a suitable site for eelgrass restoration. There you will find, among other things, detailed information on the factors that may affect eelgrass growth and survival, how they can be measured, and suggested threshold values for different variables. The chapter also describes how test plantings can be performed. Last, the most important steps in evaluating sites are summarized.

**Chapter 3** describes the regulations that may be involved in the harvesting and planting of eelgrass, which official contacts and consultations should be made, and any permits and exceptions that may be needed.

**Chapter 4** gives a brief description of the advantages and disadvantages of various restoration methods, which should be taken into account when choosing the method. The chapter also presents arguments about why restoration with shoots is recommended in Swedish waters today.

**Chapter 5** first describes various methods for eelgrass restoration with shoots. Then a detailed description of all steps in the restoration process is given, from selection of donor meadows to harvesting and planting of shoots.

**Chapter 6** describes how a planting should be monitored and evaluated. It provides a detailed description of recommended variables, how they should be monitored and what methods should be used to evaluate the results.

**Chapter 7** provides a summary of estimated costs for the restoration of eelgrass in the North Sea. **Appendix 3** presents the data and calculations for these estimates.

An important complement to this manual is the report *"Management and restoration of eelgrass in Sweden - Ecological, legal and economic background"* (Moksnes et al. 2016; The Swedish Agency for Marine and Water Management Report 2016: 8). As the report provides important background information for eelgrass restoration, this manual regularly refers to different chapters in the report.

As an introduction, some summary advice for eelgrass restoration in Sweden is given below.

## **1.4 General advice for restoration of eelgrass in Sweden**

#### It is not always possible to restore

Studies in Bohuslän show that eelgrass in some areas can no longer survive in areas where large eelgrass meadows were found in the 1980s, mainly as a result of degraded water quality, which is probably a result of increased resuspension of sediment when the eelgrass meadow no longer stabilizes the bottom. In these areas, the bottom is now covered by drifting algae mats, which further complicates natural recovery and restoration. For example, along the eastern side of the Hakefjord and the coastal area inside Marstrand down to the mouth of Nordre Älv in the municipality of Stenungsund and Kungälv, where very large areas of eelgrass have disappeared, environmental conditions have deteriorated so much that eelgrass restoration today is very difficult if at all possible in many places. Despite four years of trying to plant shoots and seeds in

these areas, very few eelgrass plantations have survived (see Tables 4.1 and 4.2). Therefore, in areas that have lost large eelgrass meadows, very costly, large-scale measures may be required that change the environmental conditions in the area before restoration of eelgrass is possible (see Appendix 2). Thus, one should not expect that it will always be possible to restore a lost eelgrass meadow. It is therefore extremely important to protect the remaining eelgrass habitat, especially in these areas, and only as a final measure to allow compensation restoration as a solution in exploitation.

### **Bigger is better**

Because eelgrass is a so-called ecosystem engineer who, when meadows are sufficiently large, creates a physical habitat and changes hydrodynamics and biogeochemistry on the seabed where it grows, a restored eelgrass meadow when it reaches a critical size can create a self-generating effect that stabilizes the bottom and improves water conditions and consequently growth conditions for eelgrass. Although there is still currently no knowledge of how large a meadow needs to be in order to achieve such an ecosystem change, studies of completed seagrass restorations show that major projects ( $\geq$ 100,000 shoots; corresponding to about 0.5 - 1 ha) have generally been more successful than those planted on a smaller scale ( $\leq$ 10.000 shoots; corresponding to about  $\leq$ 0.1 ha; van Katwijk et al. 2015). Therefore, it may be **important for a planted meadow to reach a critical size that improves the growth conditions and opportunities for the eelgrass to survive.** 

### Spread the risks

Like harvests on land can fail for some years due to adverse conditions, plantings of eelgrass can fail even if the sites are carefully selected and the planting is done correctly. As natural eelgrass meadows show great variation in growth and distribution between different years, random events such as storms, ice scraping, large freshwater outflows, blooms of algae or unusually high summer temperatures can mean that the survival of transplanted eelgrass can be very low in some places. It is therefore **important not to carry out a large-scale restoration attempt at only one point in time and one location, but instead divide the plantings into time and space.** Experience from restoration efforts in i.e. Holland has shown that restoration will be more successful if the risk is spread across different scales in time and space by, for example, divide the planting over several years and at several different sites (van Katwijk et al. 2009, 2015). However, this must be weighed against the fact that each planting reaches a critical size that can create self-generating effects (see above).

# 2 Site selection for restoration

One of the main reasons for the relatively low success in restoration of seagrass is that unsuitable sites have been chosen for planting, where environmental conditions simply do not allow the growth of seagrass. It is therefore crucial for the success of a restoration that potential sites for planting are carefully evaluated and tested before a large-scale restoration is initiated (Fonseca et al. 1998, Short et al. 2002a, Leschen et al. 2010). If the location to be restored has lost an eelgrass meadow, it is very important to try to identify the causes of the loss, why no natural recovery has taken place, and to determine if conditions have today improved so that eelgrass growth is possible. Because eelgrass meadows change the biogeochemical conditions in which they grow, the loss of an eelgrass meadow can result in such large changes in, for example, sediment and water quality that the eelgrass can no longer survive in an area where it has previously grown, even if the causes to why the meadow initially disappeared is no longer present.

There are a large number of factors that can counteract the re-establishment of eelgrass in one area. The most common problem in eelgrass restoration is poor water quality with poor light conditions in the water, but also unsuitable temperature or salinity conditions, epiphytic growth and drifting algae mats, disturbance from burrowing and grazing animals, exposure to waves and currents and unfavorable geochemical conditions can cause problems (Fonseca et al. 1998, Short et al. 2002a, van Katwijk et al. 2009). It is therefore important to carefully investigate several critical variables for eelgrass growth and also to perform test plantings at potential restoration sites.

In general, evaluation and selection of sites for restoration is done by first gathering available information on the historical and contemporary distribution of eelgrass as well as environmental changes and measures that have taken place (and potentially will) in the target area. Based on this material, a larger number of potential sites are selected that are visited in the field, where the most promising sites are sampled, monitored and test planted for a year. The results are then analyzed where the sites with high survival and growth of eelgrass are used for the large-scale restoration. There are a number of different suggestions in the literature on how different variables should be used to rank potential sites for seagrass restoration (see e.g. Fonseca et al. 1998, Short et al. 2002a).

Below, we first describe some important aspects to consider when choosing a restoration site. Then follows a detailed description of important factors and processes that affect eelgrass growth and survival, as well as variables that can be monitored to evaluate if these factors are a problem at the sites. Finally, a summary is presented of how the site selection is done (see section 2.8 and fact box 2.7). The text presents factors that affect both eelgrass seeds and plants. Factors that affect eelgrass seeds are primarily of importance if seed methods are used in restoration (which is not currently recommended in Swedish water; see section 4), but are also important to know when shoot methods are used. This is because seeds produced by planted shoots are important for the growth and survival of a restored meadow, especially in the early years.

## 2.1 Difference between ecological restoration and compensation

The method for evaluating and selecting a restoration site may differ slightly between ecological and compensatory restoration since the goals differ slightly between these restoration types. In ecological restoration, the goal is usually to recreate a lost habitat, either in a known location where the loss is documented, or to recreate a specific area of lost habitat (100 hectares) on the most suitable sites. In the latter case, the overall goal may be to achieve a Swedish environmental target or an EU directive's requirement that the environment must not be degraded (see Moksnes et al. 2016, sections 2 and 7 for details). In the case of ecological restoration, the targets are therefore usually large-scale, and you are interested in maximizing the area of eelgrass habitat that the planted eelgrass gives rise to in the long term. This is important because the area that can be restored with the methods that are available today is usually small (less than 10 ha per year) in comparison with the area that has disappeared (1000's of hectares; see Moksnes et al. 2016, section 3.3). The goal of restoration is then to enable and accelerate natural recovery of eelgrass. It is then important to avoid planting on sites that are likely to recover naturally without restoration, and instead choose sites that optimize natural spread from the restored meadow. In ecological restoration, it is therefore important to have a landscape perspective over a larger area (10s of kilometers) consisting of a large number of shallow soft bottom areas both with and without live eelgrass habitat where several restoration sites are strategically selected to maximize recovery in the entire area in the long term (see section 2.4). It is then also important to ensure that the restored area has legal protection against future exploitation or disruption of human activities, for example that the sites (both restored and natural meadows) are included in some sort of spatial protection (nature reserve, biotope protection, etc.).

In compensation restoration, the goal is usually to only compensate for the damage caused by an actor to the eelgrass habitat and the scale is therefore usually relatively small (0.1–10 ha). In this type of restoration, depending on the requirements placed on the responsible actor, it may suffice to choose a site where planted eelgrass is able to survive long term, with less concern regarding how the meadow is affected by or affects the distribution of eelgrass meadows in the immediate area. In compensation restoration, it is also important that the restored meadow be established quickly, (preferably before the meadow is destroyed) to minimize the loss of ecosystem services, and to limit the extent of compensation that may be required (see Moksnes et al. 2016, sections 2.2 and 9 and Appendix 2 for details). Since restoration using seed methods takes two years longer to evaluate than methods where vegetative shoots are planted (since seeds do not germinate until the year after they are planted), methods where shoots are used in compensatory restoration are often preferred. In Southern California where compensatory restoration of eelgrass is a long-established and well-functioning method, all restoration is done with using shoots due to, among other things, this reason (personal communication, Keith Merkel). In compensatory restoration, it is especially important to also include a reference bed (see section 2.7) which is monitored in parallel with the restored eelgrass meadow to see if, for example, poor growth of the planted meadow may be due to large-scale processes and not that the restoration is poorly performed, or that the site is not suitable for restoration. In some states in the United States, restoration success in compensation cases is evaluated by comparing survival of the restored meadow with changes in a reference bed (SCEMP 1991; see Moksnes et al. 2016, section

8.4 for details). Except for these differences, the methods for evaluating and selecting restoration sites are the same for ecological restoration and compensation restoration.

## 2.2 Causes of loss and lack of recovery

If the potential restoration site has lost an eelgrass bed, it is very important to first try to identify the causes of the loss and why no natural recovery has taken place as this may indicate that the problems persists, and that the environment is unsuitable for eelgrass growth. The optimal situation is that the cause of the loss (e.g. site eutrophication) has disappeared and the environment has recovered, where the lack of eelgrass is only due to the limited dispersal possibilities of eelgrass. Under such conditions, restoration of eelgrass can be very effective and successful, such as outside of Chesapeake Bay in the United States, where 130 ha planted eelgrass grew to more than 1,700 ha over a 10-year period (Orth et al. 2012).

Along the NW coast of Sweden, eutrophication and reduced light and oxygen supply as a result of increased growth of phytoplankton, epiphytic algae and mats of macroalgae are considered to be a main reason for the eelgrass's reduced spread since the 1980s. In Bohuslän, overfishing of cod and other large predatory fish is also considered to have reinforced the effect of eutrophication by causing changes in the food Net that reduced the presence of small algaeeating invertebrates in the eelgrass meadows (Moksnes et al. 2011; see Moksnes et al. 2016, section 3.4 for details). Since the 1990s, the supply and levels of nitrogen to NW Sweden have decreased, while the levels of phytoplankton have decreased and the water clarity has increased in many areas, so that most environmental variables indicating eutrophication today show good or high status in coastal waters (Moksnes et al. 2015, The Sea 2016). This would indicate that eutrophication's negative effect on eelgrass has decreased and that environmental conditions today would allow restoration to begin. However, few positive changes are found in shallow coastal areas in Bohuslän where the prevalence of drifting algae mats is still high, and no general recovery of eelgrass can be discerned (Swedish Agency for Marine and Water Management 2012). This is especially true in the area west of Marstrand in Kungälv municipality and in Hakefjorden where the largest losses have occurred since the 1980s and where new inventories show that large losses have also occurred in the last 10 years (see Moksnes et al. 2016, section 3.3.3 for details).

The lack of natural recovery indicates that the growth of filamentous algae today is not only controlled by the nutritional supply to the coast, but probably also by observed changes in the food Net and the release of nutrients from the sediment in shallow coastal areas (Sundbäck et al. 2003). It may also indicate that there has been an regime shift in shallow soft sediment areas, where the loss of eelgrass beds and the accompanying destabilisation of the bottom led to increased resuspension of sediment and turbidity of the water.

Vegetation in these areas is today dominated by drifting mats of perennial brown and red algae that can grow in the low-light environment. Studies in Kungälv municipality in Bohuslän show that the secchi depth has decreased with more than a meter since the 1980s in several shallow soft bottom areas that have lost large eelgrass meadows, probably due to increased wave-driven resuspension of the sediment, which means that eelgrass today cannot grow on these sites (Figure 2.1, Moksnes *unpublished data*). The drifting algae mats aggravate the problem by increasing the resuspension of sediments as they drag over the seabed, as well as by shading and tearing off new plants. If a regime shift has occurred, it can be very difficult to restore

eelgrass on the sites (see Moksnes et al. 2016, section 3.4.8 for details). Studies in the municipality of Kungälv and the eastern parts of the Hakefjord up to Stengungsund show that the environmental conditions in most of the areas today are too poor to allow the growth of eelgrass (Table 2.1). In these studied areas, eelgrass restoration cannot be recommended with currently available methods. It is therefore very important to investigate the environmental conditions in all potential restoration sites, even though eelgrass has previously appeared on the sites. For those areas that do not allow eelgrass growth today, it may be necessary to first try to change the environmental conditions in the area, or temporarily counteract some processes before restoration of eelgrass is possible (see Appendix 2 for discussion of these potential measures).

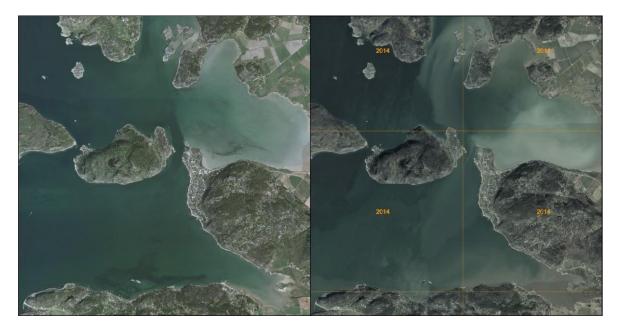


Figure 2.1 Wind-driven resuspension of sediment. The picture to the left shows Älgöfjorden in Kungälv municipality under calm weather conditions where the two bays Lökebergskile and Ödmålkile are seen on the right in the picture. These bays were covered in large parts by eelgrass beds in the 1980s, which have since disappeared. The picture to the right shows the same area after western winds as waves have caused sediment resuspension in shallow water, causing "clouds" that cover large parts of the area. Studies show that after a period of wind, the fine clay particles in the water can dramatically deteriorate the water clarity for several days. Today, the light supply in these bays is too poor for eelgrass to grow in the depths of the meadows found in the 1980s. The images are geometrically corrected aerial photographs (orthophotos) from the Swedish Land Survey.

**Table 2.1** Compilation of investigated sites in Bohuslän. Summary of information on 15 different sites in Bohuslän where various environmental variables were monitored during the eelgrass growth season (May – October) and test plantations were carried out, or sites that formed donor meadows where vegetative shoots or flower shoots were harvested (marked with D) for studies (see Figure 2.4 for map of sites). Positions indicated are WGS84 DDM. For *Status on eelgrass, Good* denotes that the extent of the meadow is estimated to have decreased by a maximum of 10% since the 1980s. For larger losses, the decrease is indicated in % and the year when a significant meadow still existed. Averages are given ± the standard deviation. A dash indicates missing data. For sites where plantings have occurred at more than one depth, variation in averages from shallow to deep is indicated. *Infections* indicate percent prevalence of protist *Labyrinthula zosterae*. *Disturbance* indicate expert-assessed levels (from *Low to High*) of physical and biological factors that can counteract the growth of eelgrass. *Test plantings* indicate perennial results of plantations with vegetative shoots on the growth of shoots in the autumn of the same year they were planted (*Growth year 1*) and survival after the first winter (*Multi-year survival*; see Table 4.2 for detailed results). \* Chlorophyll: data from only one sampling occasion. \*Salinity: salinity data based on occasional sampling in the field, other data comes from continuous measurements throughout the season. \*Water content: calculated from organic content (see fact box 2.5).

	1	2	3 <sup>D</sup>	4 <sup>D</sup>	5 <sup>D</sup>	6 <sup>D</sup>	7	8	9	10	11	12	13	14	15
Site Information															
Name	Snäckeb.	Torgestad	Lindholm.	Gåsö	Viks kile	Wallhamn	Bärby	Kyrkeby	Källsby	Lökebergsk.	Storebrorn	Lyngholmen	Ryskärsfj.	Trollö	Överön
Latitude	58 ° 21.7	58 ° 19.9	58 ° 15.8	58 ° 13.9	58 ° 3.3	58 ° 0.8	58 ° 1.6	57 ° 59.4	57 ° 59.3	57 ° 54.4	57 ° 53.9	57 ° 52.9	57 ° 49.2	57 ° 48.3	57 ° 47.7
Longitude	11 ° 34.0	11 ° 32.4	11 ° 29.7	11 ° 24.0	11 ° 34.3	11 ° 43.0	11 ° 48.1	11 ° 47.8	11 ° 47.5	11 ° 46.0	11 ° 40.9	11 ° 40.6	11 ° 42.3	11 ° 43.0	11 ° 43.7
Municipality	Lysekil	Lysekil	Uddevalla	Lysekil	Tjörn	Tjörn	Stenungs.	Stenungs	Stenungs	Kungälv	Kungälv	Kungälv	Kungälv	Kungälv	Kungälv
Water body	SE581700- 113000	SE581700- 113000	SE581570- 113040	SE581338- 112332	SE580325- 113500	SE575700- 114240	SE575700- 114240	SE575700- 114240	SE575700- 114240	SE575500- 113750	SE575500- 113750	SE574870- 113795	SE574870- 113795	SE574870- 113795	SE574650- 114360
Natural eelgrass															
Status / loss of eelgrass	Good	Good	Good	40% (1980)	Good	Good	95% (2004)	> 99% (2004)	> 99% (1980)	100% (1980)	80% (2004)	100% (2004)	100% (2004)	> 99% (1980)	95% (1980)
Maximum depth distribution. (m)	4.5	3.5	4.0	4.0	5.0	4.0	1.3	-	1.4	-	2.8	-	-	1.2	1.2
Shoot density (shoot m <sup>-2</sup> )	204 ± 59	722 ± 94	229 ± 206	-	506 ± 119	213 ± 61	-	-		-	145 ± 37	-	-	502 ± 21	721 ± 272
Leaf biomass (g m <sup>-2</sup> )	76 ± 53	202 ± 69	232 ± 52	-	103 ± 30	48 ± 22	-	-	-	-	189 ± 63	-	-	143 ± 14	207 ± 91
Light variables															
Extinction coefficient. (Kd)	0.47 ± 0.21	0.37 ± 0.12	0.43 ± 0.22	$0.32 \pm 0.13$	0.56 ± 0.25	-	$0.62 \pm 0.25$	0.94 ± 51	1.30 ± 1.0	0.82 ± 0.37	0.58 ± 0.25	0.81 ± 0.31	$0.90 \pm 0.33$	0.87 ± 0.35	1.25 ± 0.38
Max. depth distribution (m)	4.0 ± 1.5	4.7 ± 1.0	$4.0 \pm 0.5$	$5.3 \pm 0.9$	3.5 ± 2.1	-	$2.9 \pm 0.9$	$2.1 \pm 0.9$	1.9 ± 1.2	2.4 ± 1.2	3.2 ± 1.1	2.3 ± 1.0	2.1 ± 0.9	2.2 ± 1.1	$1.5 \pm 0.7$
Chlorophyll a (µg L <sup>-1</sup> )	5.35 *	5.64 *	-	-	-	-	-	-	-	3.3 ± 1.4	3.1 ± 1.3	4.5 ± 1.5	3.1 ± 1.2	2.7 ± 1.1	$3.2 \pm 0.6$
TSS (mg L <sup>-1</sup> )	-	-	-	-	-	-	-	-	-	15.2 ± 8.7	10.2 ± 5.3	9.3 ± 4.8	8.6 ± 3.0	7.0 ± 2.3	6.5 ± 2.6
Temperature (° C)	18 (13-24)	18 (11-23)	19 (15-25)	18 (14-24)	18 (12-24)	18 (10-22)	18 (16-20)	18 (15-20)	18 (15-20)	16 (14-20)	16 (13-20)	17 (14-19)	16 (14-19)	17 (14-22)	16 (14-20)
Salinity	-	20 (11-25)	-	23 (20-27)	19 (8-22)	-	-	-	-	17 (8-24)	16 (12-29) *	14 (4-24)	12 (7-20) *	11 (7-21) *	6 (0-19)
Sediment variables															
Silt and clay (%)	6.6 to 24.7	1.3 to 10.5	-	-	13.4 ± 6.5	-	37.5 ± 3.4	77.1 ± 26.3	50.8 ± 22.6	32.7 ± 13.7	30.1 ± 5.4	59.9 ± 8.0	62.1 ± 19.3	53.0 ± 12.3	33.9 ± 6.6
Organic content (%)	1.2 to 11.3	0.4-1.4	$25.7 \pm 0.7$	-	1.8 ± 0.3	-	$2.6 \pm 0.3$	3.6 ± 1.0	2.6 ± 1.9	2.6 ± 1.4	7.6 ± 1.1	$7.5 \pm 0.3$	$4.0 \pm 0.6$	$2.7 \pm 0.6$	$2.4 \pm 0.7$
Water content (%)	28.6 to 74.1	6.3 to 31.7 *	85.9 ± 2.4	-	28.7 ± 2.4	-	40.5 ± 2.5	37.8 ± 7.3	32.5 ± 9.5	36.7 ± 3.4	67.1 ± 2.2	49.4 ± 2.9	42.7 ± 5.4	45.0 *	36.5 ± 5.7
Sulfide content (µmol g <sup>-1</sup> )	1.7 to 5.4	0.5-1.7	-	-	-	-	-	-	-	$0.09 \pm 0.02$	0.24 ± 0.18	1.85 ± 2.66	1.41 ± 0.60	0.51 ± 0.30	$0.06 \pm 0.03$
Infections															
Labyrinthula (occurrence)	0%	17%	-	-	-	-	-	-	-	-	-	15%	-	-	0%
Disturbance															
Resuspension	Low	Low	Low	Average	Low	Average	Average	High	High	High	Average	Average	High	High	High
Filamentous algae	Average	Average	High	Average	Low	Average	Average	Low	Low						
Algal mats at the bottom	Low	Low	Low	Average	Low	Low	Average	Average	Average	Average	Average	High	High	Average	Average
Test plantings	1.2 to 1.5	1040		1100	1 5		1 4 1 9	1.6	1.4	2.4	2.2	2.2	2.5	10	1.0
Planting depth (m)	1.2 to 4.5	1.0-4.0	-	1.1-2.2	1.5	-	1.4-1.8	1.6	1.4	2.4	3.2	2.3	2.5	1.2	1.2
% of surface light at the bottom	54.7 to 17.6	66.2 to 29.7	-	45.6 to 82.1	45.9 ± 13.7	-	41.7 ± 11.9	22.2 ± 13.8	16.2 ± 19.9	18.9 ± 12.9	23.6 ± 11.2	20.3 ± 11.9	13.9 ± 10.0	21.8 ± 13.4	24.7 ± 12.1
Growth year -1	High	High	-	Medium-High	High	-	Low	No	No	No	High	Average	No	Low	High
Multi-year survival	High	High	-	High	High	-	-	-	-	No	No	Low	No	-	Average

## 2.3 Historical distribution of eelgrass

Sites with the best conditions for successful restoration are those that previously had an eelgrass population because many variables there are probably still favourable for growth, such as wave exposure, sediment type, depth, etc. If possible, sites should be selected where there is previously documented occurrence of eelgrass (Fonseca et al. 1998). However, the availability of eelgrass inventories prior to the 1990s along Sweden's coasts is very limited and is currently mostly available from a number of municipalities in Bohuslän. For these restricted areas, maps with historical distribution are available in GIS format at e.g. County Administrative Board of Västra Götaland (see fact box 2.1). These inventories indicate that around 60% of all eelgrass in Bohuslän has disappeared since the 1980s, corresponding to 8,000-22,000 ha, which is in the same order of magnitude as estimates of shallow (0-6 m) unpopulated soft bottom found in the area today (15 000 ha; see Moksnes et al. 2016, Appendix 1 for details). It can therefore be assumed that most shallow soft-bottom areas without vegetation found in Bohuslän today constitute historic eelgrass sites. It is therefore important to also include areas where historical data is missing when selecting potential sites for restoration in Bohuslän. For these sites, historical occurrence of eelgrass can be investigated through conversations with older site fishermen and residents in the area.

With the exception of the Kungsbackafjord in northern Halland, which was included in the socalled municipal inventories in the 1980s, there are no known historical inventories of eelgrass from Halland and Skåne's coastal waters, so there is no evidence to identify or estimate the existence of historical eelgrass sites. Possibly historical orthophotos from Lantmäteriet (see fact box 2.1) can give an indication of the eelgrass's historical distribution in these regions. In these areas, the the site selection need to a greater extent rely on field sampling of the sites. As Halland and Skåne's coastal waters in the Kattegatt largely lack an archipelago, exposed sandy beaches dominate in these areas, which probably due to wave erosion have a naturally limited distribution of eelgrass on shallow water. These exposed soft bottom areas are therefore probably less suitable for eelgrass restoration. However, this should be investigated with studies.

