

# Restoration of species rich ditch banks in agricultural peat meadows



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Bachelor research project thesis

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## Summary

Intensive land use has caused the decline of peat meadow species and ecosystems. This rapid decline caught the attention of many researchers and ordinary citizens, who have been trying to slow and reverse this downward trend. Recently some of these concerned citizens have established the Land van Ons. They recently acquired a degraded piece of land, on which they want to restore the peat meadow biodiversity as much as possible. However, there are still some knowledge gaps which will make restoring the ditch bank ecosystems a challenge. For one, there is little to no information about peat-meadow ecosystem restoration in South-Holland. Moreover, most known restoration methods focus on enlarging the existing biodiversity (with which, often, taxonomic richness is meant) of an ecosystem which is not completely degraded. Next to that, little is known about the ecological impact on species richness and composition of different types of management in peat-meadow areas. This project aims at making a start in filling some of these knowledge gaps.

The focal polder of this research, the Northern part of the Vrouwe Vennepolder, is a good model system to test which restoration method has the best effect in reversing the decline of peat-meadow species. This project focuses solely on the restoration potential of the seed and propagule banks in the ditch bank soil and dredge, but also at finding a good baseline management for ecological restoration in peat meadow areas in South-Holland.

First, we compared four different peat meadow managements from polders nearby Leiden, South-Holland. These polders were the Lakerpolder, the Boterhuispolder, the Southern part of the Vrouwe Vennepolder and the Northern part of the Vrouwe Vennepolder itself. The data used to compare these managements was gathered in September 2020 and in May 2021. From this comparison, we concluded that, based on the species richness, the Southern part of the Vrouwe Vennepolder and Boterhuispolder were best. But based on the type of species, the Lakerpolder's abiotic factors most resemble those of a peat meadow ecosystem. As such, the best management to get the peat meadow ecosystem back is the management of the Lakerpolder.

Secondly, we researched whether the local seed and propagule banks could be used to restore the vegetation community of typical peat meadow species. To this end, we gathered soil and dredge samples from fifteen locations equally distributed throughout the Northern part of the Vrouwe Vennepolder. The seeds and propagules in the soil and dredge were left to germinate for eight weeks. Every week, the number of germinated seeds and propagules was counted, and at the end all seedlings were identified to species level. In total we found 24 different plant species. After several analyses it was concluded that there were enough seeds and propagules present in the soil and dredge, but that there were almost no additional species in the seed and propagule banks compared to those in the covering vegetation. The seven species that did not show in the covering vegetation were very small in numbers and were not peat meadow species. Therefore, the seed and propagule banks of the Northern part of the Vrouwe Vennepolder cannot be used to bring back the peat meadow species to the polder.

Our conclusions indicate that for the return of peat meadow species, additional sowing and or other restoration methods are needed. Of course, this may be different for other polders in different areas because the seed bank restoration capacity heavily depends on different abiotic factors, past use and time between past and present ecosystem compositions. On the other hand, the managements can almost always be implemented in other peat-meadow polders, however they have little influence on the species composition and richness, and as such, additional restoration methods are likely needed in the Vrouwe Venne polder and beyond unless the ecosystem is not yet completely degraded.

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# Introduction

The intensive use of land in many agricultural areas has a negative impact on biodiversity around the world including in the Netherlands (Polasky, S. et al., 2011; Living Planet Report, 2019/2020). The disappearance of species and homogenization of land degrades ecosystem functions, which leads to a decrease in ecosystem services (Kandziora, M., 2013). A decrease in regulating and provisioning ecosystem services on its own turn lowers agricultural production and pushes farmers to intensify the use of land. It is a self-sustaining downward spiral of events, which indicates that a new, nature inclusive approach to agricultural land use is sorely needed.

In the preservation of species diversity in agricultural land, farmland ditches and ditch banks play an important role, particularly in lowland countries like the Netherlands. Ditches resemble miniature wetlands, and have therefore been coined ‘linear wetland’ (Blomqvist, M. M. et al, 2003). Together with the ditch banks, which form ecotones between the aquatic and grassland ecosystems, they are often the main spot for species richness and habitat diversity, especially in agricultural areas (Blomqvist, M. M. et al, 2003). Despite the importance of wetlands, their number, and as such, the abundance and distribution of typical wetland species, is decreasing (Shaw, R. F. et al, 2015; Wetlands, n.d.). Preservation and restoration of these areas is therefore important. Particularly in degraded peatland, which historically harbour a large portion of the wetland habitat at higher latitudes, there is a question of how to restore biodiversity in ditches and ditch banks. Intensive land use has pushed these ecosystems to an alternative stable state of low biodiversity (Beisner, B.E., 2012). This state has a high resilience and is therefore difficult to change (Beisner, B.E., 2012). With intensive land use come nutrient rich soil ecosystems, bearing a few dominant, nutrient-loving plant species, leaving no room for others to grow. For aquatic ecosystems, water quality is a determining factor. Aquatic plants secure the dredge with their roots and take up nutrients, which has a positive effect on water quality. But when they disappear due to excessive dredging or poor water quality conditions, the water will become turbid with every disturbance and FLAB of algae will form, shutting off the light as well as oxygen availability and thereby killing the saplings and aquatic fauna (Shaw R.F. et al., 2015). These changes typically follow a non-linear trajectory (see Fig. 1). One needs to move far to the left of the x-axis, in other words, reduce, for example, the N-content to a very low level to move back to the previous state. This means that more change is needed than seems necessary to drive the species poor ecosystem to a species rich system (Beisner, B.E., 2012). So the question is: how can heavily degraded ditch and ditch bank ecosystems in peat meadow areas be restored?

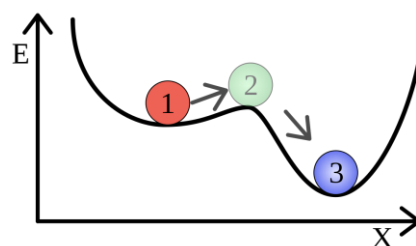


Figure 1. Adapted from: Georg Wiora

There are many projects and initiatives with the goal of ecological restoration. They often, like agri-environmental schemes (AES), try to prevent these shifts to a state of lower biodiversity, and also try to mitigate the effects of intensive agriculture by providing protection for the existing biodiversity (Science for Environment Policy, 2017; Shaw, R. F. et al, 2015), and improving ecosystem diversity and connectivity (Maes, J. et al., 2008). But there is still room for developments in our knowledge, for each ecosystem needs its own refined measure. (Science for Environment Policy, 2017). Most available knowledge on restoration is aimed at enlarging the existing biodiversity (with which, often, taxonomic richness is meant) of an ecosystem which is not completely degraded. Due to this lack of knowledge, restoring our completely degraded peat meadow ditch bank ecosystem to one with many peatland species will present a number of challenges. Based on previously used restoration methods, we can extrapolate the parts that can be used in a peat meadow polder. With these parts, we can compile our own restoration method to create better habitat conditions. Habitat conditions that favor higher species richness will also result in an increase in the number of interactions between organisms that will likely colonize the area. When an ecosystem has many species interactions, it will also have a higher tolerance against disturbances. But there are more aspects to keep in mind when restoring the ditch and ditch bank ecosystems, like connectivity to other water bodies, water level, water quality, amount of sludge, fertilization of the adjacent fields, disturbance due to animals and (timing of) management (Shaw, R.F. et al, 2015; Verberk, W. et al, 2008). On top of that, conservation and restoration strategies should take spatial and temporal biodiversity patterns into account as well (Leng, X. et al., 2011). Most of these aspects and patterns also have to be kept in mind during this case study.

Besides the temporal and spatial patterns, and the abiotic boundary conditions, the biotic legacy (seeds and propagules) will for a large part determine the success of ecological restoration. Seed and propagule banks are underground reservoirs of seeds and propagules, and their contribution to the ecological recovery capacity of an ecosystem is governed by the number of species still present. Seeds and propagules expire after some time, degrade faster in wet conditions (Bossuyt & Hermy, 2003) and the state of the abiotic factors in the soil might also form a hindrance for the germination of certain species. A low number of germinating species would indicate a low recovery capacity, whereas a high number of germinating species would indicate a high recovery capacity. To be able to adequately estimate the recovery capacity of the seed and propagule banks, the current species composition has to be known. It is, however, difficult to find out which and how many seeds and propagules are still present in the soil.

Aside from the seed and propagule banks, management also plays a role in the success of ecological restoration. There are a few management factors that are important specifically to peat meadow areas like the Vrouwe Vennepolder, like fertilization, mowing regime and water table. The current water table of the Vrouwe Vennepolder is relatively low and will be increased by around 20-30 cm, so it will end up around 20-30 cm below soil level in order to decrease soil subsidence and loss of landscape specific

species (Erkens, van der Meulen, & Middelkoop, 2016). At the same time, overfertilization and badly planned mowing schedules also lead to biodiversity loss (Socher, Stefanie, et al., 2013) and hinder our restoration goals of bringing back peat meadow ecosystems and species. However, the exact influence of different managements on species richness and diversity in peat meadow areas in South Holland is still unknown. Moreover, there are still questions around the effects of restoration methods in peat meadow areas. These knowledge gaps are too big to fill with one study. As such, we focussed solely on seed banks and four management methods in peat meadow areas. Two questions were formulated:

1. Which management(s) work best as a baseline for bringing back peat meadow ecosystems and species in the Northern part of the Vrouwe Vennepolder, while combining this with profitable agriculture?
2. Can the existing seed and propagule banks be used to improve the occurrence of peat meadow species on the ditches and ditch banks in the Northern part of the Vrouwe Vennepolder and as such push the ecosystems towards a 'healthier' peat-meadow system?

The management methods of the four different polders (Boterhuispolder, Lakerpolder, Southern and Northern part of the Vrouwe Vennepolder) will be linked to their respective species richness and diversity currently present in the plant layers. Seed banks also contribute to ecological restoration, but only if they contribute to an increase of the species richness of the current covering vegetation. Of course, the questions above both focus on improving the species richness and diversity of degraded ecosystems, but contain many components. The questions cover too large a subject to answer with certainty in one experiment, and therefore this project needs a follow-up experiment.



# The Experiment

## 1. Study Area

This study focussed, among others, on the ditches and ditch banks in a polder situated in Oud Ade, locally called the Northern part of the Vrouwe Vennepolder, hereafter called the NVV-polder. It belongs to the typical peat meadow cultural landscape. At the end of 2020, the civilian cooperation Land van Ons (hereafter ‘Land van Ons’) acquired 33 ha of land in the NVV-polder. Their goal is to improve biodiversity, to halt soil subsidence, improve the quality of the landscape and environmental factors, and then combine all the previous goals with profitable agricultural practices (Land van Ons, 2021). Before the entire polder was acquired by Land van Ons, the polder was under two management regimes. As such, part of the polder has a history of intensive use, while the other part has been under organic agriculture with meadow bird protection schemes. Its ecosystems are poor in peat meadow species, and it is therefore a good model system to test how to improve species richness of degraded ecosystems in peat meadow areas. In a ten-year study, Leiden University in coalition with Land van Ons, will, amongst others, keep track of the changes.

The second part of the study focussed on the management of the NVV polder and three other nearby polders. These polders are called the Boterhuispolder, Lakerpolder, and Southern part of the Vrouwe Vennepolder. These peat-meadow polders all have a different management and function as reference sites on which the management will not change in the coming years. The September database contains species information of these polders (p. 12). It, however, doesn’t contain information on each polder’s management and type of ditch bank. Therefore, the owners of the land were interviewed about their management, and a walk through the polders presented the information about the ditch banks. The Lakerpolder (LP) is a nature reserve and has very wet soil. The Boterhuispolder (BP) has nature-friendly ditch bank management and part of its fields have a delayed mowing schedule to promote meadow bird populations. The Southern part of the Vrouwe Vennepolder (SVV) also has a mowing schedule with the intention of promoting meadow bird populations, and, on top of that, it has ecologically certified farming. The Northern part of the Vrouwe Vennepolder (NVV) has partially organically certified farming, and partially intensely farmed. The part that has intensive farming has steeper ditch banks or no ditch banks at all. The part that was biologically farmed has more ditch bank-like structures. However, the dataset from September 2020 only contains data from the part that was not biologically farmed (p 12; Image 2.1). The BP has very broad and slowly descending ditch banks, the SVV polder has smaller descending ditch banks, and the Lakerpolder has almost no descending ditch banks, but more like just watersides. These ditch banks get trampled by the cows, which are present in all polders except the Lakerpolder.

## 2. Materials and Methods

### 2.1. Materials

All items needed to complete the field experiments:

1. Wheelbarrow
2. Shovel
3. Transparent tape
4. Duct tape
5. Scissors
6. Paper
7. Pen
8. Gardening gloves
9. Water fauna net
10. Bamboo sticks
11. Auger
12. Fruit net
13. 60x plant pots
14. 4 pegs and a 4 m cord
15. Ruler
16. Plant identification guide



*Image 1.1: Experimental setup and equipment. The right slot contains the pots with soil and the left slot contains the pots with dredge.*

All items needed to complete the dataset analysis:

17. Program RStudio
18. Oeverplanten Project Database
19. September 2020 database
20. Program QtGrace
21. Verspreidingsatlas ([www.verspreidingsatlas.nl](http://www.verspreidingsatlas.nl))

### 2.2. Methods

The goal of this study is to find out what kind of management can be used as a baseline for restoration of a ‘healthy’ peat meadow ecosystem while combining this with profitable agriculture, and to eventually tell if there is restoration potential in the seed and propagule banks for the Northern part of the Vrouwe Vennepolder to set the change towards a ‘healthier’ peat meadow ecosystem in motion. With a ‘healthy’ peat meadow ecosystem we mean a high species richness with a species composition that indicates the abiotic factors of a peat meadow. During the project, we kept the Lakerpolder in mind because this polder is labelled a peat meadow nature reserve, even though it is rather saline. We did not

use it as a reference polder. Rather, we used the Oeverplanten Project Data to determine the abiotic conditions and target species composition.

We started with an analysis of the species richness data from the September 2020 database (The Experiment 2.4, p. 12). This analysis indicated which of the four polders has the best management based on the number of different species present. But a big species pool is not the only indication for good management. As indicated, abiotic factors are also important to consider. Several abiotic factors could also be retrieved from the September 2020 database. To complete the inventarisation we also used information from the Oeverplanten Project Dataset and Verspreidingsatlas (The Experiment 2.5, p. 16) to see which management created the best ‘peat-like’ abiotic conditions. In the second part of

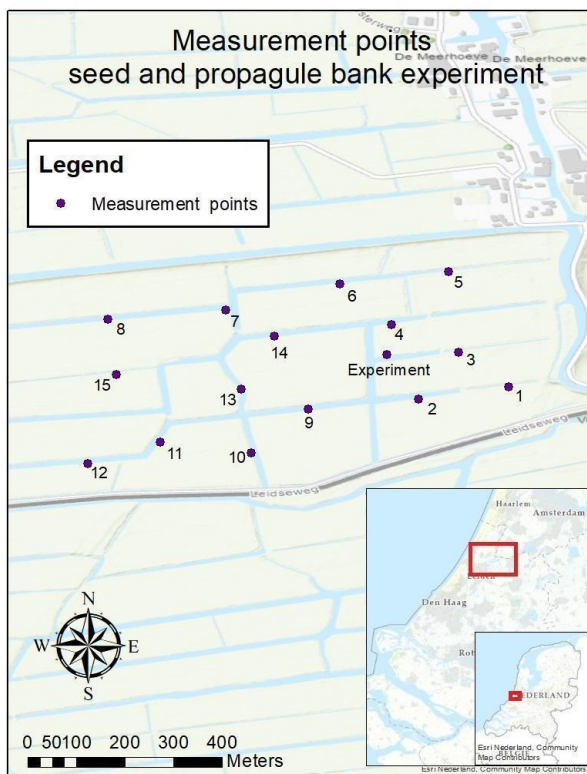


Image 1.2: Chart of the measurement points for the seed and propagule bank experiment. Some points appear to be in the middle of the ditch, but their numbers are placed on the correct side. The point with “Experiment” indicates where all the pots are stored.

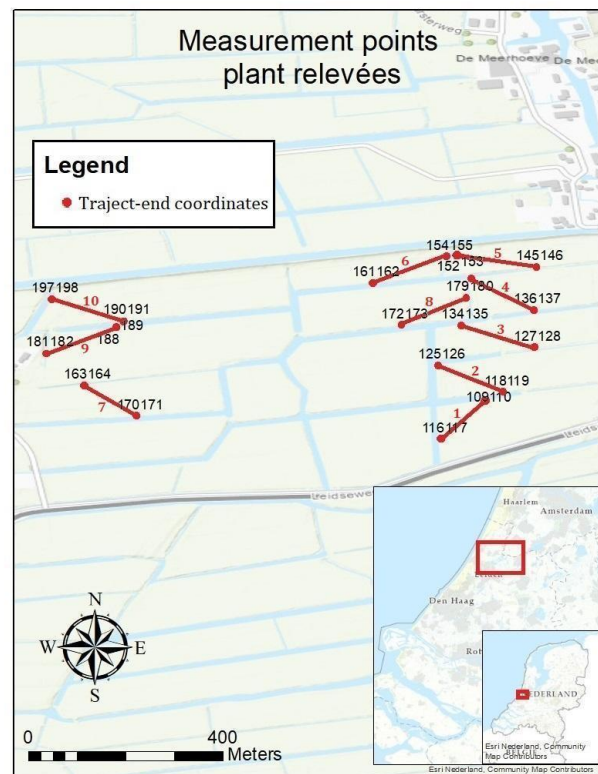


Image 1.3: Chart of Traject-end locations. These points indicate at the same time the plant relevée plot locations. The red numbers indicate the traject number, and the black numbers indicate the plot number. (Plot numbers have three digits. For example, 109110 are plots 109 and 110). The highest and lowest number per traject represent the aquatic measurement points.

the study we researched whether the seed and propagule banks in the Northern part of the Vrouwe Vennepolder had any restoration potential for bringing back the characteristic peat meadow species specified in the Oeverplanten Project Dataset. Seeds and propagules in the substrates ‘dredge’ and ‘soil’ collected from 15 sample sites throughout the polder were left to germinate and grow for eight weeks.

