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The effect of the pioneer stage of cranberry cultivation on environmental processes: vegetation diversity and carbon accumulation

Sustainable agriculture on rewetted peat meadows

BSc research report
1st of March – June 26, 2023
24 EC

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Abstract

For a long period of time, Dutch peat meadows have been drained for agricultural purposes. As a consequence, the ecosystem is slowly degrading causing problems related to ecosystem functions: emission of greenhouse gasses, soil subsidence, loss of climate regulating function and decline of biodiversity. A proposed measure for reducing these problems is: raising the water tables. But, current agricultural activities, like dairy farming, cannot be maintained under wet soil conditions, which will endanger the valuable anthropogenic function of these areas. Therefore, theoretical research has proposed sustainable agriculture in the form of 'wet crops', for example: cultivating cranberries. Nevertheless, the practical application of these crops is difficult and the effects on the ecosystem functions remain unknown yet. Therefore, this study investigates two aims that support improvement of practical application of cranberry cultivation. These questions are related to the ecosystem functions of peat meadow areas. The first aim: 'Can cranberries play a role in restoration of a stable, peat meadow ecosystem that supports peat-specific plant diversity?' was investigated at a pioneering cranberry cultivation located near Oud Ade. There were settlements of peat-specific plant species observed after transplanting peat moss (*Sphagnum sp.*) to the cranberry cultivation. This may indicate stimulation of development of a stable peat meadow ecosystem. The second aim: 'Does cranberry support the main reason of raising water tables: mitigating climate change and soil subsidence? Are cranberries able to induce the process of carbon accumulation?' was investigated at a well-developed cranberry cultivation near Krimpenerwaard. According to the soil analysis, the peat forming process seems to be activated. Bulk density was significantly lower and carbon accumulation was significantly higher when a successful cranberry cultivation was compared to a rewetted grassland (p = .003 ; p < .001). However, the elevation measurements are questioning this statement, because a decline in land surface was detected.

In general, further investigation of later successional stages is required to monitor the development of these two ecosystem functions of a cranberry cultivation at Dutch peat meadow areas.

Keywords

rewetting peat meadow area, sustainable agriculture, wet crop, cranberry cultivation, soil subsidence, climate change, biodiversity.

Contents

Introduction

The Netherlands is coping with a compounding effect related to drainage of peat. Soils are subsidizing, greenhouse gasses are emitted, soils become salinized, several ecosystem functions are under pressure and biodiversity is declining (Van den Born, 2016; Strack et al., 2022). Therefore, peat meadows need to be preserved to prevent further degradation and the loss of valuable ecosystem functions (UNEP, 2021). In order to understand the complete situation occurring at the Dutch peat meadow areas, firstly the process driving these ecosystems is explained. Followed by the valuable ecosystem functions served by a natural peat meadow. Then, their main threat: land use change is discussed. Lastly, the sustainable options for the Dutch peat meadow areas are introduced, ending with the aim of this study.

Peat Forming Process

Peat meadows in The Netherlands are recognized as wet ecosystems (Wesselingh, n.d.). These type of systems are driven by an important environmental process called: peat formation, or carbon accumulation (International Peatland Society, 2019). For centuries, large amounts of dead plant material have been accumulated under wet soil conditions (Moore, 1989). When water tables are high, two types of soil layers can be distinguished. (Also visualized at Figure 1).

A moist, anoxic, acidic and cool soil layer is called the "catotelm". This layer is usually soaked in water. Therefore, only anaerobic bacteria can perform decomposition inside this layer (Moore, 1989). As a result, organic material is processed into CH_4 and CO_2 without the use of oxygen (Partlow, 2022). Anaerobic decomposition is a very slow process. Therefore, thick layers of organic material can accumulate and be preserved for a long period of time. So, the process of peat formation is supported by the specific condition of the catotelm layer of the soil. In Figure 1, this layer can be recognized as "anoxic layer".

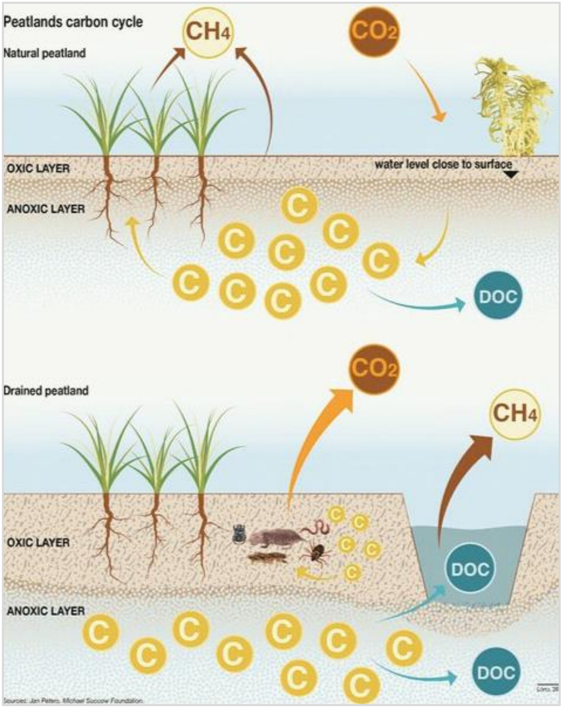

Figure 1: A natural peatland with an activated peat forming process, because of a dominant catotelm/anoxic layer. A drained peatland with a deactivated peat forming process, because of a dominant acrotelm/oxic layer. (Izquierdo, 2017)

In contrast to the catotelm, the conditions of the upper layers, called: "acrotelm", of the soil are less extreme and fluctuate constantly; humidity is lower, enough oxygen is available and temperatures are relatively higher compared to the catotelm layer (Moore, 1989). The acrotelm layer usually isn't soaked, or only during winter. The less extreme conditions of this layer outcompetes anaerobic bacteria. Several other decomposers: fungi, aerobic bacteria, and invertebrates, can decompose under oxic conditions, which is more effective than anaerobic decomposition (Clymo, 1965). The change in respiratory activity will accelerate the breakdown of organic material into CO_2 (Huang et al., 2021). Leading to lower accumulation rates, and deactivation of the peat forming process (Inglett, Reddy, & Constanje, 2005). In Figure 1, this deactivation is visualized for drained peat meadows. So, when conditions of the acrotelm dominate the system, the stored organic material slowly degrades. This results in degradation of a peat meadow ecosystem.

Ecosystem Functions of Peat Meadows

When peat formation is activated, a peat meadow ecosystem can serve multiple functions that facilitate life (Bonn et al., 2014). Because of these ecosystem functions, peat meadows are considered as valuable ecosystems that need conservation (UNEP, 2021).

The first valuable function is related to the peat forming process introduced earlier. The peat forming process can keep energy in the form of carbon inside the system for a long period of time (Odum, 1969). In case of the Netherlands, accumulation of organic material began approximately 11.700 years ago (Wesselingh, n.d). Therefore, a lot of energy has been stored since then. The ability to accumulate organic material causes peat soils to be recognized as the largest carbon storage of the world (IUCN, 2021). When soil conditions are activating the peat forming process, peat meadows can be an important inhibitor of climate change. Because, soils are then capable of assimilating emitted greenhouse gasses (Bonn et al., 2014).

Furthermore, peat soils can store large amounts of water inside thick layers of organic material, which can regulate local climate. If the atmosphere becomes dry during summer, the water stored inside peat soils evaporates and will moisten and cool down the surrounded air (IUCN, 2021).

Also, peat soils support many scales of biodiversity. The unique conditions of these type of ecosystems creates a lot of niches, which suit a wide variety of species (IUCN, 2021).