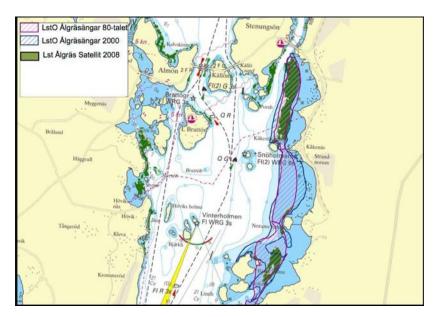
## 2.4 Current distribution and natural dispersal of eelgrass

In both ecological and compensatory restoration, it is important to avoid restoring places that would most likely recover naturally in the near future. Although the vegetative expansion of rhizome (ground stems) from nearby eelgrass meadows is very slow (16–45 cm per year; Olesen & Sand-Jensen 1994) and most seeds spread only a few meters from the meadows (Orth et al. 1994), individual seeds can spread considerably longer, for example during storms, which is believed to be the explanation for a very rapid spread of restored eelgrass in some studies (e.g. Orth et al. 2012). A rule of thumb is therefore to avoid restoring eelgrass closer than 100 m from a healthy eelgrass stock (Fonseca et al. 1998). It is therefore important to carefully examine the presence of live eelgrass in all potential restoration sites and in nearby areas.

#### Fact box 2.1 Materials for mapping the distribution of eelgrass

**Field inventories carried out by municipalities and administrative county boards.** There is no comprehensive national inventory or monitoring of eelgrass in Sweden, but eelgrass has been mapped with aqua scope by some municipalities in Bohuslän during the 1980s, which was revisited in the 2000s (Baden et al. 2003, Nyqvist et al. 2009). These inventories have been digitized and are now available in GIS format at the County Administrative Board of Västra Götaland. In the Kungsbackafjord in Halland County, the municipality carried out an inventory of the distribution of eelgrass in 1999 (Karlsson 1999). The County Administrative Board of Halland has recently conducted a new inventory of eelgrass in the county, which shows large occurrences of both eelgrass and dwarf eelgrass (personal *communication*, B. Gustafsson, 2016). The County Administrative Board and municipalities in Skåne have also carried out an inventory of eelgrass in Skåne's coastal waters during the 2000s by means of sampling in certain areas where the vegetation cover and depth distribution of eelgrass were determined (Olsson 2005, Svensson 2014).

**Remote sensing with satellite images.** The County Administrative Board of Västra Götaland has also made an attempt to map the eelgrass in the county with remote sensing analysis with satellite images from several different years (Lawett et al. 2013). Although this method is still under development, the mapping can be an important first material for the presence of eelgrass in the restoration area. The County Administrative Board of Västra Götaland now has distribution maps of eelgrass in the county in GIS format from several years of remote sensing analyses.



**Figure A**. Example of map data on the current and historical distribution of eelgrass in the northern part of the Hakefjord in the municipality of Stenungsund, prepared by the County Administrative Board of Västra Götaland. The dashed areas show the distribution of eelgrass in the 1980s (red) and in the year 2000 (blue), based on field inventory with aquascope. Green area marks where remote sensing analysis with satellite images (2008) estimates that there is a high probability of finding eelgrass. Note that estimates of prevalence using these two methods are not directly comparable, but can provide guidance on the historical and current distribution.

**Aerial photos.** If eelgrass inventories are lacking in the target area, available aerial photographs may provide a first basis for an indication of the presence of eelgrass in the area. Swedish Land Survey has been photographing Sweden since the 1930s with airplanes and can supply both contemporary and historical aerial images from Swedish coastal waters that are geometrically corrected (so-called orthophotos) from which distribution of vegetation in shallow areas can be indicated (see Figure 2.1 for example and *www.lantmateriet.se* for information and prices). Free aerial images of Swedish coastal waters can also be found on various map services online (ex. *Eniro.se* and *Google Earth*) where the distribution of eelgrass can be indicated. However, for these images, it may be difficult to obtain information regarding when the image was taken.

In addition to the aforementioned dispersal possibilities, smaller amounts of seeds can be spread over longer distances through floating reproductive shoots with seeds. Studies in Bohuslän show that eelgrass' reproductive shoots easily detach when the seeds are ripe and that they have positive buoyancy for at least four weeks when they can be spread long distances using surface currents (Källström et al. 2008). Although only a small number of seeds are spread between areas in this way and the growth of new meadows is therefore very slow, it constitutes an important mechanism for colonization of remote areas over extended periods of time. The limited genetic difference found between eelgrass stocks in Skagerrak and Kattegatt (Boström et al. 2014, Eriander et al. 2016; B. Källström, *unpubl. data*) is likely to be an effect of spread via floating reproductive shoots.

In ecological restoration over larger areas, it is important to take into account this potential spread of seeds through floating reproductive shoots. It is especially important to identify the sites that optimise the natural spread of eelgrass via this mechanism from the restored meadow to other areas in need of restoration, but also to identify potential restoration sites that should be avoided since they are likely to receive seeds from existing nearby meadows.

In order to evaluate this dispersal and connectivity (degree of interconnection) between shallow soft bottom areas (0–6 m) with and without live eelgrass, it is important to carefully catalogue the presence of these habitats within a larger area surrounding the target area to be restored.

If the distribution of now living eelgrass and potential restoration sites in the target area is known, dispersal via floating flower shoots can potentially be modelled using high resolution oceanographic circulation models to evaluate the connectivity. These types of models are currently being developed by researchers at the University of Gothenburg in collaboration with the Swedish Agency for Marine and Water Management and DHI for, e.g. Kosterhavet and the Gullmarsfjord and are expected to be available for studies on eelgrass seed dispersal within the next few years. If these opportunities are lacking for the study area, a dominant distribution direction for wind-driven floating reproductive shoots can be assumed to be northeast along the Swedish west coast due to the prevailing south-westerly winds during the period July to September (Källström et al. 2008) when the eelgrass forms seed shoots in Bohuslän (Infantes et al. 2016), and because of the Baltic surface current, which usually produces a northward coastal current along the Swedish west coast (Fonselius 1995).

## 2.4.1 Mapping eelgrass distribution in the target area

A first step in mapping the existing distribution of eelgrass in a target area is to investigate whether the municipality or the County Administrative Board has carried out inventories of eelgrass in the area. Even data based on remote analysis of satellite photos, or aerial photos (Figure 2.2) can be of great help in identifying suitable restoration sites and possible reference beds (Lundén & Gullström 2003; see fact box 2.1). Once a number of potential sites have been identified, they should be carefully investigated in the field.



**Figure 2.2** Eelgrass inventory with aerial photo. Aerial photo from *Eniro.se* showing the bay of Lökebergskile in Kungälv municipality which today lacks eelgrass. The darker area in the left part of the image is drifting mats of macro algae. The attached picture (from Baden et al. 2003) shows the distribution of eelgrass in the bay in the 1980s when an approximately 36 ha large eelgrass meadow covered the area.

Field inventory of eelgrass distribution should be carried out under good weather conditions with light winds and preferably sunshine to optimise visibility conditions in the water. In areas where wind-driven resuspension of sediments may pose a problem (Figure 2.1), there should have been calm weather conditions for at least a couple of days before the inventory is started. If possible, the inventory can advantageously start by filming the base area with a drone (see fact box 2.2, figure 2.3). If the flight takes place on assignment, permission is required from the Swedish Transport Agency (see www.transportstyrelsen.se/dronare for information). If drones are not available, aerial photos or satellite-based inventory (see fact box 2.1) can provide an overview of different habitats at the sites, although it must be taken into account that the extent of the habitat may have changed since the image was taken. With the remote image as a base, places that indicate underwater vegetation can then be examined more thoroughly with aquascope, drop video or snorkelling. With the help of GPS, the outer edges of existing eelgrass beds can be marked and then calculated in GIS their areal distribution and distance to potential restoration sites. The presence of algal mats (see section 2.5.6) should also be mapped during the field inventory. If no remote images are available, the site must be systematically scanned with aquascope, drop video or snorkelling. If water depth and water transparency (secchi depth) do not allow mapping with drones or aqua scope, drop-video or diving must be used to investigate the presence of eelgrass (Gullström et al. 2014), which makes the inventory slow and expensive. Traditional sonar and side-scan sonar can also be very helpful in identifying and mapping vegetation at the bottom which can be difficult to see from the surface. However, the mapping must be supplemented with direct observations to confirm whether vegetation is eelgrass or not.

#### Fact box 2.2 Examples of measuring instruments and prices (2015)

**PAR meters**. Instruments that measure and store light radiation in PAR have historically been quite expensive, but in recent years companies with lower prices have emerged. Dataflow systems sells a popular PAR meter (Odyssey *Photosynthetic Active Radiation Logger*) for around 2000 SEK.



**Lux meter**. Instruments that measure and store light in the lux unit are generally cheaper than PAR meters. The company *Onset HOBO data loggers* manufactures a popular lux meter (*UA-002-64, Pendant Temp / Lights*) which continuously stores data of both light radiation (in the unit lux) and temperature, which sell for about 500 SEK each.



**Salinity and temperature meters**. There are several types of instruments for continuous measurement and data storage of salinity and temperature. Generally, they are a little more expensive than the aforementioned light meters. The company Onset manufactures a supple conductivity sensor (U24-002, Conductivity logger) for around 8000 SEK, which measures and stores data on salinity and temperature continuously in a marine environment.



**Drones with cameras for estimating vegetation distribution.** Aerial photos are the considerably best way to estimate the area of planted eelgrass or algae mats if water transparency permits. Today there are surprisingly cheap small aircraft, so-called drones, which with cameras take video in HD quality, store GPS position on the drone and send real-time images to a smartphone, which can be used for this purpose. Today, there are a large number of drones on the market with built-in cameras, or where an HD camera can be attached (e.g. a GoPro camera), for around SEK 10,000 (incl. Camera).



Examples of popular drone model (DJI Phantom 3) with built-in HD camera, GPS memory, image stabilizer and autopilot.



**Figure 2.3** Eelgrass inventory with drones. Photo taken with drones about 100 m above a bay on Gåsö in Lysekil municipality. The leopard-spotted pattern is small eelgrass beds that have recently established themselves in the shallower parts of the bay. The weakly drawn square area in the middle of the image is a test plant of 10 x 10 m where eelgrass shoots have just been planted on 2.0 m deep with a shoot density of 16 shoots per square meter. Photo: E. Infantes.

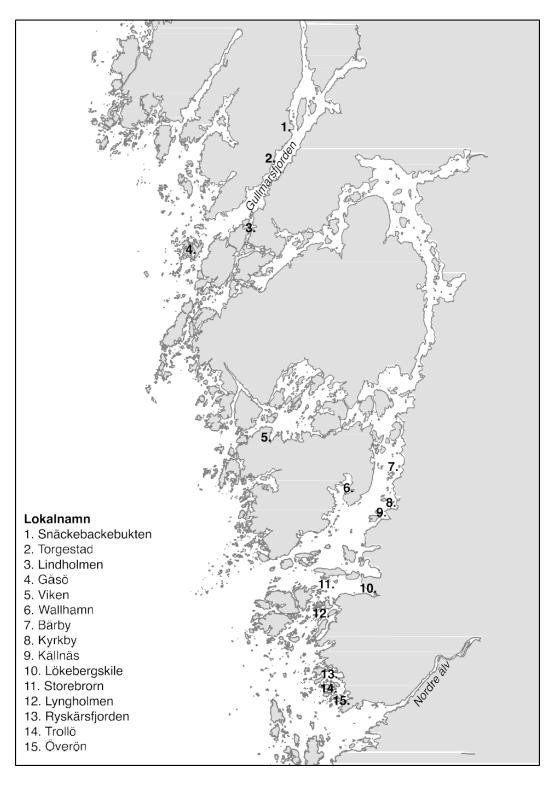
## 2.5 Important factors in choosing a restoration site

Here is a description of various key factors and variables that can affect the growth and survival of eelgrass, as well as descriptions of how to measure, test and evaluate these variables to select suitable sites in a scientific way. Many of these variables have been monitored at 15 different sites in Bohuslän (Figure 2.4) to assess the environmental conditions for eelgrass growth. Testing plantings have also been carried out at most sites. Table 2.1 summarises information and results from measurements and test plantings. This table gives an overview of how environmental variables vary in shallow soft bottom areas in southern Bohuslän and how these affect the growth of eelgrass. The table can be an important basis for evaluating new potential eelgrass restoration sites. Table 2.2 then summarizes estimated threshold values for these variables indicating whether planted eelgrass can grow or not. Many of the proposed limit values are based on studies in Bohuslän and should be representative of the Skagerrak – Kattegatt area, while others are taken from the literature and may be less representative of this area.

**Table 2.2** Threshold values for eelgrass restoration in the Skagerrak-Kattegat area. Summary of estimated threshold values (seasonal averages) for key variables indicating whether planted eelgrass is expected to exhibit positive growth (Yes) or adversely affected in terms of growth and long-term survival, or for other reasons is not an appropriate site for restoration (No). See section 2.5 for explanations.

variables	Yes	No	references
Site			
Nearest natural eelgrass meadow (m)	-	≤100	Fonseca et al. 1998
Depth (m)	1.5-2.5	<1	This report
Light and water variables			
Light extinction coefficient (Kd)	<0.92	> 1.6	This report
Light (% of light at surface)	> 25%	<20%	This report
Light (moles of photons m <sup>-2</sup> d <sup>-1</sup> )	> 7	<3	This report
TSS (mg L <sup>-1</sup> )	-	> 15	Dennison et al. 1993
Chl-a (µg L <sup>-1</sup> )	-	> 15	Dennison et al. 1993
DIN (µM)	-	> 10	Dennison et al. 1993
DIP (µM)	-	> 0.67	Dennison et al. 1993
Temperature (°C)	<20	-	Borum et al. 2004
Salinity	> 5	-	Borum et al. 2004
Physical exposure			
Current speed (cm s <sup>-1</sup> )	<15	> 50	Fonseca et al. 1998
Sediment erosion rate (mm day <sup>-1</sup> )	-	> 0.5	Merkel 1992
Wave exposure index		> 3 * 10 <sup>6</sup>	Fonseca et al. 1998
sediment Variables			
Sedimentation rate (mm day <sup>-1</sup> )	-	≥0.3	Merkel 1992
Content silt and clay (%)	<34%	> 50%	This report
Organic content (%)	<5%	-	Koch 2001
Organic content (%) *	<2%	-	Lillebø and others 2011
Water content (%) *	<40%	-	Lillebø and others 2011
Ammonium content in pore water (µmol $L^{\text{-1}})$	> 100	-	Dennison et al. 1987
Bio disturbance for eelgrass seeds			
Lugworm ( <i>Arenicola marina</i> ; no. m <sup>-2</sup> )	<10	> 50	Delefosse & Kristensen 2012

\* Limit values for increased risk of resuspension of sediments  $m^{-2} d^{-1} = per square meter and day, mg L^{-1} = milligrams per litre,$  $<math>\mu g L^{-1} = micrograms per litre, cm s^{-1} = centimetres per second, mm day^{-1} = millimetres per day, \mu mol L^{-1} = micromole per litre, m^{-2} = per square meter.$ 



**Figure 2.4** Map of study sites in Bohuslän. The map shows 15 different sites in Bohuslän where different environmental variables have been monitored, test plantings have been carried out or sites that have formed donor meadows where vegetative shoots or flower shoots have been harvested for studies (see Table 2.1 for details).

## 2.5.1 Water depth

The water depth selected for planting can influence a wide range of factors that are crucial for eelgrass survival and growth, such as light supply, wave exposure, grazing and disturbance from animals, risk of dehydration and damage from ice. The maximum depth distribution of eelgrass is determined almost exclusively by the light supply and varies widely between regions and between sites, where the depth distribution has also decreased dramatically over the last hundred years. Studies from the end of the 19th century indicate that the eelgrass was then common down to about 15 m deep in western Kattegatt (Loo 2015), while the eelgrass there today very rarely grows deeper than 8 m (Boström et al. 2014). In the Öresund, eelgrass grows on average down to about 6 m, while the maximum depth distribution in Bohuslän on average varies between 3 and 4 m (Swedish Agency for Marine and Water Management 2012). In some extreme sites with high turbidity such as at the outlet of Nordreälv in the northern Kattegatt, eelgrass is not found deeper than 1.2 m, while it can be found down to 5 m depth at other sites in Bohuslän with good light supply (Table 2.1). Due to the large variation between sites, it is very important to measure the light supply at all potential restoration sites, both to assess whether eelgrass can grow on site, and to determine the optimum planting depth (see section 2.5.2 for information on how lighting conditions are measured). Since shoot growth is very low near the maximum depth distribution (Eriander et al. 2016), restoration near these depths is generally not recommended, especially as a deterioration of the light supply can eliminate the entire planting.

Along the Swedish west coast, the upper distribution limit is determined by different factors at different sites. In exposed areas, it can be determined by wave erosion, while in more sheltered areas it can be determined by ice scraping in the winter, or by dehydration on shallow water that under high pressure can produce long-lasting low water conditions. Studies in Bohuslän where shoots planted at 1 to 5 m depth in protected and semi-exposed sites showed no negative effects of dehydration or wave erosion on planted shoots. However, some losses of shoots at 1 m depth were observed from ice scraping after ice winters, while slight ice effects were observed at 1.5 m depth (Eriander et al. 2016). Based on these studies, eelgrass should not be planted shallower than 1 m below average water level.

To reduce the risk that large parts of the plantings are eliminated due to interannual variations in e.g. light supply, storms or ice winters, it is recommended that eelgrass be planted with a suitable margin to the lower and upper distribution limits, and that the planting is concentrated near the upper distribution limit where the good light supply normally results in high lateral shoot growth and rapid establishment of the restored meadow (see below). **Generally, it is therefore recommended that eelgrass be planted between 1.5 and 2.5 m depth** (although the light supply would allow growth at greater depths). In good conditions, the eelgrass will, over time, spread to its natural distribution limits. For sites with a sloping bottom, therefore, the planting may initially take the form of a narrower band that follows the optimal depth curve (see Figure 5.7, section 5.5).

## 2.5.2 Light conditions

Water quality and light conditions in the water determine how deep eelgrass can grow in a site area and is usually the most critical variable for survival. In addition to the depth, the availability of light is also determined by how quickly the visible light is absorbed into the water, which depends on the type and amount of organic and inorganic particles in the water. Lighting conditions in coastal waters can therefore be influenced by a wide variety of factors. In addition to the content of nutrients that limit the amount of phytoplankton and organic particles in the water, light conditions are also affected by the amount of suspended sediment particles that can be suspended from the bottom in shallow areas by waves or strong currents, or carried out to the coast via watercourses, especially during spring runoff and after rain. Watercourses can also carry humus substances that can have a major impact on the lighting conditions in the sea.

According to studies from several countries, eelgrass needs about 18-21% of light supply at the surface to grow (Dennison et al. 1993). Formulated in the amount of light, eelgrass needs at least 7 moles of photons per square meter and day of photosynthetically active radiation (PAR) in order not to be light limited, and at least 3 moles of photons per square meter and day for long-term survival (Thom et al. 2008). This is in good agreement with studies in Bohuslän where planted eelgrass has shown long-term survival down to 18% of light supply at the surface, and down to 4 moles of photons per square meter and day in average values over the growing season (Eriander et al. 2016; Table 2.1).

The greatest depth that eelgrass can survive at a site can be calculated if you know how guickly the light is absorbed in the water, which can be described by the extinction coefficient of light (Kd) and calculated by light measurements from several depths. With the help of the Kd value, the maximum depth of distribution can then be estimated if you know how much of the light supply at the surface the eelgrass needs to survive (see fact box 2.3). By measuring the light continuously during the growing season at a potential restoration site, the area's average Kd value can be calculated, which provides information on both light supply and the growing season's length, which decreases with depth (Figure 2.5). In a location with relatively low light absorption in the water (Kd = 0.45), the light supply is high at 1 m depth (64% of the light at the surface) and contains enough light for the growth of eelgrass (> 7 moles of photons per square meter and day) from the beginning from March to early October. At the same location at 3 m depth (26% of the light at the surface), the growing season extends only from about mid-April to the end of August, and at 4 m depth, where on average about 17% of the light reaches the bottom, there is only enough light to survive during the summer months, but not to grow (Figure 2.5a). This means that although the light supply during the summer in this example at 3 m depth does not limit the growth of eelgrass, the growth period is several months shorter than at 1 m depth, which is why the annual growth is considerably lower. Although a planted eelgrass shoot can survive the first summer at the limit of maximum depth, the low light supply and growth over the summer make it difficult to store enough carbohydrates in the rhizome to survive the winter when it needs this energy to survive without light for 3-6 months (Eriander et al. 2016). Studies in Bohuslän where eelgrass shoots are planted at sites with different light conditions indicate that about 20% of the light supply at the surface is required for the shoots to have positive growth, and >25% of the light at the surface for the shoot growth to be >100% after three months. (Figure 2.6).

#### Fact box 2.3 Calculation of light supply at the planting depth

The maximum depth at which eelgrass can survive is determined by how quickly the portion of the visible light used in photosynthesis (PAR, *photosynthetic active radiation*) is absorbed into the water. This can be calculated if you know the light radiation at two different depths using the following ratio (Lambert-Beers law):

#### I<sub>D</sub> =I<sub>S</sub> \*e<sup>-Kd\*D</sup>

where  $(I_D)$  and  $(I_S)$ , is the light illumination measured in PAR at the deeper and shallower depths respectively and D is the depth difference in meters between the two depths. Kd is the extinction coefficient of light and describes how light is absorbed over depth. It is constant over depth and can be calculated accordingly:

#### $Kd = -In(I_D/I_S)/D$

Extinction coefficient varies over time with water quality, but if Kd has been measured on many occasions at a location, a representative mean can be calculated that describes the average light conditions in the water. With this average (Kd<sub>A</sub>), the maximum depth at which eelgrass can survive ( $D_{max}$ ) at the site can be determined if one knows the proportion of light supply at the surface (I<sub>0</sub>) required at the maximum depth for eelgrass to survive in the area, i.e. ID<sub>max</sub>/I<sub>0</sub> according to the following equation:

#### $D_{max} = -In(I_{Dmax}/I_0)/Kd_m$

If the eelgrass in an area requires at least 20% of the light supply at the surface to survive ( $I_{Dmax}/I_0 = 0.20$ ) and the average value of Kd over an entire growing season is measured to 0.50, the maximum depth that eelgrass can survive on ( $D_{max}$ ) be estimated by the equation above ( $D_{max} = -In(0.20) / 0.50$ ) to 3.2 m.

The advantage of measuring the light at two depths at a site is that the light supply can be calculated at any depth at the site, using the equations above, so that an optimal planting depth for the eelgrass can be calculated for the site.

If you only have access to one light meter per site, you can instead use the total inflow of photons per area and day (PPFD; *photosynthetic photon flux density*) by placing the PAR sensor at the depth you think is right for planting. Eelgrass requires, on average, a minimum of 3 moles of photons per square meter per day to survive (Thom et al. 2008) and the number of photons reaching the sensor in a day can be calculated by summing the total light radiation per day (which is normally measured in µmol photons per square meter and second). In the calculation it is important to compensate that the light meter normally only stores a number of values per hour. The disadvantage of using only one light meter is that one can only evaluate light supply at the depth where the PAR sensor is placed.

Since PAR sensors are relatively expensive and the cost becomes high if many sites are to be evaluated, an alternative may be to use cheaper light meters that measure illumination (illuminance) in the unit lux (lumens per square meter). Since a lux meter also includes wavelengths that are not photosynthetically active, they must be calibrated with a PAR meter at each site (since the ratio may vary in time and space). Conveniently, this is done every two weeks when the light meters are cleaned in the field.

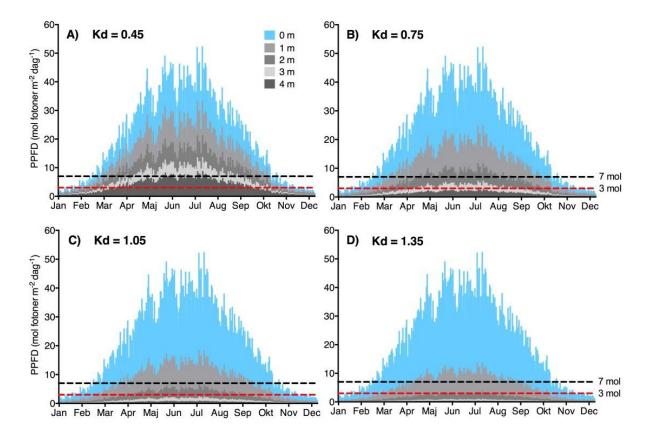
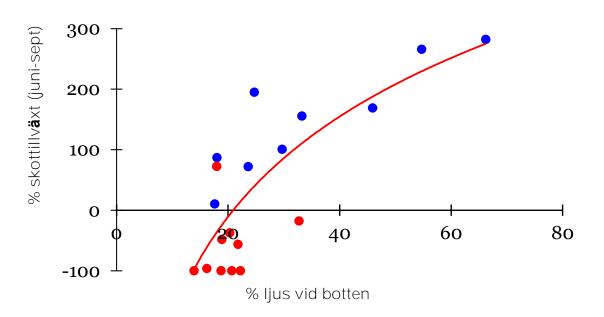


Figure 2.5 Seasonal variation in light supply at different depths and light absorption in the water. The figure shows in simplified terms how the amount of light (PPFD; moles of photons per square meter and day) varies in the air at the surface and at 4 different depths for a year at 4 sites with different light absorption and extinguishing coefficients (Kd) in the water: A) Kd = 0.45, provides ca 41% of the amount of light at the surface at 2 m depth, B) Kd = 0.75; ca 22% at 2 m, C), Kd = 1.05; ca 12% at 2 m, and D) Kd = 1.35; ca 7% at 2 m. The black dashed line marks the amount of light 7 moles of photons per square meter and day that eelgrass needs not to be light limited, and the red dotted line marks the amount of light 3 moles of photons per square meter and day that eelgrass needs to survive (Thom et al. 2008). Light data at the surface is from SMHI (average value of STRÅNG data from Gullmarsfjorden between 2010-2014) where the light extinction in the water was calculated based on empirical light measurements in different areas in Bohuslän (Eriander et al. 2016). With increased depth and Kd value, both light supply and the number of months per year that the eelgrass can grow decreases. At site A (Kd = 0.45) the light supply is relatively good and at 4 m depth it varies between 3-7 moles during the growing season. At these light values, transplanted eelgrass could survive, but grow very slowly, which is why restoration is recommended at a shallower depth. At a depth of 2 meters, the shoots have light that enables growth from March to October with an average of about 41% of the light at the surface, which better meets the light requirement when choosing a site for restoration. At site B (Kd = 0.75), the light at the same depth only allows growth from mid-April to August, while the light at site C (Kd = 1.05) is <7 moles at 2 meters throughout the year so planting at this depth cannot be recommended. At site D (Kd = 1.35), the calculated maximum depth distribution (see fact box 2.3) is only 1.2 m, which is why the sites are generally not recommended for restoration.