Analyses on the number of germinated seedlings and the species composition of the seed and propagules were conducted. From the results we can deduce whether the seed and propagule banks are a useful tool for restoration in the Northern part of the Vrouwe Vennepolder.

The answers to this study will form the first handholds towards deciding the future management for the Northern part of the Vrouwe Vennepolder.

### **2.3. Experimental Setup**

All pots were divided over two slots that were dug to accommodate 30 pots each. The slots were orientated from east to west. The slot further to the north had some more shadow, and protection from the wind from the pile of soil that lies between the two slots. The pots with dredge were put in this slot to protect them a little bit more against torrefaction than the pots with soil. Some soil was put in between the pots to lock them in place. To protect the seedlings from the birds, the setup was covered with a net. The net is held in place with bamboo sticks, which have flags at their upper ends to make them more visible and prevent injury. Because the soil that was dug out to create the slots lies in between the two slots, the nets that cover the pots make the whole look like a pyramid. (Image 1.1)

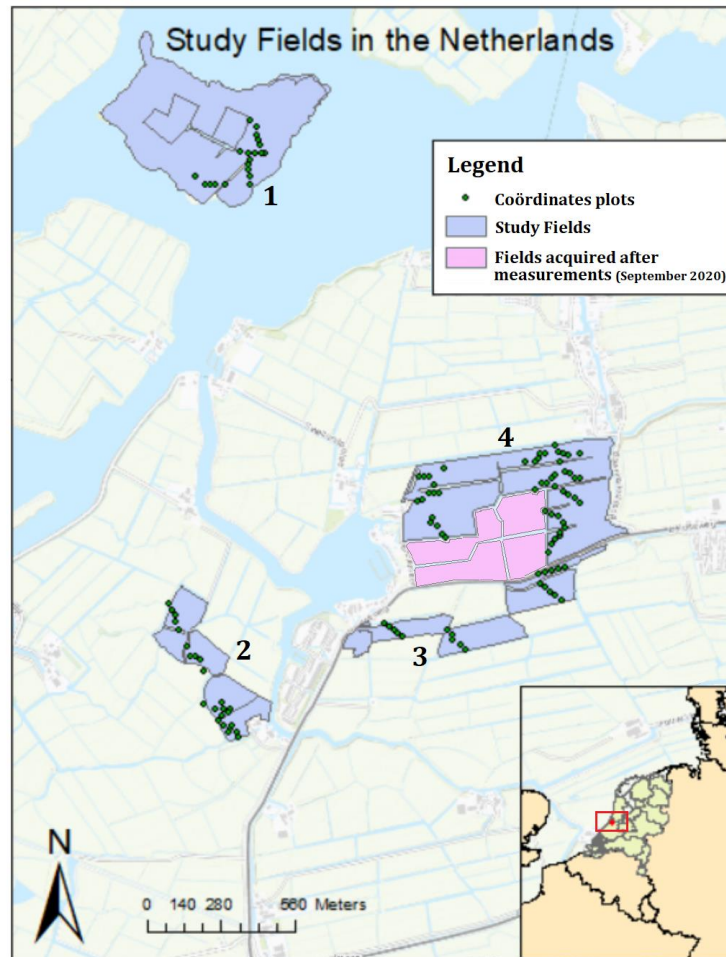
### **2.4. Plant Species Richness and the Influence of Management**

The management of the field influences not only the plant species richness, but also species richness of, among others, insects and macrofauna. The September 2020 database encompasses data from the four polders about insect diversity, aquatic, riparian and terrestrial plant diversity, aquatic macrofauna diversity, aquatic abiotic factors, greenhouse gas emissions per field, and micro-organisms in the soil. To find out which polder is under the best management, several questions about the insects, plants, abiotic factors and aquatic macroinvertebrates would have to be answered.

When analysing or using the database, it should be noted that:

- The database misses aquatic abiotic measurements and aquatic macrofauna measurements for the SVV polder. Due to a lack of time, these measurements were not taken.
- The database misses all measurements for almost half of the NVV polder. This land was acquired after all other measurements were taken. The fields are indicated in Image 2.1 with the pink colour.
- The database only contains measurements from September 2020, and gives, as such, an incomplete picture of the situation because each season has its own species diversity and abiotic milieu.
- The NVV polder is called the ‘Land van Ons’, since this is the focal polder and bought by the Land van Ons cooperation.
- The NVV polder has more measurement points than the other polders.

During the statistical analyses, the polder names are often shortened to make analysis quicker. As such, SVV stands for Southern part of the Vrouwe Vennepolder (3), NVV stands for Northern part of the Vrouwe Vennepolder (4), or LVO for Land van Ons, BP for Boterhuispolder (2), and LP for Lakerpolder (1).



*Image 2.1: Study fields from the September 2021 database. 1= Lakerpolder, 2= Boterhuispolder, 3= Southern Vrouwe Vennepolder, 4 = Northern Vrouwe Vennepolder.*

## Statistical analysis

### - Plant and macrofauna species

The first question answers whether there is a relation between the number of plant species and the number of macrofauna species. These numbers were extracted from the September 2020 database. They represent count data, are paired (because they are data from the same plot, and same ditch) and not normally distributed. As such, the new data subset was fitted in Rstudio using a poisson GLM with a 'sqrt' link function. The summary result discusses a possible correlation between the two. The strength of this relation was defined using the Spearman's Rank-Order correlation.

### - Species diversity per polder

The second question answers which polder has the most species diversity, and whether the differences between the polders are significant. Using the same subset as during the first question's analysis, boxplots were made for the macrofauna biodiversity per polder, the plant diversity per polder (bank and water data taken together as well as separated), and the total biodiversity on the banks (plants and insects) together with the total biodiversity in water (plants and macrofauna). The data is not paired, and independent, but non-normal. Therefore, sets of Wilcoxon Rank-Sum tests were done to find out which boxplots had a significant difference between them. The boxplots and their respective significant differences were explained afterwards, using each polder's management. This gave an insight into why the management resulted in a higher species diversity.

All conclusions are drawn on the assumption that the number of species (biodiversity) gives an indication for the quality of management of the polder.

## 2.5. Plant Species in the Current Vegetation

The management of a field influences the state of the ecosystems. This is reflected in the vegetation. So called indicator plants can be used to determine the range of a number of abiotic factors, without having to use measurement instruments. For example, *Urtica dioica* indicates nutrient rich soil and *Lycopus europaeus* indicates clean waters. The plants present in autumn (September) in the polder are included in the September 2020 database. These data were compared between the polders to determine which one, based on what the plant species composition says about the abiotic factors, has the best management. This "optimum", or "healthy", state of the abiotic factors in the peat-meadow area is determined through an analysis of the Oeverplanten Project Data.

To get the complete picture of plant species present, the plant relevés for the NVV polder were repeated in May 2021, following the same protocol as in September 2020. The autumn and spring data were compared to see how big the difference was. The original protocol and the fill-in sheet are added in Appendix 1.



## Field Methods

The locations for the plots can be seen in Image 1.3. Each plot was marked with pegs and cord as shown in Image 1.4. First, the total coverage of the vegetation for both the water and bank plots was determined, together with the maximum and mean plant species height of the plot. The second step was to identify all plant species and their respective coverage (10 cm x 10 cm is 1%). In the case of the aquatic plots, a net was used to take the submerged plants out of the water to be able to identify them. The total and partial coverages are only determined by the floating and emergent vegetation.



*Image 1.4: Plot parallel to the water side. # represents the plot marker (pole).*

It must be noted that the minimum value for plant coverage was put at 1%. For mosses, the coverage value can also be lower than 1%, and is divided into categories:

1. 0.0%-0.3%: A single strand of moss
2. 0.3%-0.7%: A handful of moss
3. 0.7%-1.0%: Nearly 10cm x 10cm of moss
4. Above 1%: estimate the coverage the same way as you would with plants.

When gathering the data, the different vegetation layers should be taken into account. For each layer, where applicable, the first and second steps mentioned above were repeated. The different vegetation layers were identified as:

- Mosses
- Herb layer
- Shrub layer
- Tree layer

The thickness of the litter layer was measured with a ruler.

## Autumn and Spring Data Comparison Analysis

Species lists from the Northern part of the Vrouwe Vennepolder collected in September 2020 and May 2021 were compared to see how many plant species differed. To get a better visual of the differences and similarities in species composition, Venn diagrams were made on scale in Excel. The diameter of the first circle, representing one of the two species pools (September or May), was arbitrarily chosen. The second circle would then be bigger or smaller than the first, following the same size ratio as between the two species pools. Using the circle surface, the diameter of the second circle was calculated. To calculate the percentages, four values were needed:

1. The number of species that are only found in the plant relevées
2. The number of species that are only found in the seed and propagule banks
3. The number of species occurring in both the plant relevées and seed and propagule banks

4. The total number of different species found over both species pools

The numbers belonging to group 1, 2, and 3 were divided by 4; the total number of different species found over both species pools.

### Grading of Polders Analysis

The Oeverplanten Project contains information on which plant species you can find for three specific sets of abiotic factors or managements in a peat meadow area, ordered from 'not healthy' or 'bad' (group A) to 'healthy' or 'good' (group C):

#### A) Nutrient rich soil and careless ditch bank management:

- |   |  |
|---|--|
| 1. Common nettle ( <i>Urtica dioica</i> )         | 4. Hedge bindweed ( <i>Calystegia sepium</i> ) |
| 2. Great willowherb ( <i>Epilobium hirsutum</i> ) | 5. Bittersweet ( <i>Solanum dulcamara</i> )    |
| 3. Common comfrey ( <i>Symphytum officinale</i> ) | 6. Ground-ivy ( <i>Glechoma hederacea</i> )    |

#### B) Careful ditch bank management and conditions with which a flowery vegetation can develop:

- |   |  |
|---|--|
| 1. Marsh woundwort ( <i>Stachys palustris</i> )     | 5. Purple loosestrife ( <i>Lythrum salicaria</i> ) |
| 2. Valerian ( <i>Valeriana officinalis</i> )        | 6. Gypsywort ( <i>Lycopus europaeus</i> )          |
| 3. Hemp-agrimony ( <i>Eupatorium cannabinum</i> )   | 7. Water mint ( <i>Mentha aquatica</i> )           |
| 4. True forget-me-not ( <i>Myosotis palustris</i> ) | 8. Yellow iris ( <i>Iris pseudacorus</i> )         |

#### C) Very careful ditch bank management, less nutrient rich soil and a flowery vegetation:

- |   |  |
|---|--|
| 1. Common fleabane ( <i>Pulicaria dysenterica</i> )         | 5. Marsh-marigold ( <i>Caltha palustris</i> )    |
| 2. Meadowsweet ( <i>Filipendula ulmaria</i> )               | 6. Common self-heal ( <i>Prunella vulgaris</i> ) |
| 3. Narrow-leaved-rattle ( <i>Rhinanthus angustifolius</i> ) | 7. Sneezewort ( <i>Achillea ptarmica</i> )       |
| 4. Lesser spearwort ( <i>Ranunculus flammula</i> )          | 8. Ragged-robin ( <i>Silene flos-cuculi</i> )    |

The species that are presumed 'common' on ditch banks are common reed and cattails. (see: Appendix 2 for translation to Dutch and alternative common names):

In order to assign the plants of the September relevée to one of the groups above an interval grading system was made for each group using the ecological data from Verspreidingsatlas (verspreidingsatlas.nl) for each plant mentioned above:

**Bad (1):** (Soil contains humus). (mediocre) Nutrient rich - (very) nutrient rich. Nitrogen-rich. (Weakly acidic -) calcareous soil

**Mediocre (5):** (Soil contains humus). Moderately nutrient rich- nutrient rich. (Nitrogen rich). (Acidic- ) weakly acidic - calcareous soil.

**Good (10):** (Nutrient poor-) moderately nutrient rich. Acidic-weak acidic (- calcareous). Alkaline- and nitrogen-poor

Appendix 5 contains the list with plants and their respective ecological data from NDFV Verspreidingsatlas on which this consensus is based.



As can be seen in Appendix 3, a plant could in some cases not be identified to species level. These observations were left out of the grade calculation per polder because each species has its specific abiotic environment, which can differ a lot within a genus.

Moreover, whilst making the grading, you have to weigh components against each other to determine the grade. Acidity overrules nutrient richness and nitrogen richness enhances nutrient richness but does not influence the grade if 'Moderately nutrient rich - nutrient rich' is indicated. However, when the indications are 'Moderately nutrient rich', 'Calcareous soil' and 'Moderately nitrogen rich', then it overrules 'Calcareous soil' and the plant will be rated 10. If a plant has the indication 'Moderately nutrient rich-nutrient rich', but 'Calcareous soil', it is rated as 1. On the other hand, if a plant has the indication 'Nutrient rich-very nutrient rich' but '(Slightly) acidic', it is rated as 5. Also, 'Weakly acidic-calcareous' indicates a large spectrum and is often omitted when grading the plant. When a plant is very dependent on 1 variable, and according to the other variables it would be rated as 5, it also is rated as 10.

After placing the plants in a group, points were assigned. Plants in category 'GOOD' got 10 points, the plants in category 'MIDDLE' got 5 points, and plants in category 'BAD' got 1 point.

Points were calculated per polder, and then divided by the number of plant species categorized per polder to normalize the data. The max value was 10, and the min value was 1 so that the grade ranged from 1 to 10.

Based on the grade, the polders were ranked from 1 to 4. The 'best' polder has the highest grade, and is ranked 1. This is however a relative value, and the choice of the 'best' polder will therefore be supported and explained.

## **2.6. Seed Banks**

The seed and propagule bank experiment gave an indication of which, and how many seeds or propagules are still present in the ditch bank soil and in the dredge from the ditches. The results were compared to the plant species present in the covering vegetation of the NVV polder. These data were collected beforehand (The Experiment 2.5, p. 14).

The methods used to collect soil and dredge samples are approximately the same, and will be explained further on in the text. The samples were taken from fifteen measurement points, evenly distributed throughout the polder (Image 1.2). They were stored together, in a predesignated spot in the field.

Based on the analyses, a conclusion will be drawn about whether the seed and propagule banks alone contain enough seeds to slowly turn the ecosystems towards more diverse ones, or that additional sowing would come in handy. However, not all seeds are expected to germinate. Under favourable conditions in, for example, a lab, more seeds would germinate.

## Field Methods

There were fifteen measurement points in total (Image 1.2). Per measurement point, two soil samples and two dredge samples were taken. That amounts to thirty pots for the dredge samples and thirty pots for the soil samples; in total sixty pots. These pots were, after being filled with soil or dredge, placed in two slots. These slots were as long as fifteen pots, as broad as two pots and as deep as one pot. One slot was for the thirty pots with soil, and the other was for the thirty pots with dredge.

All pots had to be numbered according to their measurement point and content. Fifteen pots would contain the top 0-5 cm of soil. Another fifteen pots would contain soil of the next five cm, so 5-10 cm below ground level. Thirty pots would contain dredge.

The soil was collected by drilling a twenty cm deep hole in the ground with an auger, between 50-60 cm from the waterside. All of the grassy bits of soil were removed, and the top 5-6 cm was put into the pot numbered #‘measurement point’, 0-5 cm. The next 5-6 cm in the pot numbered #‘measurement point’, 5-10 cm. This was repeated twice to collect a fair amount of soil, the holes being about sixty cm apart but still each being 50-60 cm from the waterside.

The dredge was scooped out of the ditch with a net. 5-7 cm of dredge was put in one pot and 5-7 cm in another pot. Afterwards, the net was cleaned.

After collecting all the soil and dredge, the pots were put in their respective slots. Every week, from 14-04-2021 to 10-06-2021 the number of seedlings were counted. To make sure our counting was correct, each seedling was marked with a little toothpick next to it. This way, it was

also easy to see whether a seedling had died. During the period in which the seedlings were counted, the grass around the experimental setup was shortened three times to prevent it from blocking the sun. The pots were watered every two days as well, except when there had been heavy rain. Every watering, the thirty soil pots got 10 L and the thirty dredge pots got 20 L.

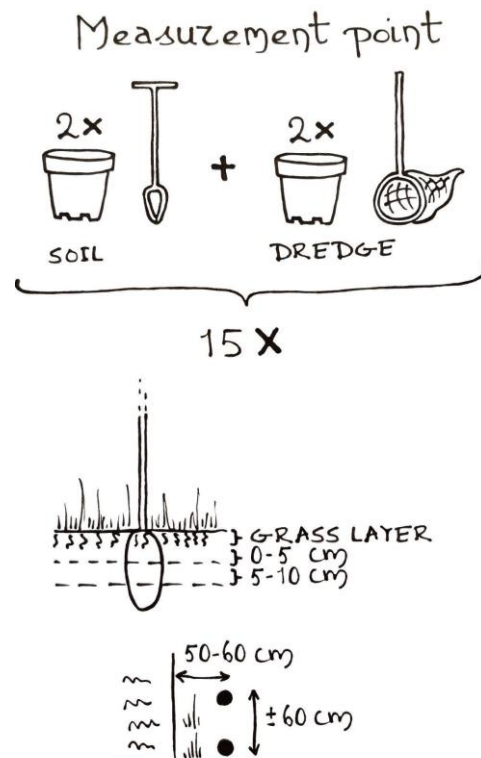


Image 1.5: Drawing of some methodical steps.