The most essential function of peat meadows for the Netherlands, is rising of land surface by expanding the soil. The accumulation process keeps creating more soil by taking up carbon out of the atmosphere. Furthermore, peat is characterized by its high moisture percentage, which will decrease the soil density, resulting in expansion of the soil. This is of great advantage for a country located under sea level. Especially, with the current acceleration of climate change (Van den Born, 2016).

To conclude, Dutch peat meadow areas take advantage of the carbon accumulating function that causes the soil to expand. This process can also inhibit climate change by storing carbon inside the soil. Furthermore, humid peat soils can regulate atmospheric temperatures by increased evaporation rates. Also, several scales of biodiversity are supported by the unique conditions of peat meadow areas.

Natural Peat Meadows

Nevertheless, these ecosystem functions are only supported when peat formation is activated. This happens when the system is in a natural state. So, it is important to take the origin of a natural peat meadow into consideration, when restoring these ecosystems. Therefore, The Different successional stages of a peat meadow ecosystem are explained in more detail and are supported by a visualization (Figure 2).

In general, peat soils start as small lakes. Large amounts of organic material are deposited on the bottom of these lakes. Here is a lack of oxygen, so the peat forming process is activated. Thick layers of accumulated material, peat, start to develop. After long periods of accumulation, the bottom of the lake will rise. Its appearance is slowly transforming into shallow meadows, called "fens" (Vitt, 2008). The hydrology of a fen is based on the ground water. It continuously feeds the area with nutrient-rich water.

As peat formation continues, the layers of accumulated soil become thicker. When the bottom of the lake expands over the ground water level, the fen becomes isolated. Now the hydrology of the system is fully derived from precipitation. The ecosystem slowly turns into a nutrient-poor and acid environment, because hardly any alkalic ions accumulate in precipitation and present nutrients will gradually leach out (Gorham, 1957). This limits the occurrence of most plant species growing in fens. Therefore, this successional stage is called "bog" instead of fen (Konvalinková, 2010; Gorham, 1957). Only plant species that are adapted to these specific conditions, can thrive in bogs. An important keystone species of a bog system is peat moss (*Sphagnum sp.*). Initially, this moss tends to develop slowly. Nevertheless, peat moss stimulates peat forming process excessively. They can absorb and store large amounts of water inside their cells, which develops an impermeable layer that closes off the soil from oxygen and nutrients out of the atmosphere (Lepp, 2008). Therefore, peat moss reinforces the peat forming process. (Craft, 2016; Clymo, 1965). Such a system is at a stable successional stage and slowly becomes self-sufficient (Beyer et al., 2021).

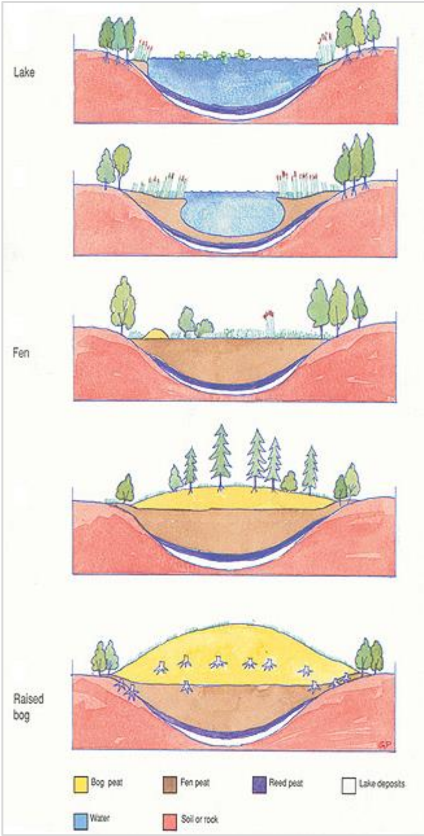

Figure 2: Successional stages of natural peat meadows. From small lake to fen until a bog. (Irish Peatland Conservation Council, 2016)

In short, natural peat meadow areas have different stages of development that all support the ecosystem functions in their own way. During all these stages, the peat forming process is the driving factor. Especially, the introduction of the keystone species: peat moss, is reinforcing the peat forming process. The introduction of peat moss leads to a stable, self-sufficient, natural peat meadow ecosystem. In the end, is the ambition of restoring the Dutch peat meadows.

Agricultural Land-Use Change

However, the natural state of the Dutch peat meadow areas is slowly disappearing. Over the past centuries, the land use of the Dutch peat meadows has changed greatly. This change has threatened the natural state of the ecosystem, and their valuable ecosystem functions. (Bos, Smit, & Schröder, 2013).

When agriculture started at the peat meadow areas, it was performed in an extensive way. Multiple methods were performed on a small scale in order to pay the costs of the farm. The landscape became very heterogeneous, because several gradients were created. For example, a nutrient gradient was created by removing biomass from a field, followed by deposition on another field. These gradients supported multiple niches for a lot of species (van der Windt, 2014). Several herbaceous plant species were able to grow at the extensive agricultural fields. Diverse vegetation also served as hiding spots for meadow bird chicks, since they are more difficult to find for raptors. Wader bird species, like: Black-Tailed Godwit (*Limosa limosa*), Redshank (*Tringa tetanus),* Lapwing (*Vanellus vanellus*) and Oystercatcher (*Haematopus ostralegus*), bred, hibernated or fed at Dutch peat meadow areas (Breeuwer et al., 2009). Flowering herbs also attracted a lot of important pollinators, like bees and ants (*Hymenoptera*), butterflies (*Lepidoptera*), flies (*Diptera*) and beetles (*Coleoptera*) (Langlois, Jacquemart, & Piqueray, 2020). In short, traditional farmers maintained a heterogeneous landscape, which promoted multiple scales of biodiversity (Henle et al., 2008).

At the end of 19th century, the demand for agricultural products increased. So, agricultural activities needed to intensify to keep up with the demand (van der Windt, 2014). This was enabled by new developments in technology that made management of large cultivations or many livestock possible (Henle et al., 2008). Large, efficient machines replaced the manual labor of farmers. Unlike human, heavy machines sunk away in wet peat, therefore a dry and firm soil was required to perform intensive agricultural activities. So, the ditches were dug and water tables have been kept low ever since. The intensive agriculture couldn't go along with small scale variation in the landscape: heterogenous landscapes. Which also caused environmental gradients to disappear (Henle et al., 2008). Several niches were disturbed, which had consequences for the biodiversity. A lot of unique flowering plant species experienced difficulties with the new environmental conditions. Plant species adapted to wet soils declined in abundance (van der Windt, 2014). As a result, several pollinators suffer from food shortages (Langlois, Jacquemart, & Piqueray, 2020). Also, there is detected a population decline of several wader bird species, because lower water tables cause soil life to move into deeper layers, which lead to food limitations for meadow bird species (Roodbergen & Teunissen, 2014). In conclusion, the accelerated development of agriculture and the essence to drain the soil are threatening several scales of biodiversity.

Beside the effects of drainage for biodiversity, many important ecosystem functions of peat meadows are also threatened by drainage of these soils. Draining activities are interrupting the process of peat formation (IUCN, 2021). The catotelm layer of the soil is exposed to oxygen, which creates conditions that accelerate decomposition activities (Moore, 1989). The accumulated carbon degrades, and is released in the form of greenhouse gasses, like; CO_2 and CH_4 (Huang et al., 2021). Thereby drained peat soils are contributing to climate change, by causing 5% of the total global emissions on only 3% of the total land surface (IUCN, 2021; International Peatland Society, 2019). This is an excessive amount, giving the fact that these soils can retain carbon instead of releasing it. Consequently, soils will subsidize instead of expanding due to degradation and compaction of peat (van den Born, 2016).