For eelgrass restoration in the North Sea, it is recommended that the average light supply during the growing season at planting depth is> 25% of the light supply at the surface, or more than 10 moles of photons per square meter per day, and that the Kd value is at most 0.92 since the light supply is <25% at 1.5 m depth if the Kd value is higher. Sites with so turbid water that the light absorption gives a Kd value above 1.6 are unsuitable for restoration as the light supply makes it difficult for the eelgrass to survive deeper than 1 m, and since ice scraping can destroy plantings at shallower depths (see section 2.5.1; Table 2.2). Although eelgrass can survive with an average light supply just under 20% of the light at the surface, restoration is normally not recommended under these light conditions as growth is very low and the risk to high of planting being eliminated by temporarily deteriorating environmental conditions.



**Figure 2.6** Relationship between light supply and growth of eelgrass. The figure shows on the y-axes percent growth of the number of vegetative shoots (from June to September) planted in early June at 12 different sites in Bohuslän (5 sites with eelgrass: blue dots; 7 sites where the eelgrass disappeared: red dots) at different years (see Table 2.1 for description) in relation to measured light conditions (percent of light at surface reaching eelgrass shoots) at the bottom. The curve describing the ratio ( $y = 238.69 \ln(x) - 725.78$ ;  $r^2 = 0.63$ ) indicates that positive growth of eelgrass is obtained when about 20% of the light reaches the bottom. At sites where eelgrass are found today, about 25% of the light at the surface is required for the planted shoots to at least double in number during the first growing season. At sites where the eelgrass has disappeared, the light conditions are generally too poor to allow growth to day, and drifting algal mats also prevent the growth of plants.

#### **Measurement of light conditions**

Since measurements with light sensors are not normally included in environmental monitoring of shallow coastal areas, below is a more detailed description of how this can be carried out.

Since the lighting conditions in the water can change quickly and unpredictably in shallow coastal areas, and since field sampling often takes place under calm conditions (when the light supply is usually highest), occasional light measurements during field visits provide a poor basis for the actual lighting conditions at a site.

Therefore, it is **recommended that the light is continuously measured at two different depths using data-storing instruments that are placed in each potential restoration site during the growing season** (May – September). The advantage of measuring the light at two different depths at each site is that the light supply can then be calculated at any depth at the site using the light's extinguishing coefficient (Kd; see fact box 2.3) so that an optimal planting depth for the eelgrass can be calculated for the site .

To measure the light at two different depths, two PVC pipes can be used with different lengths (1 and 2 m long) where light meters with PAR or lux sensors can easily be mounted on top of the pipes using cable ties (see Figure 2.7; box 2.2 for example of light meters). The two tubes with light meters are placed on the deeper part of the potential planting site by placing the tubes vertically into the sediment so that the light sensor on the respective measuring instrument points upwards. One light meter is placed about 20 cm above the bottom to measure the light supply where the eelgrass is to grow. By placing it a bit above the bottom, the impact of drifting algae

which shadow the meter can be avoided. If possible, place the second light meter at least one meter closer to the surface than the first (120 cm above the bottom) by pressing down the tube to a pre-marked depth on the tube. These light meters are used to calculate the extinguishing coefficient (Kd) of light at the site (see fact box 2.3). It is important that the shallow light meter is not placed so close to the surface that it risks being dry at low tide, and that the difference in depth between the light meters is measured precisely. The distance to the surface varies with tides, wind and air pressure and should be measured at each visit. The pipes should be spaced at 1-2 m appart so that they do not shade each other, where the light meters point in the same direction from the pipes to the west to minimize shading. If light measurements are made on live eelgrass beds, e.g. in the case of a reference bed, measurements should be made one meter outside the meadow, or above the meadow's leaves to avoid shading.



Figure 2.7 Light Meter. The picture shows a lux meter (Onset Hobo) and a PAR meter (Odyssey) mounted with cable ties on a PVC pipe at 2 m depth. Photo: P. Moksnes.

Although most data-storing light meters have battery capacity to measure for many months, they need to be visited regularly to remove growth or sedimentation on the light sensor. In order to obtain reasonably continuous light measurements, it is recommended that growth is removed at least every two weeks during the measurement period, data can also be emptied from the instruments at that time. To facilitate the analysis of light data, it is important to note the degree of growth on the light meter at each visit. As the growth of epiphytic micro- and macroalgae on the eelgrass leaves impairs light access to the plant, observations of growth can also indicate if this can be a problem at the site. Observations of loose sediment on the light meter should also be noted as it indicates sedimentation that can shade planted shoots if they accumulate on the leaves in calm areas. When analysing the light data, values indicating that fouling or

sedimentation has disturbed the measurement should be removed from the analysis. Since fouling from epiphytic growth often occurs faster on the shallow light meter where the light supply is greater, problem with fouling can be indicated by a decreasing difference in light radiation between the deep and shallow meters over time.

If healthy eelgrass populations are in the study area, light availability and maximum depth distribution can also be measured at the natural meadows, which can then be compared with light and depth conditions at potential planting sites. This can be used as a complement, but can never replace light measurements at the restoration site as large differences in lighting conditions can be found between nearby sites.

If light meters are not available, the light supply at a site can be measured with a Secchi disk, where the secchi depth can then be converted to a value of the extinguishing coefficient (Giesen et al. 1990). However, light measurements with Secchi disc in shallow areas have several serious limitations, since the depth of sight cannot be measured if the bottom depth is shallower than the depth of sight (which is often the case), and that it can only be measured during field visits and in relatively calm weather conditions.

Furthermore, studies in Denmark have shown that the relationship between Kd and Secchi depth is not constant in time and space, and that measurements with Secchi disc can overestimate the light supply in coastal waters (Pedersen et al. 2014).

### 2.5.3 Turbidity, chlorophyll and nutrients

Although measurements of light supply is the most important variable for assessing water quality at a site site, it may be desirable to investigate what causes a deterioration of light supply sitely in order to possibly implement site measures (see Appendix 2). An important variable that affects the absorption of light in the water is the total amount of inorganic and organically suspended material (Total Suspended Solids; TSS), which is a measure of the turbidity of the water. TSS is measured via water samples that are filtered and weighed, which can then be divided into an organic and an inorganic fraction by combustion of the sample. TSS can also be estimated using a turbidity meter, but that must then be calibrated against TSS measurements for each site. A large proportion of inorganic material of TSS may indicate wave-driven site resuspension of sediment or suspended sediment being discharged from a nearby watercourse. In areas affected by watercourses, humic substances in the water can also affect lighting conditions. A large part of the organic fraction of TSS normally consists of phytoplankton, the concentration of which is usually estimated by the content of chlorophyll a (Chl a) in the water, which is measured in water samples with a spectrophotometer, or estimated by fluorescence measurements.

By measuring TSS, its inorganic fraction and ChI a in different weather conditions, it is possible, for example, to deduce whether poor lighting conditions at a site are due to eutrophication problems and high concentration of phytoplankton or wind-driven resuspension of the sediment. For example, at the Lökebergskile sites in Kungälv municipality (site 10, figure 2.4), the lighting conditions are poor at 2.4 m deep where a large eelgrass meadow was found in the 1980s (Table 2.1). Regular water sampling at the site shows high levels of TSS (on average 15.2 mg per liter), which is above the recommended limit value for eelgrass restoration (Table 2.2). Analysis of the

samples show that the organic fraction was low (on average 21%), which indicates that the high turbidity was not caused by phytoplankton.

This is also supported by relatively low levels of Chl a in the water (average  $3.3 \mu g$  per litre; Table 2.1). In summary, the measurements indicate that wave-driven resuspension of sediments causes poor lighting conditions at the sites (see Figure 2.1).

#### Inorganic nitrogen

In the North Sea, nitrogen and, to a lesser extent, phosphorus, are considered to be limiting the growth of algae. High levels of dissolved inorganic nitrogen (DIN), consisting of ammonium (NH<sub>4+</sub>), nitrate (NO<sub>3</sub>-) and nitrite (NO<sub>2</sub>-) in the water can cause blooms of both phytoplankton and filamentous macroalgae, and indirectly adversely affect eelgrass. As the uptake of DIN is very rapid during the summer, winter values of DIN are normally used in environmental monitoring where levels above about 10 µmol per litre (µM) are considered to be elevated values along Bohuslän's coastal waters at normal salinity levels (Swedish Environmental Protection Agency 2007). In summer, the levels are usually considerably lower. In eelgrass meadows in the Gullmarsfjord, the concentration of DIN is normally between 1–2 µM during the summer, where the main part consists of ammonium. Experiments have shown that raising the concentration to about 4–6 µM DIN sitely can be sufficient to cause blooms of filamentous algae with negative effects on the growth of eelgrass (Moksnes et al. 2008, Baden et al. 2010). In the US, studies show that eelgrass has poor survival in areas where the DIN concentration is >10 µM (Dennison et al. 1993; Table 2.2).

Experimental studies in the laboratory have also shown that high levels of ammonium (25–125  $\mu$ M) in the body of water can produce toxic effects and dead eelgrass if the levels are maintained for several weeks. The experiments also showed that the effects were stronger at high temperatures and poor lighting conditions, and it was proposed to be able to explain eelgrass losses in confined sea areas during warm autumn periods because nutrients are released from degrading algae (van Katwijk et al. 1997).

In Chesapeake Bay in the USA, via multi-year field studies, threshold values (based on seasonal averages) of levels for several of the variables mentioned above have been produced that predict whether eelgrass can grow in the area (Table 2.2; Dennison et al. 1993). If the threshold value for any of these variables is exceeded, the probability is low that eelgrass can survive in the study area. These values could also serve as a benchmark for Swedish conditions.

### 2.5.4 Salinity, temperature and oxygen conditions

Eelgrass can grow in anything from full salinity (35 psu) to brackish water environments with salinity around 5 psu, and also withstands large variations in salinity over shorter periods (Borum et al. 2004). For most eelgrass populations, the optimal salinity for growth and survival is between 10 and 25 psu (Nejrup & Pedersen 2008), while eelgrass beds adapted to lower salinity in the Baltic Sea grow optimally in salinities between 6 and 20 psu (Salo et al. 2014). In the Baltic Sea, the distribution of eelgrass is limited at the isohaline (salinity line) 5.3 psu, found at the Stockholm archipelago and the Åland Sea (Boström et al. 2003). Studies in Bohuslän show that both shoots and seeds picked from relatively high and constant salinity conditions in the Gullmarsfjord ( $25 \pm 3$  psu) could survive at very low (6.4 psu in seasonal averages) and varying (0.04-21.0 psu) salinity conditions at the outlet of the Northern River (Table 2.1). This indicates that the eelgrass of the

North Sea is relatively tolerant of variation in salinity and can be successfully transplanted between different salinity environments. However, low salinity can cause seeds to germinate during a disadvantageous part of the season. Laboratory studies indicate that if planted seeds are exposed to low salinity (5 psu) during some warm (15 ° C) autumn weeks, more than half of the seeds may germinate before winter (Infantes et al. 2016), which may cause the young plants to die during winter (normally eelgrass seeds germinate in spring in Swedish water). It may therefore be appropriate to measure salinity if the sites are affected by watercourses, especially if eelgrass seeds are used as a restoration method.

Eelgrass is generally adapted for a relatively cold climate from a water temperature of -1 °C in winter to 25 ° C in summer (Borum et al. 2004). In Chesapeake Bay in the United States, high summer temperatures (up to 30 ° C in the water) are considered to be an important cause of the reduced propagation of eelgrass in the area as well as the failure of large-scale planting trials with eelgrass (Goshorn 2006). Since eelgrass in Swedish water is not close to the limit for the species' southern or northern distribution in terms of temperature, direct effects of climate-driven temperature increases are not expected to limit the growth of eelgrass in Swedish water. However, high temperatures can cause serious indirect effects on eelgrass by increasing the growth of algae and reducing the oxygen content of surrounding water while increasing the metabolism and oxygen demand of eelgrass during nightly respiration (Rasmusson 2015). At night, the eelgrass is dependent on oxygen in surrounding water to oxygenate the tissue in leaves and roots and to counteract intrusion of toxic sulfide from the sediment (see Moksnes et al. 2016, section 3.1.1). High summer temperatures in combination with hight growth of epiphytic algae in eelgrass meadows are considered to be main cause to rapid losses of whole eelgrass meadows in Denmark for instance (Greve et al. 2005). Therefore, it is recommended that temperature is measured continuously at all potential restoration sites, especially during summer months to investigate whether high temperatures can cause problems for planted eelgrass.

### Measurement of temperature and salt content

As with light supply, temperature and salinity can change quickly and unpredictably in coastal waters and are best measured with instruments placed in the field that continuously measure and store data. Temperature sensors are included in many instruments, such as light meters, while continuous measurement of salinity in the marine environment requires special instruments (see fact box 2.2). The salinity meter can advantageously be mounted with cable ties inside a larger PVC pipe with drilled holes for water flow which is inserted into the sediment at a suitable depth.

The salinity meter should be placed about 20 cm above the bottom at the intended restoration site. The conductivity sensor should be cleaned from growth at least once a month. Since the salt content varies less than e.g. light and temperature it may be sufficient to measure salinity in one of several nearby sites if none of the sites is located at the outlet of a watercourse.

### 2.5.5 Sediment conditions and physical exposure

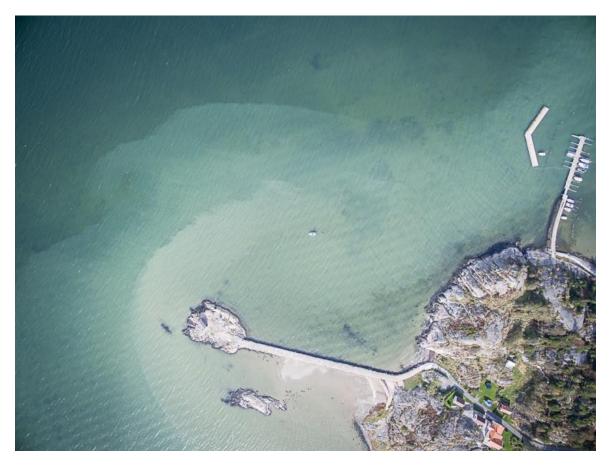
In Sweden, eelgrass is naturally found in environments with variable sediment conditions, from exposed sites with gravel or sandy bottom, to highly protected environments where fine sediment with high organic matter content (up to 25%) and water (up to 90%) dominates (Table 2.1, Jephson et al. 2008).

This shows that the eelgrass is very adaptable to different exposure and sediment conditions. However, the different environments present different types of challenges for eelgrass restoration. An analysis of simple sediment variables can give a good indication of which processes can interfere with the planting, and whether a site is suitable.

Recommendations in the literature regarding beneficial sediment for eelgrass restoration are not consistent. Although most studies recommend that soil with gravel and rock should be avoided, the recommendations vary with regard to how fine grained the sediment can be for eelgrass to grow (Fonseca et al. 1998). In a compilation of different seagrass species, requirements for the content of clay and silt in the sediment (<63  $\mu$ m grain size), a threshold value of less than 20% silt and clay is proposed (Koch 2001). In a large-scale restoration study of eelgrass in Boston Harbour, USA, no test plantings survived in areas where the proportion of silt and clay in the sediment was above 57%, while good growth was found in areas with a content below 35% (Leschen et al. 2010). However, other studies have suggested significantly higher threshold values for eelgrass restoration, below 70% silt and clay (Short et al. 2002a).

Eelgrass test plantings in Bohuslän show a similar result to that in Boston harbour where eelgrass generally showed good growth in areas where the clay content is below 34%, while very few plantings survived in sediment with a clay content of more than 50% (Table 2.1) . Studies in Bohuslän further show that the light supply in the water decreases with an increased proportion of silt and clay in the sediment, at the same time as the growth of planted eelgrass is reduced (see Fact box 2.4), which suggests that resuspension of the fine-grained sediment may be an explanation for the poor growth in areas with high levels of silt and clay (Moksnes et al. unpublished data). Fine-grained sediment also reduces the exchange of pore water in the sediment, which can lead to the accumulation of sulfides and other toxic substances, with negative effects on growth (Koch 2001). Therefore, it is **recommended that sites with a content of silt and clay of more than 50% is to be avoided for restoration of eelgrass** (Table 2.2), or that light and growth conditions are investigated extra closely at the sites.

In Bohuslän, glacial clay (i.e. fine-grained clay sediment deposited from the glacier's melt water) can be found at the sediment surface in some areas, especially in shallow soft sediment areas that have lost large eelgrass beds and have been exposed to erosion. There is a lack of studies on whether eelgrass can grow on glacial clay, but this type of clay is very compact and has low water permeability, which probably makes it unsuitable as eelgrass substrate. The fine clay particles are also easily suspended into the water by waves, which gives very poor light conditions in the water for long periods (Figure 2.8). **Restoration on bottoms where glacial clay is found <5 cm from the sediment surface should therefore be avoided**.



**Figure 2.8** Wave-driven resuspension of glacial clay in the Hakefjord. The picture shows Källsby headland in the Hakefjord (site 9, figure 2.4) 2015 where a western breeze causes site resuspension of the bottom sediment. In the 1980s, the shallower parts of the image were covered by an eelgrass meadow, which today is replaced by clay bottom without vegetation. Due to erosion, glacial clay is now found at the surface of the sediment, which is very easily stirred up by waves in shallow water, which is seen as a grey plume closest to land in the picture. The visibility in the water in the plume is <0.5 m. Photo: E. Infantes.

#### Exposed, sandy environments - exposure index and grain size

A sediment with a high proportion of gravel and sand, and low organic matter content (see fact boxes 2.4. and 2.5.) indicates a seabed that is exposed to waves or strong currents. In such areas, erosion of sediment can cause problems where plants and seeds are washed away. Sites with a current velocity exceeding 50 cm per second should not be used for restoration as these velocities can cause both sediment and plants to be flushed away. At velocities below 15 cm per second, negative effects of water currents are not expected to affect eelgrass distribution, but here wave exposure is more important (Fonseca et al. 1998), unless the sediment has high water content (see below). As the tidal difference along the Swedish west coast is small (less than 30 cm), the currents may appear, for example, in straits. In general, however, wave exposure is a more important factor to take into account when choosing a location in Swedish water.

At high wave exposure, erosion of bottom sediment and plants can make restoration impossible. The wave exposure at a site can be estimated by calculating a wave exposure index based on stroke length or fetch, i.e. the length of the stretch of open water on which the wind can form waves (see fact box 2.4.). A threshold value appears to be at a wave exposure index above 3\*10<sup>-6</sup> when eelgrass begins to erode away (Fonseca et al. 1998). However, this is a relatively rough estimate of wave exposure that does not take into account bottom conditions and wave reflection (Koch 1999). Today there are more advanced model tools for estimating wave and current exposure at a site and its effect on seagrass (see, e.g. Infantes et al. 2009). Eelgrass that grows on sites with high wave exposure often have a naturally uneven and patchy distibution unlike a sheltered areas that may have a continuous meadow. Therefore, if the objective is to replace 1 hectare of meadow from a protected site by planting eelgrass on an exposed site, then a larger area must be planted as wave erosion is most likely to remove a large proportion of the planted shoots (Fonseca et al. 1998).

Another measure of wave exposure is erosion and sedimentation rates. On the west coast of the United States, eelgrass cannot survive if the sediment erodes by more than 0.5 mm per day or if the sedimentation rate is higher than 0.3 mm per day (Merkel 1992). Wave exposure can also lead to resuspension of the sediment and thus impaired light supply.

#### Fact box 2.4 Grain size and exposure index

#### Grain size

The grain size of sediments is determined by sorting the sediment into sieve with different mesh sizes. Sediments remaining in a 2.0, 0.55 and 0.063 mm sieve are classified as gravel, sand and fine sand, respectively, while sediments passing through the 0.063 mm sieve are classified as silt and clay.

In Bohuslän, the light supply in the water and the growth of eelgrass decrease with increased proportion of silt and clay in the sediment decreases, where growth is very poor in sediment with a content of silt and clay of more than 50% (figure A, Moksnes et al., Unpublished data). The proportion of silt and clay in the sediment can therefore give a good indication of the growth conditions for eelgrass in an area

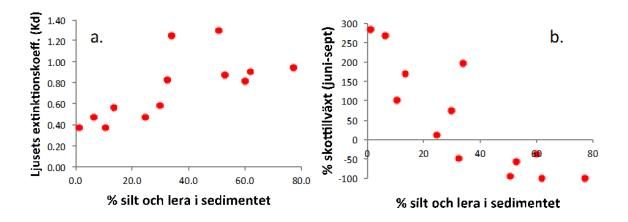


Figure A Relationship between the proportion of silt and clay in the sediment and the extinction coefficient of light (Kd) that reflect how quickly the light is absorbed in the water (a), and the proportion of silt and clay in the sediment to the growth of planted eelgrass shoots (b).

#### Wave exposure index

A wave exposure index can be calculated at each site according to the following equation (Murphey and Fonseca 1995):

Wave exposure = 
$$\sum_{i=1}^{8} (V_i * P_i * F_i)$$

i = compass direction from direction i (1-8), V = maximum wind speed in m / s, (average per month of the maximum wind speed per day), P = percent frequency of wind direction from direction i, and F = effective fetch from direction i (i.e. the length of the stretch of open water on which the wind can form waves)

#### Fact box 2.5 Organic content, water content and erosion of shoots

#### Organic content and water content

The water content in sediment is determined by comparing the weight before and after the sample has been dried at 105° C for 12 hours in an oven. The content of organic matter in the sediment is determined by comparing the weight before and after the sample is incinerated at 520 ° C for 5 hours in an oven.

Studies in Bohuslän (Moksnes *unpublished data*) show that there is an exponential relationship between the water content and the content of organic matter in sediment. Therefore, knowing the proportional water content of a sample, the proportion of organic matter can be calculated according to:

#### Organic content = 0.003294 \* e <sup>5.108 \* Water content</sup>

Similarly, the proportional water content can be approximated if one knows the proportion of organic matter according to:

#### Water content = In (Organic content / 0.003294)/5.108

#### **Erosion of seedlings**

Studies in flume aquariums have found an exponential relationship between the water content of the sediment and the current velocity required to release a young eelgrass seedling (Lillebø et al. 2011), which can be used to approximate the current velocity that seedlings can erode:

#### U = 653.06 \* e -0.056 \* Water content

Where U is the current velocity (cm per second) that results in young seedlings eroding

Although the ratio of water content, organic content and erosion rates can vary between different sediments, depending on e.g. at the grain size, these conditions can be useful for approximating these variables and the risk of erosion of shoots at a site (Figure A).

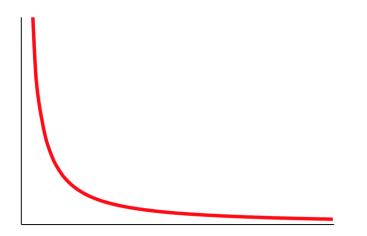


Figure A. Relationship between organic content in the sediment and the current velocity in the water required for a young eelgrass seedling to be pulled up and eroded away, based on Danish studies and the equations above from (Lillebø et al. 2011). Under these conditions, anchoring of young shoots can be problematic at an organic content above 10% (corresponds to a water content >67%) since erosion of shoots can occur already at current speeds around 10 m/s.

In Bohuslän, however, most eelgrass meadows are found in relatively sheltered sites where erosion is usually not a major problem. In the most exposed site examined for restoration in Bohuslän, where the sediment consisted of 98.7% sand and 0.4% organic content at 1–1.5 m depth, planted eelgrass shoots showed very high survival and growth (Eriander et al. 2016), suggesting that erosion does not pose a major problem on sites with these sediment conditions.

In contrast, the loss of planted eelgrass seeds was very high in this environment (>99%), which is largely caused by the erosion of seeds (Infantes et al. 2016). Similar high losses of eelgrass seeds have been reported from shallow sites in Denmark (Delfosse & Kristensen 2012). Seed restoration in shallow sandy sediment areas should therefore be avoided unless measures are taken to reduce the loss of seeds.

If erosion from waves or currents is suspected to cause problems at a site site also for planted eelgrass shoots, this can be investigated during test planting by including treatments where the shoots are anchored by special methods (see section 5.1.2). Since planted shoots are relatively poorly achored in the sediment the first days before the sediment has been packed around rhizomes and roots (see fact box 5.1), it is important not to perform large-scale planting the days before unstable weather has been forecasted.

#### Protected environments - organic content, water content and sediment stability

Although eelgrass is adapted to grow in oxygen-poor sediments with high levels of organic matter, high organic content in the sediment can cause problems for eelgrass restoration. The content of water and organic matter in the sediment affects its properties. Among other things, the organic content affects the sulfide content in the sediment (see below) and possibly also the availability of nutrients. Therefore, it has been suggested that an organic content above 5% in the sediment limits the growth of seagrass, although eelgrass can grow in sediment with an organic content of up to 16% in environments with good light conditions (Koch 2001). However, in Bohuslän's healthy eelgrass meadows are found in protected areas where the organic content of the sediment can be up to 25% (Table 2.1), which indicates that the organic content does not limit the distribution of eelgrass in Swedish water.