On 10-06-2021, the species that had germinated and survived were identified. The number of germinated seedlings were put in a dataset as follows:

Measurement Point	Type of substrate	{date} # seedlings
1	Soil 0-5 cm	...
1	Soil 5-10 cm	...
1	Dredge	...
2	Soil 0-5 cm	...
...	...	...

Table 1.1: Example of the dataset ordering for the germinated seedlings per pot.

And the different species that were identified were organized as below:

Measurement Point	Type of substrate	Number of species	Species names
1	Soil 0-5 cm	...	
1	Soil 5-10 cm	...	
1	Dredge	...	
2	Soil 0-5 cm	...	
...	...	...	

Table 1.2: Example of the dataset ordering for the different species found per pot.

## Statistical Analysis

The first step to assessing the restoration potential of the seed and propagule banks is to see which and how many seeds and propagules are still present. As such, the experiment described on page 18-19 was conducted.

### - Seedling accumulation

After finishing the experiment, the accumulation of seedlings was plotted in a diagram for each substrate ('dredge', 'soil 0-5 cm' and 'soil 5-10 cm') separately. The data distribution has an unknown upper limit and a lower limit on zero. This upper limit represents the estimate for the seed richness in the soil and can be extrapolated after fitting the data. Using the program QtGrace, the measurement points were fitted with an upside-down Gaussian curve, because a Gaussian distribution describes a natural distribution and approximates the curve of the measurement points best (Robert Ferréol, 2019, mathcurve.com). The data points were loaded into QtGrace. Then, using the non-parametric curve fitting tab, an inverse standard Gaussian model was entered for fitting the data points:

$$f(x) = a_0(1 - \exp(-(x \cdot x)/a_1))$$

The peak (Estimate) lies on  $a_0$ , and from  $a_1 (=2 \cdot (\text{STD} \cdot \text{STD}))$  the inflection point can be inferred.

The program fitted the curve and returned several computed values, including  $a_0$ ,  $a_1$ , the resulting formula and the correlation coefficient. The estimated total number of seedlings, which is the limit of the function, were compared to find out which substrate type contains the most seedlings.

For the inflection point (STD), which is the point of maximum seedling germination, the formula for  $a_1$  needs to be rewritten:

$$a_1 = 2\text{STD}^2$$

$$\text{STD} = \sqrt{a_1/2}$$

The correlation coefficient is a measure for the goodness-of-fit of the Gaussian curve fitted through the data points. According to Jim Frost on his website Statistics by Jim, the limit should be based on your study area and the number of variance that can be explained. Since the seed- and propagule bank experiment contained many factors causing variance, like abiotic factors, insects, animals, watering and seedling fertility, a limit of 0.9 for the goodness-of-fit was chosen (Statistics by Jim). The correlation coefficients were compared to this value to estimate the accuracy.

To be able to compare the number of seeds per substrate, the number of seedlings on 1 m<sup>2</sup> of soil or dredge was extrapolated using the estimate. A low number of seeds, or seed density won't add much to ecosystem recovery, because not all seedlings survive until they are all full-grown. Moreover it also implies a slower recovery towards a more diverse ecosystem, and indicates most probably a low number of species.

#### - Species accumulation

The seedlings were determined purely on morphological characters. DNA sequencing would have taken too much time and would have been too expensive. When the seedlings were identified, the resulting numbers of observed seedling species per pot were organized in a species frequency table. Using R-studio's package `vegan` and its function `specaccum` (method = "exact"), species accumulation curves were made per substrate type. The function `specpool` includes several non-parametric estimator types for extrapolating the total species diversity. This species diversity is the asymptote of the fitted curve through the measurement points. The data type was used as an argument to choose between the different estimators. The dataset presents incidence data, because plots were used to sample the species pool. Per sample, the species occurrence frequencies were counted. The non-parametric estimator Jack2 was chosen. According to Carlos Martínez-Sanz et al. (2010), the Jack2 estimator needs the least number of samples to find the most accurate value for the total species richness (gamma

diversity). The extrapolated values for the total species richness per substrate types were compared to find out which substrate type contains most species.

To find out how many samples one needs to have most probably encountered all species at least once (How high does the sampling effort need to be?), the slopes of the accumulation curves on the last point were compared. These slopes can be extracted using

```
> slopes <- with(data, diff(richness)/diff(sites)) (stackoverflow.com)
```

To find out whether we made many identification mistakes, the curve of the species accumulation graph was extracted, and thereafter, the points of the curve were imported into QtGrace. These points were fitted with the 'expected model'. When the offset between the perfect fit (exactly through the points) and the fit with the 'expected model' (not perfectly through the points, but shows exactly the trend you would expect) is very big, we made a lot of identification mistakes. Whereas if the offset is very small, we made very few or no mistakes.

### **Comparison of Species Analysis**

The species found in the seed- and propagule banks were compared to the species in the plant relevées done in September 2020 (included in the September 2020 database) and in May 2021. To get a better visual of the differences and similarities in species composition, a Venn diagram was made using Excel, following the same steps as in The Experiment 2.5 Autumn and spring data comparison analysis (p. 15).

The same method was used to compare the species compositions between the substrate types from the seed and propagule bank experiment.

### **Species Abundance Analysis**

The frequencies of occurrence per plant species per substrate were extracted from the seed and propagule bank dataset. The results were put in a stacked bar chart, which visualizes the difference in total seedling counts, seed counts per species and species composition. The top three most abundant species and the rarest species were recorded per substrate layer.

# Results

## 1. The Influence of Management on Species Diversity

### 1.1. Is the Number of Aquatic Macrofauna Species Related to the Number of Aquatic Plant Species?

There is a positive relation between the number of aquatic plant species and aquatic macrofauna (p-value = 6.86e-06), as can also be seen in the output of the GLM in figure 2.2. See page 12 for the used statistical methods. In order to show that the relation is not an artefact, we test the strength of the relationship per polder and for all polders combined (named Total) with the Spearman Rank-Order test ( $\alpha = 0.05$ ):

Polder name	Rho	P-value
Boterhuispolder:	0.675	0.066
Lakerpolder:	0.828	0.011
Northern Vrouwe Vennepolder:	0.471	0.036
Total	0.559	0.000399

The calculation for each polder separately shows that only in the Boterhuispolder the correlation is not significant. Calculating Total gives a strong significant relation (Rho=0.559, P-value=0.000399).

Because more points give a more dependable outcome, only the 'Total: rho' was used in the conclusion. Note that as rho differs significantly from 1, there might be another influential factor.

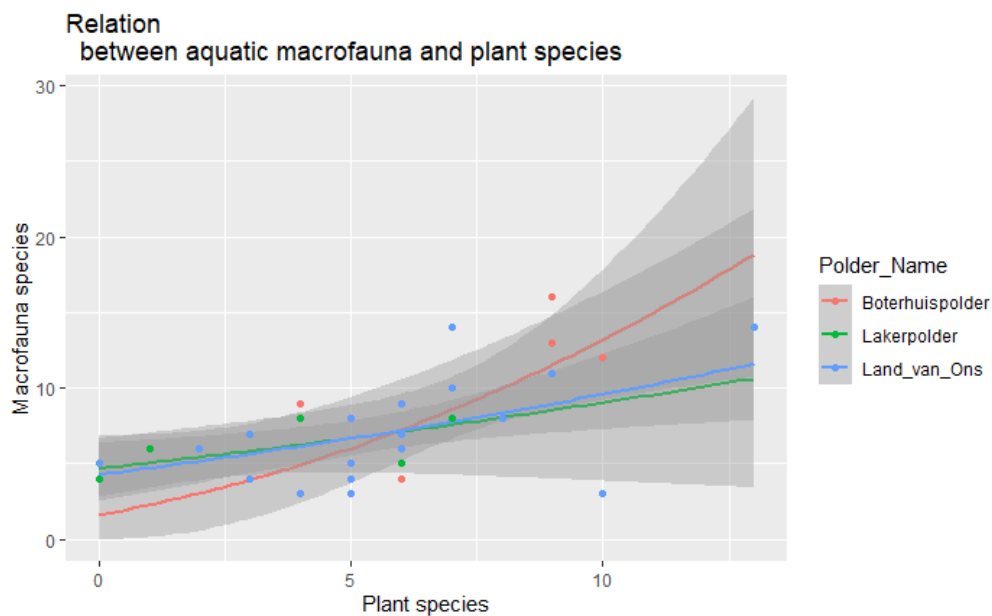


Figure 2.2: Line plot showing the relation between the number of aquatic plant species and the number of aquatic macrofauna species per polder.

## 1.2. What is the overall Plant, Insect and Aquatic Macrofauna Species Richness per Polder?

### Macrofauna Species Richness per Polder

There is no significant difference between the found aquatic macrofauna species richness between all polders, as can also be seen in figure 2.3. (See page 12 for the used statistical methods.)

The Boterhuispolder has the highest median in species richness, followed by the NVV. The Lakerpolder has the lowest median in species richness, though Rstudio shows no significant difference between the Lakerpolder and NVV. At the same time, the Lakerpolder also has the lowest variation in its data, and the Boterhuispolder the most.

The NVV macrofauna outliers from plot 127 and 145 are potentially explained by the equally high aquatic plant species richness.

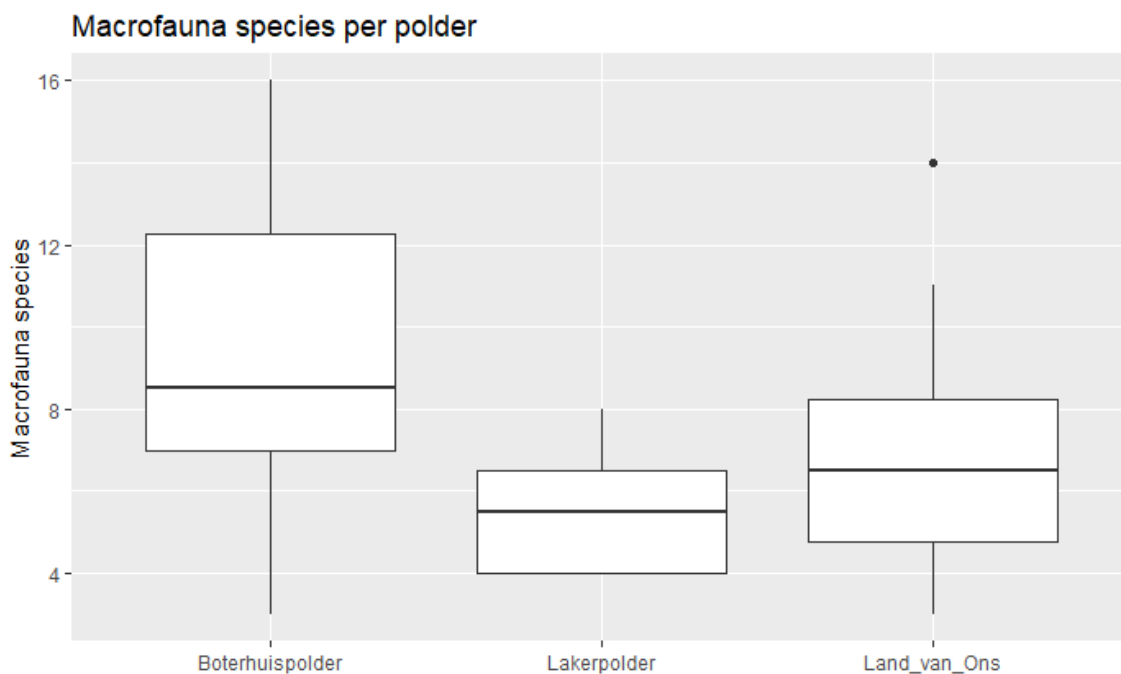


Figure 2.3: Boxplot of the aquatic macrofauna species plotted for each polder.

Comparison polders	P-value
NVV polder : Boterhuispolder	0.2106
NVV polder : Lakerpolder	0.3973
Boterhuispolder : Lakerpolder	0.1096

## Total Biodiversity of Plant Species per Polder; Riparian and Aquatic Data Pooled

The Boterhuispolder, now together with the SVV, came out on top. There is no significant difference between these two, but both do have a significant difference with the Lakerpolder and NVV. This can be seen in the boxplots in figure 2.4. (See page 12 for the used statistical methods.)

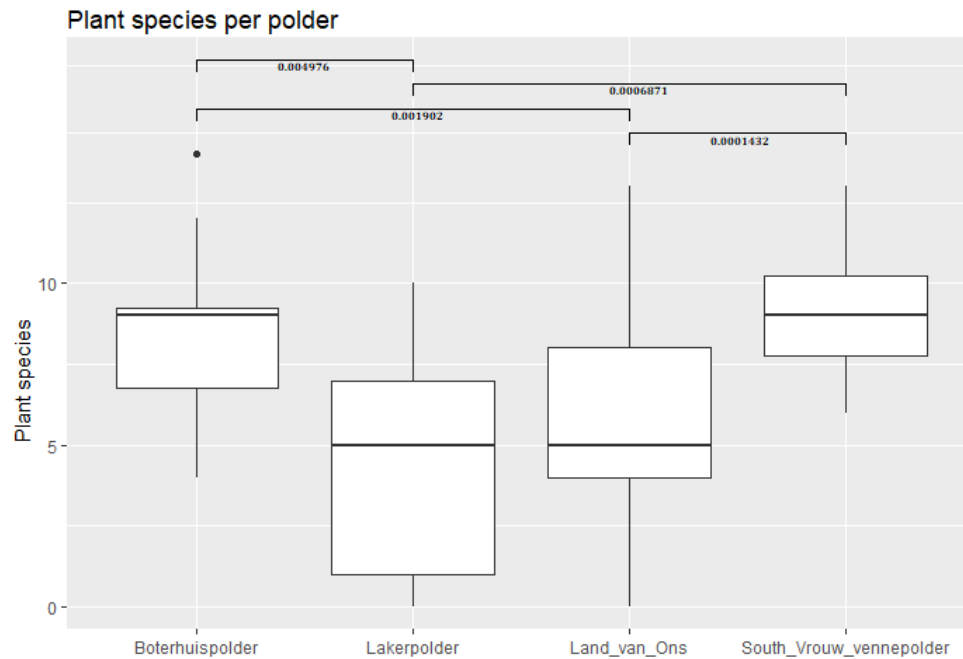


Figure 2.4: Boxplots for the pooled riparian and aquatic plant species richness per polder. The black lines above the boxplots indicate a significant difference between the respective boxplots.

Comparison polder	P-value
Boterhuispolder : Lakerpolder	<b>0.004976*</b>
Boterhuispolder : NVV	<b>0.001902*</b>
Boterhuispolder : SVV	0.4933
Lakerpolder : NVV	0.2506
Lakerpolder : SVV	<b>0.0006871*</b>
NVV : SVV	<b>0.0001432*</b>

\* Indicates a significant difference at  $\alpha < 0.05$



## Unpooled Riparian and Aquatic Plant Species Diversity per Polder

To interpret the above mentioned results, we have separated the pooled plant species richness in riparian (B) and aquatic (W) plant species richness. The results can be seen in figure 2.5. This analysis shows that the significant difference between the polders is mainly due to the riparian species in the case of the Boterhuispolder and the NVV, the aquatic species in the case of the Boterhuispolder and Lakerpolder and to both species in case of the NVV and SVV, and the SVV and Lakerpolder. The Lakerpolder still has the biggest variation in species. The ditches (riparian species) contain very few species in general, which explains the big variation in the total dataset of figure 2.4. The aquatic species data from the Lakerpolder and NVV differ significantly as well, but doesn't show up in the total dataset in Figure 2.4.

The Boterhuispolder hasn't got an outlier anymore. The riparian plant species data from the SVV has. For the riparian plant species richness, the SVV has the highest median, followed closely by the Boterhuispolder, which differs significantly only with the NVV. As the SVV differs significantly with both the NVV and the Lakerpolder, it is deemed best. The aquatic plant species richness shows the same trend. The SVV has the highest median, but no significant difference with the Boterhuispolder, which in this case differs significantly only with the Lakerpolder. This shows that even when the pooled plant species dataset is divided, the SVV has still the highest median followed closely by the Boterhuispolder. However, drawing definite conclusions based on these numbers is a bit tricky as explained in Results 1.3.

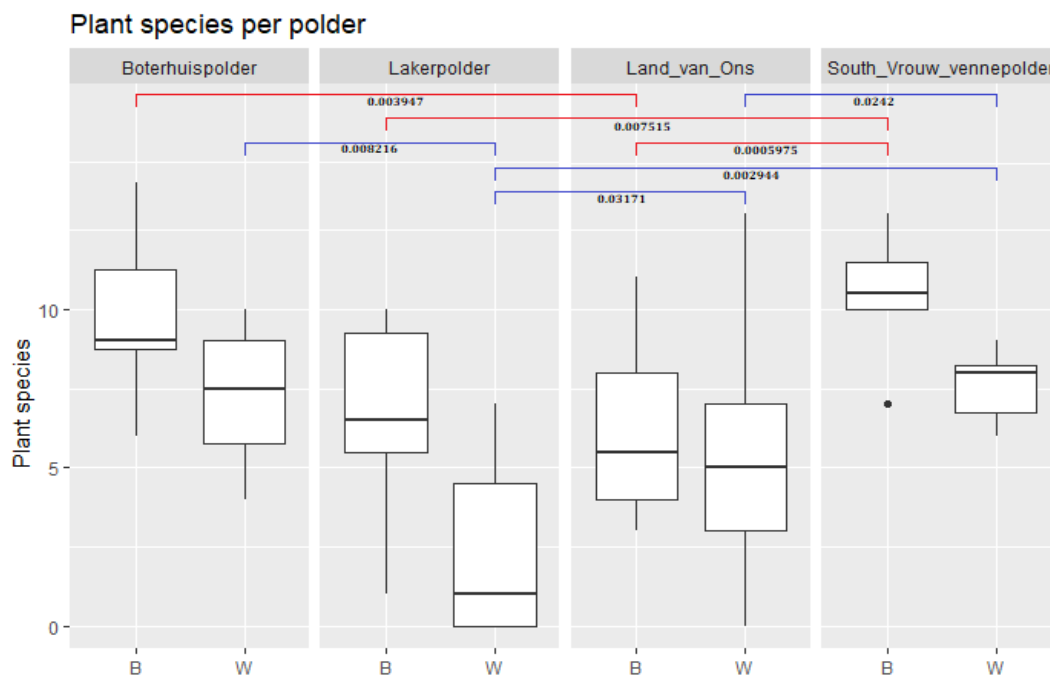


Figure 2.5: Boxplots for the riparian (B) and aquatic (W) plant species richness per polder. The x-axis indicated if the data is from the banks (B) or the water (W). The y-axis is the plant species richness. The blue lines indicate a significant difference in the water data and the red lines in the bank data.