To summarize, the change of agricultural activities at Dutch peat meadow areas are causing problems that reinforce each other. This leads to an unstable ecosystem, that negatively effects life on earth, instead of providing valuable functions. Therefore, sustainable agriculture is needed to conserve both ecosystem functions and anthropogenic functions of the Dutch peat meadow areas.

Sustainable peat meadow areas

To strike the problems related to drained peat soils caused by land use change, several measures have been proposed to restore the natural state of these type of ecosystems and preserve its ecosystem functions (Rienks & Gerritsen, 2005; UNEP, 2021).

One proposed option is raising the water table (van den Born, 2016). Rewetting peat soils induces the peat forming process, because soil conditions of the catotelm are supported. Therefore, aerobic decomposition cannot occur, which inhibits the degradation of peat. This option may combat the loss of ecosystem functions and biodiversity (Beyer et al., 2021). Although, the development of the ecosystem is not supported by science, therefore these assumptions remain uncertain.

Certainly, the environmental conditions of peat meadow areas will change. Drained peat will develop into a wetland ecosystem again. Which has consequences for the anthropogenic function of the Dutch peat meadow areas. Current methods of intensive agriculture, like dairy farming, cannot be maintained at a wet state of peat soils. Therefore, sustainable agricultural methods that can be managed under wet conditions are required, in order to preserve this anthropogenic value. Some studies have proposed alternative crops (van Belle et al., 2022; Bestman et al., 2019). For example: Bulrush (*Typha sp.*), Common Reed (*Phragmites australis*), Peat moss (*Sphagnum sp.*), Silvergrass (*Miscanthus*), Willow (*Salix*) and Cranberry (*Vaccinium macrocarpon*). This study is focused on cranberries.

Theoretically, cranberry cultivation at Dutch peat meadow areas is a potential replacer of current agricultural methods, because nutrient-poor, acidic and humid conditions are all met on rewetted peat meadows. Cranberries have multiple advantages towards current crops and other wet crops. Where most of the current used crops are annual plants, cranberries are a perennial. Meaning that, once planted, they can be productive for multiple years. Also, cranberry products contribute to the food chain, while other wet crops are used as construction materials (Timmer & Balkhoven-Baart, 2006; Jukem a, Netjes, Zimmermann, & Prins, 2006; Bestman et al., 2019). Furthermore, the demand for these fruits is increasing, supposedly because several health claims have been researched (Jukema, Netjes, Zimmermann, & Prins, 2006).

While there is enough theoretical support for cranberry farming. The practical application remains complicated. And its effect on the ecosystem lacks scientific foundation.

Study aim

To increase the feasibility of cranberry farming at Dutch peat meadow areas, a couple of unknowns need scientific foundation first; (1) 'What cultivation method leads to the highest vitality, and is the most feasible for farmers?', (2) 'Can cranberries play a role in restoration of a stable, peat meadow ecosystem that supports peat-specific plant diversity?', (3) 'Does cranberry support the main reason of raising water tables: mitigating climate change and soil subsidence? Are cranberries able to induce the process of carbon accumulation?' During this study, these questions are investigated on a just-started cranberry cultivation at Oud Ade.

Designing a cranberry cultivation

The design of the cranberry cultivation at Oud Ade was based on another cultivation on peat soil near Krimpenerwaard. This cranberry cultivation started in 2016 and is now very productive (The Cranberry Company, 2016). The Cranberry Company (2016) and Kaptein (2020) encountered some discussion points through investigation at this cranberry cultivation. Kaptein (2020) concluded that competition of dominating plant species, like: Common rush (*Juncus effusus*) are a problem for the vitality of the cranberries at Krimpenerwaard. Especially, at the pioneer stage of a cultivation, the slow-growing cranberry plants are easily outcompeted by these fast-growing vegetation species (Praktijkcentrum Islandberry, n.d.). So, combatting dominant weeds need further research to improve the feasibility of cranberries cultivation. Therefore, three treatments are applied related to combatting weeds:

(1) Addition of an anti-root-fabric before planting the cranberries. This fabric will inhibit the growth of dominant plant species.

(2) Soil transplantation of a successful cranberry cultivation. Transplantation of soil may support the colonization of Ericoid mycorrhizal fungi, which stimulates the growth of cranberries. (Praktijkcentrum Islandberry, n.d.).

(3) Planting cranberries in combination with peat moss (*Sphagnum sp.*). Peat moss covers the open spaces between the cranberry plants. Therefore, the moss serves as a natural method of combatting dominance of other vegetation.

(4) Cranberries planted without any addition. This serves as a control measurement.

The area description and design of the cranberry cultivation at Oud Ade can be found at Box 1**.**

Vegetation diversity

Furthermore, Kaptein (2020) observed a significant change in vegetation diversity compared to grassland at Krimpenerwaard. But, Kaptein (2020) only investigated a later stage of succession, so the development of the vegetation diversity in the pioneer stage remains unknown. Therefore, this study monitors the composition of the vegetation community of the beginning stage. Especially, in disturbed soils, like the cranberry cultivation at Oud Ade, seedbanks provide the origin of the growing vegetation (Luzuriaga, Escudero, Olano, & Loidi, 2005). These seedbanks derive from the surrounding or are introduced by external factors, for example: potting soil or transplantations. That is why, the addition of peat moss is expected to introduce a seedbank including peat-specific vegetation species, like: Common Sundew (*Drosera rotundifolia*), Lesser-Butterfly Orchid (*Platanthera bifolia*) and Devil's Bit Scabious (*Succisa pratensis*) ("Flora van Nederland - Veen," n.d.). Also, the soil transplantation may provide a seedbank to the cranberry cultivation (Brown & Bedford, 1997). Introduction of seedbanks may induce the development of a stable peat meadow ecosystem, when the seeds are derived from peat-specific plants (Isbell et al., 2017).

In the end, the unknown: (2) 'Can cranberries play a role in restoration of a stable, peat meadow ecosystem that supports peat-specific plant diversity?', is covered

Carbon accumulation

Unfortunately, research at the cranberry cultivation near Krimpenerwaard, didn't include the activity of the peat forming process. While, this process is driving peat meadow ecosystems and is supporting valuable ecosystem functions. Also, farmers can earn some extra income, when their method contribute to mitigate climate change by storing carbon inside their soils. This may increase the feasibility of cranberry cultivation (Carbon Farmers, 2023).

Therefore, this study examines the state of the soil organic carbon at the Krimpenerwaard. Which is analyzed by performing soil analysis combined with elevation measurements. Soil analysis will provide information about the soil bulk density and soil organic carbon content, which both provide insight in the activity of the peat forming process. When, the peat forming process is activated, the soils may expand, leading to an increasing elevation of the soil surface. The data from Krimpenerwaard may in the end provide a future perspective for Oud Ade. Wherefore, a null measurement is conducted that would support further research. It is expected that the peat moss treatment will induce the activation of carbon accumulation, because peat moss can absorb large amounts of water, resulting in extended catotelm conditions (Lepp, 2008).

Ultimately, this will fill in the unknown: (3) 'Does cranberry support the main reason of raising water tables: mitigating climate change and soil subsidence? Are cranberries able to induce the process of carbon accumulation?'

The cranberry cultivation is located at the Vrouw Venne polder near Oud Ade, The Vrouw Venne polder is a good representation of a peat meadow area in the Netherlands. All major problems of the Dutch peat meadow areas described earlier, are occurring at this area.