However, a high content of organic substances and water can also reduce the stability of the sediment, which increases the risk of erosion and resuspension of the sediment with deteriorating light conditions as a consequence (Valdemarsen et al. 2014). Since eutrophication enriches organic material in the sediment (Zimmerman & Canuel 2002), many protected areas in Bohuslän today have elevated organic levels where the sediment is easily stirred up in the water during windy conditions. The reduced stability of the sediment can also cause problems for the eelgrass anchoring, especially for young seedlings with poorly developed root/ rhizome system and relatively large leaf area. Studies in flume aquariua indicate that the stability of the sediment is already affected at 2% organic content and shows an exponential relationship between the water content of the sediment and the flow rate required to root a young seedling. At 2% organic content (corresponds to about 40% water content) a flow rate of about 74 cm per second is required to uproot and wash away the shoot, and at 10% organic content (about 72% water content), a current of only 12 cm per second is sufficient (Lillebø et al. 2011; fact box 2.5).

Therefore, it is valuable to investigate the content of organic material and water in the sediment to determine if wind-driven resuspension of the sediment can cause a problem for water quality, and

if planted shoots can be pulled up by the currents. The latter mainly applies to seedlings. Water content and organic content can be measured relatively easily in sediment samples, and since there is an exponential relationship between these values, one value can be approximated from the other (see Fact box 2.5).

If the organic content is >2% in the sediment (approx. 40% water content), the risk of winddriven resuspension increases and the light conditions in the water at the sites should be investigated extra carefully. If the organic content is >10% in the sediment (water content about 67%) seedling will have problems if the flow rate is >12 cm per second. Therefore, current conditions should be investigated and test plantings performed if the restoration is to be done using seed methods (Table 2.2). However, it should be added that small-scale planting with seeds has been successfully carried out in Bohuslän in a protected site with an organic content of >11% in the sediment (Eriander et al. 2016, Infantes et al. 2016), so only a high organic content material is not enough to disqualify a site from restoration.

#### Oxygen deficiency and hydrogen sulfide

In sheltered environments where the sediment has a fine grain size and a high organic content, the sediment is usually only oxygenated in the upper millimetres. In these oxygen-poor environments, hydrogen sulfide can be formed which is toxic to all organisms and can kill the eelgrass if it enters the plant through roots and rhizomes. However, eelgrass is well adapted to grow in this type of sediment and has special vessels that carry oxygen from the leaves down to the roots where they form an oxygenated area around the roots that prevents the hydrogen sulfide from entering (Holmer & Bondgaard 2001). This works well as long as the environment has good light conditions for oxygen production and oxygenated water at night so that oxygen can be absorbed by the leaves when no photosynthesis occurs. If the light supply deteriorates due to e.g. shading from rapidly growing algae, dock structures or increased resuspension of the sediment, the photosynthesis and oxygen production can however be reduced below a critical level so that hydrogen sulfide enters the plant and causes poisoning. This is especially a problem if there is also lack of oxygen in the water around the eelgrass, e.g. due to algal mats and high water temperatures (Goodman et al. 1995, Holmer & Bondgaard 2001, Holmer et al. 2005). Poisoning of hydrogen sulfide has been found in several studies to be the direct cause of reduced growth and mortality in eelgrass (Orth et al. 2006, Holmer & Nielsen 2007), and is considered to be a contributing cause of observed mass mortality in eelgrass where whole populations disappear from an area in a short period of time. This usually occurs during the late summer when calm and warm weather can cause rapid growth and degradation of macroalgae mats with resulting oxygen deficiency in the bottom water (Greve et al. 2005).

In Bohuslän, where eelgrass often grows in protected environments, the level of hydrogen sulfide in the sediment is relatively high (Holmer et al. 2005), and may therefore be a problem in restoration. However, the studies carried out in Bohuslän to date cannot show any connection between the growth or survival of planted eelgrass and hydrogen sulfide in sediment or plant. Both planted seeds and shoots have shown surprisingly good growth and survival in sediments with high organic content even when light supply has been limited, without having problems with sulfide intrusion. Areas that have lost large meadows of eelgrass, e.g. Kungälv Municipality, have today relatively low sulfide levels in the sediment (Table 2.1). Eelgrass shoots planted at these unvegetative sites show a lower sulfide intrusion into shoots compared to shoots planted in areas with eelgrass, despite poorer lighting conditions in areas that have lost eelgrass. (Moksnes et al., Published data). Sulfide levels in the sediment therefore do not appear to be a decisive factor in selecting restoration sites in this area. One likely reason for the relatively low sulfide levels in the sediment may be that sediments containing high levels of organic matter and sulfide have eroded away when the eelgrass bed is lost.

#### Nutrient supply in sediment

Studies of natural eelgrass in other parts of the world have shown that the growth of plants can be affected by the nitrogen content of the sediment, where levels of ammonium (NH<sub>4+</sub>) below 100 µmol per litre in the sediment's pore water can limit growth (Dennison m (1987). In some areas, it has even been recommended to add nutrients to the sediment during eelgrass restoration (Kenworthy & Fonseca 1992). However, studies conducted in the Kiel Bay in the Baltic Sea in nutrient-poor sediment found no support for the growth of eelgrass in this area being restricted by nutrients, possibly due to a lower nutritional need for eelgrass in the Baltic (Worm & Reusch 2000). Based on this study and considering that sediments in shallow coastal areas in the North Sea are considered to have elevated levels of organic substances and nutrients due to eutrophication, nutrients probably do not constitute a limiting factor for the growth of planted eelgrass in Swedish water.

#### Sampling of sediment conditions

Sampling of grain size, water content and content of organic material is done by means of sediment cores that are either taken from a boat with suitable equipment, or by diving or snorkelling. The cores should be taken down to 6 cm depth, which is the normal depth distribution of the eelgrass roots, and the greatest depth from which the eelgrass seed sprouts can grow up to the surface. The samples should be divided into two depth fractions in order to better assess the exposure level at the sites. At least five random samples per site should be taken at one time. If the depth variets at the site, the samples should be divided into different depth strata.

In order to investigate whether erosion or sedimentation of sediment can be a problem, the PVC tubes used in light measurement can be accurately placed in sediment to a pre-marked mark on the pipe. Changes between the sediment surface and the mark can then be measured at each visit to give a rough estimate of these processes.

### 2.5.6 Epiphytic algae and drifting algal mats

In Bohuslän, the prevalence of fast-growing filamentous algal mats has increased dramatically in shallow coastal areas since the 1980s (Pihl et al. 1995, 1999, Swedish Agency for Marine and Water Management 2012). The increase is considered to be due to eutrophication in combination with overfishing which, through a trophic cascade, has increased the number of small predators and predatory crustaceans, and reduced the number of small algae-eating crustaceans (Moksnes et al. 2008, Baden et al. 2012). The algal mats are dominated by filamentous green algae (*Ulva* spp., *Cladophora* spp.) that often float on the water surface, or filamentous brown algae (*Ectocarpus* sp.) that often grow entangled in perennial vegetation (Figure 2.9). These annual algae cover many eelgrass meadows during the summer and are believed to be an important cause of the eelgrass's dramatic decline in the area (Baden et al. 2003, Baden et al. 2010). **Filamentous algal mats accumulate should be avoided for restoration**. In the Netherlands, floating mats of green algae has caused eelgrass plantings to fail in the intertidal

zone on protected sites (van Katwijk et al. 2009). In Finland, transplantation studies of eelgrass shoots in exposed sites have shown that shading from mats of filamentous brown algae had a strong negative effect on eelgrass growth and survival (Gustafsson & Boström 2014).

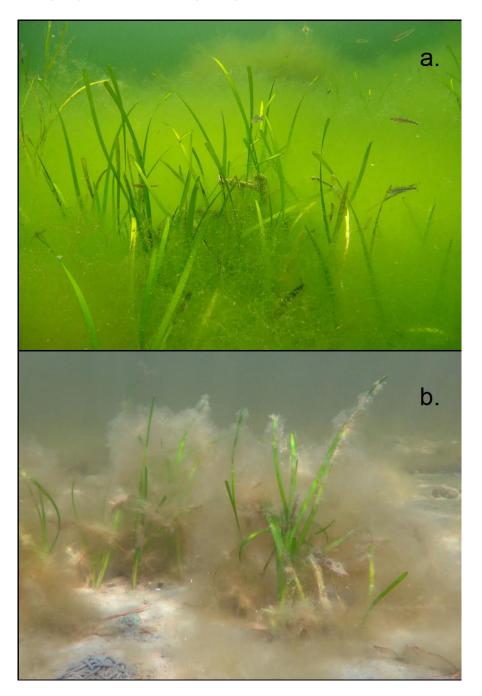


Figure 2.9 Filamentous, annual algal mats. Annual, filamentous algae often form thick mats during the summer that can cover eelgrass meadows and stifle the plants, and therefore make restoration efforts difficult. Image (a) shows an eelgrass meadow in the Gullmarsfjord, which is covered by a mat of filamentous green algae of the genus *Ulva* (formerly called *Enteromorpha*). Image (b) shows eelgrass shoots planted 3 months earlier and covered by the filamentous brown alga *Ectocarpus siliculosus*. Photo: P. Moksnes.

Even algae and sessile animals that grow epiphytically on the leaves can cause severe shading and weight of the eelgrass leaves, which reduces the growth of the plants (Duffy et al. 2014). In Bohuslän, for example, epiphytic microalgae form thick mats on leaves in some areas. In protected sites, tube-building amphipods can also become so numerous that their tubes cover a large proportion of the eelgrass leaves' surface (Moksnes, unpublished data). **If test plantings** are performed, the presence of epiphytes on the leaves can be analysed to determine if this may cause a problem for the restoration.



**Figure 2.10** Drifting, perennial algal mats. In many areas that have lost eelgrass meadows, the bottom vegetation has been replaced by loose perennial brown algae (dominated by *Fucus serratus*; pictured) and red algae (dominated by the red alga *Furcellaria lumbricalis*) that can form several hectares of mats. These drifting algae mats easily tear off or suffocate eelgrass shoots and therefore make it difficult for both natural establishment and restoration of eelgrass at these sites. The algal mats also increase resuspension of the bottom sediment as they drift on the bottom, thereby also degrading the lighting conditions in the water. Photo: E. Infantes.

Studies in Bohuslän show that algal mats (10–1000 meters in diameter) consisting of perennial brown algae (dominated by Fucus serratus) and red algae (dominated by the red alga Furcellaria lumbricalis) today cover large parts of the areas where eelgrass meadows grew in the 1980s (Figure 2.10, 2.11). These algae seem to thrive in these low-light environments where they grow and accumulate at the bottom of 1-3 m deep, and drift around with currents and waves. As the mats move, especially during storms, they can tear off or shade eelgrass with high mortality as a result. These algal mats can therefore potentially destroy large eelgrass plantations. Trials in Bohuslän show that in some sites only shoots that are protected from drift algae by means of cages survive (Moksnes, unpublished data). Studies in Denmark show that drifting algae can account for 40% of the mortality of young seedlings (Valdemarsen et al. 2010) and that the algae also stir up the sediment as they drift along the bottom with increased turbidity and deteriorating light conditions in the water as a result (Canal-Verges et al. 2010). It is therefore important to map the occurrence of drifting algal mats along the bottom of potential restoration sites. If algal mats cover a large part of the seabed, the site should be avoided, especially if the planting is to be done with seeds. If algae mats cover a smaller part of the bottom, the spread of the mats can be mapped. As algae mats often move and accumulate in the same areas year after year, these areas can be avoided to reduce the risk of disturbance.



**Figure 2.11** Inventory of algal mats with aerial photo. The picture shows a shallow bay (<2.5 m) in Ryskärsfjord in Kungälv municipality in 2012, which in the 1980s was almost completely covered by an eelgrass meadow over 200 ha. The eelgrass meadow, which was one of Bohuslän's largest connected meadows, has disappeared today. The dark areas on the bottom are drifting, perennial brown and red algae. Picture from the Swedish Land Survey.

#### Estimation of distribution of algae mats and epiphytes on leaves

Distribution of floating mats of filamentous algae is best estimated using aerial photos. Even the distribution of bottom-drifting perennial algae can be estimated with aerial photos if the weather is calm and the water transparency is greater than the bottom depth. Drones with cameras that take high-resolution images with GPS positions (see fact box 2.3) can be used to estimate the spread of both algae mats and planted eelgrass in shallower areas (see Figure 2.3). Older aerial photos from e.g. The Swedish Land Survey can be useful for estimating the general occurrence of algae mats, although the picture is not from the same year (Figure 2.11).

If aerial photos are not possible, the distribution of floating algal mats can be estimated from a boat where the outer edges of the mats are followed by a boat and marked with GPS points from which the surface can be calculated in the same way as for eelgrass (see section 2.4.1). If test planting of eelgrass is done, it is important to note the coverage of filamentous algae or perennial algae mats on the planted shoots at each visit. Random shoots can also be picked to analyse the biomass epiphytic algae and sessile animals on the leaves. There are no threshold values for when algae mats and epiphytes pose a problem for eelgrass plantings, but if algae mats cover >10% of the potential planting area, they are likely to have a negative impact on the plantings.

### 2.5.7 Disturbance from burrowing and grazing animals

Somewhat surprisingly, biological disturbance from burrowing organisms (bioturbers) and grazers is one of the biggest problems for restoration of seagrass (Paling et al. 2009). Before newly planted eelgrass shoots or seedlings have developed new roots, they are susceptible to disturbances in the sediment, and activities from a wide range of organisms can cause the shoots to become loose or buried, thus leading to very high losses of planted seagrass, often 100% mortality. Also, grazing of leaves and rhizomes from especially seabirds, fish and urchins can also cause major damage to plantings (Fonseca et al. 1998, Short et al. 2002a).

Grazing by seabirds is especially a problem in tidal areas where the birds reach the seagrass at low tide and can have great negative effects on eelgrass plantations (Short et al. 2002a). Many crustaceans and fish species can also consume eelgrass seeds (Wigand & Churchill 1988; Sumoski & Orth 2012).

#### Damage to shoots

In Swedish waters, fish species that can graze seagrass are lacking, but potentially shore crabs (*Carcinus maenas*), swans (*Cygnus* spp.), geese and sea urchins (*Strongylocentrotus* spp.), all of which are common along the Swedish west coast and part of the Baltic Sea, can cause grazing damage to eelgrass plants. Swans and geese could primarily affect shallow plantings that they can reach from the surface. However, there are no studies on how grazing from these species affects eelgrass shoots in Swedish water. Shore crabs along with lugworm *Arenicola marina* and sandworm *Nereis virens* can also interfere with planted shoots with their digging activities (Philippart 1994, Davis & Short 1997, Davis et al. 1998), but there are no experimental studies in Swedish waters. However, test plantings show that individually planted shoots have high growth and survival even at high densities of lugworm (18 individuals per square meter; Eriander et al. 2016), suggesting that bioturbation from lugworm does not pose a major problem in restoration with shoots.

In Swedish waters, shore crabs in particular appear to be a potential problem for eelgrass restoration. Studies in the Northwest Atlantic, where the shore crab is an invasive species, have shown that they can both dig up planted plants and graze on the plant itself, which can lead to both natural and restored eelgrass meadows being eliminated (Davis et al. 1998, Malyshev & Quijón 2011, Garbary et al. 2014). In Sweden, there are no studies of how shore crabs affect eelgrass shoots, but in some sites shore crabs have been observed to rip the leaves on planted shoots to eat from the lower part of the plant, leading to characteristic damage to the shoots (Garbary et al. 2014; Figure 2.12). In these sites, digging activity and grazing from shore crabs are suspected to be an explanation for the failure of test plantings (Moksnes, unpublished data). Since shore crabs are normally very abundant in shallow soft bottom areas and are believed to have increased in numbers in the North Sea over the last 30 years, including as a result of reduced predation from cod (Eriksson et al. 2011), they could potentially cause extensive damage to eelgrass plantations. However, there is no knowledge as to the conditions and densities that shore crabs may pose as a problem when planting eelgrass shoots. At many sites where test plantings have been carried out the shoots were not damaged despite high densities of shore crabs. However, the following observations may indicate that shore crabs are a problem at a site:

- Shore crabs are common
- Characteristic pits formed by burrowing crabs are common among planted shoots (see Figure 2.12).
- A large proportion of planted shoots disappear quickly for no apparent reason
- Planted shoots show characteristic damage (see Figure 2.12)



**Figure 2.12** Bioturbation and grazing from shore crabs. Shore crabs can interfere with the planting of eelgrass by digging up newly planted shoots, or by tearing the leaves and eating the lower part of the shoots. Problems from beach crabs can be indicated if pits from burrowing crabs are numerous in the planting (picture on the left) or if characteristic crab damage occurs on the shoots (picture on the right). Photo: P. Moksnes.

If shore crabs appear to adversely affect plantings, other sites should be considered for restoration. If restoration is still executed at such a site, it is recommended that the shoots be planted with a density of at least 16 shoots per square meter, as studies suggest the crabs will then only be able to injure a smaller portion of the shoots, allowing the planting to survive (Moksnes, *unpublished data*).

#### Damage to eelgrass seeds

Factors that can cause damage to eelgrass seeds are particularly relevant if restoration is to be done with seed methods. However, because seeds produced from planted shoots are important for planting growth even when shoots are used as a restoration method, seed predation of shore crabs and bioturbation of lugworms can also adversely affect shoot planning.

Shore crabs (>10 mm in spine width) are effective predators of eelgrass seeds and one crab can consume over 20 seeds per day and reduce seed plantings by> 70% in one week (Infantes et al. *in review*). Also hermit crabs (*Eupagurus* spp.) and sea urchins can consume eelgrass seeds, but to a lesser extent than shore crabs (Infantes et al. *in review*). Seed predation from crabs probably explains a significant portion of the large losses of planted seeds found in Bohuslän (see Table 4.1). An effective way to reduce seed predation from shore crabs is to bury the seeds 2 cm below the sediment surface (Infantes et al. 2016 *in review*). However, there are currently no

cost-effective methods for Scandinavian conditions to bury the large number of seeds needed for large-scale restoration.

Studies in Denmark show that the bioturbation of lugworm (*A. marina*) can bury eelgrass seeds deeper than 6 cm in the sediment where the sprouts cannot reach the sediment surface, causing the young plants to die. These studies show that lugworms at high densities (80 lugworms per square meter) can bury 60% of planted seeds below this critical depth in one month, suggesting that just over 10 lugworms per square meter is sufficient for a majority of planted seeds should fall below the critical depth over an 8-month winter period. However, at low densities ( $\leq$ 5 lugworms per square meter), lugworms can have a positive effect on seed plantings by burying the seeds just deep enough to levels where losses caused by seed predation or erosion of sediment are minimized (Valdemarsen et al. 2011, Delefosse & Christiansen 2012). Therefore, if seeds are to be used as a planting method, it is important to assess the abundance of lugworms at the sites.

#### Sampling of the abundance of lugworms

The density of the lugworms is tested by placing a number of 0.25 m<sup>2</sup> sampling quadrats at random on the bottom of the sampling area. Within each quadrat, the sediment and all faeces piles of lugworm are smoothed out by hand. After an hour, the quadrats are revisited and the number of faeces piles is counted. The faeces of the lugworm are recognised by the fact that the faeces are several millimetres wide, unlike other burrowing polychaetes.

The method excludes the possibility that the same lugworm produced more than one faeces pile. Each quadrat should be provided with rope and float to the surface to facilitate the reading. This sampling can advantageously be done with snorkelling.

The sampling of lugworms is best done in the autumn (August - September) when eelgrass seeds naturally release from the flower shoots. For lugworms, at least 10 test quadrats per site are recommended on at least two occasions. If the abundance of lugworm is >10 individuals per square meter, this can have negative effects for seedling and if the abundance is >50 individuals per square meter, restoration with seeds is not recommended for the sites.

# 2.6 Test planting

A very important part of the process of selecting suitable sites for restoration is to perform a test planting of eelgrass at the sites that are considered most suitable based on measurements of the above mentioned physical and biological variables. This step is important to test if the potential sites and planting methods really allow the growth of eelgrass, and to select the best sites for the project. Certain factors such as physical exposure, varying lighting conditions and drifting algal mats are difficult to evaluate via monitoring of variables. In several sites in Bohuslän, test planting has shown that areas are unsuitable for eelgrass restoration, although monitored variables indicated that environmental conditions could allow growth at planting depth (see Fig. 2.6). **Therefore, test planting should always be performed in all large-scale restoration projects before the very expensive restoration work is started on a full scale (Fonseca et al. 1998, Short et al. 2002a).** 

#### 2.6.1 Test planting of shoots

A test planting of shoots should be carried out according to the recommendations given for largescale restoration of harvesting and planting methods (see section 5). **The study is should be started in early June and monitored until May of the following year** before being evaluated. It is important to evaluate winter survival, which can be very low in Swedish water, before the choice of sites is made. In order for the evaluation of sites to take no more than one year, test planting of shoots can be carried out the same year as the monitoring of variables is carried out. If the monitoring of variables is started in early May, data from one month's sampling can be used to select the most suitable sites for test planting before it is started in June. If possible, test planting should be carried out in more sites than is necessary for the restoration, so that the results can be used to select the best sites.

In general, it is recommended that restoration is only performed at sites where test plantations show positive shoot growth after one year.



Figure 2.13 Test planting with shoots. Test planting using the single shoot method within a planting quadrat at 1.8 m deep in the Gullmarsfjord. Photo: E. Infantes.

The test plantings are carried out by planting shoots in smaller groups that are replicated within each site being examined. Normally, the shoots are planted in 0.25 - 1.0 m<sup>2</sup> large squares at predetermined densities (Figure 2.13). Different types of treatments can be included in the test plant depending on which issues are relevant to the project. A particular question may only be relevant to a site and then only tested there. However, it is important that there are comparable treatments at all sites so that the results can be compared. For example, eelgrass from the same donor meadow(s) must be used at all sites. It is also important that there are replicates of all treatments so that the results can be statistically tested.

An important issue to investigate in most projects is what planting density can be used as this has a strong impact on restoration costs. This can be investigated by including a treatment with different planting densities (for example, 4 and 16 shoots per square meter). If depth or light availability is expected to affect survival, treatment with different planting depths can be included. If conditions at a site indicate that e.g. wave exposure and erosion can affect the plantings, a treatment with different anchoring methods (see section 5.1.2 and fact box 5.1) can be included in this site. If more than one donor meadow is planned to be used (see section 5.3), the shoots from different sites should be evenly distributed between different treatments and potential restoration sites. See fact box 2.6 for an example of how a test plant with different treatments can be designed and performed.

The test plantings should be sampled on three occasions: after one month to see if site conditions have caused rapid losses of shoots, after 2–3 months (in August – September) to estimate survival and growth after the first growing season, and after 11 months (in May the following year) to assess winter mortality. For the first two sample time points, only non-destructive methods should be used to measure the eelgrass variables shoot density , number of leaves per shoot and maximum leaf length per shoot, which can be sampled in the field by divers (see section 6 for details on planting monitoring). If the number of leaves per shoot decreases over the summer and is below 4 leaves per shoot on adult plants, this indicates that the plant is stressed (Carr et al. 2012, Eriander et al. 2016). It is also important to estimate the amount of epiphytic algae and drifting algae mats that can affect the plantings. The occurrence and bioturbation of beach crabs, as well as typical damage to shoots from crabs (see Figure 2.12) should also be estimated.

Large-scale restoration should only be carried out at a site if the test planting shows positive growth after one year, and where other variables indicate good conditions for a long-term re-establishment of eelgrass. If the test plantatings survive at several sites, the sites that show the highest growth can be selected for the full-scale restoration.

#### Fact box 2.6 Example of design of test planting with shoots

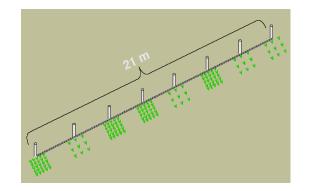
In the example below, there are 4 potential restoration sites where 2 should be selected for a large-scale restoration project. Two suitable donor meadows have also been identified within the same study area. Initial light measurements indicate that eelgrass can survive down to 2.5 m at the sites. Because the bottom slopes sharply at all sites, eelgrass needs to be planted at different depths in order for the meadow to reach sufficient size. It is therefore decided to test plant at two different depths at each site: 1.5 and 2.2 m. Furthermore, two different planting densities will be tested at all sites (0.25 and 0.5 m distance between shoots, corresponding to a shoot density of 4 and 16 shoots per square meter). Finally, shoots should be used from two different donor meadows.

The three different treatments are combined in every conceivable way at the 4 sites with 2 replicates of each treatment, giving 16 planting squares (1 m<sup>2</sup> in size) at each site, and 64 squares in total for 4 sites (Table A). As no major differences are expected between shoots from the two equivalent donor meadows, only two replicates are used to limit the scope of the study. Nine and 25 shoots per planting plot are used for the lower and higher planting densities (see Figure A). Therefore, a total of 1088 vegetative eelgrass shoots are needed to perform the planting, and to include 5% waste during harvesting and transport, approximately 600 shoots are needed from each meadow (Table A).

Table A Summary of the number of treatments in the test planting as well as the total number of planting squares and shoots needed for the study.

Sites	4
Depth	2
Shoot density	2
Donor meadow	2
Replicate	2
No. of planting plots No. of eelgrass shoots	64 1088

During the planting, 21 m long transect ropes are used at each depth, anchored to the bottom with reinforcing iron, and where 8 numbered PVC-pipes are placed every three meters where the eelgrass is to be planted (Figure A). The eelgrass is planted using a 1 m<sup>2</sup> planting frame that is temporarily placed at the markings and helps the diver to place the shoots at the correct distance. The 4 different treatments are randomized at each transect.



**Figure A** Transect rope with planting plots placed at marking every three meters with two different densities and two difference origins. The planting squares are placed approximately 1 m from the marking pipe to avoid drifting algae, accumulating around pipes, from interfering with plantings.