The output for the riparian plant species. (See page 12 for the used statistical methods):

<b>Comparison polder</b>	<b>P-value</b>
Boterhuispolder : Lakerpolder	0.08928
Boterhuispolder : NVV	<b>0.003947*</b>
Boterhuispolder : SVV	0.3674
Lakerpolder : NVV	0.5721
Lakerpolder : SVV	<b>0.007515*</b>
NVV : SVV	<b>0.0005975*</b>

\* Indicates a significant difference at  $\alpha < 0.05$

The output for the aquatic plant species:

<b>Comparison polder</b>	<b>P-value</b>
Boterhuispolder : Lakerpolder	<b>0.008216*</b>
Boterhuispolder : NVV	0.09626
Boterhuispolder : SVV	0.8725
Lakerpolder : NVV	<b>0.03171*</b>
Lakerpolder : SVV	<b>0.002944*</b>
NVV : SVV	<b>0.0242*</b>

\* Indicates a significant difference at  $\alpha < 0.05$

## Total biodiversity on the banks (insects and plants) and in the water (aquatic macrofauna and plants)

Based on the total species biodiversity in the water (macrofauna + plants) the Boterhuispolder ranks highest according to its median, however there is no significant difference with the NVV. The results are shown in figure 2.6.

The data set for the SVV contains only aquatic plant species data and no macrofauna data, so the aquatic data (W) boxplot of the SVV cannot be compared to the other aquatic data boxplots.

Deciding which polder ranks highest based on the total species diversity on the banks (insects + plants) is difficult. According to the median, the SVV ranks highest, followed closely by the Lakerpolder and Boterhuispolder, the NVV definitely ranks lowest. However, the Lakerpolder has a lot of variance in biodiversity per plot, so both the SVV and the Boterhuispolder score the highest in this regard.

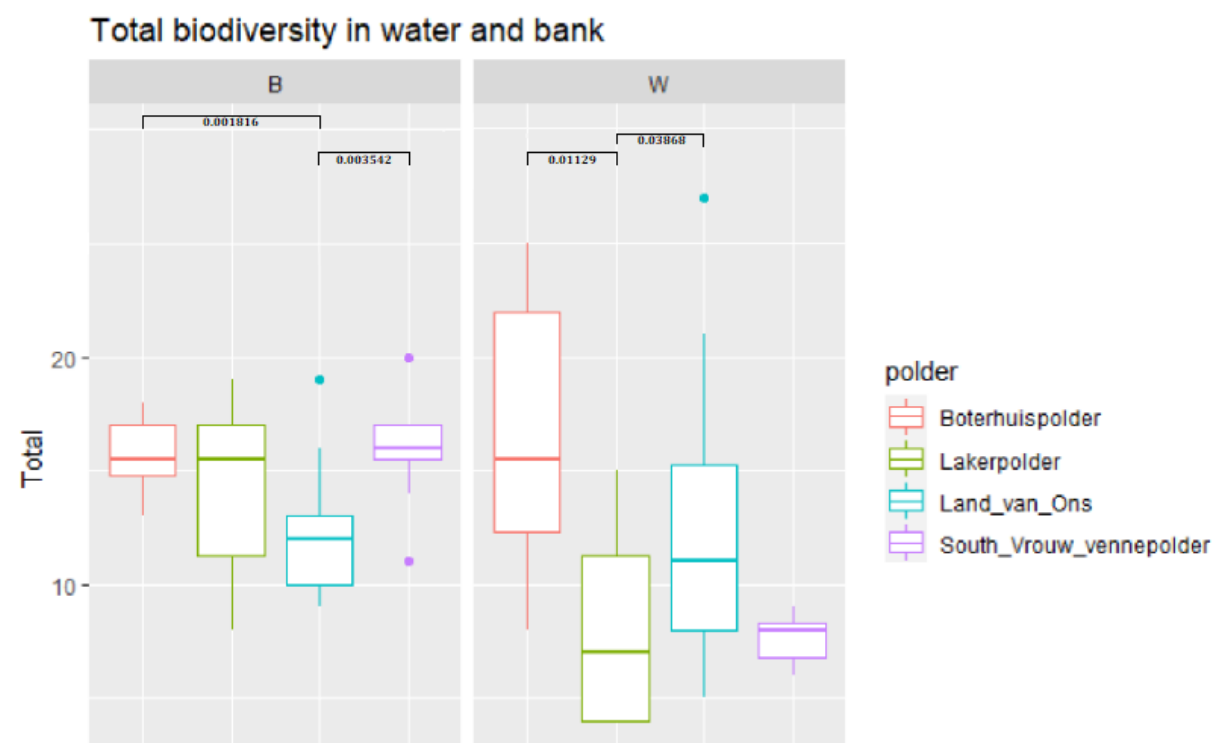


Figure 2.6: Boxplots of the total biodiversity (insects and plant species diversity) on the ditch banks (B) per polder, and boxplots of the total biodiversity (aquatic macrofauna and aquatic plant species diversity) in the ditches (W) per polder. The black lines above the boxplots indicate a significant difference between the respective boxplots.

The output for the water (W). See page 12 for the used statistical methods.:

Comparison polder	P-value
Boterhuispolder : Lakerpolder	<b>0.01129*</b>
Boterhuispolder : NVV	0.1021
Boterhuispolder : SVV	<b>0.002987*</b>
Lakerpolder : SVV	0.7908

Lakerpolder : NVV **0.03868\***

SVV : NVV **0.01208\***

\* Indicates a significant difference at  $\alpha < 0.05$

The output for the banks (B):

<b>Comparison polder</b>	<b>P-value</b>
Boterhuispolder : Lakerpolder	0.671
Boterhuispolder : SVV	0.7895
Boterhuispolder : NVV	<b>0.001816*</b>
Lakerpolder : SVV	0.5224
Lakerpolder : NVV	0.1919
SVV : NVV	<b>0.003542*</b>

\* Indicates a significant difference at  $\alpha < 0.05$

### 1.3 Connecting Species Richness to Management

The managements that are currently implemented in each polder are important to determine the cause for the current species richness and composition. On page 9 the management and situation in each polder are illustrated in detail.

To see which management is the best baseline for restoration of the NVV polder, the species richness values were compared per polder in Results p. 22-28.

	<b>Highest median</b>	<b>Lowest median</b>
<b>Macrofauna</b>	Boterhuispolder	Lakerpolder
<b>Total plant species (pooled)</b>	Boethuispolder & SVV	Lakerpolder & NVV
<b>Total plant species (aquatic)</b>	SVV	Lakerpolder
<b>Total plant species (riparian)</b>	SVV	NVV
<b>Total biodiversity (aquatic)</b>	Boterhuispolder	Lakerpolder
<b>Total biodiversity (riparian)</b>	SVV	NVV

Table 2.2: Summary of the boxplots

As you can see in the table above (Table 2.2), the Boterhuispolder and SVV have the highest species richness of the four polders in every analysed species group, never differing significantly. This is partly due to the management that is being implemented.

The Boterhuispolder has a high species richness median due to its variation in ditch bank management, which results in a big micro- and macro-habitat range and a lot of variation in abiotic factors, making some areas less liveable than others and creating habitats with many types of species. Moreover, cows sometimes trample the ditch banks, killing the plants and creating opportunities for new species to settle. The SVV polder has a high species richness median due to biological farming, which resulted in a more diverse habitat range with a wider range of biotic and abiotic factors and less dominated by a few or a single species.

In almost every case the Lakerpolder scores significantly lower than the Boterhuispolder and SVV, the NVV only marginally better, except in case of the total plant species (aquatic) and total biodiversity (aquatic) where the NVV scored better.

Even though the NVV and Lakerpolder both score lowest based on species richness, the reason behind this low score is absolutely not comparable. The NVV polder has often the lowest species richness median due to dominance of nutrient loving species (*Lolium perenne* and *Holcus lanatus*) because of the frequent appliance of slurry. The management of the NVV polder did not take biodiversity into account.

In contrast, the Lakerpolder is managed as a nature reserve, with less differentiated habitats, a smaller abiotic factor range and no cow-trampled ditch sides which create all kinds of new habitats. Moreover, it has rather wet conditions suitable for only a few species, like common reed (*Phragmites australis*), that easily dominate less suited plants. Interestingly this significant difference with de SVV and the Boterhuispolder does not show excessively in fig. 2.6 (total biodiversity), which indicates that the Lakerpolder has a very high insect biodiversity and a healthy ecosystem even though all other data points in the opposite direction. It can therefore be concluded that the management of the Lakerpolder is good.

## **2. Plant Species Community of the Current Vegetation**

Most plant species can tell you something about their abiotic environment. From the NVV's past use, we know that the soil and water are both nutrient rich, but what does the vegetation say? To determine the state of the environment per polder, we collected data on the plant species present on the ditch banks and in the water, and compared their ecological data using the interval grading system (The Experiment 2.5) .

### **2.1. Grading of the Ditches and Ditch Banks per Polder for “Healthy” Peat-Meadow Environment.**

The plants from the relevées that were represented in the Oeverplanten project database were:

- 1: *Epilobium hirsutum*, *Symphytum officinale*, *Urtica dioica*
- 5: *Iris pseudacorus*, *Lycopus europaeus*, *Lythrum salicaria*, *Mentha aquatica*, *Stachys palustris*

10: *Filipendula ulmaria*, *Ranunculus flammula*

Others were not represented in the database, and were graded following the interval grading system.

A) Nutrient rich soil and careless ditch bank management (1):

<i>Acorus calamus</i>	<i>Festuca arundinacea</i>	<i>Ranunculus acris</i>
<i>Agrostis stolonifera</i>	<i>Galium palustre</i>	<i>Ranunculus repens</i>
<i>Alopecurus pratensis</i>	<i>Glyceria maxima</i>	<i>Ranunculus sceleratus</i>
<i>Anthriscus sylvestris</i>	<i>Lemna minuta</i>	<i>Rumex acetosa</i>
<i>Bidens tripartita</i>	<i>Lysimachia nummularia</i>	<i>Rumex crispus</i>
<i>Cardamine hirsuta</i>	<i>Nasturtium officinale</i>	<i>Rumex palustris</i>
<i>Cerastium fontanum</i>	<i>Persicaria amphibia</i>	<i>Sonchus arvensis</i>
<i>Cerastium glomeratum</i>	<i>Persicaria maculosa</i>	<i>Sparganium erectum</i>
<i>Ceratophyllum demersum</i>	<i>Phalaris arundinacea</i>	<i>Spirodela polyrhiza</i>
<i>Cirsium arvense</i>	<i>Plantago major</i>	<i>Stellaria media</i>
<i>Elodea nutallii</i>	<i>Polygonum aviculare</i>	

B) Careful ditch bank management & conditions in which a flowery vegetation can develop (5):

<i>Alisma plantago-aquatica</i>	<i>Hydrocharis morsus-ranae</i>	<i>Potamogeton trichoides</i>
<i>Berula erecta</i>	<i>Juncus effusus</i>	<i>Potentilla anserina</i>
<i>Cardamine pratensis</i>	<i>Lathyrus pratensis</i>	<i>Rumex hydrolapathum</i>
<i>Eleocharis palustris</i>	<i>Lemna minor</i>	<i>Taraxacum officinale</i>
<i>Epilobium montanum</i>	<i>Lemna trisulca</i>	<i>Trifolium repens</i>
<i>Equisetum fluviatile</i>	<i>Myosotis scorpioides</i>	<i>Vicia cracca</i>
<i>Ficaria verna subsp. verna</i>	<i>Phragmites australis</i>	
<i>Geranium molle</i>	<i>Plantago lanceolata</i>	

C) Very careful ditch bank management, less nutrient rich soil and a flowery vegetation (10):

<i>Apium inundatum</i>	<i>Juncus subnodulosus</i>	<i>Persicaria hydropiper</i>
<i>Angelica sylvestris</i>	<i>Montia arvensis</i>	<i>Scutellaria galericulata</i>
<i>Bidens connata</i>	<i>Oenanthe aquatica</i>	

For the ecological information from Verspreidingsatlas, which was used to classify each plant, see Appendix 5.

With this grouping, we could calculate a grade for each polder, after which a rank could be assigned.

Polder	Water (W)	Rank	Banks (B)	Rank	W+B	Rank
Boterhuis	2,892857143	4	3,424242424	1	3,158549784	3
Lakerpolder	3,941176471	1	2,761904762	4	3,351540616	1
NVV polder	3,451612903	2	2,952380952	3	3,201996928	2
SVV polder	2,933333333	3	3,34375	2	3,138541667	4

Table 2.3: Grade and rating per polder, for water plots and bank plots apart and together.

The ditches of the Lakerpolder and the banks of the Boterhuispolder resemble a peat meadow abiotic environment and species composition best, and the ditches in the Boterhuispolder and banks of the Lakerpolder the least. Combining ditches and ditch banks the Lakerpolder resembles a peat meadow abiotic environment and species composition the best, and the SVV the least. These ranks and grades are, of course, relative numbers. To help support and explain the conclusions, the number of plants from the Oeverplanten Project Database and website that were present in the polders were recorded.

<b>Water</b>	Boterhuispolder:	2x 5 points
	Lakerpolder:	1x 1 points, 3x 5 points, 1x 10 points
	NVV polder:	2x 1 points, 2x 5 points
	SVV polder:	1x 1 points, 1x 10 points
<b>Banks</b>	Boterhuispolder:	1x 1 points, 1x 5 points
	Lakerpolder:	3x 5 points
	NVV polder:	2x 1 points, 4x 5 points
	SVV polder:	1x 1 points, 3x 5 points, 1x 10 points

## 2.2. Results Plants Relevée Data, does Biodiversity in Spring differ from that in Autumn

For the NVV, plant relevées were also done on the 19th, 20th and 23rd of May 2021. Combining these data with the September relevées, the NVV was found to contain 58 different species. In spring (May 2021) we found 30 plant species and 49 plant species were found in autumn (September 2020). Species pools were compared to check for plant species differences between the two seasons. Nine new species were found in May:

- *Cardamine hirsuta*
- *Alopecurus pratensis*
- *Stellaria media*
- *Iris pseudacorus*
- *Cerastium glomeratum*
- *Potentilla anserina*
- *Ficaria verna subsp. verna*
- *Ranunculus sceleratus*
- *Carex hirta*

The twentyeight plant species that were found in autumn and not in spring are:

- *Agrostis stolonifera*
- *Alisma plantago-aquatica*
- *Apium inundatum*
- *Berula erecta*

- *Bidens connata*
- *Bidens tripartita*
- *Ceratophyllum demersum*
- *Epilobium montanum*
- *Geranium molle*
- *Hydrocharis morsus-ranae*
- *Juncus subnodulosus*
- *Lemna minor*
- *Lemna minuta*
- *Lemna trisulca*
- *Lythrum salicaria*
- *Myosotis scorpioides*
- *Nasturtium officinale*
- *Persicaria hydropiper*
- *Phalaris arundinacea*
- *Phragmites australis*
- *Polygonum aviculare*
- *Potamogeton trichoides*
- *Ranunculus acris*
- *Rumex crispus*
- *Rumex palustris*
- *Scutellaria galericulata*
- *Sparganium erectum*
- *Spirodela polyrhiza*
- *Stachys palustris*

For a list of the plant species found on the ditch banks and in the water per polder in September, see Appendix 3. For a list of the plant species found on the ditch banks and in the water in the NVV polder in May, see Appendix 4.

From figure 2.7 it can be concluded that, when plant relevés are done in May approximately 51.7% ( $= (30/58) * 100\%$ ) of the total plant species pool will be found whereas 84.5% ( $= (49/58) * 100\%$ ) will be found in autumn (September). Sampling in September will give the best impression of the gamma species diversity in the ditches and on the ditch banks of the NVV.

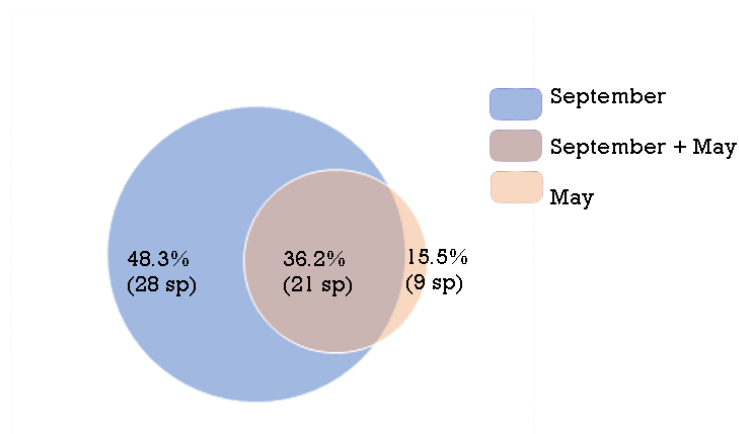


Figure 2.7: Venn diagram with percentages and number of species indicated per circle colour. The red percentage and species number gives the values of the blue and purple circle parts taken together (=the number of species found in September).