The cultivation has started in March 2023. The cranberry cultivation is about 400 m^2. Each treatment (1,2,3,4) is applied on about 100 m^2 of the cultivation Figure 3. In order to reduce the impact of inflowing nutrients, a buffer zone has been planted around of about 1 meter width. Before planting the cranberries, several preparations have been to make the cultivation at Oud Ade a success. In December 2022, the top layer of the soil, composed of a thick layer of sea clay, has been excavated. This removed top layer composed of sea clay included a seedbank and nutrients. Furthermore, a pipe has been laid to transport the exceeding water to the nearby ditch.

On March 18, 2023, cranberries of the breed: Early black, were planted. This breed is introduced in 1850 and has evolved by natural selection (Timmer & Balkhoven-Baart, 2006). Within the treatments, two groups of cranberries can be distinguished: expensive, well-developed P9 plants and cheap, young plugs. The purchase of plugs is lower than that of P9 plants, therefore plugs would be more feasible for farmers to use. But the actual difference in their vitality and productivity remains unknown. Therefore, the groups are investigated to improve the application of cranberry cultivation for farmers. They are taken into account when answering the first aim: (1) 'What cultivation method leads to the highest vitality, and is the most feasible for farmers?' (Klazema, 2023). However, they are also included at the vegetation diversity experiment, because plugs and P9 plants differ in the coverage. Which may influence the growth of other vegetation. Especially, during the pioneer stage of the cranberry cultivation, competition of vegetation is an important factor for the vitality of the plants.

Figure 3: Cranberry cultivation at Oud Ade in pioneer successional stage.

Box 1: Brief description of the study area and an introduction to the design of the cranberry cultivation of this experiment. Also the plug and P9 groups are explained.

Methods

During this study, the effect of land use change, changing a current grassland to a cranberry cultivation, on environmental processes driving the ecosystem was examined. It focused on two main ecosystem functions: plant diversity and carbon accumulation.

Vegetation Biodiversity

This study investigates the effect of vegetation diversity on the cultivation of cranberries. The effect of succession can only be seen after a couple of years. But in order to do so, the pioneer stage need to be monitored. To acquire a representation of current agricultural activities, the surrounded grasslands were used as a control measurement. The cranberry cultivation measurements show the effect of sustainable agricultural methods on the plant diversity in the area, when comparing them. Monitoring the plant diversity was done by noting the species and their coverage. For determination of the plant species, Heukels' Flora van Nederland (Duistermaat, 2020) and the ObsIdentify app were consulted. For the coverage of the species, the Tansley (1946) vegetation scale was used.

On the grassland, two plant releves of 1 m^2 were conducted. On the cranberry cultivation four releves were performed per treatment. Of which two were conducted on the plug group as well as the P9 group. Besides this distinction, the locations of the releves were chosen randomly, as shown in Figure 4.

After monitoring the species composition of the area, the species lists and their coverage were entered into the software: Turboveg. Then, the data was exported to Microsoft Excel. Furthermore, the Tansley (1946) vegetation scale was converted into percentages to perform calculations. Species richness was noted, and Simpson's index (D) was calculated by the following equation.

$$D = \frac{\sum n(n-1)}{N(N-1)}$$

Statistical analysis was computed using R. On the species richness data, a GLM, family: Poisson, was performed. While on the Simpson's index an ANOVA and TukeyHSD posthoc was done.

Afterwards, the data was visualized by creating a stacked bar plot, and a boxplot.

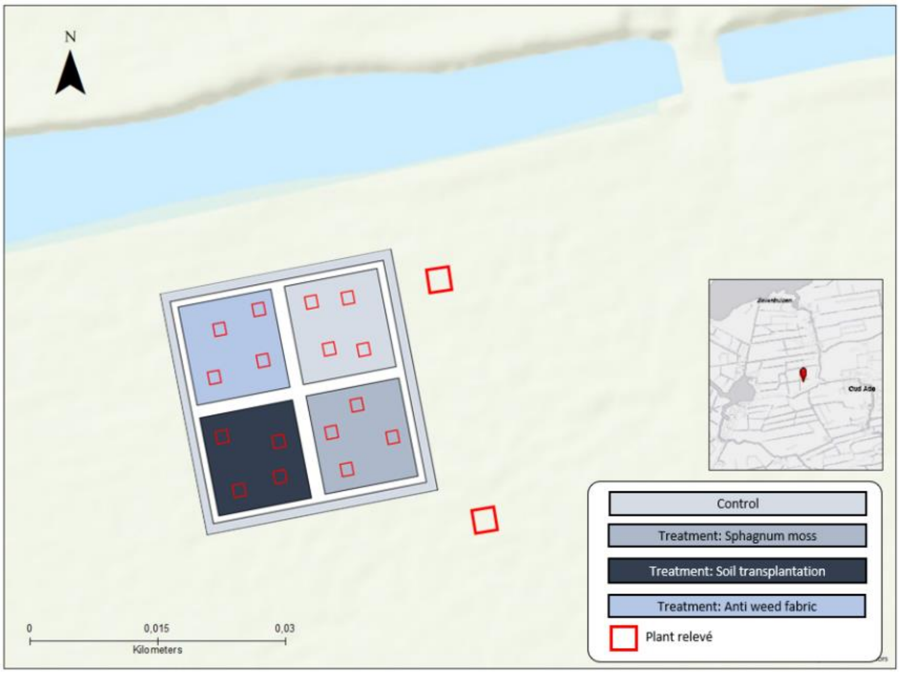

Figure 4: Design of vegetation diversity experiment at Oud Ade.

Carbon Accumulation at Krimpenerwaard

Carbon accumulation is a slow process that can be seen after long period of consistent carbon gain. At the cranberry cultivation at Oud Ade this effect cannot be seen yet. Therefore, a successful cranberry cultivation on peat soils located near Gouda was used to detect the activity of the carbon accumulation process. These measurements can then be used to draw a future perspective for the cranberry cultivation at Oud Ade.

At first, the elevation of the area was determined by an external company: Geovalk. They already did measure the height of the area near Gouda in August 2016. February 2023, Geovalk measured heights of the area again. After obtaining the data, the average height per field was determined, using Arc GIS. This was done for 2016 and 2023. In the end, the average elevation per field could be seen. Then, the difference in elevation between grasslands and cranberry fields could be detected.

In order to show the origin of the elevation, the carbon concentration of the soil were investigated. To detect a significant difference between rewetted grassland, unsuccessful cranberry fields and successful cranberry fields. On both grasslands and (unsuccessful) cranberry fields 15 soil samples were taken, see Figure 5. Therefore, Holes of 15x15x30 centimeter were dug to sample the soil in the top layer (5 centimeter dept) and the deep layer (20 centimeter dept). Bulk density rings were used to take samples that represent 100 ml soil. Then, the soil samples were dried and burned using a furnace (See Box 2 for the complete protocol). In between, the weights of the samples were measured a couple of times by using a scale. This allowed us to calculate the bulk density (g/cm^3), soil moisture (%), soil organic material (%) (SOM), soil organic carbon, and minerals (%). In the end, some of these variables were used to calculate the soil organic carbon (SOC) using the following equation.

$$SOC = 100.000\ m^2 \times horizon\ dept\ \times bulk\ density\ \times carbon\ percentage$$

After calculating the variables, a statistical analysis was performed using R. The data was separated into the different horizon depts. Bulk density was analyzed by performing an ANOVA and a TukeyHSD posthoc. This was the same for the carbon percentage of 5 centimeter dept. But, carbon percentage on 25 centimeter dept turned out to be non-parametric, so a Kruskal Wallis test was conducted instead.