# 2.7 Reference meadows

In order to be able to evaluate the results of an eelgrass restoration, reference meadows (natural unaffected meadows as close to the restoration area as possible) are very important for assessing whether changes in planted eelgrass are due to conditions at the planting site and methods used in the restoration, or to natural variations in distribution of eelgrass between different years. Multi-year inventories in Bohuslän have shown that the distribution of eelgrass can be very dynamic and can vary widely within a site from year to year (Nyqvist et al. 2009). Reference meadows are especially important for evaluating the results of large-scale restorations, but are also valuable for assessing the results of test plantings, and should therefore already be included when evaluating potential sites. When carrying out compensating restoration, it is especially important to include reference beds as they can be crucial in assessing responsibility for a failed restoration.

In order to compare restored meadows and reference meadows, the same variables must be monitored (for example, shoot density, biomass above and below the sediment, etc., see section 6), and the reference meadows should be followed for as long as monitoring of the restored area is ongoing. In addition to these variables, the maximum propagation depth and the areal distribution of eelgrass in the reference meadow should be monitored. To avoid variations in time and space affecting evaluation, sampling must be carried out at restored areas and reference meadows during the same period, and at least two reference meadows should be used (Short et al. 2000).

# 2.8 Summary – the execution of site selection for restoration

In fact box 2.7, the most important steps in evaluating and selecting a site for eelgrass restoration are summarised when shoot methods are used. In this schedule, it is suggested that evaluation is done for only about 1.5 years (including only one field season), which may be desirable in environmental compensation cases where time is often limited). If more time is available, sampling 1 can start a year earlier so that the distribution of natural eelgrass and filamentous algal mats can be sampled in the middle of summer when its distribution is greatest.

### Evaluation of available information

Evaluation and selection of sites for restoration begins early in the year by contacting the relevant authorities, possibly reporting or consulting the study. At the same time, information and data are collected on historical and current distribution of eelgrass, as well as on environmental changes and measures that have taken place in the target area. Surveys in the form of aerial photos should also be collected. Based on this data, 10–12 potential restoration sites should be selected (Selection 1; see fact box 2.7).

#### Fact box 2.7 Schedule for evaluation of potential restoration sites

Table A. Working schedule for the evaluation of potential restoration sites

	Task to do
January – April	
Information and permits	<ul> <li>Inform the County Administrative Board and the municipalities. concerned about projects and hear if the work needs to be notified or given permission.</li> </ul>
Collect background information	<ul> <li>Monitoring data on environmental variables in the target area.</li> <li>Data from eelgrass inventories in the target area.</li> <li>Aerial photos from the target area.</li> </ul>
Selection 1	<ul> <li>Identify the causes of eelgrass reduction, possible measures, and assess whether environmental conditions today allow eelgrass growth.</li> <li>Identify historic and current eelgrass meadows, and areas with shallow sediment bottom without vegetation.</li> <li>Choose the 10–12 most promising sites for restoration and 4-5 potential reference meadows in the target area.</li> </ul>
Мау	
Sampling 1	• Visit the selected sites in the field and sample depth, bottom type of, and distribution of eelgrass, drifting algae mats, turbid water, etc.
Selection 2	Select the 5-6 most promising potential restoration sites and 2 reference meadows
Sampling 2	<ul> <li>Sample sediment in the potential restoration sites.</li> <li>Place instruments for measurement of light, temperature and salinity at sites.</li> </ul>
Sampling 3 (2-3 weeks after sampl. 2)	<ul> <li>Clean and read instruments in the field.</li> <li>Observe water conditions, algal mats, etc</li> </ul>
Analysis	Analyse sediment samples and field data.
Selection 3	<ul> <li>Estimate distance to nearest eelgrass meadow (&gt;100 m)</li> <li>Assess the distribution of the suitable bottoms for planting (1-3 m)</li> <li>Assess the presence of algae mats.</li> <li>Assess grain size, water and organic content in sediment.</li> <li>Assess the light supply at potential planting depth.</li> <li>Select the 3-4 most promising restoration sites.</li> </ul>
June	
Sampling 4 Start test planting	<ul> <li>Collect eelgrass shoots from the two reference / donor meadows.</li> <li>Perform test planting at the 3-4 selected sites.</li> <li>Note the presence of drifting algae, water turbidity, etc.</li> <li>Clean and read instruments in the field.</li> </ul>
July	
Sampling 5 (1 month after planting)	<ul> <li>Sampling shoot density, leaf number and length, if any. epiphytic algae</li> <li>Note the presence of drifting algae, crabs, turbid water, etc. Clean and read instruments in the field.</li> </ul>
August – September	
Sampling 6 (2.5 months after planting)	<ul><li>Sampling shoot density, leaf morphology, epiphytic algae.</li><li>Note the occurrence of drifting algae, turbid water, etc.</li></ul>
May (next year)	Retrieve and read instruments, and analyse field data.
Sampling 7 (11 months after planting)	Sample remaining plants and analyse survival and shoot growth.
Selection 4	<ul> <li>Assess light supply and risks from algae mats and resuspension.</li> <li>Assess survival (≥50%) and growth of test plantings.</li> <li>Choose the 2 most promising restoration sites.</li> </ul>
June	

#### Monitoring of sites and test planting

Evaluation of potential sites in the field starts at the beginning of May when 10-12 sites are visited by boat to inventory the bottom depth, sediment type, distribution of possible eelgrass, drifting algal mats, etc. (*sampling 1*). The inventory is carried out using aqua scope or drop video and snorkelling where images from drones or aerial photos are used to more easily find different types of vegetation. The collected data is analysed and evaluated so that the 5–6 most promising sites can be selected for monitoring of variables during the period May – September (*Selection 2*; see fact box 2.7). These sites are visited as soon as possible after the first sampling in order to place isntruments for continuous measurement of light, temperature and salinity, and to take sediment samples (*sampling 2*). This work is best done by diving.

After 2–3 weeks, at the end of May, the sites are visited for observation and reading and cleaning of the instruments, which can be done with snorkelling (*sampling 3*). Sediment samples are analysed in the laboratory, and all data collected is analysed and evaluated to select the three to four most promising sites and determine appropriate methods for test planting (*selection 3*; see fact box 2.7). The selection should take into account that filamentous algal mats have the highest prevalence in June-August (Pihl, et al. 1999) and areas that indicate that algae mats can become a problem should be avoided or followed up during the summer. At the selected sites, test plantings of shoots are then carried out in early June, where eelgrass is harvested at reference sites (future donor meadows) and planted by different methods in the selected sites (see fact box 2.6), which is done with diving (*sampling 4*).

The plantings are then sampled in July and at the end of August - September, when the instruments are also cleaned and read. During these visits, observations are also made on the presence of algal mats, signs of resuspension of sediments, crab damage, etc. At the last sampling in August – September (*sampling 6*) the instruments are picked up and the field data is analysed. The sites are then visited for the last time in May the following year for sampling when winter survival is assessed. Thereafter, all collected data are analysed and evaluated and summarised in a report, after which the two most promising sites are selected for restoration, in consultation with the relevant authorities. This should be done by the end of May at the latest so that any large-scale restoration can begin in early June of the same year (fact box 2.7).

# **3** Consultation and permits

Restoration of eelgrass habitat aims to restore and enhance the status of the environment. Despite this, there may be requirements for the person responsible for the restoration to consult or report the operation to the authority or even apply for a permit before it is started. The purpose of these requirements is to minimize possible risks of interference. What determines whether consultation, notification, permit or exemption may be needed is the type and extent of the environmental impact the restoration may entail, as well as any type of spatial protection.

Before the planning of an eelgrass restoration is started, a first contact should be made with the County Administrative Board and the relevant municipality, with a purpose to inform regarding the plans and obtain views on the project as well as information about any consultations, notifications, permits and exemption that they believe may be needed. It is also good to contact a property owners early.

Eelgrass restoration according to the methods recommended in this manual does not affect the water depth and usually does not have a significant impact on outdoor recreation or marine habitats. Restoration in these cases is not classified as "water operations" (see 3.3) and neither a permit nor a notification for it will then have to be done. The eelgrass restoration usually does not conflict with the *shore protection* or with regulations for marine protected areas, and exemption from these protections is therefore not needed. Thus, the starting point is that the described type of eelgrass restoration will only require a notification of consultation with the county administrative board. But it is the person responsible for the restoration who is obliged to obtain knowledge if additional permits, exemptions or notifications are needed.

However, the notification or permit for water operations may be relevant in cases that plan to modify the environment to improve the conditions for eelgrass restoration, according to the methods discussed in Appendix 2. These methods can also have a significant impact on recreational activities and the environment, and could then also require exemption from *shore protection* or from regulations for marine protected areas if the restoration is to be carried out in, for example, a Natura 2000 area.

Below is a summary of what effect eelgrass restoration can have on the environment, outdoor recreation and various activities. Subsequently, an account is made of circumstances that affect the need for consultation, notification, permit or exemption and the requirements that are made in connection therewith.

# 3.1 Impact on the environment, outdoor recreation and various activities

Harvesting of shoots or seeds has a small effect on the eelgrass meadow where the harvesting takes place, but with recommended methods this effect is only temporary (see section 5.4 and box 5.2) and cannot be seen as a disturbance. If, on the other hand, the harvest of reproductive shoots with seeds should be done with harvesting machines (see Appendix 1), the impact on the environment can be significant, and therefore consultation with the supervisory authority or permit may be required. The restoration work itself may result in a temporary restriction for outdoor activities such as swimming and boating, as well as for fishing (both commercial and recreational

fishing). However, the work of harvesting and planting vegetative eelgrass shoots by hand takes place only during a relatively limited period of time where shoots for one hectare of eelgrass can be harvested and planted by divers in about 10–40 working days (depending on planting density; see fact box 5.3). Harvesting usually involves only a minor restriction in the outdoor recreation, as it takes place in a smaller area in an eelgrass meadow where outdoor activities are usually limited.

If the planting work takes place in such shallow areas that may be of interest for bathing and boating activities and for various forms of fishing, it is recommended to restrict access to the area for the public during the planting work and a few months afterwards to avoid that bathing, turbulence from boat engines or fishing gear damaging the newly planted shoots. If this closure affects individuals and the public's ability to use the area, it may require exemption from the *shore protection* (see further section 3.4 below). However, the low water depth in areas that are restored (1.5–3 m) means that commercial shipping will rarely be disturbed.

# 3.2 Consultation with the County Administrative Board

Even though the restoration is not a water operation, the person responsible for the restoration may be obliged (according to Chapter 12. Section 6 of the Swedish Environmental Code) to consult with the supervisory authority, which in these cases is usually the county administrative board. A decisive factor in whether this consultation is needed is whether the restoration can lead to a significant change in the natural environment. There is no clear limit to what is meant in this case by "significant" which is why the **recommendation is to report the restoration plans even in uncertain cases.** 

A notification for consultation must be in writing and include a map, a description of the planned restoration activities, as well as information about the property owners and usufructuaries affected. The notification should also contain information on how different types of disruptions from the restoration should be handled. If necessary, the notification must also include an environmental impact statement (EIA). If EIA is lacking and the county administrative board considers it necessary, the county administrative board may require one to be established.

When the county administrative board receives a notification for consultation, it this shall determine whether the business is allowed on the site. If the county administrative board has no comments on the planned restoration activities, the notification can be submitted without any restrictions. The County Administrative Board can also submit precautionary measures and even prohibitions when there is a need to limit or completely counteract damage to the area to be restored.

For more detailed information on consultations, see the Swedish Environmental Protection Agency's Handbook 2001: 6.

### 3.3 Permits for water operations

An activity or measure that in some way affects the water depth constitutes a so-called water operation (Chap. 11 Section 3 of the Swedish Environmental Code). The starting point is that all *water operations* are subject to a permit (Chapter 11 § 9), but there are exceptions when it is instead required that the activity be notified and also cases where neither notification nor

permission is needed. Exceptions to the duty to notify or getting a permit, applies *inter alia* when it is not obvious that neither general nor individual interests are damaged. A prerequisite for conducting a *water operation* is that the operator has right of disposal to the water (Chapter 2, § 1 of the Act (1998: 812) with special provisions on *water operations*). In the first place, it is the property owner who has right of disposition to the water that is within the person's property. The right of disposition can also be transferred to another through an agreement. The state, municipalities and water conservation associations, and in some circumstances even individuals can have right of disposition for *water operations* that are desirable from general points of view. Along Swedish coasts, the water areas are mainly public water and the State's representative for these water areas is the Legal, Financial and Public Procurement Agency (Kammarkollegiet). Private water is water areas that extend 300 meters out of the mainland (or island that is more than 100 m long) or out to three meters deep. Within the coastal area from the Gullmarsfjord to the Hakefjord, individual water can never extend beyond 300 meters from land no matter how deep it is.

Therefore, before an application for a permit or notification of *water operations*, one should have an agreement with the property owners to carry out the measure on their land. The state, municipalities and water conservation associations can be granted by the environmental court the right to conduct water activities on another's land that are desirable from a general environmental or health point of view or to promote fishing.

# 3.4 Exemptions

### Exemption from shore protection

The *shore protection* means that it is not allowed to build or take measures that impede outdoor recreation or that substantially change the living conditions for animal and plant species (Chapter 7 15 Swedish Environmental Code), unless it applies to buildings that are needed for an land based industry or when an exemption from the protection has been granted. Exemption can only be granted if there are special reasons (which are stated in Chapter 7 Section 18c of the Environmental Code) and if wildlife and plant life are not unacceptably affected and public access to beach areas is not impaired. The *shore protection* extends 100 meters from the shoreline up on land and 100 meters into the water, but can be extended to a maximum of 300 meters on each side of the shoreline. Under certain conditions, the shore protection may have been completely removed (especially in densely populated areas).

Eelgrass restoration aims to have a positive impact on plant and animal life and is only a temporary obstacle to outdoor recreation. Usually, restoration will not occur in the type of areas where many people reside, e.g. at beaches. However, restoration can lead to a significant, albeit positive, change in living conditions for a number of species. The person responsible for the restoration is obliged to apply for an exemption if needed.

Should an exemption be needed, the county administrative board or the municipality will try if there are special reasons for granting such (Chapter 7 Section 18 of the Environmental Code). To the extent that permits are tried, e.g. according to Chapter 11, the issue of exemption from the *shore protection* will also be considered. If it is a question of notification of *water operations* or consultation in accordance with Chapter 12 section 6, however, is not considered for shore protection is not considered, but an exemption may then have to be sought separately.

#### Exemption from regulations for marine spatial protection

Regulations for national parks and nature reserves are issued in order to maintain the purpose of the protection. These regulations may impose different types of restrictions on the use of land and water areas. Should the regulations for an area constitute an obstacle to the restoration of eelgrass, an exemption may be granted by the county administrative board (in some cases the municipality) if there are special reasons. Some regulations require permission for certain measures. **Therefore, find out if there are any special protections in the area to be restored and what regulations apply.** This information can be found, for example, in the Swedish Environmental Protection Agency's map tool "Skyddad natur" http://skyddadnatur.naturvardsverket.se/ . Contact the person who will decide on an exemption for information on what an exemption application should contain.

Eelgrass restoration according to the methods recommended in the manual will not negatively affect a Natura 2000 area in any significant way and therefore does not need any special permit according to the Natura 2000 rules. Similarly, such restoration will not need an exemption in accordance with the rules on biotope protection (Chapter 7 Section 11 and Appendix 3 of the Regulation on biotope protection (1998: 1252). However, if other plant and animal species within the protected biotope *Large shallow inlets and bays* are affected, exemption may need to be sought.

# 4 Selection of restoration method

Restoration of eelgrass includes both techniques where vegetative plants are harvested from a donor meadow and transplanted at a new site as well as methods where seeds from a donor meadow are spread over the area to be restored. Historically, restoration with adult shoots has dominated, and most of the restoration efforts that have been carried out at present have been carried out by using adult plants (Fonseca et al. 1998, van Katwijk et al. 2009, 2015, Fonseca 2011). Restoration methods with seeds have also been developed over the past 20 years, which in some areas have proven to be very successful (Pickerell et al. 2005, Orth et al. 2012). However, it is important to note that methods that are cost-effective in one area may be ineffective in another because of regional differences in physical factors such as wave exposure ate, water quality, salinity or temperature, or due to ecological differences in, for example, seed production, germination, winter survival of eelgrass, or occurrence of grazing organisms (Fonseca et al. 1998). It is therefore very important that different methods of restoration are initially evaluated for each specific region (Moksnes 2009). Based on multi-year studies in Bohuslän, it is recommended that the restoration of eelgrass in Swedish water is done by transplanting adult shoots without sediment (the single shoot method). Seed methods today cannot be recommended due to high losses and uncertain results. However, these recommendations may change in the future if new seed methods are developed.

# 4.1 Pros and cons of seed methods

In recent years, the use of eelgrass seeds has received attention for its potential to be costeffective in restoration of large areas (Marion & Orth 2010). The advantage of the method is that large amounts of seeds can be collected relatively easily and with small negative effects on the donor population, after which the seeds can be spread over large areas at a relatively small cost. The major advantage of using seeds during restoration is that the workload and cost of seed storage and sowing with mature seeds from a boat (see section 7 and Appendix 1 for details) only increase to a small extent with the size of the project, which is why seed planting in theory is better suited for large-scale projects than planting with shoots. By far the most successful largescale restoration of eelgrass to date has been carried out with seeds, where 125 ha of unvegetated soft bottom in Virginia in the United States were sown with almost 38 million seeds, which after a 10-year period had expanded to a fantastic 1700 ha (Orth m. fl. 2012). Thus, under the right conditions, restoration with seeds can be very effective.

The disadvantage of seed methods is the lack of knowledge about the factors that control the dormancy, survival and germination of seeds (Orth et al. 2000), and the proportion of sown seeds that develop into shoots is very variable (0.1–28% on average; Pickerell et al. 2005, Goshorn 2006, Marion & Orth 2010, 2012, Orth et al. 2012). In comparison with restoration with shoots, seed restoration is generally more uncertain. In a large-scale restoration effort in Chesapeake Bay, USA, the overall survival of seeds after storage and planting was so low (0.06%) that restoration largely failed (Goshorn 2006). Furthermore, the number of seeds produced in a meadow varies widely between different sites and years (Marion & Orth 2010), which also makes the availability of seeds and the cost of harvesting uncertain.

Along the Swedish west coast, intensive research has been conducted on developing seed methods for eelgrass restoration since 2011, and a large number of attempts have been made to

plant eelgrass seeds on different sites in Bohuslän (Table 4.1). Although methods are now available for efficiently harvesting reproductive shoots and extracting seeds in Swedish water, and seedlings have shown very good growth in some areas (Infantes et al. 2016; Figure 4.1), high and varying losses of eelgrass seeds constitute a major problem with the usage of seeds in restoration with the methods available today. Comprehensive experiments with planting eelgrass seeds in Bohuslän have shown very variable survival between sites and year, and generally very high losses of seeds, especially at depths shallower than 3 m where on average less than 1% of the seeds developed into shoots, also in areas with relatively good environmental conditions. In several attempts, no shoots have been developed at all despite 10,000's of seeds being sown. Furthermore, the survival of the seedlings has been very low, especially in affected areas that have lost eelgrass, where only two of 19 trials have resulted in long-term survival of planted seeds (Table 4.1). In total, about 295,000 seeds have been sown for 4 years over about 1100 m<sup>2</sup> in Bohuslän, which only gave rise to an estimated approximately 20 m<sup>2</sup> eelgrass that has shown long-term survival. The large losses in Swedish water are probably related to the fact that many seeds are washed away or eaten up during the long winter period when the seeds are dormant until they germinate in the spring, and that the sensitive seedlings are disloged or shaded by drifting algae mats, or die of light deficiency in the turbid waters (see Appendix 1 for details). Therefore, with the methods currently available for harvesting and sowing of eelgrass seeds in Swedish water, the use of seeds in eelgrass restoration cannot be recommended as the large amount of seeds that would be required makes the method many times more expensive than planting vegetative shoots by hand (see section 7), and more uncertain. However, at a depth of over 3 meters in protected sites, seed survival is relatively high (1.7–6.4%), and although restoration of such deep sites is generally not recommended due to very low shoot growth (see section 2.5.1), seed restoration in these environments is possibly a better alternative than planting shoots.

If methods can be developed that reduce the loss of seeds or reduce the cost of producing seeds, seed-based restoration can be an important method for large-scale eelgrass restoration even at shallower depths along the Swedish west coats. When this handbook was written, work is underway in Denmark to try to develop cost-effective methods for large-scale restoration with seeds (see www.NOVAGRASS.dk). So even though it is difficult to recommend large-scale restoration with seeds at shallower depths in Bohuslän today, due to the high losses and costs, we present detailed methods for how seeds should be harvested, stored and sown in Appendix 1, with the hope that the problems with today's high losses will be solved by time.

**Table 4.1** Summary of seed planting studies in Bohuslän 2011–2015. A total of 23 different experiments have been carried out at 8 different sites, in Gullmarsfjorden (site 1-2), Gåsö outside Gullmarsfjorden (site 4) and inside Marstrand in Kungälv municipality (site 7-12; see Figure 2.4 for a map of the sites). Large losses of eelgrass have been documented since the 1980s in the municipality of Kungälv (>98%), and in Gåsö (>40%), while losses in the Gullmarsfjord have been minimal. In the studies, eelgrass seeds were planted with different densities (40–4200 seeds per square meter) within experimental plots of varying sizes (0.12–20 m<sup>2</sup>) using different methods (Net = net boxes with seed shoot, Hand = by hand from boat or diver, S = seeds covered with sand or stones, C = Cage that excludes seed predators; see Appendix 1 for explanation). In total, over 295,000 seeds have been planted (Total seeds) in 378 experimental plots (No. of plots) over a total of 1,132 m<sup>2</sup> (Area). On average, only 0.77% of sown seeds have given rise to seedlings in the spring (% seedl). Of the seedlings that developed in the spring, only one-third of the shoots survived in the autumn (% Surv Fall), and only about half of the shoots survived until the following year (% Surv. Yr 2), most of which were found in the Gullmarsfjord.

Site	Plant. date	Depth (m)	Method	No. of plots	Area (m²)	Seed- density	Tot. seeds	% seedl	% Surv Fall	% Surv Yr 2
1	Sep 2011	1.1 to 4.6	Net	6	6	400	2 400	1.63	100	100
	Oct 2012	5.0	Hand + S	15	1.8	4200	7 500	3.79	100	100
	Apr 2013	5.0	Hand + S	15	1.8	4200	7 500	6.44	100	100
2	Jun 2011	1.2-4.0	Net	6	6	400	2 400	0.50	67	67
	Oct 2012	1.0-3.0	Hand + S	45	5.4	4200	22 500	0.60	100	100
	Apr 2013	1.0-3.0	Hand + S	45	5.4	4200	22 500	1.04	100	100
4	Oct 2012	1.2-1.6	Net	5	50	300	15 000	1.40	0	0
	Oct 2012	1.2-1.6	Hand	5	0.12	4200	2 500	2.40	0	0
	Oct 2013	1.5	Hand + C	32	8	40-4000	9 680	0.58	0	0
10	Oct 2012	2.5	Hand	5	0.12	4200	2 500	0	0	0
	Apr 2013	1.3-1.5	Hand	18	4.5	40-4000	6 660	0.14	0	0
	Sep 2014	1.0-2.0	Hand + S	36	9	2000	18 000	0.01	0	-
11	Oct 2012	3.5	Hand	5	0.12	4200	2 500	1.40	100	0
12	Oct 2012	2.2	Net	5	50	300	15 000	0	0	0
	Oct 2012	2.2	Hand	5	0.12	4200	2 500	0	0	0
	Oct 2012	2.2	Hand + S	15	1.8	4200	7 500	5.91	100	67
	Apr 2013	2.2	Hand + S	15	1.8	4200	7 500	1.29	67	0
	Oct 2013	1.5	Hand + C	32	8	40-400	9 680	0	0	0
	Oct 2013	2.2	Hand	2	800	40	32 000	0	0	0
	Sep 2014	1.4-1.8	Hand + S	24	6	2000	12 000	0	0	-
	Apr 2015	1.8	Hand	4	80	40-400	35 200	0	0	-
13	Oct 2012	2.0	Hand	5	0.12	4200	2 500	0.8	0	0
	Sep 2014	1.2-1.6	Hand + S	24	6	2000	12 000	0.03	0	-
	Apr 2015	1.2-1.4	Hand	4	80	40-400	35 200	0	0	-
15	Oct 2012	1.2	Hand	5	0.12	4200	2 500	5.40	100	100
				378	1132		295 220	0.77		



**Figure 4.1** Growth of seedlings. The picture shows the growth of a seed shoot which has grown at 1.5 m depth at the Torgestad (Site 2) sites in Gullmarsfjorden in May 2013 and which after 14 months developed into a mat of rhizome with around 30 shoots. Although only 0.4% of the seeds sown in the autumn remained and could grow, the growth of the few surviving seedlings was very high. Photo: L. Eriander.

# 4.2 Pros and cons of shoot methods

The advantage of using adult shoots as a restoration method for eelgrass is that there is a long experience and detailed guidelines for different planting methods (see Moksnes 2009 for a summary in Swedish). Another advantage is that the transplanted shoots constitute a habitat more quickly and that the eelgrass ecosystem services are thereby recovered more quickly, in comparison with seed methods. In addition, it is possible to more quickly get indications of how transplanted material survives initially at shoot planting, while it takes longer to be able to do an initial evaluation at seed sowing in northern Europe, because the seeds do not germinate until the spring, the year after they are planted. Evaluating sites for restoration with seeds therefore takes twice as long as evaluating sites for shoot planting.

The disadvantage of planting adult shoots is that harvesting and planting must be done by hand, which takes time and entails high costs for large-scale restoration projects, especially in Swedish areas where small tidal differences require divers for planting the shoots. The time-consuming work that also requires calm weather conditions **limits the size of areas that can be restored with shoot planting to less than 5 hectares per season for a dive team of 6 people** (see section 5).