### **3. Can Seed and Propagule Banks be used for Ecological Restoration**

The seed and propagule bank experiment started on 14-04-2021 and ended on 10-06-2021. The seeds have had time to grow for eight weeks. Extra information was gathered on:

1. Visual observations of the soil composition.
2. Accumulation of the number of seedlings per substrate type.
3. Influence of weather on seedling germination.
4. Accumulation of the number of species per substrate type.
  - a. Which soil layer; 0-5 cm or 5-10 cm contains most of the species?
  - b. How many samples are needed for a good representation of the population?
5. Species composition
  - a. How do the species present in the different substrates relate to each other?
  - b. How do the species present in the soil and propagule bank differ from the species present in the covering vegetation?
  - c. Which species are the most prominent, and which are rare?

#### **3.1. Visual Observations of the Soil Composition**

We had to dig between thirty and fifty cm to reach the peat layer. Whilst digging the slots for the pots, we encountered a lot of worms. This means that this top layer is very 'alive'. The soil composition differed greatly between the measurement sites. In four pots the soil felt clay-like, while two others contained some sand particles. Disregarding the remaining root systems, the soil was rather lumpy and did not actually crumble, causing four of our pots to contain lumps of soil instead of a nice homogenous layer. The other pots had a more homogenous layer of soil. This difference, however, did not affect the number of sprouting seeds.

The dredge compacted easily into a small disk, leaving a white layer behind. This white layer could be the cause of a compound, like salt, in the water or dredge itself. The soil pots did not show a white layer after watering.

#### **3.2. Seedling Accumulation over Time, per Substrate Type**

To answer our main question, we needed to know whether the soil and dredge contained enough seeds and propagules. The question that was answered:

- *What is the maximum number of germinating seeds?*
- *How long does it take to reach this number?*

Seed accumulation diagrams were made using the program QtGrace for each substrate type separately. The measurement points were fitted with an upside-down Gaussian distribution, which approximated the curvature of the germinating seedlings the most (The Experiment 2.6, p. 19-20).

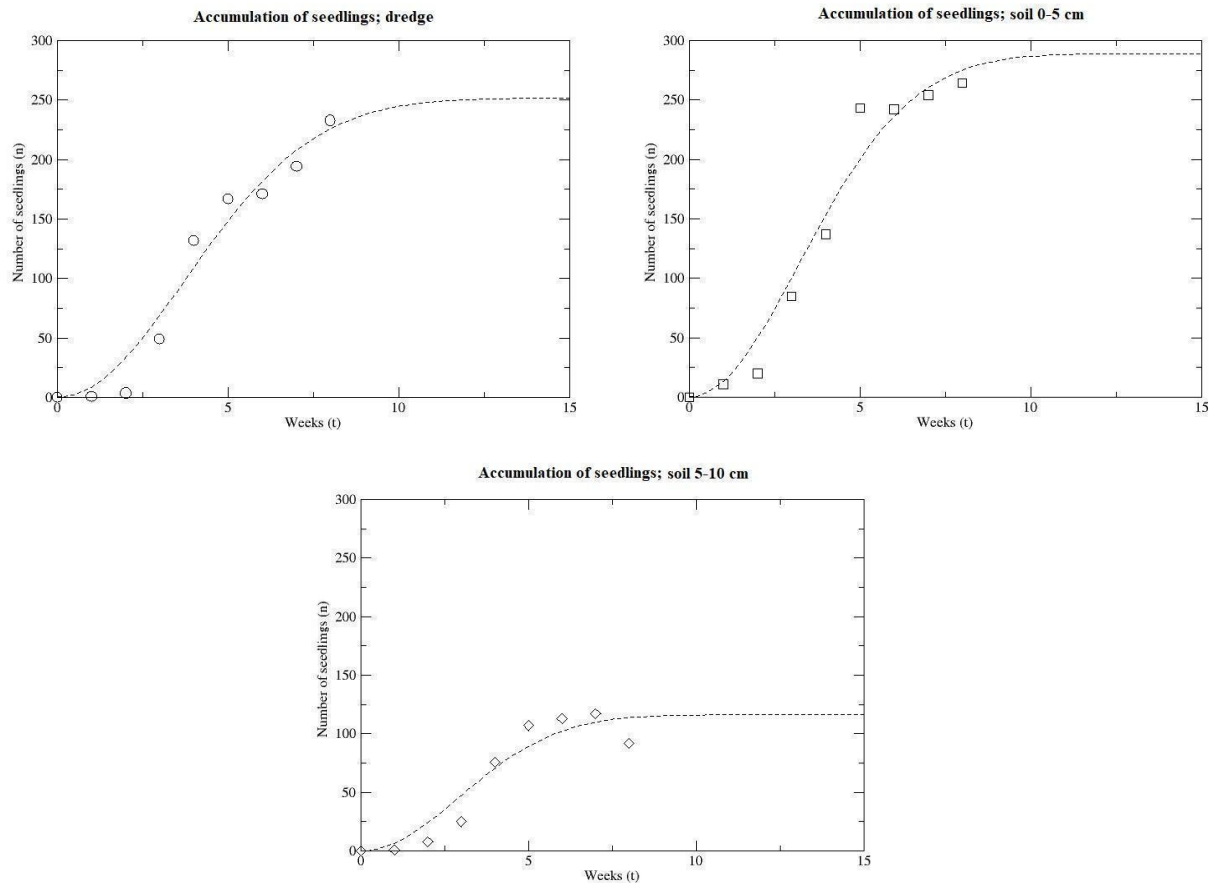


Figure 2.8: Diagram of the accumulation of seedlings over a time span of 9 weeks. The circles indicate the measurement points (one per week). Dotted line is the fitted curve through the measurement points

As can be seen in figure 2.8, the top 0-5 cm layer of soil contained most seeds, and the lowest number of seeds was found in the 5-10 cm layer of soil. The extrapolated number of seedlings in the dredge lies between the 0-5 cm layer and 5-10 cm layer of soil, but corresponds more to the 0-5 cm layer. QtGrace predicted the following:

Substrate type	Estimate (a0)	Inflection point ( $\sqrt{a1/2}$ )	Corr. coef.
- Dredge:	252 seeds	3.76 weeks	0.983
- Soil 0-5 cm:	289 seeds	3.25 weeks	0.985
- Soil 5-10 cm:	117 seeds	2.95 weeks	0.961

The inflection point is equal to the STD according to the formula used (see methods). The inflection point represents the point in time where the maximum germination speed is achieved. Remarkably, the 'soil 5-10 cm' has the lowest value for this inflection point, and as such, reaches its optimum germination speed fastest. Following 'soil 5-10 cm' is the 'soil 0-5 cm' and lastly the 'dredge'.

The correlation coefficients (R-squared, value for goodness-of-fit) for the substrates ‘soil 0-5 cm’ and ‘dredge’ are both above the 0.9 limit. The curve through soil 5-10 has a lower R-squared than the other two substrate types due to a rapid decline in seedlings during the last (8th) week.

As can be inferred from figure 2.8, the accumulation of seedlings reaches its maximum after approximately 10 weeks for the ‘dredge’ and ‘soil 0-5 cm’ substrates, but the ‘soil 5-10 cm’ substrate reaches its maximum in approximately 7.5 weeks.

Now that the number of seedlings is estimated, this number can be related to the measured surface area, which is the surface area of soil or dredge in the pots.

	Mean diameter (m)	Mean surface area (m <sup>2</sup> )	Total (m <sup>2</sup> )	Seedlings per 1 m <sup>2</sup>
Dredge	0.112	0.010	0.296	851
Soil 0-5 cm	0.130	0.013	0.120	2408
Soil 5-10 cm	0.130	0.013	0.120	588

Table 2.4: Calculation of the number of seedlings per 1 m<sup>2</sup>.

### 3.3. Influence of the Weather on Seed Germination

Weather influences germination rates and seedling survival. As such, it is important to keep this in mind when drawing conclusions or explaining strange measurements.

The weather has varied a lot. Most of the time the temperature wavered between 10-15 °C, and it was raining or cloudy. Those days that the sun did shine, it was around 20-25 °C. The rain, wind and temperature measurements in figure 2.9 were taken by the weather station in Leiderdorp. This is the closest weather measurement station to our research area. The data was collected from the site: Het Weer Actueel; Weerstatistieken van weerstation Leiden. Between the 29th of April and the 20th of May, heavy rainfall up to 17 mm was recorded. This coincides with the growth period of the seedlings (experiment was started on 14th of April). The green vertical lines indicate that the pots were watered. On the 31th of May and 2nd of June, wind speeds reaching 1380 km/h were recorded. During these days there was no rain and the temperature rose above 20 °C, increasing the evapotranspiration rates. The 30th of April recorded wind speeds up to 184 km/h, no rain but the temperature only reached 9 °C. Next to that, the seedlings had just started to grow.

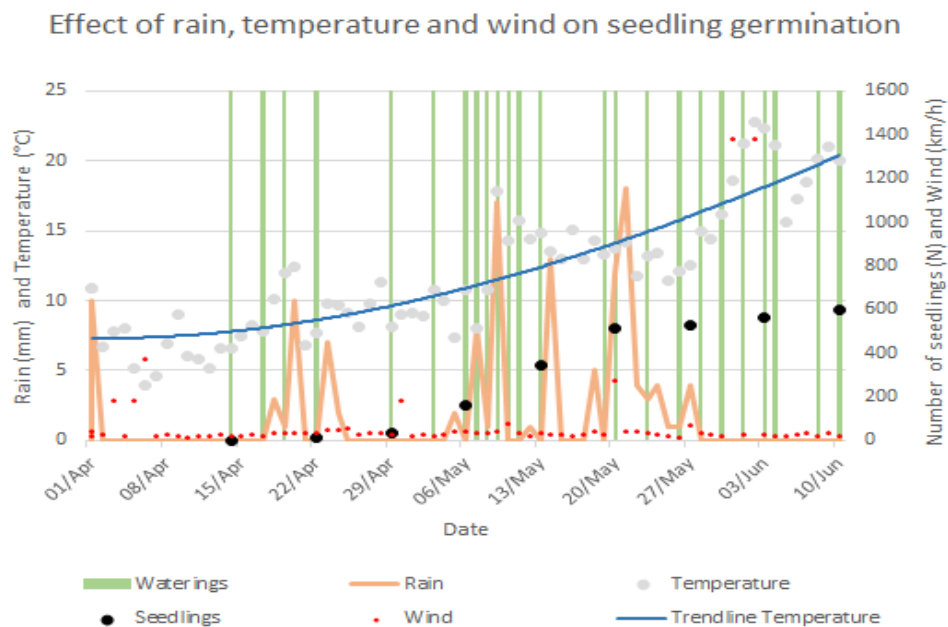


Figure 2.9: The diagram shows the weather conditions during our experiment. The green lines indicate when people watered the experiment. The orange line is the volume of rain (mm) that has fallen. The grey dots are the temperature measurements (°C), and the blue line is the trendline of the temperature. The red dots are wind measurements (km/h). The black dots are the total number of seedlings present per measurement day.

### 3.4. Species Accumulation over Time, per Substrate Type and Compared to the Plant Relevées

The goal is to see if the seed and propagule bank can be used for restoration. Therefore, the difference in species between the covering vegetation and the banks must be investigated. In Results 2.1 (p. 29-31), we saw that the species composition of the NVV polder would be considered ‘average’, ranking second for ditches, third for banks and second when combined. If the difference in species is very small, the restoration potential will be small as well. Whereas if the difference is big, and the species in the seed and propagule banks are said to occur in healthy peat-meadow systems, the restoration potential is also big. Except if the number of species or the number of frequent occurring species is very small.

#### Germination results

- Which substrate type contains most species?
- How many samples do you need to take before you reach the estimated diversity?

The sprouted seedlings that survived the eight-week growing period were identified to species level on 10-06 (end of the test period). However, five different seedlings could only be identified to genus level

and some others were still showing their first leaves. Twenty different species were found in total. The species that could only be identified to genus level were:

- *Alopecurus sp.*
- *Poaceae sp.*
- *Lysimachia sp.*
- *Lythrum sp.*
- *Taraxacum sp.*

To see the complete list of the species, see Appendix 6.

Species accumulation curves and estimates for the total species diversity were made per substrate type. It must be noted that the species numbers in the diagrams of figure 2.10 exclude the unknown dicots, but include the seedlings that could be identified to genus level. Figure 2.10 shows these species accumulation curves per substrate type. The pots with dredge contained 22 different seedlings, of which 18 could be identified to species level, and 4 to genus level. For the substrate type 'dredge', a total of 25 species was predicted.

Soil 0-5 cm contained 19 different seedlings, of which 16 could be identified to species level, and 3 to genus level. Using the same methods as for the dredge substrate, a total of 30 species was predicted.

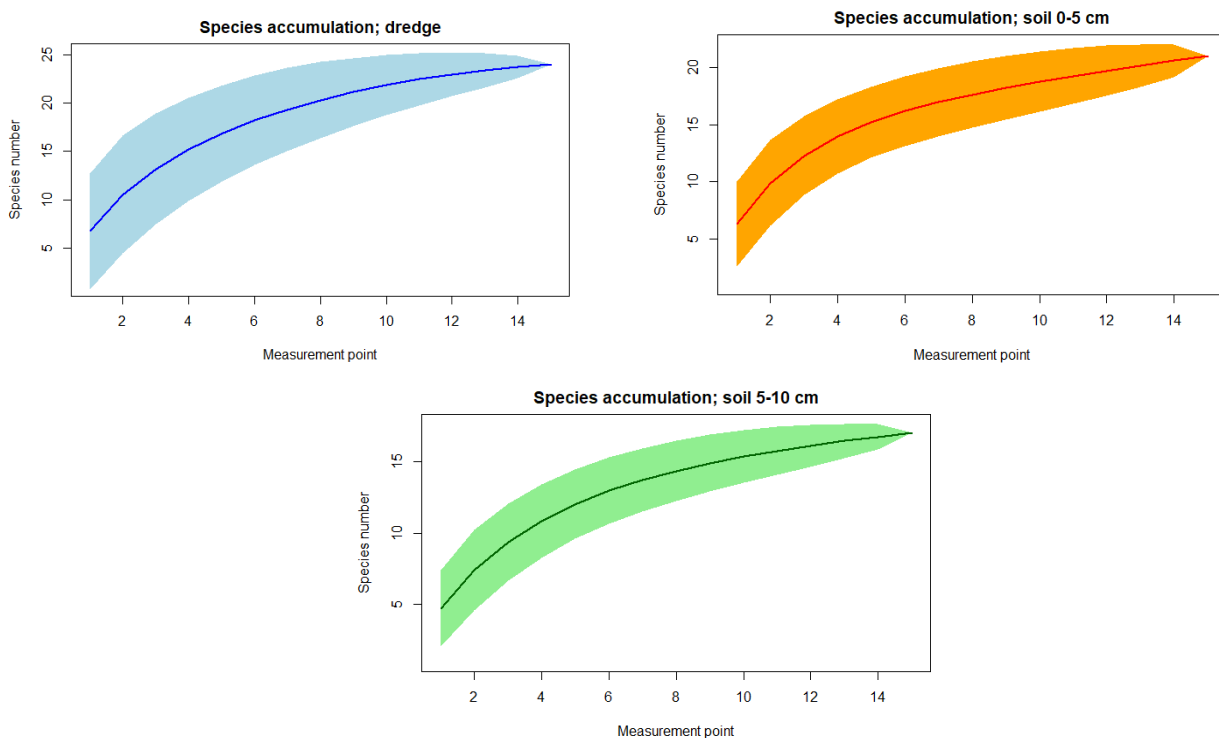


Figure 2.10: Diagrams showing the accumulation of the number of species per type of substrate: dredge, soil 0-5 cm and soil 5-10 cm. The x-axis supports the number of measurement points, and the y-axis the number of species found. The

Soil 5-10 cm contained 15 different seedlings, of which 13 could be identified to species level, and 2 to genus level. A total of 22 species was predicted.

Since the total number of species found in all substrates was lower than the estimated species richness, a sampling effort of 15 measurement points is not enough. However, as can be seen in figure 2.4, the curves are clearly approaching their asymptote, their slopes on measurement point 15 being 0.26 for 'dredge', 0.40 for 'soil 0-5 cm', and 0.26 for 'soil 5-10 cm'. Both substrate types 'dredge' and 'soil 5-10 cm' reached quite a flat slope, but for 'soil 0-5 cm', this slope was almost twice as big. Therefore, for the top 5 cm of soil, more measurement points are needed for a more accurate species diversity prediction.

The offset between the 'perfect model' and the actual curve through the points is very small, and therefore we estimate our identification mistakes (if any) negligible.

### 3.5. Species Composition

#### Differences Between the Substrates

Species occurring in the dredge but not in soil are *Lysimachia sp.*, *Lythrum sp.*, *Rumex acetosa* and *Senecio vulgaris*. Species occurring in the soil but not in the dredge are *Alopecurus sp.* and *Glechoma hederacea*.