To deliver evidence of a successful cranberry field, a root was collected. These roots were checked for the colonization of Ericoid Mycorrhizal fungi by using the protocol also used by (Slusarek, 2022). This protocol was fully written down by (Klazema, 2023).

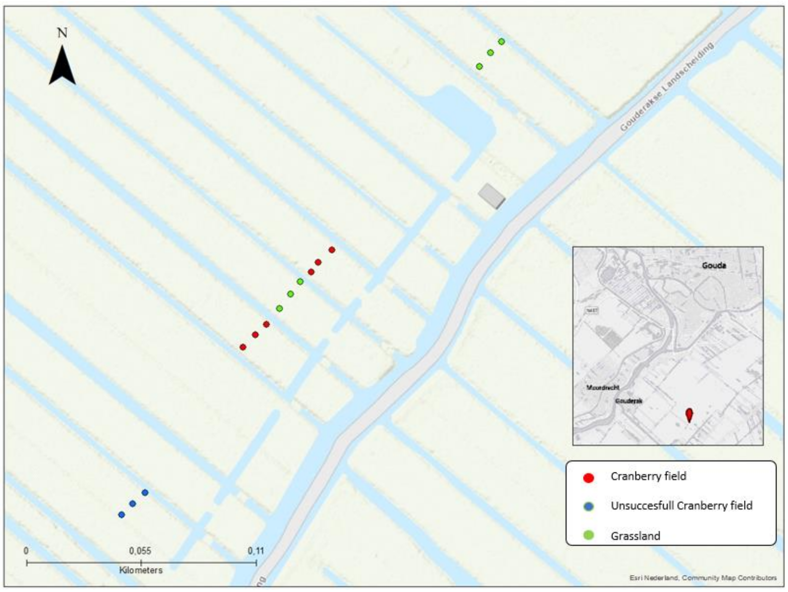

Figure 5: Design of carbon accumulation experiment at Krimpenerwaard.

Carbon Accumulation at Oud Ade

The carbon accumulation process of the cranberry cultivation at Oud Ade cannot be indicated at the pioneer stage. Therefore, this study provides a null measurement for further research. The sampling method is the same as was used at Krimpenerwaard. At every treatment two holes were dug. Each hole was sampled twice, on 5 centimeter dept and on 25 centimeter dept. Furthermore, the surrounded grassland was sampled at four different depts; 5, 25, 40 and 60 centimeters. The design is visualized at (Figure 6).

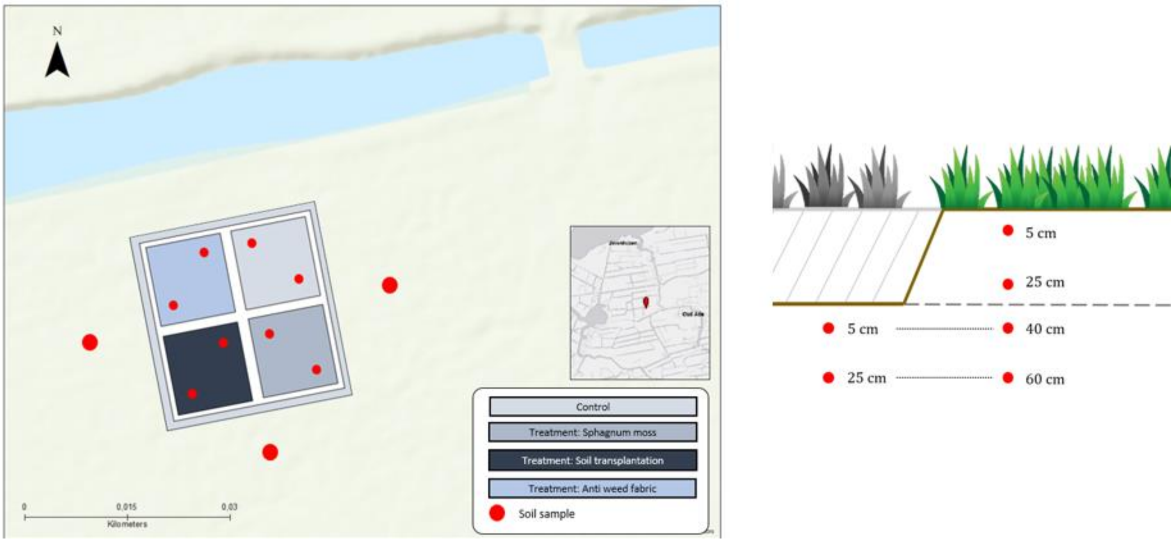

Figure 6: Design of carbon accumulation null measurement on cranberry cultivation at Oud Ade. Including a transection of the sample depts at both cranberry fields and grasslands.

Abiotic variables

Beside the main experiments, including plant diversity and carbon accumulation, several abiotic factors have been investigated as well. An eye was kept on atmospheric temperature and precipitation events. Also, pH, parts per thousands, milli Siemens and soil temperature have been measured regularly by using a waterproof pH combo measure tool of Hannah instruments. Furthermore, the nitrate and phosphate concentrations were determined by using a colorimetric chemical test kit. Nitrate and phosphate were both samples five times per treatment. The locations were chosen randomly (Figure 7). For the full protocol, see (Box 3). These abiotic factors were used to create interpolated ArcGIS maps that visualized the situation.

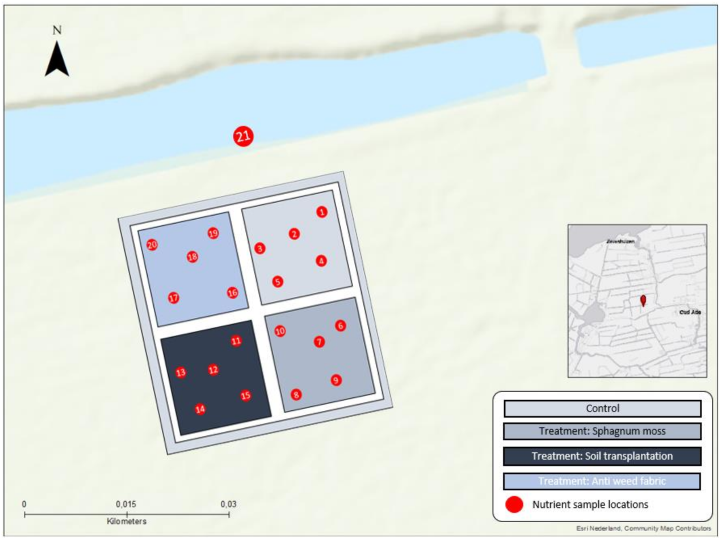

Figure 7: Design of the nutrient concentration experiment at Oud Ade.

Results

Vegetation diversity

This section provides results for the aim: 'Can cranberries play a role in restoration of a stable, peat meadow ecosystem that supports peat-specific plant diversity?'.

Species richness and species composition

The species richness at the different treatments of the cranberry fields was between 2 and 8 species, while the surrounded grassland had a species richness of approximately 12. According to the ANOVA performed with a Poisson GLM, the difference between grassland and treatments was significant, $z = 3.15$, $p = .002$. The species: Perennial ryegrass (*Lolium perenne*) and Creeping bentgrass (*Agrostis stolonifera*), thriving on the surrounded grassland, were also occurring at some treatments of the cranberry fields (Figure 8). Beside the species richness of grassland, the ANOVA with a Poisson GLM provided a significant difference in species richness of both peat moss treatments, P9 and plug, $z = 2.00$, $p = .046$; $z = 2.15$, $p = .032$.