However, in Sweden transplantation with vegetative shoots in Bohuslän, unlike seed methods, has produced good results in areas where environmental conditions allow eelgrass growth. For example, in the Gullmarsfjord, eelgrass shoots planted in small experimental planting quadrats totalling approximately 12 m<sup>2</sup> has grown and spread to a small meadow of around 100 m<sup>2</sup> over a 4-year period (see Table 4.2, Figure 4.2). In an ongoing large-scale planting experiment on Gåsö outside the Gullmarsfjord, 600 m<sup>2</sup> eelgrass was successfully harvested and planted by three divers for three days in June 2015, where the number of planted shoots increased by over 220% on average after three months (Moksnes, *unpublished data*). **Today, it is therefore** 

#### recommended that eelgrass restoration in Swedish water be carried out with shoot

**planting.** In areas where environmental conditions have deteriorated as a result of the disappearance of large eelgrass meadows, also planting with shoots has provided poor survival (Table 4.2), which highlights the importance of evaluating the suitability of sites before large-scale restoration is initiated (see section 2).

In chapter 5, we first summarize some of the most common methods for restoring eelgrass with adult shoots and then provide recommendations for the shoot methods that have proven to work best along the Swedish west coast coast, both in terms of growth and cost-effectiveness in harvesting and planting. Detailed descriptions of seed methods developed for Swedish conditions are presented in Appendix 1. Studies in Bohuslän have shown that eelgrass can no longer grow in some areas that have lost large eelgrass meadows because the environment has changed as a result of the disappearance of eelgrass. Appendix 2 discusses future measures that could be developed to improve the environment and the possibilities of restoring eelgrass in these areas.



**Figure 4.2** Growth of planted vegetative shoots. The picture on the left shows the planting of 9 vegetative shoots with the single shoot method within a 0.25 m<sup>2</sup> quadrat at 1.5 m depth at the Torgestad (Site 2) sites in Gullmarsfjorden in June 2011. The picture to the right shows the same quadrat 14 months later in August 2012 when more than 200 shoots were found. In an inventory in August 2015, all 24 planting squares (a total of 12 m<sup>2</sup>) had grown into a small meadow of over 100 m<sup>2</sup> that grew down to 4 m depth. Rapid propagation has probably occurred both with vegetative rhizome growth and with seed production. Photo: E. Infantes.

**Table 4.2** Summary of shoot planting studies in Bohuslän 2011–2015. A total of 24 different experiments have been performed at a total of 13 different sites, in the Gullmarsfjord (sites 1-2), Gåsö outside the Gullmarsfjord (sites 4), in the Stigfjord (sites 5), Hakefjorden (sites 7-9) and inside Marstrand down to the Nordreälv estuary in Kungälv municipality (sites 10-15; see Figure 2.4 for a map of the sites). Large losses of eelgrass have been documented since the 1980s in Kungälv municipality (> 98%), Hakefjord (> 75%) and Gåsö (> 40%), while losses in the Stigfjord and Gullmarsfjorden have been minimal. In the studies, vegetative eelgrass shoots were planted at different depths using different methods (Method: SS = single shoot method, P = plug method, A = shoot anchored with bamboo sticks, B = shoot with cages that protect against drift algae; see section 5 for details), different shoot densities (Shoot no. m<sup>-2</sup>) within experimental plots of different sizes (Plot m<sup>2</sup>). In total, approximately 18,000 shoots have been planted in 432 experimental plots (number of plots) over approximately 1,300 m<sup>2</sup> (Area m<sup>2</sup>). The percentage survival of shoots in September the same year that the shoots were planted (% Surv Yr 1) and in September of the following year (% Surv Yr 2), as well as the total area of eelgrass in the fall of 2014–2015 (Area 2015; m<sup>2</sup>) are indicated in the last columns. Survival values above 100% indicate growth in the number of shoots (ex: 111% indicate that the number of shoots increased by 11%). Dash indicates missing data.

Site	Plant date	Depth (m)	Method	Shoot no. m <sup>-2</sup>	Plot m <sup>2</sup>	No. plots	Area m²	% Surv Yr 1	% Surv Yr 2	Area 2015
1	Jun 2011	1.2-1.5	SS, P	16, 32	0.25	24	6	366	650	≈30
		4.0-4.5	SS, P	16, 32	0.25	24	6	111	60	3
2	Jun 2011	1.2-1.3	SS, P, A	16, 32	0.25	30	7.2	365	628	≈80
		3.0-4.0	SS, P	16, 32	0.25	24	6	201	151	≈20
4	Jun 2015	1.1-2.2	SS	4, 16	10	6	600	221	-	≈400
5	Jun 2012	1.5	SS	16	0.25	12	3	269	596	-
7	Jun 2015	1.4-1.6	SS	16	0.25	6	1.5	82	-	-
8	Jun 2015	1.6	SS	16	0.25	3	0.75	0	-	0
9	Jun 2015	1.3	SS	16	0.25	3	0.75	4	-	-
10	Jul 2011	2.4	Р	12-36	0.25	3	0.75	52	0	-
	Jun 2012	2.4	SS	16	0.25	12	3	0	0	0
	Jun 2013	1.2-1.8	SS	4, 16	10	6	600	0	0	0
	Jun 2014	1.0-2.0	SS, A, B	16	0.25	72	18	9	0	0
11	Jul 2011	3.2	Р	12-36	0.25	3	0.75	172	-	-
	Jun 2012	3.2	SS	16	0.25	12	3	187	0	-
12	Jul 2011	2.3	Р	12-36	0.25	3	0.75	63	-	-
	Jun 2012	2.2	SS	16	0.25	12	3	172	370	-
	Jun 2014	1.4-1.8	SS, A, B	16	0.25	72	18	8	0	0
13	Jul 2011	2.3	Ρ, Β	12-36	0.25	3	0.75	0	0	0
	Jun 2012	2.3	SS	16	0.25	12	3	0	0	0
	Jun 2014	1.2-1.6	SS, A, B	16	0.25	72	18	3	0	0
14	Jul 2011	1.2	Ρ, Β	12-36	0.25	3	0.75	44	0	-
15	Jul 2011	1.2	Р	12-36	0.25	3	0.75	295	-	-
	Jun 2012	1.2	SS	16	0.25	12	3	256	248	-
						432	1296			≈530

# 5 Restoration with vegetative shoots

# 5.1 Description of different methods

A variety of methods have been used to transplant adult eelgrass shoots from a donor meadow to an area without vegetation for the purpose of restoring or compensating historic eelgrass losses. Restoration with vegetative eelgrass shoots has been carried out in the US (e.g. Davis & Short 1997, Orth et al. 1999, Short et al. 2002b), Europe (e.g. van Katwijk et al. 2009) and Asia (e.g. e.g., Park & Lee 2007, Li et al. 2010). In Scandinavia, only small-scale attempts to restore eelgrass have been carried out in the Limfjord in Denmark (Christensen et al. 1995). In the Baltic Sea, only experimental studies have been carried out today in the Gulf of Kiel (Worm & Reusch 2000). In Finland, eelgrass shoots have been transplanted together with other aquatic flowering plants for scientific studies of how growth and survival are affected by shading (Salo et al. 2009, Gustafsson & Boström 2009, 2013) amongst other things. The methods previously used can be divided into two main categories: planting shoots **with sediment** and **without sediment**. The following is a brief description of the most common methods. For a more detailed description in Swedish, refer to Moksnes (2009). **For Swedish conditions, planting vegetative shoots without sediment is recommended using the single shoot method** (see below).

### 5.1.1 Planting with sediment

The method that has long been the most common and according to the literature has been considered least invasive to the plant is to transplant shoots inside sediment from the donor meadow. The most common technique is the so-called "plug method" where eelgrass shoots and accompanying sediment are harvested by means of a tube that is pressed down into the meadow (Fonseca et al. 1998; Figure 5.1). However, the method is very labour intensive and costly and is most suitable for transplantation at low tide in tidal areas. It also produces greater negative effects in the donor meadow when holes are left in the sediment. Studies in Bohuslän showed that the plug method can also work with diving, but growth was lower than when the shoots were planted without sediment (Eriander et al. 2016). The time required for harvesting, transporting and planting plugs with sediment was also several times longer and the **method is therefore not recommended for large-scale restoration in the North Sea.** 

### 5.1.2 Planting without sediment

Several different successful techniques have been developed for planting shoots without sediment. Most methods use different types of anchoring to avoid that the shoots are pulled up by water movements (Davis & Short 1997, Fonseca et al. 1998, Calumpong & Fonseca 2001, Short et al. 2002b), but also methods without anchoring have been developed (Orth et al. 1999). The most common methods of anchoring shoots are by using staples or planting frames. An example of the use of staples is the so-called "staple method", where small bundles of shoots with rhizome are tied together with degradable lines and anchored in the sediment with a U-shaped staple. The staple anchors the plants in the sediment, and enables them to withstand current speeds up to 50 cm per second (Fonseca et al. 1998). A simplified version of this method uses only two eelgrass shoots per planting unit and the plants are placed parallel to the shoots in the opposite direction and anchored to the sediment using biodegradable bamboo barbecue skewer that is bent in the middle and pressed down over the rhizome (Davis & Short 1997).



**Figure 5.1** Plug method. Restoration using the "plug method" where eelgrass shoots and accompanying sediment are harvested by means of a tube that is pressed down into the meadow and which is then placed in a prepared hole in the area to be restored. The method is very labour intensive and slow, and is not recommended for restoration in Sweden. Photo: E. Infantes.

A transplantation method that has been developed to be able to perform eelgrass restoration with the help of volunteers and to minimize the need for diving is the so-called "TERFs™ Method" (Eng. "Transplanting Eelgrass Remotely with Frame Systems") where the shoots attached by degradable strings to a metal frame which is lowered from a boat at the restoration sites and left on the sediment surface until the shoots have rooted in the sediment (Chumpong & Fonseca 2001 Short et al. 2002b). However, the method is not suitable for large-scale restoration due to high costs.

A very fast and efficient restoration method is the "single shoot method" (SSM) where individual shoots are planted by hand without anchoring. By pushing the rhizome of the plants into the sediment in a 45° angle (approx. 3-4 cm) an undisturbed layer of sediment is kept on top of the rhizome, which reduces the risk of the shoot being dislodged. This method has the advantage

that the number of shoots that need to be harvested is lower than with methods where several shoots are planted together, and the planting process is much faster as no anchoring material need not be used (Orth et al., 1999; Figure 5.2). Trials in the US (Orth et al. 1999) and Korea (Park & Lee 2007) have shown good survival and growth comparable to methods that use anchoring. In Bohuslän, restoration with SSM has proven to work very well, with a survival and growth that was even higher than when shoots were planted with sediment or with anchorage support (Eriander et al. 2016). Thus, planting without anchoring is generally recommended with the single shoot method for transplanting adult eelgrass shoots in Sweden.



**Figure 5.2** Single shoot method. A vegetative eelgrass shoot on the way to be planted with the single shoot method in a shallow bay in Bohuslän. By pushing the rhizome into the sediment in a ca 45° angle, an undisturbed layer of sediment is deposited on top, which reduces the risk of the shoot being detached. A skilled diver can plant 300-400 shoots per hour using this method. Photo: E. Infantes

### 5.1.3 Plant density and design of large-scale restoration

For large-scale restoration where 1,000 to 10,000 square meters of eelgrass is planted at the same site, the shoots need to be distributed to optimize survival, growth and propagation of the planted meadow. Since the cost of a restoration increases almost linearly with the number of shoots planted per unit area, it is desirable to use as few shoots as possible, without causing reduced growth or survival due to too low shoot density.

Studies have shown that higher planting density can increase the survival of shoots in exposed environments by the shoots protecting each other from physical forcing and dislodging. Studies in the Gulf of Kiel in the Baltic Sea found that eelgrass shoots planted at 20 cm intervals had higher shoot growth and higher survival after autumn storms than shoots planted at 40 cm intervals (corresponding to approximately 25 and 6 shoots per square meter, respectively; Worm & Reusch 2000). In a similar study in the Wadden Sea, Bos and van Katwijk (2007) found that the survival rate was higher in eelgrass with a planning distance of 30 cm than with 50 cm (corresponding to about 14 and 4 shoots per square meter, respectively), but only in exposed areas and not in protected environments. At densities above about 60 shoots per square meter, competition between the shoots may limit the growth of eelgrass (van Katwijk et al. 1998), so a planting distance less than 13 cm should be avoided. In Bohuslän, where eelgrass usually grows in sheltered environments, studies in semi-exposed shallow sites showed that shoot growth

increased when planting density was lowered from 32 to 16 shoots per square meter without adversely affecting survival (Eriander et al. 2016).

In the literature, the planting distance between eelgrass shoots (or groups of shoots) for largescale restoration varies from 15 cm to 2 m (corresponds to densities of 44 to 0.5 shoots per square meter) where a planting distance of 0.5-1.0 m between groups of shoots dominates (Davis & Short 1997, Fonseca and others 1998, Orth and others 1999, van Katwijk and others 2009). In Bohuslän, ongoing large-scale planting studies in a protected bay show a higher survival and shoot growth for shoots planted with 25 cm distances (corresponding to 16 shoots per square meter) than shoots planted with 50 cm distances (4 shoots per square meter), possibly due to interference from shore crabs (see section 2.5.7). Preliminary results therefore indicate that eelgrass can be restored with a planting distance of 50 cm under good conditions, but that higher densities may be required when disturbances occur. Since the cost of eelgrass restoration is directly proportional to the number of shoots planted (see Section 7), **it is recommended that planting distances of 50 cm (4 shoots per square meter) be tested with test plantings and used whenever possible.** 

The design of planting units for large-scale restoration also varies widely in the literature where both rows and different patterns with squares are used (Davis & Short 1997, Fonseca et al. 1998, Orth et al. 1999, Leschen et al. 2010). For practical reasons, plantings are often carried out over smaller areas (4 to 20 m in size) where planting densities etc. are easier to handle. These guadrats can then be placed in a row, or spread out in specific patterns. For example, when smaller squares were used, squares were often placed with empty squares between them so that a check pattern is obtained within the restored area (Orth et al. 1999, Leschen et al. 2010). However, small planting units should be avoided as studies of natural eelgrass show that mortality is increasing in small units with less than 36 shoots due to decreased protection from erosion (Olesen & Sand-Jensen 1994). In exposed areas where the eelgrass naturally grows more spotted, it is recommended that the plantings be designed in a similar manner. Fonseca et al. (1998) recommend a planting distance of no more than 50 cm between shoots and that the shoots are collected in groups that are approximately 5 to 10 m in diameter for areas with flow rates above 30 cm per second or areas with fetch lengths over 1 km. However, there appear to be a lack of studies evaluating how different planting patterns affect the success of more protected areas, and since large-scale restoration studies in Scandinavian waters are still lacking, it is difficult to make recommendations for this area. It is therefore important that all Swedish restoration efforts are carefully monitored and evaluated so that knowledge can increase.

Below is an in-depth description of the entire restoration process based on the single shoot method being used in conditions that dominate in Bohuslän.

## 5.2 When to carry out a restoration

The growing season for eelgrass in Scandinavian waters is relatively short and extends approximately from April to September, but varies depending on temperature and light conditions. In Scandinavia, the growth and biomass of eelgrass is greatest during July to September (Boström et al. 2014). To maximize shoot growth in the first year (and winter survival; see below), **it is recommended that transplants of eelgrass shoots be executed from late May to early July.** It is also important to note that the growing season varies with the light conditions at the restoration site, which means that deeper and lower water quality sites have shorter growing seasons (see Figure 2.5).

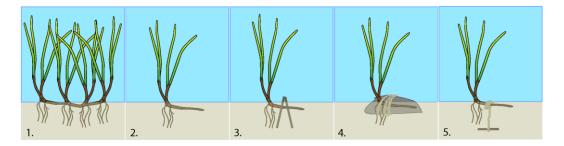
The reason why one should avoid transplanting shoots during late summer or fall is that the carbohydrates stored in the rhizome during the growing season are essential for the eelgrass to survive long periods of poor light such as during the winter season (Zimmerman et al. 1995, Govers et al. 2014). It is therefore important that the transplant is performed at a time that allows maximum shoot and rhizome growth before the first winter, especially when shoots are moved to a site with poorer lighting conditions than at the donor meadow. For the same reason, restoration should be avoided during early spring, as depleted carbohydrate reserves can make plants transplanted with a short rhizome sensitive to the low light conditions prevailing during spring.

Other reasons why the spring and autumn period should be avoided during restoration is that the storm rate is higher, which can make restoration work difficult and cause planted shoots to be detached from the sediment. The date for the restoration should be flexible enough to be able to choose a period of at least 1 week with calm wind conditions that do not create waves in the restoration site. Studies have shown that shoots planted according to the single shoot method have anchoring capacity comparable to natural meadows less than 10 days after planting (see Fact box 5.1.).

#### Fact box 5.1 Methods for anchoring shoots

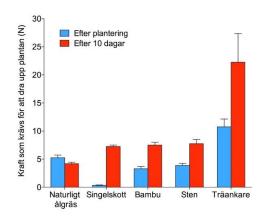
If pilot studies indicate that plant dislodgement may be a problem, the shoots may need to be anchored in the sediment. There are a wide variety of methods for anchoring eelgrass shoots described in the literature that provide different degrees of anchoring and which entail different amount of work and thus increase costs to varying degrees. Below a smaller study is described that compared the anchoring of 4 different planting methods, with or without anchoring (E. Infantes, *unpublished data*).

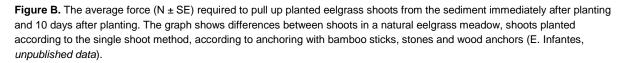
**Anchoring Study.** The study was conducted in 2014 at a depth of one meter in a semi-exposed site in the Gullmarsfjord where transplanted shoots anchored in the sediment were examined directly after planting and again after ten days by measuring the force required to pull the shoot vertically out of the sediment. Shoots were planted according to 3 different anchoring methods and according to the single shoot method and compared with the anchoring of shoots in natural eelgrass beds (Figure A).



**Figure A.** Planting methods investigated in the study. In the study, anchoring of natural eelgrass (1) was compared with 4 different planting methods: (2) Single shoot method where no anchorage is used, (3) anchorage with bamboo stick pressed down over the rhizome (Davis & Short 1997), (4) anchorage with stones (Zhou et al. 2014) and (5) anchorage with a wooden stick as anchor that were attach to the rhizome (Merkel 1988).

The study showed that shoots planted according to the single shot method are easily dislodged from the sediment immediately after planting, while the anchoring methods gave the shoots an anchorage that was comparable or stronger than natural eelgrass (Figure B). Ten days after planting, when the sediment was packed by wave motion and roots may have been formed, the shoots without artificial anchoring were also well anchored in the sediment and the forces required to pull them up were comparable to those required to pull up natural and shoots planted with anchoring methods. However, the method with wood anchors gave twice as much anchoring as the other methods (Figure B). These results demonstrate the need to choose a period of calm wind conditions when large-scale restoration is to be performed according to the single-shoot method, in order to avoid that newly planted shoots are pulled up. However, in areas with frequent wave exposure or strong currents leading to erosion, some type of anchorage can increase the ability of the shoots to remain in sediment.





## 5.3 Selection of donor meadows

When restoring, it is of great importance to select the donor meadow that contribute with shoots for the restoration in a way that minimizes the risk of damage to the donor meadow and maximizes the potential for shoot survival and growth at the restoration site. During environmental compensation, when there is a meadow that will be destroyed during exploitation, if possible, these meadows should be used primarily for the collection of transplant material.

### Match the conditions

The morphology of eelgrass shoots show large variation depending on the physical and chemical environment in which they grow. In general, the shoots are longer in deeper and more protected sites, compared to shoots that grow shallow or exposed (Figure 5.3). General advice when choosing donor meadows is that the environmental conditions at the donor meadow should be as similar as possible to those at the restoration site in terms of exposure rate and depth (Fonseca et al. 1998, van Katwijk et al. 2009). However, studies in Bohuslän have shown that transplanted eelgrass shoots have a great ability to adapt their morphology and growth strategy to new environmental conditions, so **shoots with a leaf length of 20–50 cm can be recommended for restoration in several environments at depths between 1.5–3,5 m** (Eriander et al. 2016). However, transplantation between highly diverse environments should be avoided as high mortality rates are observed when short shoots (20 cm) are planted at deep sites (over 4 m) and when long shoots (70–90 cm) are planted at shallow (1 m) more exposed sites (Eriander et al. 2016).



Figure 5.3 Eelgrass' varied morphology. The leaf length and shoot density of eelgrass varies greatly depending on wave exposure and light supply. In shallow, exposed sites, the leaves are short (<30 cm) and the shoot density high (left image), while the leaves are long (up to one meter) and the shoot density is low in deep, light-poor environments (right image). Studies have shown that this difference is not genetic. If a shoot is transplanted between these environments, it changes shape in a few months (Eriander et al. 2016). Photo: E. Infantes.

Another reason why shoots larger than 50 cm should be avoided is that the handling time of these types of shoots is increases and that they are bulkier during storage and transport and since long shoots from protected environments are generally more fragile and break more easily. Donor meadows where the shoots are an average size of 20–50 cm are usually found in areas with a depth of between 1–3 meters, but the depth at which these shoots are found can vary with exposure and lighting conditions at the sites.

### Minimize impact

Although studies in Bohuslän show that up to 40% of shoots in a meadow can be harvested without adversely affecting the meadow (if the shoots are harvested by hand thinning the meadow; see fact box 5.2), donor meadows should not be selected in areas where the eelgrass is threatened. For example, harvesting for large-scale restoration should be avoided in Kungälv municipality in Bohuslän as more than 98% of the meadows that were mapped there in the 1980s have disappeared today (only about 13 ha remain) and the trend is still negative (see Moksnes et al. 2016, section 3.3.3).

To minimize the impact, donor meadows that grow in moderately exposed sites should be selected and the shoots should be picked in relatively shallow water with good lighting conditions. This is because growth and branching of the eelgrass rhizome (which is important for recovery) are generally higher there compared to sheltered (Fonseca and Kenworthy 1987) or deeper environments with poorer light conditions (Eriander et al. 2016). Sheltered sites where the sediment has a high water content should also be avoided as the harvesting is made more difficult as the loose sediment easily leads to high turbidity and poor visibility for a long time.

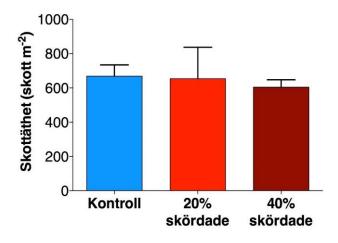
To avoid negative effects on the donor meadow, **no more than one-third of the adult shoots should be harvested within the harvest area, which in turn should account for less than half of the total meadow.** In addition, **meadows less than 0.25 hectare** (50x50 m) **should be avoided** as smaller meadows may be more susceptible to disturbance.

## Genetic aspects

The ongoing loss of eelgrass in the North Sea could potentially deplete genetic diversity within the species. Managers should therefore strive to maximize genetic diversity by selecting donation plants from genetically diverse donor meadows, and from several different, geographically and genetically distinct populations. If all "donation plants" are taken from the same meadow, a high degree of genetic similarity may be obtained within the restored meadow, which could inhibit sexual reproduction and make the population more susceptible to disease or other disturbances (Fonseca et al. 1998, Borum et al., 2004). Studies in the Netherlands have shown that low genetic diversity in planted eelgrass reduces growth and fitness (genetic adaptation to the environment) in plants (Williams 2001). This is especially true in areas where the eelgrass do not reproduce sexually with seeds as the genetic difference is large between different areas (for example in the central Baltic; Boström et al. 2014).

#### Fact box 5.2 Harvesting of vegetative shoots - effects on donor meadows

To study the possible negative effects of harvesting vegetative shoots during restoration using the single-shoot method, shoots were harvested within marked 1 m<sup>2</sup> large areas of approximately 1.5 m depth within a healthy eelgrass meadow identified as a potential donor population on northern Tjörn (Viks kile, see site 5, Figure 2.4). The shoots were harvested by divers by picking them by hand one by one so that the meadow "thinned" according to the recommendations in chapter 5.4. Two different harvest intensities were investigated: 100 and 200 shoots per m<sup>2</sup> (corresponding to about 20 and 40% of the shoot density at the time of harvest), which were compared with control areas where no shoots were harvested (n = 3). The harvest was carried out at the beginning of June 2012 and the areas were sampled after about 4 months at the end of September when shoot density, biomass and leaf morphology were measured.



**Figure A**. Average shoot density (+SE) per  $m^2$  in three different treatments: control, 100 shoots (20% harvest) and 200 shoots (40%) harvested per  $m^2$  4 months after shoots were harvested.

Four months after the harvest, no significant differences in shoot density were found between harvested and unharvested areas (Figure A). Nor did the biomass of leaves and rhizomes (per m<sup>2</sup>) show any significant effects of the harvest. The only parameter that differed between harvested areas and controls was the leaf length, which was significantly longer (about 74 cm) within control areas compared to areas where shoots were harvested (52-57 cm long shoots). The difference in leaf length is probably due to the fact that the plants in the harvested areas have set many new lateral shoots and therefore consist of a larger proportion of young shoots that have not yet reached maximum leaf length in comparison with the control areas. Since surrounding plants are lower in these treatmnets, the shoots do not have to grow as high to compete for light. Studies in Bohuslän have shown that eelgrass shoots quickly adapt their morphology to e.g. lighting conditions in the environment (Eriander et al. 2016).