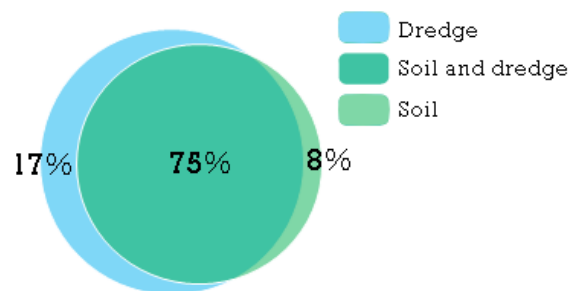


Figure 2.11: Venn diagram of the distribution of species over the substrates 'dredge' and 'soil' (soil 0-5 cm and 5-10 cm measurements taken together). The percentages are calculated through division with the number of species found in the soil and dredge together.

Figure 2.11 indicates that 75% ( $= (18/24) * 100\%$ ) of the species were found in both the soil and dredge, 17% ( $= (4/24) * 100\%$ ) of the species were only found in the dredge and 8% ( $= (2/24) * 100\%$ ) of the species were only found in the soil.

## Frequencies of Occurrence per Species for the Seed- and Propagule Banks

The number of species does not tell how often these species occur. Whether they are prominent, or can only be encountered once or twice. And in which layer or substrate the seeds of certain species are.

Species ratios per substrate type

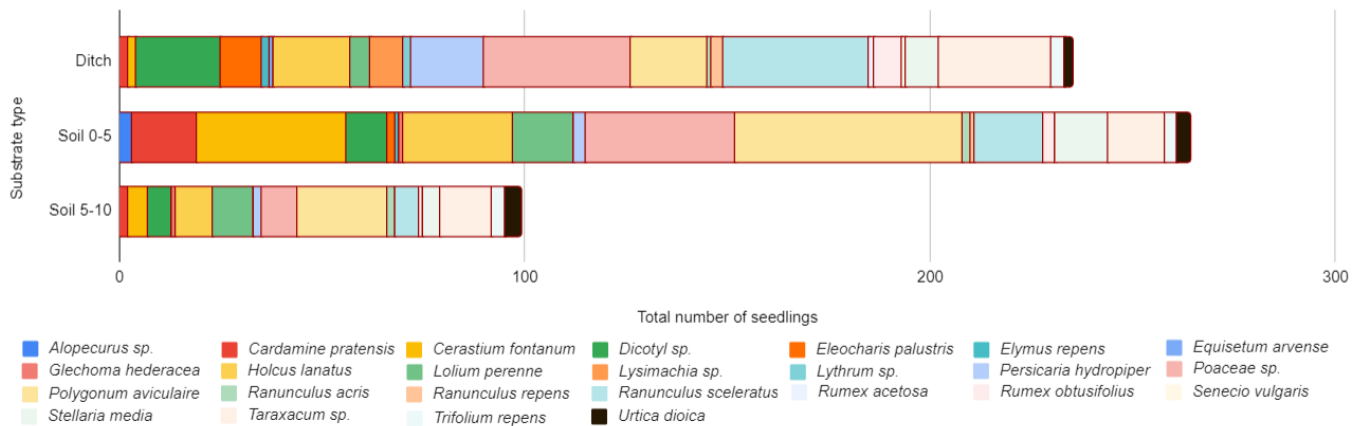


Figure 2.13: The diagram shows the abundance per plant species in each type of substrate.

As can be seen in figure 2.13, *Holcus lanatus*, *Taraxacum sp.* and *Polygonum aviculare* are in both 'Ditch' and 'Soil 5-10 cm' the three most prominent species. In 'Soil 0-5 cm', *Holcus lanatus* is substituted by *Lolium perenne*.

*Cardamine pratensis* and *Cerastium fontanum* are much more abundant in 'Soil 0-5 cm' than in the other two substrates. *Persicaria hydropiper* and *Ranunculus sceleratus* are both more abundant in the 'ditch' pots. *Eleocharis palustris* is seen more often in the ditch pots than in 'Soil 0-5 cm', and is not seen in 'Soil 5-10 cm'.

Some plant species have very low frequencies in the sample data. Such a plant species is labelled 'rare', when it is encountered 1 to 3 times in total. The following observations cover the rare plants:

- *Ranunculus repens* is seen in the 'ditch' and 'soil 0-5 cm', but not in 'soil 5-10'
- *Rumex acetosa*, *Senecio vulgaris* and *Equisetum arvense* are only seen in the ditch pots
- *Elymus repens* is only seen in the 'ditch' and 'soil 0-5'
- *Glechoma hederacea* is only seen in the 'soil 0-5 cm' and 'soil 5-10 cm' pots
- *Ranunculus acris* is seen in all three of the substrate types

*Alopecurus sp.* is only seen in 'Soil 0-5 cm', and *Lysimachia sp.* and *Lythrum sp.* are only seen in the ditch pots.

After comparing the species to the grading system (p. 29), it was evident that slightly more than half of the species belonged to the group 'Bad', and the vast majority of the other half belonged to the group

‘Mediocre’. The three most prominent species, *L. perenne*, *H. lanatus*, and *P. aviculare*, are rated as ‘Bad’.

### Difference Between the Plant Relevées and Seed- and Propagule Banks

The plant relevées (September 2020 and May 2021) contained 58 species, which is significantly more species than the seed- and propagule banks, which contained 24 species. However, comparing them reveals that their species compositions are mostly the same (see figure 2.11). The plant species that were not represented in the plant relevées are *Senecio vulgaris*, *Rumex obtusifolius*, *Equisetum arvense*, *Elymus repens*, *Taraxacum sp.*, and *Lysimachia sp.* The seed- and propagule bank experiment also introduced *Lythrum sp.*, but *Lythrum salicaria* was found in the plant relevées. To conclude, 4 new plant species were found in the seed- and propagule banks. This number could potentially rise by 3 species; the seedlings which could only be determined to genus level. Of these 7 species that were not found in the covering vegetation of the ditch banks and ditches, 6, except *Equisetum arvense*, were found in the plant relevées of the fields.

Figure 2.12 states that 10,8% ( $= (7/65) * 100\%$ ) of the species can be found in the seed- and propagule banks, 27,7% ( $= (18/65) * 100\%$ ) of the species can be found in both samples, and 61,5% ( $= (40/65) * 100\%$ ) in the plant relevées.

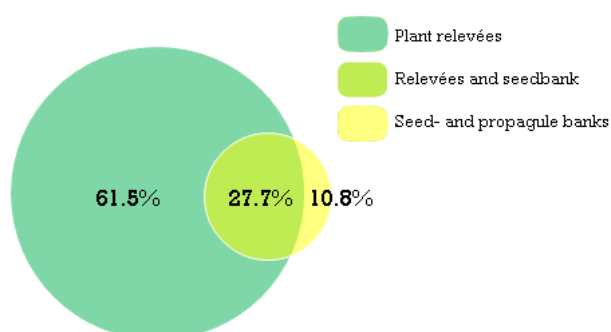


Figure 2.12: Venn diagram of the distribution of species found in the plant relevées (September '20, and May '21) and the seed- and propagule bank experiment. The percentages are calculated through division with the number of species found in the plant relevées and seed- and propagule banks together.



## Discussion and Conclusion

This project was aimed at filling the knowledge gap around ecological restoration of ditches and ditch banks in peat meadow areas in South Holland. We chose to do research on two subjects, management and seed and propagule banks, using the area around Leiden as a research site. For the first subject we analysed which of four managements would work best as a baseline for bringing back peat meadow ecosystems and species in the Northern part of the Vrouwe Vennepolder, and comparing this with profitable agriculture. Out of these four managements, the Southern part of the Vrouwe Vennepolder's and Boterhuispolder's managements were found best for a high species richness, but the Lakerpolder's management was found best for a peat meadow species composition. Rare ecosystems often have a low species richness but are rich in rare species (pers. comm. Krijn Trimbos, July 2021). Therefore, the goal of restoration has to be clear when choosing the management. For the second subject we did research on whether the existing seed and propagule banks could be used to improve the occurrence of peat meadow species in the ditches and on ditch banks of the Northern part of the Vrouwe Vennepolder. From our field experiments we could conclude that the existing seed and propagule banks contain enough seeds, but do not have the species composition needed to increase the occurrence of typical peat meadow species on the ditch banks and in the ditches of the Northern part of the Vrouwe Vennepolder. However, the seed bank experiment only approximated the actual size and composition.

The experimental setup to determine the size and diversity of the seed and propagule bank was located in the field. As such, there were a number of practical limitations. Firstly, the negative influence of animals and the weather was reduced as much as possible by installing fences and nets and digging the slots from east to west. Due to time restrictions, a large-scale experiment was not achievable because the identification of all seedlings based on morphological characters would have taken too long and molecular identification would have been too expensive. Moreover, because the eight-week growing period was barely enough to let most seeds in the substrates 'Soil 0-5 cm' and 'Dredge' germinate (p. 34), identification of most seedlings based on morphological characters was sometimes difficult. And identification mistakes can lead to a wrong estimate of the total number of seedlings. Luckily, these potential mistakes are compensated by statistics because in reality you can only approximate the perfect model. Comparing the found species accumulation curve to the 'perfect' model (p. 21), we estimated the influence of identification mistakes sufficiently small to be neglected. Moreover, eight weeks is a long enough growing period, because seedling death (visual observation, not recorded) then starts to outweigh seed germination. So, despite these limitations, we have made a useful size and composition determination of the seed and propagule bank.

During this thesis we also researched the number of seeds and plant species in the soil and dredge, which, in our case, represents the realised seed and propagule bank, being the number of germinated seeds and associated species that would have been found under the natural conditions of the field (Panufnik-Mędrzycka, D. et al, 2001). Due to favourable weather conditions during the growing period of our seedlings, many seeds germinated (p 39). However, to find the actual size and diversity of the seed and propagule banks (Panufnik-Mędrzycka, D. et al, 2001), optimal ideal growing germinating circumstances are needed which can be only be achieved in a lab or greenhouse. The missed fraction could potentially contain some peat meadow target species (p 11). However, as the seed and propagule bank composition is dominated by non-target species rated as ‘Bad’ by our grading system (discussed in the next paragraph) and the land has a long history of intensive use, the chance of finding a target species is estimated to be very small, which was also concluded in previous studies (Bossuyt & Hermy, 2003). And as such, we conclude that the results of our analysis are a good indication for the seed and propagule bank composition.

A proper conclusion on the restoration potential of the seed and propagule bank should take the frequencies of occurrence per species into account. The Venn diagrams, on which our conclusions are based, lack a considerable part of this information. We chose not to dig too deep into species frequencies, due to our observations during the plant relevés in May. In most locations *Holcus lanatus* and *Lolium perenne* were found to be the dominant species, often covering about 80% of the total surface of the relevée plot (results not included in the thesis). This was in line with Fleur van Duin’s (2021) findings. van Duin did her research on ‘Species Rich Hay Meadows’ in the same time period and polder, and labeled the ecosystem states dominated by these plant species as the lowest grade, categorised as ‘worst’. Her analysis showed that the *L. perenne* association was the predominant vegetation type. The same observations were made when analysing the September 2020 dataset and even in the seed and propagule bank, *L. perenne* and *H. lanatus* were among the top three dominant species (p. 39). Both plants are classified as indicators for an ‘unhealthy’ peat meadow area (p. 29). As such, when the frequencies of occurrence per species is taken into account, the conclusion about the restoration potential of the seed and propagule banks, which is in line with the findings of M. Blomqvist et al. (2003), stays the same: there is hardly any restoration potential. This implies that, if the species richness of these polders is to be enhanced, more rigorous measures may need to be taken, such as sowing with hay from species-rich grasslands, top soil removal or creating ditch banks with tiers.

Management has an influence on many biotic and abiotic factors like species richness, composition and nutrient richness. Where possible, other outside influences on the ecosystem, like air quality, weather and soil type, were eliminated from the equation. Depending on which factor is used for determining the best management, different suggestions for optimal management come forward. This was visible in our analysis. Species richness determines the management of the SVV and Boterhuispolder as better

than that of Lakerpolder and NVV (Table 2.2). Species composition puts the management of Lakerpolder on top and that of SVV as lowest (Table 2.3). This seeming discrepancy is explained by the fact that the Lakerpolder is a nature reserve, which often has a low species richness but a high number of target or other rare species. A possible drawback is that, due to the high water level, this land cannot be used for a suite of different types of profitable agriculture (e.g. arable farming, orchards). In contrast, the NVV is species poor due to a history of intensive farming and even though the analysis suggests that they are comparable, this is not the case. Meanwhile, the SVV has a high species richness. Unfortunately, not the desired target species. However, there are many stages between peat and meadow ecosystems. So, choosing the right management depends on your goals. Our goal for the NVV is to bring back typical peat meadow species and therefore a good solution is to start with the Lakerpolder's management, but with a somewhat lower water table under the fields.

In short, we want to combine bringing back a peat-meadow ecosystem on the ditch banks and in the water with profitable agriculture in the field. The first step is to find the right management for bringing back the peat-meadow ecosystem. Our analysis above shows that the management of both the SVV and Boterhuispolder can be used to increase species diversity and that the management of the Lakerpolder is best to sustain a target species composition. The management of the SVV is well suited to be combined with profitable agriculture. The drawback of the method is that it will not increase the number of peat meadow species. In contrast, the management of the Lakerpolder is profitable to peat meadow species but cannot be combined with profitable agriculture. Therefore, in order to reach our goal in the NVV, both managements have to be combined.

One possible restoration method to bring back peat meadow species in our ecosystem uses existing seed and propagule banks. However, our project shows that even though there are enough seeds in the soil and dredge, the difference with the current NVV vegetation pool, which has no peat meadow composition, is very small. Thus, it was concluded that the existing seed and propagule banks cannot be used for peat-meadow restoration. However, it should be noted that this conclusion cannot be generalised to other polders, as seed bank restoration capacity is influenced by abiotic factors, past use and seed deterioration with time. On the other hand, our conclusions on management can be generalised to be used for other peat meadow polders. However, the investigated managements have little influence on species composition and richness, and as such additional restoration methods are needed unless the ecosystem is not yet completely degraded.

In order to fill more existing knowledge gaps, follow-up experiments can be done. One interesting project would be to determine the influence of water and soil quality on the growth of peat-meadow species in South-Holland peat meadows. For example, the Lakerpolder, is surrounded by the rather eutrophic water of the Kaag but still contains rare species and ecosystems. A nice follow-up experiment in the NVV, or elsewhere, could explore the seed banks through top soil removal in a small area on the

ditch bank itself. Seed germination would be in situ which might possibly improve seedling growth or otherwise influence the outcome of the experiment. Another option is to explore the seed bank composition deeper in the soil of the NVV to see whether deeper layers, in contrast to the upper layers, do contain seeds from target peat meadow species. However, for future research on seedbanks, we recommend putting the pots with dredge with their bottoms in a layer of water, which keeps the dredge from compacting and makes the germination circumstances more realistic.

# Appendix

## Appendix 1: Original Plant Relevée Protocol and Fill-in Sheet

### Monitoring Plant Diversity

#### Goals and Objectives

Register the plant diversity in 1 m<sup>2</sup>, without damaging the plants.

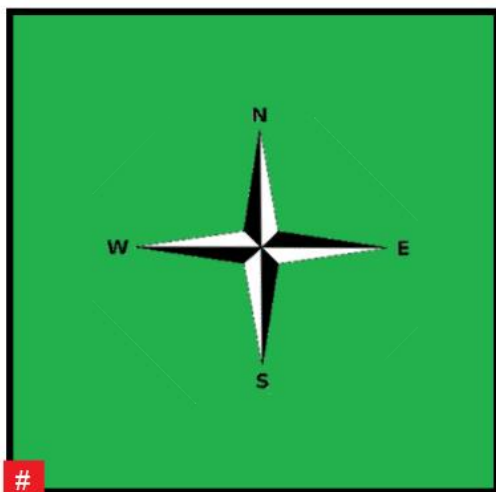
#### Equipment

- 4 pegs with 1 meter x 1 meter cord
- 4 pegs with 2 meter x 0.5 meter cord
- Water fauna net
- Magnifying glass
- Camera
- Ruler
- A4 paper
- Reference book (If you use an app to identify, always check with literature!)

#### Methodology

Step 1: Find the plot.

Step 2: Mark the plot with the pegs and cord as shown below. Make sure all the plants starting inside the plot are entirely inside the plot and vice versa for plants starting outside of the plot.



^Above: Plot **parallel** to the bank of a ditch

<Left: Plot in open field, pointed to north

# = Plot marker

Step 3: Take an overview photo of the plot and determine the total plant coverage in percentage.

Step 4: Identify and optionally photograph all plant species inside the plot and determine their coverage (10 cm x 10 cm is 1%). The minimum value for plant coverage is 1%. If in doubt make sure to take a picture and try to collect the same plant species outside of the plot to use for identification.

Fold the collected plant into the A4 paper.

For mosses, the coverage value can also be lower than 1% and is divided into categories:

0.0%-0.3%: A single strand of moss

0.3%-0.7%: A handful of moss

0.7%-1.0%: Nearly 10cm x 10cm of moss

Above 1%, estimate the coverage the same way as you would with plants.

Step 5: Determine the maximum height of the plot and the amount and average height of plant layers inside the plot.

- mosses: If different mosses are present in the area, take a sample and give it a name. Keep it in a folded A4 paper.
- herb layer
  - ground covering plants
  - lower herb layer
  - upper herb layer
- shrub layer
- tree layer

Step 6: Determine the amount of litter (thickness of the layer), by measuring the height using a ruler.

Step 7: In case of the water plots, take the aquatic plants out of the water with the net and identify them. First determine the emergent and floating aquatic plants and their coverage, then identify the submerged plants.

Step 8: Determine the relief of your plot e.g. slope, microrelief. Use the stick of the net to determine the slope of the banks.