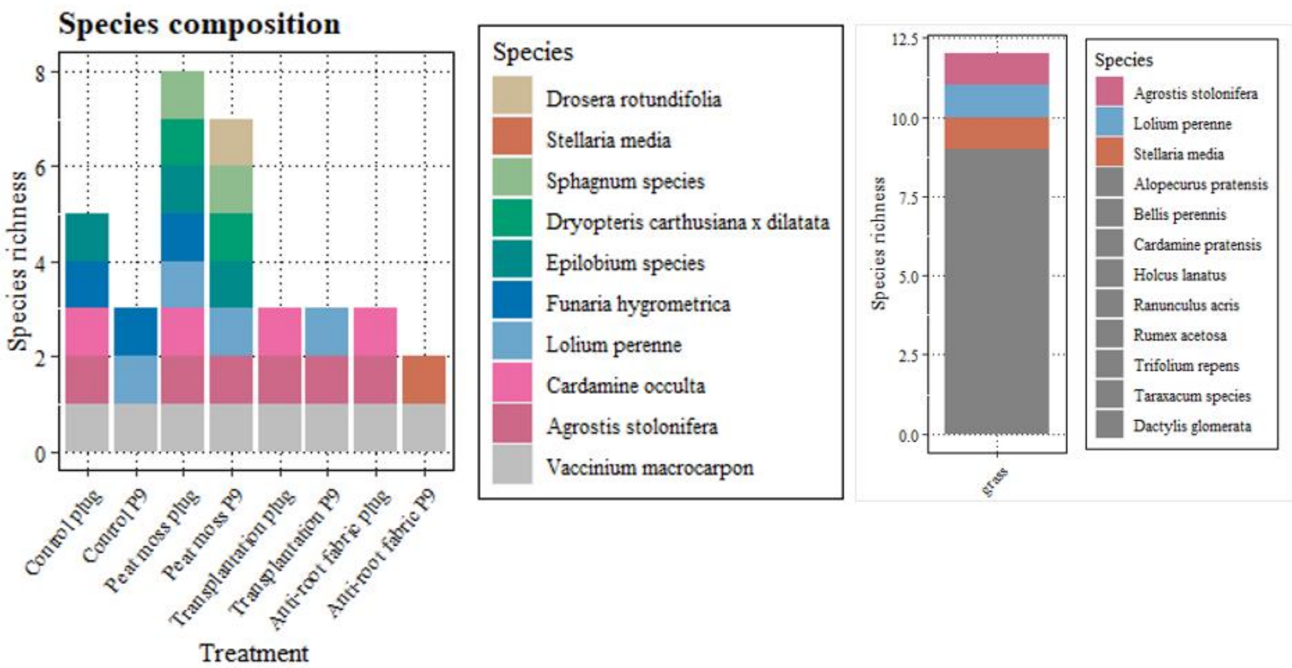

Figure 8: Species composition and species richness of the treatments at the cranberry cultivation (left graph), and their species overlap with the surrounded grassland (right graph).

Furthermore, Figure 8 shows the species composition of all treatments. Some outstanding results are being discussed. At first, Asian wavy bittercress (*Cardamine occulata*) was typically found on the plug groups of all treatments. Asian wavy bittercress was growing next to the cranberry plugs, as shown Figure 9a for the anti-root fabric treatment. Also, some unique species were observed at the peat moss treatment. Narrow buckler-fern + broad buckler-fern (*Dryopteris carthusiana x dilatata*) (Figure 9b) was colonizing multiple places of the peat moss treatments, therefore its Tansley score was locally frequent: lo. Common sundew (*Drosera rotundifolia*) was observed ones (Figure 9c), and had a Tansley score of sporadic, s. Both species were growing within the peat moss (*Sphagnum sp.*).

Figure 9: (a) Asian wavy bittercress (Cardamine occulata) growing next to cranberry plug at the anti-root fabric treatment. (b) Narrow buckler-fern + Broad buckler-fern (Dryopteris carthusiana x dilatata) growing on the peat moss treatment. (c) Common sundew (Drosera rotundifolia) growing on the peat moss treatment.

Diversity: Simpsons reciprocal Index

Figure 10 shows the plant diversity per treatment using the Simpson reciprocal index. Simpson's index is taking the abundance of the species into account. Therefore, increased dominance of certain species can lead to a lower diversity. The lower the value of the index, the lower the plant biodiversity is. For example, the P9 group of the anti-root fabric treatment had a Simpson index of 1, which was significant after computing an ANOVA, $t = 2.37$, $p = .042$. This ANOVA also detected a significant difference of the plug group of the anti-root treatment, $t = 2.86$, $p = .019$.

Furthermore, the diversity index of the plug group of the peat moss treatment seemed relatively high towards the rest of the treatments. After computing an ANOVA, it became clear that this was a significant difference, $t = 5.56$, $p < .001$.

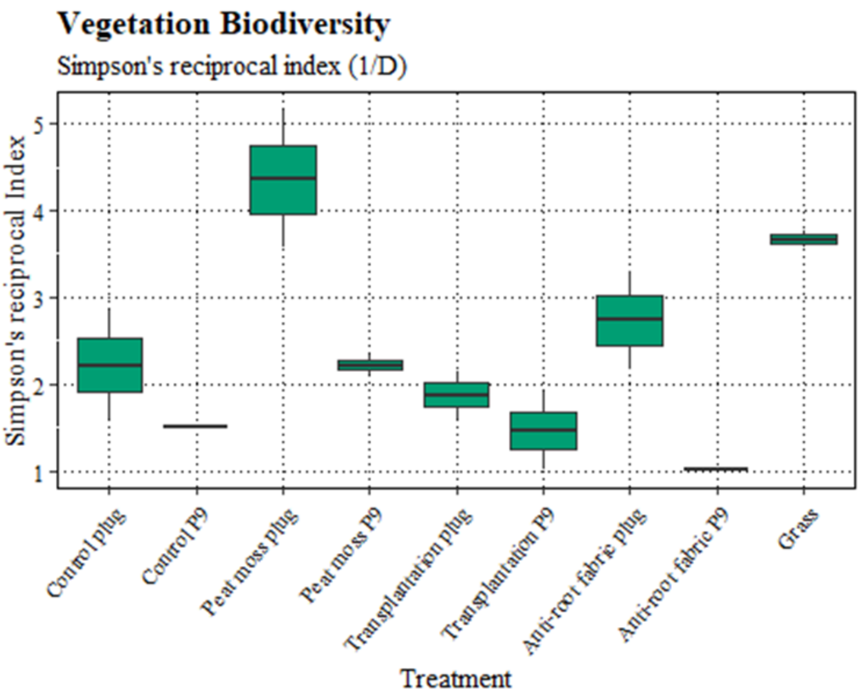

Figure 10: Simpsons reciprocal index (1/D) per treatment.

Carbon accumulation at Krimpenerwaard

This Section supports results for the aim: : 'Does cranberry support the main reason of rewetting grasslands: mitigating climate change and soil subsidence. Are cranberries able to induce the process of carbon accumulation?'. Therefore, this study examines the activity of the peat forming process at a 7 year old cranberry cultivation located near Krimpenerwaard. Field 1, 4, 5, 6 and 10 were compared to each other. Field 4 and 6 represent successful cranberry cultivations, since these plants relatively have had high yield between 2016 and 2023. Figure 11, shows the high colonization of Ericoid mycorrhizal fungi on a cranberry root from field 4, indicating a well-developed cranberry plant. In contrast, field 1 is recognized as unsuccessful field, because the growth and yield lag behind. As a control the grasslands (Field 5 and 10) were used, because grass represents the current agricultural methods under wet conditions.