The results of this study, conducted in a seemingly healthy eelgrass meadow, in an area with good water quality show that **up to 40% of all shoots can be harvested without any negative effects found in the meadow after 4 months.** 

Along the Swedish west coast, however, the degree of genetic isolation between different populations of eelgrass is relatively small (B. Källström & C. André, unpublished data ; Eriander et al. 2016), which indicates that the populations are not isolated but have an frequent genetic exchange with each other, probably through the spread of floating reproductive shoots with seeds. This reduces the risk of genetic depletion if only plants from a donor meadow are used. Nevertheless, **at least two donor meadows are recommended for large-scale restoration.** 

### Logistic aspects

If possible, meadows near the restoration area should be chosen first and foremost as this facilitates transport times, stress on shoots and planting costs. The donor meadow can also be a good reference meadow to compare the result from the restoration (see section 2.7) and as such it should also be as close to the restoration area as possible. In this case, the donor meadow must be large enough that the harvested area represents only a small proportion (<25%) of the meadow, so unaffected areas of the meadow can be used as a reference.

## 5.4 Harvesting and transport

## Permits, personnel and equipment

Before commencing harvesting and planting, it is important that appropriate notifications and possible permits have been granted for harvesting and planting of shoots (see chapter 3). For the harvesting work, a fast-moving boat is needed that can take at least four people, diving equipment and the harvested material. In the described restoration studies, a 5 m plastic boat with a 50 hp outboard motor was used, which worked well for this purpose. The harvesting of shoots can in some cases be effective with snorkelling, if the water depth is so shallow that the shoots can be reached from the surface, but in other cases diving is recommended as it is more efficient. For diving work, the current Swedish dive rules for work diving should be followed (AFS 2010: 16), whereby a dive leader and boat driver are needed in addition to the divers in the water. As diving takes place in shallow water (<3 m), air rather than bottom time is limiting, so harvesting can be performed for many hours by the same people.

### Minimize impact

When harvesting, it is important to avoid as much as possible unnecessary negative impact on the meadow when the work is done. In order for this to happen, it is important not to anchor in the eelgrass meadow and that the divers move carefully in the meadow and try to minimise disturbance by being well-balanced in the water and avoid standing up in the meadow. In order to ensure that the harvest is done in the correct extent (less than 1/3 of the shoots are harvested in maximum half of the meadow), marking buoys should be used to identify harvested areas. It is also important not to harvest more than what can be planted in the next few days to minimize wastage. Since harvested eelgrass should be planted as soon as possible and not stored for longer than 2-3 days in submerged net bags (see below), good planning is required with regard to weather forecasts and vacant days. Optimally, harvested eelgrass should be planted the same or following day, since it should be taken into account that the planting work takes about 25% longer than the harvesting work (see fact box 5.3).

#### Fact box 5.3 Harvesting and planting

#### Calculation of number of shoots needed.

The number of shoots that need to be harvested for a planting is determined by the total area to be planted, the distance between each planted shoot and the amount of shoot loss during transport and planting. When planting eelgrass shoots with the single shoot method, it is recommended that individual shoots are planted at intervals of 25 to 50 cm (4 to 16 shoots per square meter; m<sup>2</sup>) depending on physical exposure, disturbance from beach crabs, etc. and expected shoot growth, which is best investigated in test planting (see section 2.6). The number of shoots needed total can be calculated according to the following ratio:

$$N_{shoot} = (sqrt(A)/D)^2$$

where  $N_{shoot}$  = number of shoots needed in total, A = total area to be planted (in square meters) and D = distance between each shoot (in meters). The number of planted shoots per square meter is then obtained by:

Shoot per 
$$m^2 = N_{\text{shoot}}/A$$

For example, when restoring one hectare (10,000 m<sup>2</sup>) with shoots planted at 50 cm intervals, 40,000 shoots will be needed. Including approximately 5% spillage of plucked shoots, approximately 42,000 shoots need to be harvested (see table below).

**Calculation of the harvested area.** In shallow water (0.5-1.5 m depth), the shoot density of eelgrass is often 400-1,000 shoots per m<sup>2</sup> (Boström et al. 2003, table 2.1). Based on an average number of 700 shoots per m<sup>2</sup> and that 25% consists of adult shoots suitable for transplantation, each square meter contains approximately 175 suitable shoots. If one-third of these are harvested, about 58 shoots can be harvested per square meter. To harvest enough shoots to plant one hectare of eelgrass with a shoot distance of 50 cm (4 shoots per m<sup>2</sup>), approximately 720 m<sup>2</sup> of the donor meadow needs to be harvested (corresponding to an area of about 27 × 27 m). At a planting distance of 25 cm (16 shoots per m<sup>2</sup>), about 4 times as many shoots and 4 times as large harvest area is needed (see table below).

**Calculation of the working hours.** Harvested eelgrass shoots should be planted as soon as possible in order to not lose quality. To avoid the destruction of harvested material due to uncertain weather conditions it is therefore recommended for the work to be planned in 2-day sessions, where shoots that are harvested on day 1 are planted on day 2 (fact box 2). Since it is about 20% faster to harvest shoots by hand than to plant them with the single shoot method, more dive hours must be planned for planting than harvesting.

Experienced divers or snorkelers can pick about 420 shoots per hour on average, so a dive pair that harvests 5 hours a day can harvest a total of 4,200 shoots in one day, and

42,000 shoots over a total of 10 working days. An experienced diver can plant about 335 shoots per hour on average with the single shoot method. A dive pair can therefore plant about 4,000 per day for 6 hours of diving work and a total of 40,000 shoots during 10 working days. Therefore, in order to harvest and to plant one hectare with 4 shoots per m<sup>2</sup> (40,000 shoots), a total of 20 working days are required during a period of just under 5 weeks (if work is not performed on weekends). To plant one hectare with 16 shoots per m<sup>2</sup> the time required is 4 times as long (Table A).

**Table A.** Shoot and work requirements for harvesting and planting one hectare of eelgrass with two different shoot densities. Calculation of shoot density of planted shoots, number of harvested shoots needed (including 5% loss), harvested area in donor meadow, and number of working days for *2 divers* to harvest and plant one hectare of eelgrass with a mutual planting distance between shoots of 50 and 25 cm, using the above-mentioned assumptions of shoot density, harvest speed and dive hours per day.

Planting distance (m)	Planting shoots per m <sup>2</sup>	Harvest shoots	Harvest m <sup>2</sup>	Harvest days	Planting shoots	Planting days
0.50	4.0	42 000	720	10	40 000	10
0.25	16.0	168 000	2880	40	160 000	40

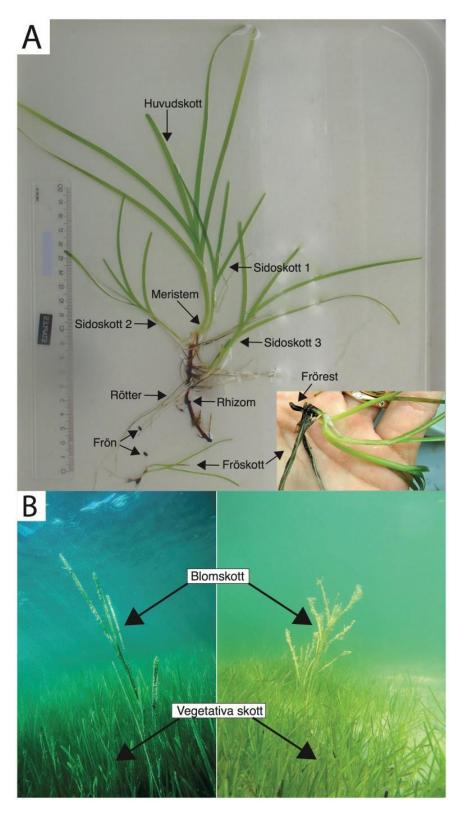
### Identification of vegetative shoots

When transplanting, only adult vegetative shoots should be used and it is therefore important that divers are well-versed in how they are identified and separated from other types of shoots that can be found in a meadow (see Figure 5.4). Since shoots of widgeon grass (tasselweed; Ruppia spp.) can grow mixed with eelgrass, it is also important to be able to distinguish this species. In the eelgrass meadow, the shoots grow vegetatively through the branching of the stems in the sediment (rhizomes). Each rhizome has a fully developed head shoot (apical shoot) with a frontal growth zone (meristem), which is in the direction that the rhizome spreads as it grows. It is this shoot that should be harvested and used in restoration to enable vegetative spread at the restoration site. As the plant grows, a large number of branches with lateral shoots are often formed along the rhizome, which are also branched so that a mat of rhizome is formed with hundreds of shoots from the same individual. When a new branch is formed, the lateral shoots are smaller in size (5–10 cm) than the adult shoots and should then be avoided as transplant material because they have poorer survival and growth. Adult shoots (20-50 cm long leaves depending on the physical conditions of the site) at the end of each branch along the main rhizome are suitable for transplantation (Figure 5.4a). During early spring (March – May), seedlings (i.e., small young shoots developed from seeds) can also be found in the eelgrass meadow. These are initially small and have not yet developed rhizome, and the remains of the seed can often be found at the root threads (Figure 5.4a). These young shoots are very sensitive to handling and should therefore not be used in transplantation. During the summer (June -August), reproductive shoots can also be found in the meadow. These are often easy to discern as they are taller than the rest of the meadow and have spathes with flowers and seeds that gives the leaves a branched appearance (Figure 5.4b). It is these reproductive shoots with seeds that are harvested during restoration with seeds, but should be avoided during shoot planting as they wither after the seeds are released.

## Harvesting

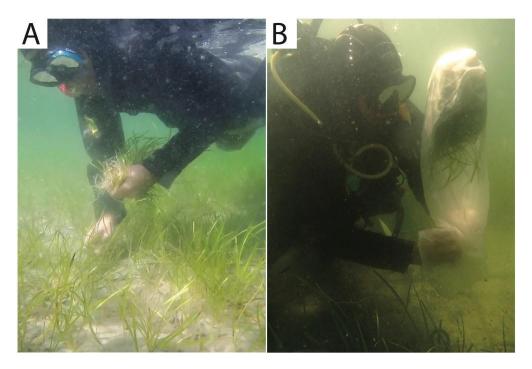
Before harvesting work, a suitable spot in the meadow is marked with buoys where the density of 20–50 cm long shoots is high. If the sediment has a high content of clay or organic material that is easily stirred, it is important that the divers work against the stream so that the visibility is maintained when the shoots are shaken clean from sediment. The divers, who work in pairs, should pick about **one-third of all adult shoots** at regular intervals so that the meadow "thins". At normal shoot densities, a dive pair can harvest over 5000 shoots during 6 hours of diving work (see fact box 5.3).

Marking buoys are left in the corners of the harvested area so that no sub-area is harvested more than once and so that areas are left that have not been harvested. To restore one hectare of eelgrass where the shoots are planted at 50 cm intervals, approximately 42,000 shoots are needed, which can be harvested from a 0.072 ha large area of one or more donor meadows (corresponding to an area of about  $27 \times 27$  m in total; fact box 5.3). Therefore, the total area affected by the harvest in a donor meadow is relatively small in relation to the area planted at this planting density. Although large losses of eelgrass have been documented in Bohuslän, there is still a lot of eelgrass (a total of between 5,000 and 13,000 ha of eelgrass; Moksnes et al. 2016, Appendix 1), where eelgrass is found in almost all water bodies. Access to donor meadows should therefore be good in most areas.



**Figure 5.4** Vegetative, seed and reproductive shoots. Different types of eelgrass shoots that can be found in a meadow. Figure A) shows the structures of a vegetative shoot consisting of a branched stem (rhizome) with roots where a main shoot and 3 smaller lateral shoots grow up. On the main shoot, the growth zone (meristem) is highlighted. A two-month-old seed shoot is also shown, where the seed is still stuck to the root zone. Figure B) shows a deep and shallow eelgrass meadow during the month of July where the taller, branched reproductive shoots are clearly visible among the vegetative shoots. For eelgrass restoration with shoots, only adult (20-50 cm) vegetative, adult shoots should be used. Photo: L. Eriander and E. Infantes

During the harvest work, vegetative head shoots are picked one by one by following the shoot's leaves with the fingers down into the sediment to the rhizome, which is then broken off about 5-10 cm from the meristem. Each picked shoot thus contains an adult end shoot with a rhizome length of about 5–10 cm where smaller lateral shoots may occur (Figures 5.2 and 5.4a). Harvested shoots are shaken free of sediment underwater and stored in the hand until about 50 shoots are counted, after which they are bundled using rubber bands placed 2–3 cm above the meristem (Figure 5.4a). Bundles with collected shoots are stored in net bags (approx. 70x50 cm; approx. 1 mm mesh size) which are attached to the diver with carabiner hook (Figure 5.5). Because eelgrass has positive buoyancy, it is important that the bags are fastened in a long enough rope to reach the surface so as not to interfere with the work. When the net bag is full of bundled shoots, they are taken to the boat where the boat driver transfers them to a larger storage bags that is hung from the boat, and provides the diver with a new empty bag. It is important to ensure that harvested shoots are kept moist as the leaves can dry out quickly. The larger storage bags should therefore be hung on the shadow side of the boat at sunshine and turned periodically so that all shoots are kept moist (Figure 5.6), or weighed down below the water surface.



**Figure 5.5** Harvesting of vegetative eelgrass shoots. A) Harvesting can be done with snorkelling (if it is possible to reach the shoots from the surface), but should be done with diving in deeper water. The shoots are picked one by one and placed in bundles of 50 shoots joined together with rubber bands. B) Bundles with shoots are moved to net bags that are stored in the water during most of the restoration process. Photo: E. Infantes.



Figure 5.6 Storage of eelgrass shoots in large net bags during collection. Photo: E. Infantes.

## Transportation

When transported to the restoration site, the net bags are picked up in the boat where they are kept moist, protected from dehydration by means of a tarpaulin or inside boxes with lids. To ensure that the shoots that are harvested are viable, it is important to handle the shoots carefully throughout the restoration process. This is especially true of the plants' growth zone (meristem) which is the most sensitive part of the shoot. Therefore, avoid storing eelgrass bags too densely packed or stacked on one another in a way that can cause pressure damage or that the leaves are folded off during transport.

If transplantation cannot take place on the same day as the harvest, the shoots should be kept completely submerged in the water, if possible at the restoration area. If the shoots are stored in tanks on land, it is important that the water is oxygenated properly during the night as lack of oxygen can kill the shoot. As eelgrass shoots float, nets stored in the field must be weighed down with weights to prevent them from lying on the surface where dehydration or sunlight can damage the shoots. When storing shoots that are densely packed in bags submerged in the water, the shoots are viable for up to 72 hours after harvesting (Davis & Short 1997), but should preferably be planted as soon as possible after the harvest (Fonseca et al. 1998).

## Samples for monitoring

Prior to planting work, representative shoots should be taken from collected eelgrass (n = 40) to determine different eelgrass variables at the start of restoration. These can then be compared with how the shoots and planting develop during monitoring (see section 6). Variables that should be measured on all shoots collected are: (1) number of side-shoots per harvested plant, (2) number of leaves per shoot, (3) the length of the longest leaf, (4) and the length and number of internodes on the rhizome (from the meristem and back; see Figure 5.4a). In addition, dry weight of (4) leaves and (5) rhizome and roots (leaves divided from the rhizome at meristem) can also be taken. Dry weight is determined by drying the eelgrass for 48 hours at 60  $^{\circ}$  C.

## 5.5 Planting

## Information and planning

Since the plants are susceptible to disturbance and easily detach from the bottom the first week before the sediment has been packed (see fact box 5.1), it is important to mark the planting area well with buoys and to set up signs where boat activities, fishing, swimming, etc. are excluded within the planting area during the first summer. The County Administrative Board may issue temporary regulations for the protection of a restoration area. It is also important to contact landowners and inform residents in the area about this. For the same reason, planting should only be carried out under calm wind conditions, especially on more exposed sites where waves may otherwise cause large losses of planted shoots.

Since planting takes place directly with the harvest, it must be carefully planned so that harvested shoots can be planted less than three days after they are harvested. To avoid the destruction of harvested material due to uncertain weather conditions it is therefore recommended that the harvested shoots and planted in the same day, or for the work to be planned in 2-day sessions, where shoots that are harvested on day one are planted on day two (fact box 5.3). Since planting should be performed at depths greater than one meter (see section 2.5.1), diving is recommended during planting as snorkelling at this depth becomes ineffective.

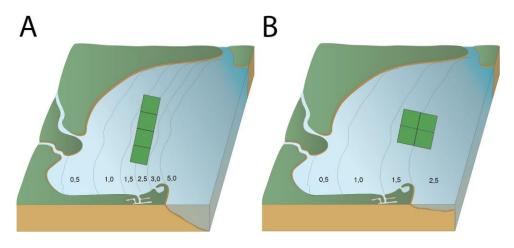
To spread the risk of, for example, a storm destroying the entire restoration attempt at one time (see section 1.2), it is recommended, if possible, that large-scale restoration work be up using at least two different sites within the same target area that are restored in parallel over two or more different years. However, in dividing the work, any planting area should not be less than approximately 0.1 ha large if possible (see section 1.2).

## Methods and design of planting area

Based on studies in Bohuslän, it is recommended that eelgrass shoots be planted using the single shoot method where the shoots are planted by hand one by one without anchoring. If test planting indicates that wave exposure or current conditions can lead to dislogment and loss of planted shoots, the shoots may be anchored. For moderate physical exposure, anchoring with curved bamboo sticks is recommended, which is the least labour-intensive and thus the least costly anchoring method. In case of high exposure, anchoring with wood anchors may be needed, which provides a strong anchorage immediately after planting (see fact box 5.1). However, this method entails considerable additional work in planting as these anchors must be manufactured and tied to each individual shoot, so the cost associated with anchoring must be weighed into the equation when selecting a site (see section 2). Planting distances between shoots also have a significant impact on cost and should be evaluated in the site selection studies (see section 2.6).

For practical reasons, it is recommended that plantings take place in rectangles or squares with a length of about 10–25 m along a certain depth curve within the planting area. The size of each planting unit is not central and can be decided by what a diver is expected to be able to plant in a day. Since an experienced diver can plant about 2000 shoots during 6 hours of diving work, a planting unit of 500 m<sup>2</sup> (20 × 25 m) is suitable when the shoots are planted at 50 cm intervals (4 shoots per square meter), and a unit of about 125 m<sup>2</sup> (approx. 11 ×11 m) when 16 shoots per square meter should be used. These planting units are placed in a row or in groups depending on

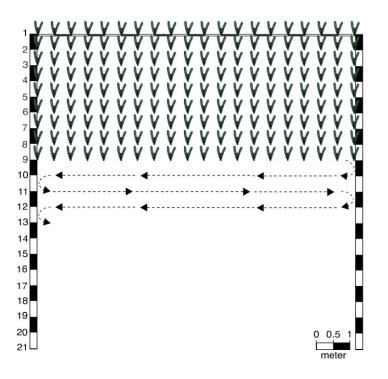
the lighting conditions and the topography of the selected restoration site. For Scandinavian conditions, it is recommended that eelgrass should not be planted at a depth of shallower than about 1.5 meters, and generally not deeper than 2.5 m (section 2.5.1). Many coves along the Swedish west coast are narrow with sloping bottoms, which means that the planting units can be narrower than 25 meters and must be placed in a row to ensure that the shoots are planted at optimum depth, which means that the planting area can get a long narrow shape (Figure 5.7a). In other areas with more homogeneous bottom depth, the planting units can be placed in the centre of the bay where the depth conditions are optimal (Figure 5.7b). We recommend that several planting units be placed at close intervals so that a planting area of at least 1,000 m<sup>2</sup> (0.1 ha) is achieved. When the shape and design of the planting is determined, it is important to mark the planting area with surface buoys. This facilitates both the planting work itself but also when the surface is to be relocated under surveillance and to protect the plantings from, for example, boat traffic.



**Figure 5.7** Examples of design of planting surfaces in two bays. A) The example shows a bay with a sharply sloping bottom where optimum planting depth has been set to between 1.5 and 2.5 meters. Due to the sharp slope, the planting units are therefore placed in a row along the bay. B) The example shows a site with very slight slope of the bottom where the planting units are instead planted more centred in the bay.

## **Execution of planting**

In order to identify possible causes of damage or death of the shoots, monitoring of planting should start, and instruments that continuously measure light, temperature and salinity be placed out (see section 6) before planting begins. The planting work starts by marking all the corners of the planting units with a rod with buoy to the surface and with measuring tape or marking lines with half meter markings along all 4 sides (Figure 5.8). These tape measures act as a guide for the person planting to ensure that the shoots are planted at the correct distance. The tape measure and the ropes are removed after the planting is completed and then moved to mark the next square.



**Figure 5.8** Description of the planting process with shoots. The picture shows as an example a planting unit of 100 m<sup>2</sup> where shoots are planted at 50 cm intervals. 10 m long marker lines are deployed along all sides to facilitate navigation and placement of shoots. After the first row is planted, the shoots on this row are used as a reference when row two is planted, etc.

The diver performing the planting brings the eelgrass shoots in a net bag with ca 10 bundles of 50 shoots each, where the net bag is attached to the diver with a carabiner via a rope of suitable length so that the net bag does not interfere with the work. One bundle at a time is picked up from the net bag and the rubber band is removed. The bundle with shoots is held in one hand and the planting is performed with the other until all shoots in the bundle are planted, when the next bundle is picked up from the bag. The shoots are planted one by one at marked distance on the measuring tape by gently pushing the rhizome down in an angle into the sediment using 2-3 fingertips placed along the length of the rhizome (Figure 5.2). This causes undisturbed sediment to fall on top of the rhizome, which increases the anchoring of planted shoots compared to if the shoot had been pushed straight down into the sediment surface. The technique should be practiced by the divers before the start, until the planting can be performed correctly by all practitioners. The first row of shoots is planted along the tape measure that runs along the side of the planting unit where the tape measure helps the diver to place the shoots at the correct distance. When the diver reaches one of the vertically positioned bands of the planting unit, the person turns and plants shoots on the return path, using the first row of planted shoots as guide for row two and so on until the entire planting unit is filled. The vertically placed tape measures are used as a guide to start the next line at the correct distance from the previous line (Figure 5.8).

Since shoots are susceptible to disturbance immediately after planting, it is important to minimize any movement over planted surface. The diver should therefore always place the body outside the planted area when planting and avoid swimming over shallow plantings. For this reason, it is best that only one diver works with each planting unit, and that divers in pairs work with two adjacent units. If the depth is greater than 1.5 m, the divers should be provided with surface buoys to facilitate the positioning of the divers.

# 6 Evaluation of restoration

## 6.1 The importance of monitoring and evaluating the results

The goal of eelgrass restoration is to try to restore a viable habitat and its ecosystem services, similar to what has been lost historically or as a result of direct human impact. Therefore, in a restoration, the goal is not only to recreate a certain area of the habitat, but also to achieve a quality that delivers all ecosystem functions and services that an eelgrass meadow normally produces. An important part of all restoration projects is therefore to follow up and evaluate whether these goals have been achieved after the restoration has been completed. Evaluating whether the habitat's ecosystem functions and values have been restored is a key question within management and should have a scientific foundation based on the quantitative data. In addition, you need to define what success means and how this is assessed so that the goals are measurable and can be monitored.

A well-designed monitoring program is therefore necessary to evaluate whether the goal of the restoration has been achieved and that the measure has given the desired improvement of the environment. This is also central to ensuring that e.g. an operator has met the requirements in an environmental compensation case. Regular sampling of the planting is also important to increase the knowledge of different eelgrass restoration methods. Even a failed restoration can be valuable if monitoring allows the causes of the failure to be identified so that they can be avoided in future projects. Frequent sampling also allows any problems and disruptions to the planting to be quickly detected and possibly remedied before it is too late, or to demonstrate the need to redo the planting to reach the goals in time. **Monitoring of planted eelgrass is therefore a very important part of the restoration work and should be an obvious part of the budget for each project, and should be demanded as a requirement in compensation restoration.** 

## 6.2 Variables and criteria for evaluation of restoration

The goal of the restoration of eelgrass should be to recreate a meadow with the lost ecosystem functions, which could be met if the sparsely planted shoots grows and expand to natural densities within the planting area, and preferably expanding beyond it. The planted meadow should therefore not only achieve a certain area, but also achieve structural (shoot density, biomass and coverage, etc.) as well as functional characteristics (abundance and biodiversity of plants and animals, carbon and nitrogen sequestration, improved water quality, etc.) of natural eelgrass beds found in the area.

A variety of variables have been used to evaluate whether planted eelgrass has achieved structural and functional goals; for example, shoot density, percentage coverage, leaf length, biomass, abundance and diversity of leaf and infauna, light conditions, nutrient content, chlorophyll concentration in the water, etc. (Fonseca et al. 1998, Short et al. 2000, Orth et al. 2012). Sampling of, for example, fauna can provide information about the recovery of ecosystem functioning and services to humans, but can be difficult to test regularly because sampling and, above all, the analysis of fauna can be very costly. Therefore, the variables that are regularly monitored should be inexpensive but at the same time be able to give good indications of how well functional properties have been recreated. In the United States, it has been proposed that evaluations of eelgrass restoration should primarily focus on how plants survive and grow as

seagrass structure correlates strongly with other biological and physical variables that constitute important ecosystem functions (e.g. sediment stabilization, nutrient uptake, etc.; Fonseca et al. 1998). These studies therefore suggest that one primarily should measure, for example, shoot density, vegetation coverage and extent of the planted meadow to assess whether a restoration has been successful since the variables together describe the status of the planting and its function.

Although many useful variables have been developed to describe the quality of the eelgrass, it is rarely specified how these are to be assessed with criteria and threshold values. A method proposed by Short et al. (2000) is to use reference meadows to identify these threshold values. There, the value of e.g. shoot density from the restored meadow is compared with the average shoot density from reference meadows to calculate a "quality ratio", which is then assessed against a threshold value based on the variation in the reference meadows (see Moksnes 2009 for a summary in Swedish; fact box 6.2 for calculation examples). The method is comparable to the "ecological quality ratios" used in status classification according to the Water Framework Directive. The advantage of this type of evaluation is that it is based on quantitative data and allows an objective assessment of whether the goals have been achieved with the restoration (see section 6.5 for details).