Step 9: Make a picture of the surroundings and determine the environmental influences on the composition of plant species e.g. wind.

### Researchers

- 1 person marks all the species and their coverage down.
- 1 person identifies the species, while the others find new ones.

### Schedule

20 min/plot.

### Paperwork

- |                                    |                      |
|------------------------------------|----------------------|
| ● Total coverage                   | ● Height of the plot |
| ● Coverage per plant species       | ● Amount of layers   |
| ● Sum of all species coverage      | ● Amount of litter   |
| ● Latin names of the plant species |                      |

Original fill-in sheet for the plant relevées from September 2020

Area (Length*With) [m^2]		Place	
Total coverage [%]		Picture number	
Sum coverage [%]		Date	
Total height of plot [cm]		Group number	
Average height litter [cm]		Notes	
Average height ground covering plants [cm]		Coordinates	
Average height lower herb layer [cm]			
Average height upper herb layer [cm]			
Average height shrub layer [m]			
Average height tree layer [m]			
Relief/angle			
Coverage emergent vegetation [%]			
Coverage floating vegetation [%]			
Latin name	(Common name)	Coverage %	Notes

## Appendix 2: Translation Plant Common Names English to Dutch, and Other Names

<b>English</b>	<b>Dutch</b>	<b>Latin</b>
Ash	Es	<i>Fraxinus excelsior</i>
Bird vetch	Vogelwikke	<i>Vicia cracca</i>
Bittersweet	Bitterzoet	<i>Solanum dulcamara</i>
Blunt-flowered rush	Paddenrus	<i>Juncus subnodulosus</i>
Broadleaf cattail	Grote lisdodde	<i>Typha latifolia</i>
Broadleaf plantain	Grote weegbree	<i>Plantago major</i>
Broad-leaved willowherb	Bergbasterdwederik	<i>Epilobium montanum</i>
Cattail	Lisdodde	<i>Typha sp.</i>
Celery-leaved buttercup	Blaartrekkende boterbloem	<i>Ranunculus sceleratus</i>
Chickweed	Vogelmuur	<i>Stellaria media</i>
Commen skullcap	Blauw glidkruid	<i>Scutellaria galericulata</i>
Common comfrey	Gewone smeewortel	<i>Symphytum officinale</i>
common duckmeat	Veelwortelig kroos	<i>Spirodela polyrrhiza</i>
Common duckweed	Klein kroos	<i>Lemna minor</i>
Common fleabane	Heelblaadjes	<i>Pulicaria dysenterica</i>
Common knotgrass	Gewoon varkensgras	<i>Polygonum aviculare</i>
Common marsh bedstraw	Moeraswalstro	<i>Galium palustre</i>
Common Nettle	Grote brandnetel	<i>Urtica dioica</i>
Common rush	Pitrus	<i>Juncus effusus</i>
Common self-heal	Gewone brunel	<i>Prunella vulgaris</i>
Common sorrel	Veldzuring	<i>Rumex acetosa</i>
Common spike-rush	Gewone waterbies	<i>Eleocharis palustris</i>
Cow parsley	Fluitenkruid	<i>Anthriscus sylvestris</i>
Creeping bentgrass	Fioringras	<i>Agrostis stolonifera</i>
Creeping buttercup	Kruipende boterbloem	<i>Ranunculus repens</i>
Creeping thistle	Akkerdistel	<i>Cirsium arvense</i>
Cuckoo flower	Pinksterbloem	<i>Cardamine pratensis</i>
Curly dock	Krulzuring	<i>Rumex crispus</i>
Dandelion	Paardenbloem	<i>Taraxacum officinale</i>
Dove's-foot Crane's-bill	Zachte ooievaarsbek	<i>Geranium molle</i>
European water-plantain	Grote waterweegbree	<i>Alisma plantago-aquatica</i>
Field milk thistle	Akkermelkdistel	<i>Sonchus arvensis</i>
Fineleaf water dropwort	Watertorkruid	<i>Oenanthe aquatica</i>
Frogbit	Kikkerbeet	<i>Hydrocharis morsus-ranae</i>



Goat willow	Boswilg	<i>Salix caprea</i>
Great manna grass	Liesgras	<i>Glyceria maxima</i>
Great water dock	Waterzuring	<i>Rumex hydrolapathum</i>
Great willowherb	Harig wilgenroosje	<i>Epilobium hirsutum</i>
Ground-ivy	Hondsdrif	<i>Glechoma hederacea</i>
Gypsywort	Wolfspoot	<i>Lycopus europaeus</i>
Hairlike pondweed	Haarfonteinkruid	<i>Potamogeton trichoides</i>
Hairy bittercress	Kleine veldkers	<i>Cardamine hirsuta</i>
Hedge bindweed	Haagwinde	<i>Calystegia sepium</i>
Hemp-agrimony	Koninginnenkruid	<i>Eupatorium cannabinum</i>
Hitchhikers	Kleefkruid	<i>Galium aparine</i>
Hogweed	Gewone berenklaauw	<i>Heracleum sphondylium</i>
Hornwort	Grof hoornblad	<i>Ceratophyllum demersum</i>
Lady's thumb	Perzikkruid	<i>Persicaria maculosa</i>
Least duckweed	Dwergkroos	<i>Lemna minuta</i>
Lesser marshwort	Ondergedoken moerasscherm	<i>Apium inundatum</i>
Lesser spearwort	Egelboterbloem	<i>Ranunculus flammula</i>
Lesser water-parsnip	Kleine watereppe	<i>Berula erecta</i>
Longroot smartweed	Veenwortel	<i>Persicaria amphibia</i>
Marsh dock	Moeraszuring	<i>Rumex palustris</i>
Marsh woundwort	Moerasandoorn	<i>Stachys palustris</i>
Marsh-marigold	Gewone dotterbloem	<i>Caltha palustris</i>
Meadow buttercup	Scherpe boterbloem	<i>Ranunculus acris</i>
Meadow foxtail	Grote vossenstaart	<i>Alopecurus pratensis</i>
Meadow soft grass	Gestreepte witbol	<i>Holcus lanatus</i>
meadow vetchling	Veldlathyrus	<i>Lathyrus pratensis</i>
Meadowsweet	Moerasspirea	<i>Filipendula ulmaria</i>
Moneywort	Penningkruid	<i>Lysimachia nummularia</i>
Mouse-ear chickweed	Gewone hoornbloem	<i>Cerastium fontanum-subsp. vulgare</i>
Narrow-leaved-rattle	Grote ratelaar	<i>Rhinanthus angustifolius</i>
Perennial ryegrass	Engels raaigras	<i>Lolium perenne</i>
Pilewort	Gewoon speenkruid	<i>Ficaria verna subsp. verna</i>
Purple loosestrife	Grote kattenstaart	<i>Lythrum salicaria</i>
Purplestem beggarticks	Smal tandzaad	<i>Bidens connata</i>
Ragged-robin	Echte koekoeksbloem	<i>Silene flos-cuculi</i>
Reed canary grass	Rietgras	<i>Phalaris arundinacea</i>
Reed	Riet	<i>Phragmites australis</i>

Ribwort plantain	Smalle weegbree	<i>Plantago lanceolata</i>
Sea clubrush	Heen	<i>Bolboschoenus maritimus</i>
Silverweed	Zilver schoon	<i>Potentilla anserina</i>
Simplestem bur-reed	Grote egelskop	<i>Sparganium erectum</i>
Sneezewort	Wilde bertram	<i>Achillea ptarmica</i>
Star duckweed	Puntkroos	<i>Lemna trisulca</i>
Sticky mouse-ear chickweed	Kluwenhoornbloem	<i>Cerastium glomeratum</i>
Sweet flag	Kalmoes	<i>Acorus calamus</i>
Tall fescue	Rietzwenkgras	<i>Festuca arundinacea</i>
Three-lobed beggarstick	Veeldelig tandzaad	<i>Bidens tripartita</i>
True forget-me-not	Moeras vergeet-mij-nietje	<i>Myosotis palustris</i>
Valerian	Echte valeriaan	<i>Valeriana officinalis</i>
Water blinks	Groot bronkruid	<i>Montia arvensis</i>
Water horsetail	Holpijp	<i>Equisetum fluvatile</i>
Water mint	Watermunt	<i>Mentha aquatica</i>
Water pepper	Waterpeper	<i>Persicaria hydropiper</i>
Watercress	Witte waterkers	<i>Nasturtium officinale</i>
Western waterweed	Smalle waterpest	<i>Elodea nuttallii</i>
White clover	Witte klaver	<i>Trifolium repens</i>
Wild angelica	Gewone engelwortel	<i>Angelica sylvestris</i>
Yellow iris	Gele lis	<i>Iris pseudacorus</i>

## Appendix 3: September 2020 Database; Plant Species per Polder in the Water and on the Banks

### Water

#### Boterhuispolder

<i>Agrostis stolonifera</i>	<i>Glyceria maxima</i>	<i>Plantago lanceolata</i>
<i>Alisma plantago-aquatica</i>	<i>Iris pseudacorus</i>	<i>Ranunculus repens</i>
<i>Bidens tripartita</i>	<i>Lemna minor</i>	<i>Ranunculus sceleratus</i>
<i>Ceratophyllum demersum</i>	<i>Lemna minuta</i>	<i>Rumex acetosa</i>
<i>Cirsium arvense</i>	<i>Lemna trisulca</i>	<i>Rumex crispus</i>
<i>Eleocharis palustris</i>	<i>Lythrum salicaria</i>	<i>Sparganium erectum</i>
<i>Elodea nuttallii</i>	<i>Myosotis scorpioides</i>	<i>Spirodela polyrhiza</i>
<i>Equisetum fluviatile</i>	<i>Nasturtium officinale</i>	<i>Spirogyra</i> sp.
<i>Galium palustre</i>	<i>Oenanthe aquatica</i>	<i>Trifolium repens</i>
<i>Glechoma hederacea</i>	<i>Persicaria maculosa</i>	

#### Lakerpolder

<i>Acorus calamus</i>	<i>Lycopus europaeus</i>	<i>Salix caprea</i>
<i>Bolboschoenus maritimus</i>	<i>Mentha aquatica</i>	<i>Sparganium erectum</i>
<i>Filipendula ulmaria</i>	<i>Persicaria hydropiper</i>	<i>Symphytum officinale</i>
<i>Glyceria maxima</i>	<i>Phalaris arundinacea</i>	<i>Symphytum</i> sp.
<i>Iris pseudacorus</i>	<i>Phragmites australis</i>	<i>Vicia cracca</i>
<i>Lemna minor</i>	<i>Plantago lanceolata</i>	
<i>Lemna minuta</i>	<i>Ranunculus repens</i>	

#### Northern Vrouwe Vennepolder

<i>Agrostis stolonifera</i>	<i>Fraxinus excelsior</i>	<i>Nasturtium officinale</i>
<i>Apium inundatum</i>	<i>Galium palustre</i>	<i>Persicaria amphibia</i>
<i>Berula erecta</i>	<i>Glyceria maxima</i>	<i>Persicaria hydropiper</i>
<i>Bidens connata</i>	<i>Holcus lanatus</i>	<i>Phalaris arundinacea</i>
<i>Bidens tripartita</i>	<i>Hydrocharis morsus-ranae</i>	<i>Phragmites australis</i>
<i>Bolboschoenus maritimus</i>	<i>Juncus subnodulosus</i>	<i>Polygonum aviculare</i>
<i>Cardamine pratensis</i>	<i>Lemna minor</i>	<i>Sparganium erectum</i>
<i>Ceratophyllum demersum</i>	<i>Lemna minuta</i>	<i>Spirodela polyrhiza</i>
<i>Eleocharis palustris</i>	<i>Lolium perenne</i>	<i>Stachys palustris</i>
<i>Epilobium hirsutum</i>	<i>Mentha aquatica</i>	<i>Urtica dioica</i>
<i>Epilobium montanum</i>	<i>Myosotis scorpioides</i>	

#### Southern Vrouwe Vennepolder

<i>Agrostis stolonifera</i>	<i>Alisma plantago-aquatica</i>	<i>Bidens tripartita</i>
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<i>Bolboschoenus maritimus</i>	<i>Hydrocharis morsus-ranae</i>	<i>Ranunculus acris</i>
<i>Carex</i> sp.	<i>Lathyrus pratensis</i>	<i>Ranunculus flammula</i>
<i>Ceratophyllum demersum</i>	<i>Lemna minor</i>	<i>Sonchus arvensis</i>
<i>Eleocharis palustris</i>	<i>Lemna minuta</i>	<i>Sparganium erectum</i>
<i>Elodea nuttallii</i>	<i>Lemna trisulca</i>	<i>Spirodela polyrhiza</i>
<i>Galium mollugo</i> -sp.	<i>Lolium perenne</i>	<i>Spirogyra</i> sp.
<i>Galium palustre</i>	<i>Montia arvensis</i>	<i>Symphytum officinale</i>
<i>Glechoma hederacea</i>	<i>Nasturtium officinale</i>	<i>Symphytum</i> sp.
<i>Glyceria maxima</i>	<i>Oenanthe aquatica</i>	<i>Taraxacum officinale</i>
<i>Heracleum sphondylium</i>	<i>Persicaria amphibia</i>	
<i>Holcus lanatus</i>	<i>Persicaria hydropiper</i>	

## Banks

### Boterhuispolder

<i>Agrostis stolonifera</i>	<i>Hydrocharis morsus-ranae</i>	<i>Persicaria maculosa</i>
<i>Apium inundatum</i>	<i>Juncus subnodulosus</i>	<i>Plantago major</i>
<i>Berula erecta</i>	<i>Lemna minor</i>	<i>Ranunculus acris</i>
<i>Bidens tripartita</i>	<i>Lemna minuta</i>	<i>Ranunculus repens</i>
<i>Cardamine pratensis</i>	<i>Lemna trisulca</i>	<i>Ranunculus sceleratus</i>
<i>Eleocharis palustris</i>	<i>Lolium perenne</i>	<i>Sparganium erectum</i>
<i>Equisetum fluviatile</i>	<i>Lysimachia nummularia</i>	<i>Spirodela polyrhiza</i>
<i>Galium palustre</i>	<i>Mentha aquatica</i>	<i>Stellaria media</i>
<i>Glechoma hederacea</i>	<i>Montia arvensis</i>	<i>Taraxacum officinale</i>
<i>Glyceria maxima</i>	<i>Nasturtium officinale</i>	<i>Trifolium repens</i>
<i>Holcus lanatus</i>	<i>Persicaria hydropiper</i>	<i>Urtica dioica</i>

### Lakerpolder

<i>Acorus calamus</i>	<i>Iris pseudacorus</i>	<i>Plantago lanceolata</i>
<i>Agrostis stolonifera</i>	<i>Juncus effusus</i>	<i>Prunella</i> sp.
<i>Angelica sylvestris</i>	<i>Lycopus europaeus</i>	<i>Ranunculus acris</i>
<i>Anthriscus sylvestris</i>	<i>Lysimachia nummularia</i>	<i>Rumex acetosa</i>
<i>Bolboschoenus maritimus</i>	<i>Mentha aquatica</i>	<i>Rumex hydrolapathum</i>
<i>Festuca arundinacea</i>	<i>Mentha aquatica</i>	<i>Sparganium erectum</i>
<i>Glechoma hederacea</i>	<i>Persicaria maculosa</i>	<i>Symphytum</i> sp.
<i>Glyceria maxima</i>	<i>Phragmites australis</i>	<i>Urtica dioica</i>

### Northern Vrouwe Vennepolder

<i>Agrostis stolonifera</i>	<i>Berula erecta</i>	<i>Bolboschoenus maritimus</i>
<i>Alisma plantago-aquatica</i>	<i>Bidens tripartita</i>	<i>Carex</i> sp.

*Cerastium fontanum*-subsp.  
*vulgare*

*Ceratophyllum demersum*

*Eleocharis palustris*

*Epilobium hirsutum*

*Epilobium montanum*

*Galium palustre*

*Geranium molle*

*Glechoma hederacea*

*Glyceria maxima*

*Holcus lanatus*

*Lemna minor*

*Lemna minuta*

### **Southern Vrouwe Vennepolder**

*Agrostis* sp.

*Agrostis stolonifera*

*Bolboschoenus maritimus*

*Cardamine pratensis*

*Cerastium fontanum*

*Ceratophyllum demersum*

*Epilobium hirsutum*

*Galium mollugo*-sp.

*Galium palustre*

*Geranium molle*

*Glechoma hederacea*

*Glyceria maxima*

*Heracleum sphondylium*

*Lemna trisulca*

*Lolium perenne*

*Lycopus europaeus*

*Lythrum salicaria*

*Mentha aquatica*

*Nasturtium officinale*

*Persicaria amphibia*

*Persicaria hydropiper*

*Persicaria maculosa*

*Phalaris arundinacea*

*Polygonum aviculare*

*Potamogeton trichoides*

*Ranunculus acris*

*Holcus lanatus*

*Holcus lanatus*

*Iris pseudacorus*

*Juncus subnodulosus*

*Lathyrus pratensis*

*Lemna minor*

*Lemna trisulca*

*Lolium perenne*

*Lysimachia nummularia*

*Lythrum salicaria*

*Mentha aquatica*

*Myosotis scorpioides*

*Nasturtium officinale*

*Ranunculus repens*

*Rumex acetosa*

*Rumex crispus*

*Rumex hydrolapathum*

*Rumex palustris*

*Scutellaria galericulata*

*Sparganium erectum*

*Spirodela polyrhiza*

*Stachys palustris*

*Trifolium repens*

*Typha latifolia*

*Urtica dioica*

*Oenanthe aquatica*

*Persicaria amphibia*

*Plantago lanceolata*

*Ranunculus flammula*

*Ranunculus repens*

*Rumex acetosa*

*Sonchus arvensis*

*Sparganium erectum*

*Spirogyra* sp.