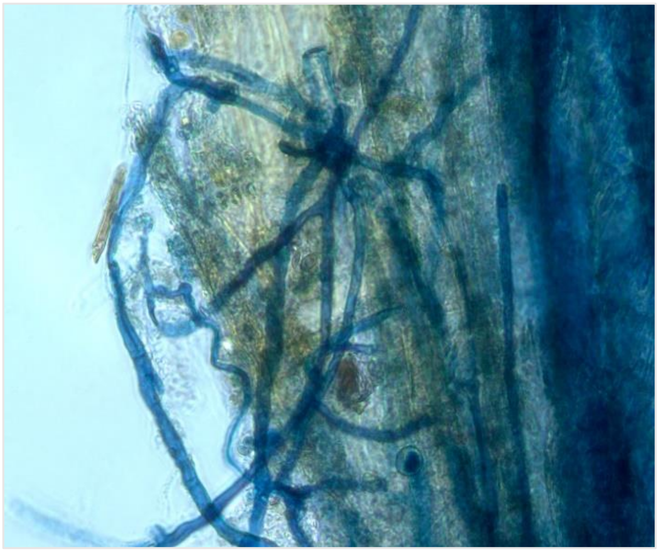

Figure 11: Ericoid mycorrhiza on cranberry root of field 4 for cranberry cultivation at Krimpenerwaard (10X10 magnification).

To detect the activity of the peat forming process in the cultivation at Krimpenerwaard, the equation introduced at the Methods was used. All variables of this equation are been discussed separately. Starting with the soil characteristics: carbon percentage and bulk density and ending with the elevation map.

Soil Organic Carbon percentage

Figure 12 shows the carbon percentage of the different fields at Krimpenerwaard. The soil samples were taken at two different horizon layers: top layer (5 centimeter dept) and deeper layer (25 centimeter dept), therefore, Figure 12 is separated into these two categories. The soils of the cranberry fields (4 and 6) had a carbon percentage of approximately 33%, in deeper layers even up to 40% Carbon. Whereas the unsuccessful cranberry field (1) and the grasslands (5 and 10) had a percentage around 23%, 20% at deeper layers.

An ANOVA provided information about the differences in type of field at both soil depts (5 and 25 centimeter). For 5 centimeter dept, a significant effect was detected for the type of field on the carbon percentage of the soil, F (2,12) = 11.91, p < .001. After performing a posthoc: TukeyHSD, the individual effects became clear. Cranberry and grasslands differed significantly, p < .001. Also, cranberry fields and unsuccessful cranberry fields differed significantly, p = .016. But, no significant difference was found between grasslands and unsuccessful cranberry fields, p = .886. For 25 centimeter dept, the residuals didn't turn out parametric, so a Kruskal Wallis test was performed to detect any significant effects. The main effect was significant, X^2 (2) = 12.15, p = .002. The difference between cranberry and grasslands was significant after computing a pairwise Wilcoxon test, p = .007. Also, the differences between unsuccessful cranberry and successful cranberry, and between unsuccessful cranberry and grassland were significant, p = .024 , p = .024.

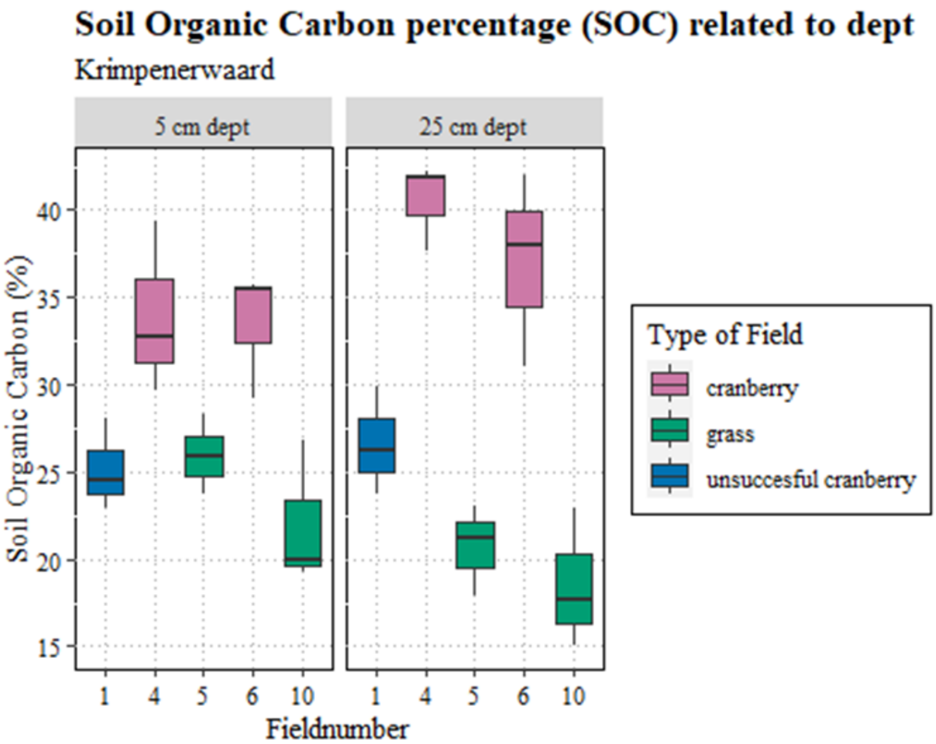

Figure 12: Soil organic carbon (SOC) percentage of 5 cm and 25 cm dept of different fields of cranberry cultivation at Krimpenerwaard.

Bulk density

Bulk density is correlated with the soil moisture percentage. The higher the bulk density, the more compacted the soil is. Compacted soils have relatively low moisture percentages.

Figure 13 shows the bulk density of the different fields at Krimpenerwaard. Field 5 and 10 had a bulk density between 0.43 g/ml and 0.56 g/ml. Cranberry fields had a bulk density between 0.20 g/ml and 0.30 g/ml. The unsuccessful cranberry field had a bulk density more closely to grasslands, between 0.35 g/ml and 0.39 g/ml. After computing an ANOVA, the main effect of bulk density in the top layer of the soil was significant, F (2, 12) = 8.97, p = .004. The cranberry fields had a significant lower bulk density than grassland, p = .003. Unsuccessful cranberries did not differ significantly between both grassland and cranberries, p =.487 , p = .103. Also, the deeper soil layer was analyzed by computing an ANOVA. The main effect of the bulk density at this deeper layer was significant, F (2, 12) = 35.94, p < .001. After a Tukey HSD test, all type of fields differed significant. Cranberry differed significant with grasslands and unsuccessful cranberries, p < .001 and p = .029. Also, unsuccessful cranberry differed significantly with grassland, p = .029.