In this manual it is recommended that the restoration be evaluated by testing and comparing the restored meadow with at least two reference meadows. In order to take into account the naturally large variation of eelgrass between years, and that the recovery of the habitat's functions takes at least 5-10 years (Fonseca et al. 1998, Marba et al. 2015), it is recommended that eelgrass plantings are monitored for 10 years. In the first instance, it is recommended that simple eelgrass variables (shoot density, biomass and meadow distribution) are monitored annually, and that meadow ecosystem functions are evaluated after 5 and 10 years.

## 6.3 Recommended design of monitoring

For Swedish conditions, it is recommended that monitoring focuses on variables that reflect eelgrass health and growth during the first years, when the risk of loss is greatest, and that sampling of various ecosystem functions takes place only 5 and 10 years after planting. In Box 6.1, a sampling design is presented for monitoring a restored eelgrass meadow for 10 years. If the monitoring only lasts for 5 years, the sampling is excluded for years 7 and 10, but otherwise it is the same. All monitoring data is collected during the eelgrass growth season (May - September), where sampling is mainly carried out when the eelgrass has the largest biomass in August. It is important that the sampling is carried out at the same time each year in order for comparisons between years to be considered reliable.

### Fact box 6.1 Schedule for monitoring eelgrass restoration

 Table A. Working schedule for monitoring of the eelgrass restoration

Timetable	Task to do				
Year 0					
June (at planting start)	Take morphological measurements and biomass of planted shoots. Place instruments for continuous measurement of light and temperature.				
July (1 month after planting)	Sample shoot density, leaf number and length, extent of the planting. Clean and read instrument				
August (sample reference meadows )	Sample shoot density, biomass and extent of meadow.				
Aug-Sept (ca 2.5 months after planting)	Sample shoot density, leaf number and length, biomass of whole shoots, extent of the planting. Record and read instruments				
Year 1					
June	Sample shoot density, leaf number and length, and exter of the planting.				
August	Sample shoot density, leaf number and length, biomass of whole shoots, and of extent planting.				
Year 2					
August	Sample shoot density and extent of planting.				
Year 3					
August	Sample shoot density and extent of planting.				
Year 4					
August	Sample shoot density and extent of planting.				
Year 5					
August (sampling is performed in both planted meadow and reference meadows)	Place light instruments. Sample shoot density, biomass and extent of meadows Sample sediment Sample fish and macro invertebrates.				
Aug-Sept (2 weeks later)	Collect and read light instrument Assess the variables according to Fact Box 7.2				
Year 7					
August	Sample shoot density and extent of planting.				
Year 10					
August (sampling is performed in both planted meadow and reference meadows)	Place out light instruments Sample shoot density, biomass and extent of meadow. Sample sediment Sample fish and macro invertebrates				
Aug-Sept (2 weeks later)	Collect and read light instrument Assess the variables according to Fact Box 6.2				

Sampling takes place at a higher frequency during the first summer, as the first months are most critical when eelgrass shoots must recover from the transplant itself and establish themselves in a new environment (Figure 6.1). During the first few weeks the shoots are also poorly anchored in the sediment and sensitive to strong currents and wave erosion. In the Nordic latitudes, the first winter is also a critical period for the survival of transplanted shoots, since the long period without light can lead to high mortality in low-light areas, and ice scraping can damage shallow plantings (see section 2). It is therefore normal for the shoot density to decrease over the first winter, and then to increase rapidly again next summer. By having frequent sampling during the first year, it is possible to identify the cause behind any loss of shoots, which leads to important knowledge and opportunities to correct certain types of problems. It is therefore important to document all types of damage to plants, the presence of algae mats, burrowing or grazing animals, washed-up plants on the beach, etc. which may indicate causes of loss of shoots (see section 2.5). Even a failed planting can be of value if the cause can be identified and the knowledge increased. After the first year, the monitoring does not need to occur as frequently, as the meadow can be considered to have established itself in the new sites and the monitoring that follows aims to assess the establishment of the meadow's structural and functional characters.



**Figure 6.1** Eelgrass planting. The picture on the left shows a planting of eelgrass in June 2015 at a depth of 2.2 m on Gåsö outside the Gullmarsfjord where eelgrass shoots were planted a week earlier with the single shoot method at 25 cm intervals (16 shoots per square meter). The picture to the right shows the same planting in September when the shoot density increased to over 90 shoots per square meter in 3 months. In September, large parts of the meadow were covered by the filamentous brown alga *Ectocarpus siliculosus*. Photo: P. Moksnes.

## 6.4 Recommended variables and methods

In this manual, monitoring is initially focused on three eelgrass variables: shoot density, leaf morphology and the areal extent of the meadow, which can be easily sampled in the planted meadow without damaging the shoots. These variables are tested at each field visit in the meadow during the first and second year. At the end of the first summer, biomass samples of whole shoots from both the planted meadow and the reference meadow are also taken. After the second year, only shoot density and distribution are tested once a year until year 5 when biomass samples are also taken in planted meadows and reference beds (fact box 6.1). **These samples are quick and cost-effective and constitute a minimum of monitoring that should be required for all restorations, including environmental compensation cases.** It is also recommended that light conditions are measured at years 0,5 and 10 in planted meadows, and

that sediment structure / organic content and macrofauna are measured in years 5 and 10 in both planted meadows and reference meadows (fact box 6.1) to investigate whether eelgrass's various ecosystem functions have been recreated.

### **Reference Meadows**

As discussed above, reference meadows are an important part of the monitoring of eelgrass restoration as the success of a restoration is measured in relation to changes in reference meadows. The reference meadows should be as unaffected as possible by human activity and be close to the restoration area (see section 2.7 for details) and tested at the same time as the restored meadows to avoid variations in time and space affecting the evaluation (Short et al. 2000). The reference meadows should also be as similar to the restoration meadow as possible in terms of exposure and depth so that the variables being measured are comparable. For this reason, a depth interval should be tested in reference meadows reflecting the depth of the planted meadow. These meadows should be tested in August in year 0, and in year 5 and 10. By measuring these variables in the reference meadows for several different years, natural variations in e.g. meadows. Recommended monitoring variables are presented in detail below.

### Shoot density and leaf morphology

Shoot density and leaf morphology are quick and easy to measure in the field, and at the same time are important variables that indicate the survival, health and growth of planted shoots. These variables are measured by diving and randomly sampling at least 20 sampling quadrats in the meadow. The size of the sampling quadrats is adapted to the planting density of shoots, where a  $1 m^2$  quadrat is suitable for the first sampling when shoots are planted at 50 cm intervals (4 shoots per square meter) and a 0.25 m<sup>2</sup> quadrat is suitable when shoots are planted at 25 cm intervals (16 shoots per square meter; Figure 6.2). As the shoot density increases in the meadow, the size of the quadrats can be reduced (Figure 6.1). Within each quadrat, the number of shoots is counted, and on up to 5 randomly selected shoots, the number of leaves and the longest leaf is measured. The number of leaves per shoot can provide important information as a low number of leaves on an adult shoot (<4 leaves per shoot) can indicate that the eelgrass is stressed (Carr et al. 2012). Changes in the leaf length of the shoots may also indicate that the shoots adapt to light and exposure conditions in the new environment. Leaf morphology is only measured for the first two years, after which only shoot density is measured (fact box 6.1).



**Figure 6.2** Sampling of shoot density. The picture shows a test plant in the Gullmarsfjord at 1.5 m depth where 9 planted shoots grow to over 100 shoots in 14 months. The sampling box in the picture is  $0.25 \text{ m}^2$ . Photo: E. Infantes.

At the end of the growing season years 0 and 1 (fact box 6.1), where possible, 20 random samples are also taken where whole eelgrass shoots are carefully excavated with rhizome and roots (Figure 6.3). On each individual plant, the number of side shoots, total length and the number of internodes on the rhizome, and, on up to 5 lateral shoots, the number of leaves and maximum leaf length are measured as well as. Dry weight (60 °C for 48 hours) is also taken on the biomass above and below the sediment. The same variables are also taken on shoots collected at the time of planting (see section 5.5), from which very precise measures of shoot growth can be obtained. These metrics are very valuable for forecasting plant growth and expansion over time.



Figure 6.3 Shoot growth after 14 months. The picture shows the growth of a shoot with about 5 cm long rhizome planted at 1.5 m depth in the Gullmarsfjord 14 months earlier. By carefully digging up the entire plant with older rhizomes and lateral shoots, very precise measures of growth in standing biomass can be obtained. Photo: E. Infantes.

### Areal distribution and depth distribution of eelgrass

Perhaps the most important variable for assessing whether a restoration was successful is the areal extent of the planted meadow, which is measured at all field visits. When assessing the areal distribution, the eelgrass coverage on the seabed must be at least 5% to be included (if it is assessed from a boat with aqua scope; Baden et al. 2003) or have a maximum of 1 m between each eelgrass shoot (if it is assessed with diving). Spots in the meadow with <5% coverage of eelgrass of no more than  $5 \times 5$  m are considered part of the meadow (NOAA 2014). If larger holes in the meadow occur, they should be excluded from the total area.

If the water transparency allows, the aerial distribution is most easily measured by photographing the planting from the air with the help of a drone (see section 2.4.1 for details; Figure 6.4). The marking tubes in the corners of the planting units can be used to geometrically correct the photos and calculate the area more accurately.

Note, however, that shoot densities <16 per  $m^2$  are difficult to see from the air, so observations in the water may be necessary during the initial sampling (compare Figure 2.3 and Figure 6.4). If remote photography is not possible, the distribution can be estimated by aquascope or drop video from a boat with GPS, or by snorkelling (see section 2.4.1). Side-view sonar can also be used as a supplement to estimate the distribution if the depth of sight is poor.

It is also important to measure the maximum depth distribution of the planted meadow and see if it changes over time. **The maximum depth distribution is defined as the deepest part of the meadow with at least 5% coverage**, and is most easily measured by divers swimming along the deepest edge of the meadow. Areal extent and depth distribution are also measured in the reference meadows years 0, 5 and 10. Note that shallow sites may not allow a planted meadow to reach its maximum propagation depth over a 5 to 10-year period and therefore cannot be compared with the depth distribution of the reference meadows.



**Figure 6.4** Assessment of areal distribution with drones. Photo taken with drones from about 100 m above a bay on Gåsö in Lysekil municipality. The red circle marks a test planting of 10x10 m where eelgrass shoots were planted 3 months earlier at 2.0 m depth with a shoot density of 16 shoots per square meter. At the time of the shoot, the shooting density was over 90 shoots per square meter on average. The darker spots in the picture are small patches of natural eelgrass. Photo: J. Stenström.

### Eelgrass biomass

Eelgrass biomass is sampled by divers with a small quadrat (0.06 - 0.25 m<sup>2</sup> depending on shoot density) where all eelgrass within the quadrat (including rhizome and roots) is excavated and taken to the laboratory. There, the shoots are rinsed thoroughly to remove sediment and loosely seated epiphytes, after which leaves and rhizomes with roots are dried separately at 60 °C for 48 hours to determine dry weight per unit area (g per square meter) above and below the sediment surface. Samples of biomass are taken years 0,5 and 10 in both planted meadows and reference meadows (fact box 6.1).

## Light supply and temperature

The availability of light is critical for planted shoots to survive and grow, and variation in light supply may explain losses of newly planted shoots (see Figure 2.6). Similarly, high temperature can have a negative impact on eelgrass by causing oxygen deficiency. Therefore, in order to detect any problems with light supply and temperature, it is recommended that the light is continuously measured at two depths next to the planting during the first summer in order to calculate the extinction coefficient of light (Kd) and the proportion of light at the surface reaching the shoots (see fact box 2.2 and section 2.5.2 for methods). Furthermore, it is recommended that temperature is measured at the deep edge of the planting.

### Variables that indicate ecosystem functions and services

Since the main goal of eelgrass restoration is to restore the meadow ecosystem functions and services, it is important to try to estimate these in the restored meadow. In Swedish waters, eelgrass meadows are considered to contribute several important ecosystem functions and services, including increased biodiversity, increased production of fish and seafood, improved water quality and increased uptake and long-term storage of carbon and nitrogen (see Moksnes et al. 2016, section 3.2). It is therefore recommended that the following three variables are measured during years 5 and 10 in both the planted meadow and in reference meadows (fact box 6.1).

**Sediment variables**. Changes in grain size and organic content in the sediment may indicate increased sedimentation and storage of carbon and nitrogen. Therefore, it is recommended that 10 random sediment samples be taken from the top 12 cm of the sediment in both planted meadows and reference meadows. The sediment cores should be divided and analysed into three fractions (0–4, 4–8, 8–12 cm) to see changes in storage over time (see fact box 2.5 for methods). If resources are available, carbon and nitrogen content should also be analysed in the sediment samples.

*Light*. To investigate whether the planted meadow has improved the water quality and light conditions at the sites, it is recommended that continuous light measurements be made at two depths within the planted meadow in year 0, and years 5 and 10 both within and at least 50 m outside the planted meadow.

*Fish and crustaceans*. An important function of eelgrass is to provide animals and plants with a habitat. To estimate part of this feature, it is recommended that the abundance and diversity of day-active fish and larger decapod crustaceans (crabs and shrimp) be tested by adding 5 random transects to the meadow (20 m long) where the fauna is visually quantified by divers who slowly

count all major animals found at a certain distance from the transect (Figure 6.5). Studies show that visual dive transects sample more species than e.g. sample fishing nets in dense vegetation (Pratt & Fox 2001). Based on this data, the number of individuals per square meter and taxa (for selected species) and the number of species of fish and crustaceans per meadow are calculated



**Figure 6.5** Juvenile cod nursery. One of the eelgrass meadows' most important ecosystem functions is to provide juvenile fish with a nursery habitat. The number of day-active fish and larger crustaceans in an eelgrass meadow can be quantified using divers who count the animals along a transect. Photo: P. Moksnes.

## 6.5 Assessment of results

Assessment of how well the restoration achieved set goals in terms of area and quality is performed 5 and 10 years after planting by comparing the restored meadow with reference meadows. In the assessment, the size of the restored meadow (the areal distribution) is evaluated separately from variables that reflect structural and functional qualities (see fact box 6.2).

When assessing whether the planted meadow has reached set targets in terms of areal distribution, natural variations in distribution between years are taken into account. This is done by adjusting the measured area with the proportional variation in the distribution that occurred in the reference meadows during the same period. However, this adjustment is only made if the reference meadows are reduced in distribution (fact box 6.2).

#### Fact box 6.2 Assessment of restoration results

In restoration, it is important to be able to assess whether the measure has succeeded or not, so that decisions can be made if new measures are needed. This is especially true in the case of compensatory restoration as it may be required that the restoration should be redone if the goals are not met. In the assessment, the size of the restored meadow (area) and the quality are evaluated separately with different methods after 5 and 10 years.

#### 1. Assessment of the areal extent of the restored meadow

The areal distribution of planted eelgrass is measured as the area with at least 5% coverage of eelgrass (see section 6.4 for details). In order to take into account natural annual variations in areal distribution, the measured area of the planted meadow is adjusted in years 5 and 10, with the proportional average variation in areal distribution of the reference meadows from year zero. This adjustment is only made if the reference meadows are reduced in distribution.

**Example**: The goal of the restoration was to recreate 2.0 ha of eelgrass, which was planted in year 0. In year 5, a total of 1.7 ha of eelgrass was measured. At the same time, reference meadows 1 and 2 were estimated to have decreased by 17% and 23% in areal distribution since year 0 (proportional average change = 0.80). Objectives of the restoration: 2.0 ha  $\times$  0.8 (adjustment for natural variation) = 1.6 ha eelgrass Results: 1.7 ha of eelgrass.

Assessment: The target areal distribution was reached by a margin of more than 6%.

#### 2. Assessment of the quality of the restored meadow

When assessing the quality of the restored meadow, values of selected variables are compared between the planted meadow and reference meadows by calculating a quality ratio. This value is then assessed against a threshold value that takes into account the natural variation of the variable in the reference beds (based on Short et al. 2000). The following calculation is made separately for each of the variables shoot density, biomass, maximum depth distribution, sediment variables and the abundance and diversity of fish and crustaceans.

#### a. Calculation of the quality ratio

The quality ratio is calculated by dividing the variable value in the planted meadow by the average value in the reference meadows.

Quality ratio = value in the planted meadow / average in the reference meadows

#### b. Calculation of the threshold value

The threshold value is calculated by using the standard deviation (SD) for the variable in the reference meadows.

Threshold value = (average in reference meadows - 1 SD) / average in reference meadows

#### c. Assessment

If the quality ratio is greater than the threshold value, the goal has been achieved.

**Example:** The shoot density in the planted meadow after 5 years was averaged to 470 shoots per square meter. Shoot density in reference meadow 1 and 2 was measured at 490 and 640 shoots per square meter, which gives an average and standard deviation of 565 and 106 shoots per square meter, respectively.

Quality ratio = 470/565 = 0.83

Threshold value = (565-106) / 565 = 0.81

Assessment: Quality ratio> Limit value (0.83> 0.81)

In this example, it was therefore estimated that the shoot density in the restored meadow achieved set targets.

In assessing structural and functional qualities of the restored meadow, values of shoot density, biomass, maximum depth distribution, sediment variables, light, and the abundance and diversity of macrofauna from both the planted meadow and reference meadows are used to calculate a quality ratio for each variable. These values are then compared with a threshold value based on the variable's variation within the reference meadows. If the quality ratio is higher than the threshold value, the restored meadow has achieved the quality target for that variable (Short et al. 2000, see fact box 6.2). For the analysis to be reliable, the variation (measured as standard deviation) should not exceed 20% of the variable's average value in the reference meadows (SD / Mean  $\leq$  0.20), which corresponds to a threshold value of  $\geq$ 0.80 (Short et al. 2000). If only one representative reference meadow is available for evaluation, or if the threshold value is not reliably assessed, it is recommended that a fixed limit value of 0.80 be used, i.e. that the reference meadow should reach 80% of the shoot density, biomass, etc. found in the reference meadow (NOAA 2014).

# 7 Cost of eelgrass restoration in Sweden

## 7.1 Introduction

The following is a summary of estimated costs for restoring one hectare of eelgrass in Bohuslän with shoot and seed methods. Cost calculations are based on the fact that the work is carried out in accordance with the recommendations in this manual, and that they are performed by professional personnel with the right expertise. The working hours used in the calculations are based on measured times for various work steps in studies described in this manual. Average values of working hours from staff with different experience were then used. When calculating costs for work and equipment, prices offered by marine biological consulting firms in Bohuslän 2015 have been used as data, as it is this type of company that can be expected to perform large-scale restoration work (see Appendix 3 for a detailed description of the cost calculations).

Restoration of eelgrass consists of three important parts: (1) evaluation and selection of sites for restoration, (2) harvesting and planting of eelgrass, and (3) evaluation of the restoration. In order to succeed in a restoration and to be able to evaluate this, all three parts are equally important, and funds must be allocated to all parts when planning a restoration project.

## 7.2 Summary of results

## Single shoot method

The site selection and the evaluation of the restoration are very little affected by the size of the restoration project or the planting density of shoots. For shoot methods, the cost is about SEK 380,000 for site selection and about SEK 390,000 for a 10-year monitoring and evaluation of the restoration (including report writing; Table 7.1). If the evaluation is carried out after 5 years, the cost is approximately SEK 270,000. In contrast, the cost of harvesting and planting shoots is directly proportional to the area and shoot density of the planting. For the single shoot method, which is the fastest and cheapest method of the surveyed restoration methods, the total cost of harvesting and planting shoots to restore a hectare of eelgrass is about SEK 435,000 at a planting density of 4 shoots per square meter, and about 1,728,000 at a planting density of 16 shoots per square meter. In total, therefore, the total cost of restoring one hectare of eelgrass (including site selection and evaluation) will be between SEK 1.2 and 2.5 million, depending on the shoot density used in planting (Table 7.1). In addition to the cost being affected by planted shoot density, it also depends on the planting technique.

For example, if the site requires the eelgrass shoots to be anchored, the cost of planting doubles (see Appendix 3). It is therefore very important to evaluate optimal planting densities and methods when evaluating and selecting the restoration site.

**Table 7.1** Costs of eelgrass restoration. Summary of costs for evaluation of restoration site, harvest and planting (per hectare), and monitoring and evaluation during 10 years of the restoration, when planting with the single shoot method and seed from boat at two different planting densities.

	Shoot		Seeds	
Cost (SEK)	4 shoots m <sup>-2</sup>	16 shoots m <sup>-2</sup>	4 shoots m <sup>-2</sup>	16 shoots m <sup>-2</sup>
Site selection	379 800	379 800	446 700	446 700
Cost of Labour	295 200	295 200	343 200	343 200
Boat and transport	27 600	27 600	34 500	34 500
Fees and materials	57 000	57 000	69 000	69 000
Harvesting and planting	435 000	1 728 000	1 638 188	6 361 649
Cost of Labour	404 500	1 618 000	1 414 485	5 525 189
Boat and transport	26 500	106 000	74 013	285 701
Material	4 000	4 000	149 690	550 759
Monitoring and evaluation	388 950	388 950	408 000	408 000
Cost of Labour	337 200	337 200	352 800	352 800
Boat and transport	51 750	51 750	55 200	55 200
Material	0	0	0	0
Total cost	1 203 750	2 496 750	2 492 888	7 216 349

m-2= per square meter

#### Seed method - broadcasting from boat

The cost of selecting a site and evaluating the restoration is slightly higher for seed methods than for shoot methods. This is because it takes an extra year for planted seeds to form adult shoots, which affects the cost of both site selection (about SEK 447,000) and the planting result (about 408,000) which is about 18% and 5% higher compared to shoot methods (Table 7.1). However, the major difference between seed and shoot methods is the amount of work and the cost of harvesting and planting. Due to the fact that very few of the seeds that are planted survive and form shoots (on average 0.9% of the depths recommended for restoration), very large quantities of flower shoots (approx. 130,000 to 530,000 shoots) and seeds are needed (approx. 4.6–18.2 million seeds depending on the density of surviving shoots) to restore one hectare of eelgrass. In comparison, only about one-third as many vegetative shoots need to be harvested (42,000 to 168,000 shoots) to achieve the same density of restored shoots. This difference means that it is more than four times as expensive to plant with seeds (about SEK 1.64 - 6.36 million per ha) compared to shoots. The total cost of restoring one hectare of eelgrass with seeds (including site selection and evaluation) is between SEK 2.5 and 7.2 million (Table 7.1).

Furthermore, it is almost 5 times faster to plant with shoots than with seeds. At a planting density of 4 shoots per square meter, 4 divers can harvest and plant one hectare of eelgrass with the single shoot method in 10 working days. The same dive team would need 28 working days just to harvest the flower shoots that are needed to restore one hectare of seeds, after which 4 people need an additional 19 working days to extract the seeds, while planting can be done in just one day (a total of 48 working days; Table 7.2). In order for seed methods to compete with shoots in cost, the proportion of broadcasted seeds forming shoots must increase to over 3% (see

Appendix 3). Today, this high seed survival is only found on deep (4-5 m) sheltered soils that are generally not recommended for restoration due to low shoot growth.

**Table 7.2** Comparison of costs (SEK) of harvesting and planting one hectare of eelgrass with shoots and seeds. Comparison of number of shoots needed, number of working days at harvest, seed production and planting, and different types of restoration with shoots and seeds at two different planting densities, where the establishment of planted seeds is assumed to be 0.88%.

	Shoot		Seeds		
	4 shoots m <sup>-2</sup>	16 shoots m <sup>-2</sup>	4 shoots m <sup>-2</sup>	16 shoots m <sup>-2</sup>	
No. of shoots	42 000	168 000	133 690	534 759	
No. of days harvests	5	20	28	111	
No. of days seed production	-	-	19	69	
No. of days planting	5	20	1	1	
Cost (SEK)					
Harvest	205 750	817 000	1 242 263	4 963 051	
Cost of Labour	190 000	760 000	1 169 700	678 800	
Boat and transport	13 750	55 000	70 563	282 251	
Material	2 000	2 000	2 000	2 000	
Seed Production	0	0	372 475	1 375 148	
Cost of Labour			226 785	828 389	
Material and fees			145 690	546 759	
Planting	229 250	911 000	23 450	23 450	
Cost of Labour	214 500	858 000	18 000	18 000	
Boat and transport	12 750	51 000	3 450	3 450	
Material	2 000	2 000	2 000	2 000	
Total cost	435 000	1 728 600	1 636 188	6 359 643	
2					

m<sup>-2</sup> = per square meter

In summary, the cost of restoring one hectare of eelgrass with the single shoot method is estimated to vary between SEK 1.16 and 2.34 million, which includes one year's evaluation of potential sites, harvesting and planting of shoots, and 10 years of evaluation of the restoration. The same restoration would be over 2-3 times as expensive (SEK 2.49-7.22 million) and take two years longer before an established eelgrass meadow is recovered if available seed restoration methods are used. Today restoration is therefore recommended with the single shoot method in Swedish waters.

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# Handbook for restoration of eelgrass in Sweden

## National guidelines

The manual provides detailed guidelines for eelgrass restoration and addresses all important steps in the restoration process, from site evaluation and selection, consultation and permitting, harvesting and planting, to monitoring and evaluation of the results. The methodology is primarily developed for the Swedish NW coast, but parts can also be applicable in the Baltic Sea after the methods have been investigated there.

Although well-functioning methods for eelgrass restoration are now available for Swedish conditions, restoration of eelgrass is time-consuming, expensive and associated with uncertainties. Consequently, it is of the utmost importance that the management primarily focuses on protecting the remaining eelgrass meadows, and only as a final measure allows compensation restoration as a solution in exploitation.

It is the hope of the Swedish Agency for Marine and Water Management that the Handbook can provide support for supervisory and review authorities, but also operators and consultants in the work on eelgrass restoration.

The manual has been produced in a collaboration between the Swedish Agency for Marine and Water Management, the County Administrative Board of Västra Götaland and the University of Gothenburg.

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