*Symphytum* sp.

*Trifolium repens*

Appendix 4: May 2021 Database; Plant Species on the Ditch Banks and in the  
Ditches of the NVV Polder

**Water**

*Holcus lanatus*  
*Mentha aquatica*  
*Bolboschoenus maritimus*  
*Ranunculus sceleratus*  
*Spirodela polyrhiza*  
*Typha latifolia*  
*Glyceria maxima*  
*Eleocharis palustris*  
*Rumex hydrolapathum*  
*Urtica dioica*  
*Iris pseudacorus*  
*Rumex acetosa*  
*Persicaria amphibia*  
*Lycopus europaeus*

**Banks**

*Alopecurus pratensis*  
*Bolboschoenus maritimus*  
*Cardamine hirsuta*  
*Cardamine pratensis*  
*Carex hirta*  
*Cerastium fontanum*  
*Cerastium glomeratum*  
*Eleocharis palustris*  
*Epilobium hirsutum*  
*Ficaria verna verna*  
*Fraxinus excelsior*  
*Galium palustre*  
*Glechoma hederacea*  
*Glyceria maxima*  
*Holcus lanatus*  
*Lolium perenne*  
*Lycopus europaeus*  
*Mentha aquatica*  
*Persicaria amphibia*  
*Persicaria maculosa*  
*Potentilla anserina*  
*Ranunculus repens*  
*Ranunculus sceleratus*  
*Rumex acetosa*  
*Rumex hydrolapathum*  
*Stellaria media*  
*Trifolium repens*  
*Typha latifolia*  
*Urtica dioica*

**Plants from the Oeverplanten Project Database; 'Guidelines for Grading'**

These are the plants on which the consensus and the guidelines for grading on page 29 are based.

**D) Nutrient rich soil and careless ditch bank management:**

1. *Urtica dioica*: Soil contains humus. Nutrient rich-very nutrient rich. Namely nitrogen rich.
2. *Calystegia sepium*: Nutrient rich. Sometimes in a brackish environment.
3. *Epilobium hirsutum*: Nutrient rich-very nutrient rich. Weakly acidic-calcareous. Tolerates a little bit of salt
4. *Solanum dulcamara*: Moderately nutrient rich-nutrient rich. Sometimes nitrogen rich, often calcareous.
5. *Symphytum officinale*: Nutrient rich. Nitrogen rich.
6. *Glechoma hederacea*: Soil contains humus. Moderately nutrient poor - very nutrient rich.

**E) Careful ditch bank management and conditions with which a flowery vegetation can develop:**

1. *Stachys palustris*: Soil contains humus. Nutrient rich-very nutrient rich. Often weakly acidic.
2. *Lythrum salicaria*: Moderately nutrient rich - nutrient rich. Weakly acidic-calcareous.
3. *Valeriana officinalis*: Moderately nutrient rich - nutrient rich. Weakly acidic- calcareous
4. *Lycopus europaeus*: Moderately nutrient rich - nutrient rich.
5. *Eupatorium cannabinum*: Moderately nutrient rich - nutrient rich. Weakly acidic - calcareous.
6. *Mentha aquatica*: Soil contains humus - peat-like soil. Moderately nutrient rich - nutrient rich. Weakly acidic- calcareous
7. *Myosotis palustris*: nothing on Verspreidingsatlas
8. *Iris pseudacorus*: Moderately nutrient rich - nutrient rich. Nitrogen rich.

**F) Very careful ditch bank management, less nutrient rich soil and a flowery vegetation:**

1. *Pulicaria dysenterica*: Soil contains humus. Moderately nutrient rich. Moderately nitrogen rich. Calcareous
2. *Caltha palustris*: nothing on Verspreidingsatlas
3. *Filipendula ulmaria*: Slightly - not fertilized lands, soil contains humus. Moderately nutrient rich - nutrient rich. Weakly acidic- calcareous. Disappears when phosphate increases.
4. *Prunella vulgaris*: Moderately nutrient rich - nutrient rich. Weakly acidic-calcareous
5. *Rhinanthus angustifolius*: Slightly - not fertilized lands. Moderately nutrient poor - moderately nutrient rich. Weakly acidic soil.
6. *Achillea ptarmica*: Often slightly fertilized lands. Moderately nutrient rich. Nitrogen poor. Mostly moderately acidic soil.
7. *Ranunculus flammula*: Nutrient poor-moderately nutrient rich. Alkaline and nitrogen poor. Acidic-neutral pH soil, often rather lime-poor.
8. *Silene flos-cuculi*: Prefers peat-like soil. Moderately nutrient rich.

## Plants From the September 2020 Database; 'Reasons for Grading'

Ecological information from the plants which was compared to the consensus on page ???. When a description could not be directly placed in one of the three categories, the abiotic state that greatly determined the decision was underlined.

*Holcus lanatus* and *Lolium perenne* both get a 1, because these are sown.

1. *Apium inundatum* (<https://www.verspreidingsatlas.nl/0077>)  
**10 Soil:** Moderately nutrient poor. Weakly acidic. **Water:** Moderately nutrient poor - moderately nutrient rich. Neutral pH.
2. *Acorus calamus* (<https://www.verspreidingsatlas.nl/0007>)  
**1** Nutrient rich. Weakly acidic - calcareous soil.
3. *Agrostis stolonifera* (<https://www.verspreidingsatlas.nl/0018>)  
**1** Disturbed environment. **Soil:** Moderately nutrient rich. **Water:** Very nutrient rich.
4. *Alisma plantago-aquatica* (<https://www.verspreidingsatlas.nl/0028>)  
**5 Water:** Fairly nutrient poor - nutrient rich. Weakly acidic- calcareous.
5. *Alopecurus pratensis* (<https://www.verspreidingsatlas.nl/0042>)  
**1** Nutrient rich and often fertilized soil.
6. *Angelica sylvestris* (<https://www.verspreidingsatlas.nl/0060>)  
**10** Moderately nutrient rich - nutrient rich. Often weak acidic soil.
7. *Anthriscus sylvestris* (<https://www.verspreidingsatlas.nl/0070>)  
**1** Moderately nutrient rich - very nutrient rich. Nitrogen rich soil.
8. *Berula erecta* (<https://www.verspreidingsatlas.nl/1215>)  
**5** Moderately nutrient rich - nutrient rich. Weakly acidic - calcareous soil.
9. *Bidens connata* (<https://www.verspreidingsatlas.nl/0142>)  
**10** Moderately nutrient poor - nutrient rich. Nitrogen rich. In particular weakly acidic soil but also slightly calcareous soil.
10. *Bidens tripartita* (<https://www.verspreidingsatlas.nl/0144>)  
**1** Often nutrient rich, sometimes nutrient poor. Nitrogen rich. Weakly acidic - calcareous soil.
11. *Cardamine hirsuta* (<https://www.verspreidingsatlas.nl/0203>)  
**1** Tilled soil. Moderately nutrient poor - very nutrient rich. Nitrogen rich. Neutral (not strongly acidic) soil.
12. *Cardamine pratensis* (<https://www.verspreidingsatlas.nl/0205>)  
**5** Slightly or not fertilized grasslands. Moderately nutrient rich - nutrient rich. Alkaline - weakly acidic.
13. *Cerastium fontanum* (<https://www.verspreidingsatlas.nl/0296>)  
**1** Humus rich soil, fertilized grasslands. Moderately nutrient rich - nutrient rich. Weakly acidic - calcareous soil.



14. *Cerastium glomeratum* (<https://www.verspreidingsatlas.nl/0546>)  
**1** Tilled soil. Nutrient rich - very nutrient rich. Weakly acidic - calcareous soil.
15. *Ceratophyllum demersum* (<https://www.verspreidingsatlas.nl/0299#>)  
**1** Nutrient rich. Hard water.
16. *Cirsium arvense* (<https://www.verspreidingsatlas.nl/0331>)  
**1** Tilled soil. Nutrient rich. Weakly acidic - calcareous soil.
17. *Eleocharis palustris* (<https://www.verspreidingsatlas.nl/0437>)  
**5** Moderate nutrient rich - nutrient rich. Moderately acidic- calcareous soil
18. *Elodea nutallii* (<https://www.verspreidingsatlas.nl/0442>)  
**1** Nutrient rich. Weakly acidic - calcareous soil. Hard, sometimes contaminated water.
19. *Epilobium montanum* (<https://www.verspreidingsatlas.nl/0454>)  
**5** Humus rich soil. Moderately nutrient rich - nutrient rich. Mostly weakly acidic - sometimes calcareous soil.
20. *Equisetum fluviatile* (<https://www.verspreidingsatlas.nl/0463>)  
**5** Moderately nutrient rich -nutrient rich. Moderately nitrogen - nitrogen rich. Phosphate-poor.
21. *Festuca arundinacea* (<https://www.verspreidingsatlas.nl/0514>)  
**1** Disturbed soil. Nutrient rich. Mostly alkaline rich. Weakly acidic- calcareous soil.
22. *Ficaria verna subsp. verna* (<https://www.verspreidingsatlas.nl/1047>)  
**5** Moderately nutrient rich - nutrient rich. Nitrogen rich. Weakly acidic to weakly alkaline soil.
23. *Galium palustre* (<https://www.verspreidingsatlas.nl/2376>)  
**1** Moderately nutrient poor - very nutrient rich. Weak acidic - alkaline, calcareous soil.
24. *Geranium molle* (<https://www.verspreidingsatlas.nl/0571>)  
**5** Moderately nutrient rich- nutrient rich. Nitrogen poor. Moderately acidic- calcareous soil.
25. *Glyceria maxima* (<https://www.verspreidingsatlas.nl/0585>)  
**1 Soil:** Nutrient rich - very nutrient rich. Weakly acidic-calcareous. **Water:** Nutrient rich.
26. *Hydrocharis morsus-ranae* (<https://www.verspreidingsatlas.nl/0640>)  
**5** Moderately nutrient rich - nutrient rich. Moderately nitrogen rich. Weakly acidic- alkaline soil.
27. *Juncus effusus* (<https://www.verspreidingsatlas.nl/0680>)  
**5** Disturbed soil. Moderately nutrient rich - nutrient rich. Nitrogen poor - moderately nitrogen rich. Weak acidic- alkaline, calcareous poor - no calcareous soil
28. *Juncus subnodulosus* (<https://www.verspreidingsatlas.nl/0688>)  
**10** Moderately nutrient rich. Nitrogen poor - moderately nitrogen rich. Calcareous poor- calcareous rich soil.
29. *Lathyrus pratensis* (<https://www.verspreidingsatlas.nl/0715>)

- 5** Little to no fertilized soil. Moderately nutrient rich - nutrient rich. Moderately nitrogen rich. Weakly acidic- alkaline soil.
30. *Lemna minor* (<https://www.verspreidingsatlas.nl/0723>)  
**5** Moderately nutrient poor - nutrient rich.
31. *Lemna minuta* (<https://www.verspreidingsatlas.nl/2426>)  
**1** Nutrient rich water.
32. *Lemna trisulca* (<https://www.verspreidingsatlas.nl/0724>)  
**5** Moderately nutrient rich - nutrient rich. Weak acidic - calcareous, neutral - alkaline soil.
33. *Lysimachia nummularia* (<https://www.verspreidingsatlas.nl/0782>)  
**1** Disturbed ecosystem. Moderately nutrient rich - nutrient rich. Not-acidic soil
34. *Montia arvensis* (<https://www.verspreidingsatlas.nl/0835>)  
**10** Moderately nutrient poor - moderately nutrient rich. Low-limescale
35. *Myosotis scorpioides* (<https://www.verspreidingsatlas.nl/1494>)  
**5** Nutrient rich. Along flowing water more nutrient poor.
36. *Nasturtium officinale* (<https://www.verspreidingsatlas.nl/0860>)  
**1** Moderately nutrient rich - nutrient rich. Calcareous soil.
37. *Oenanthe aquatica* (<https://www.verspreidingsatlas.nl/0868>)  
**10** Moderately nutrient rich - nutrient rich. Weakly acidic - calcareous soil. Moderately nutrient rich water. (We hebben m alleen in water gevonden!)
38. *Persicaria amphibia* (<https://www.verspreidingsatlas.nl/0967>)  
**1** Disturbed ecosystem. Nutrient rich water. Mineral rich soil.
39. *Persicaria hydropiper* (<https://www.verspreidingsatlas.nl/0972>)  
**10** Soil contains humus. Nutrient poor. Nitrogen rich. Often lime-poor.
40. *Persicaria maculosa* (<https://www.verspreidingsatlas.nl/0977>)  
**1** Tilled soil. Nutrient rich. Often low-limescale
41. *Phalaris arundinacea* (<https://www.verspreidingsatlas.nl/0930>)  
**1** Nutrient rich - very nutrient rich. Weakly acidic-calcareous.
42. *Phragmites australis* (<https://www.verspreidingsatlas.nl/0933>)  
**5** Moderately nutrient rich - nutrient rich. Weakly acidic-calcareous.
43. *Plantago lanceolata* (<https://www.verspreidingsatlas.nl/0946>)  
**5** Fertilized grasslands. Nutrient poor - moderately nutrient rich. Weakly acidic-calcareous.
44. *Plantago major* (<https://www.verspreidingsatlas.nl/2320>)  
**1** Compacted soil. (No information on Verspreidingsatlas)
45. *Polygonum aviculare* (<https://www.verspreidingsatlas.nl/0968>)  
**1** Compacted soil. Nutrient rich.
46. *Potamogeton trichoides* (<https://www.verspreidingsatlas.nl/1003>)

- 5** Moderately nutrient rich - nutrient rich. Nitrogen poor - moderately nitrogen rich, sometimes polluted. Weakly acidic-calcareous.
47. *Potentilla anserina* (<https://www.verspreidingsatlas.nl/1006>)  
**5** Often compacted/disturbed soil. Moderately nutrient rich-nutrient rich. Weakly acidic-calcareous.
48. *Ranunculus acris* (<https://www.verspreidingsatlas.nl/1040>)  
**1** Soil contains humus, fertilized grassland. Nutrient rich - very nutrient rich. Moderately acidic-calcareous.
49. *Ranunculus repens* (<https://www.verspreidingsatlas.nl/1056>)  
**1** Compacted/disturbed soil. Moderately - very nutrient rich. Not too acidic.
50. *Ranunculus sceleratus* (<https://www.verspreidingsatlas.nl/1058>)  
**1** Organic, sometimes disturbed soil. Nutrient rich. Nitrogen rich. Often calcareous.
51. *Rumex acetosa* (<https://www.verspreidingsatlas.nl/1093>)  
**1** Fertilized soil. Moderately nutrient rich - nutrient rich. Often weakly acidic.
52. *Rumex crispus* (<https://www.verspreidingsatlas.nl/1098>)  
**1** Disturbed soil. Nutrient rich. Mineral rich. Not - weakly acidic soil.
53. *Rumex hydrolapathum* (<https://www.verspreidingsatlas.nl/1099>)  
**5** Moderately nutrient rich - nutrient rich.
54. *Rumex palustris* (<https://www.verspreidingsatlas.nl/1102>)  
**1** Nutrient rich. Nitrogen rich.
55. *Scutellaria galericulata* (<https://www.verspreidingsatlas.nl/1173>)  
**10** Soil contains humus. Moderately nutrient rich. Somewhat calcareous-rather acidic soil
56. *Sonchus arvensis* (<https://www.verspreidingsatlas.nl/2324>)  
**1** Heavily fertilized, cultivated soil. Nutrient rich - very nutrient rich. Weakly acidic-calcareous.
57. *Sparganium erectum* (<https://www.verspreidingsatlas.nl/1229>)  
**1** Moderately nutrient rich - very nutrient rich. Nitrogen rich. Weakly alkaline, weakly acidic-calcareous.
58. *Spirodela polyrhiza* (<https://www.verspreidingsatlas.nl/1241>)  
**1** Nutrient rich water. Hard water. More or less organic soil
59. *Stellaria media* (<https://www.verspreidingsatlas.nl/1250>)  
**1** Often on fertilized and tilled soil. Nutrient rich. Nitrogen rich.
60. *Taraxacum officinale* (<https://www.verspreidingsatlas.nl/2430>)  
**5** Fertilized grasslands. Nutrient poor - nutrient rich. Weakly acidic-calcareous.
61. *Trifolium repens* (<https://www.verspreidingsatlas.nl/1306>)  
**5** Condensed/disturbed soil. Moderately nutrient rich. Moderately nitrogen rich. Weakly acidic-alkaline.

62. *Vicia cracca* (<https://www.verspreidingsatlas.nl/1369>)

**5** Soil contains humus. Moderately nutrient rich - nutrient rich. Lime-poor-calcareous.

## Appendix 6: List of Species Found in the Seed and Propagule Banks

1. *Alopecurus* sp.
2. *Cardamine pratensis*
3. *Cerastium fontanum*
4. *Dicotyl* sp.
5. *Eleocharis palustris*
6. *Elymus repens*
7. *Equisetum arvense*
8. *Glechoma hederacea*
9. *Holcus lanatus*
10. *Lolium perenne*
11. *Lysimachia* sp.
12. *Lythrum* sp.
13. *Persicaria hydropiper*
14. *Poaceae* sp.
15. *Polygonum aviculare*
16. *Ranunculus acris*
17. *Ranunculus repens*
18. *Ranunculus sceleratus*
19. *Rumex acetosa*
20. *Rumex obtusifolius*
21. *Senecio vulgaris*
22. *Stellaria media*
23. *Taraxacum* sp.
24. *Trifolium repens*
25. *Urtica dioica*

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