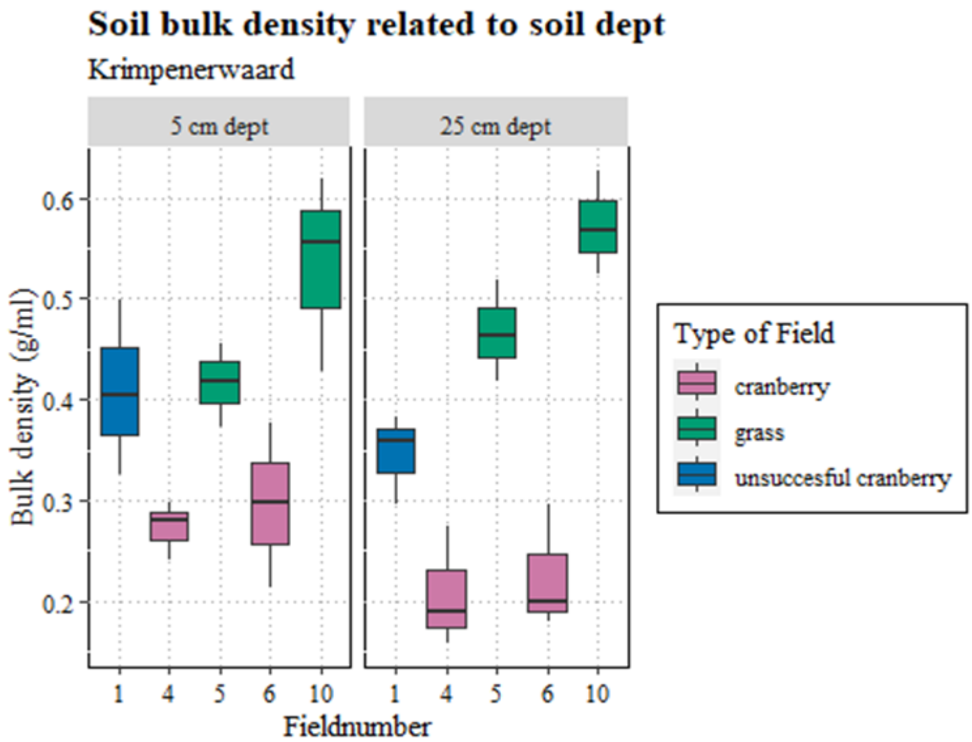

Figure 13: Soil bulk density on 5 cm dept and 25 cm dept of different fields of cranberry cultivation at Krimpenerwaard.

Elevation of soil surface

The effect of carbon accumulation is expansion of the soil. If bulk density remains equal, and soil carbon percentage increases, the process of carbon accumulation seems to be activated. This results in slow increase of soil surface.

In August 2016, the height of the soil surface of the cranberry cultivation located near Krimpenerwaard was measured. These measurements were performed with an eye on excavation. After conducting the results of these height measurements, the area was excavated for on average 13 centimeters, except for the grasslands fields (2, 5 and 10).

In February 2023, these soil surface measurements were conducted again. This time, all fields were included into the dataset. A map was created using ArcGIS, which shows the height difference between 2016 and 2023 (Figure 14). The soil surface at the red colored areas declined about 15 centimeters, whereas the soil surface of the green and yellow colored areas stayed equal or had a small increase of roughly 7 centimeters. The measurements of 2016 lack information about the heights of the grass fields, therefore no difference was obtained for these fields (2, 5).

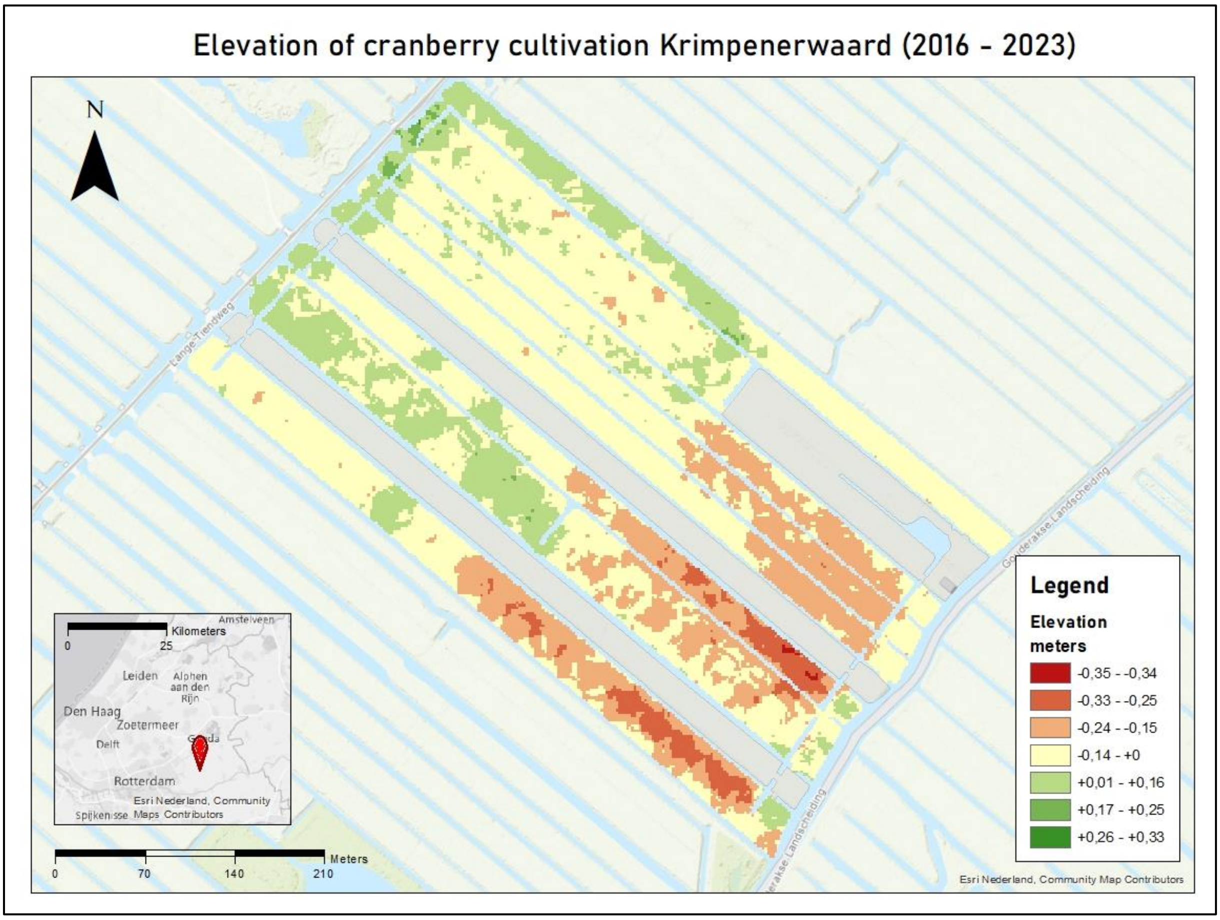

Figure 14: Soil surface elevation of the cranberry fields at Krimpenerwaard between 2016 and 2023. Field 1 to 10 are numbered and coloured from NW to NO. The category -0.14 - +0 is considered as constant, because approximately 13 centimeters was excavated inbetween 2016 and 2023.

Carbon content Oud Ade

This study also monitored the pioneer stage of the cultivation at Oud Ade. Therefore, soil analysis of Oud Ade is included.

Figure 15 shows the bulk density of the soil of different treatments at Oud Ade. The grass has a top layer consisting of clay, which is been removed at all treatments. The dept that is presented in Figure 15, is relative to the grassland, since the first 35 centimeter of the soil of the treatments is been removed. The 40 centimeter and 60 centimeter at the treatments are equal to the 5 centimeter and 25 centimeter as measured on the cultivation at Krimpenerwaard.

After computing an ANOVA, the treatments was significant different, F (4, 20) = 53.70, $p < .001$. When performing a Tukey HSD test, only grassland differed significantly with all other treatments, $p < .001$. Between the other treatments no significance was found.

Also, the different soil depts were differing significantly, F (3, 20) = 61.34, $p < .001$. Only the depts: 5 – 25 centimeter and 40 – 60 centimeter, didn't differ significantly after performing a Tukey HSD test.

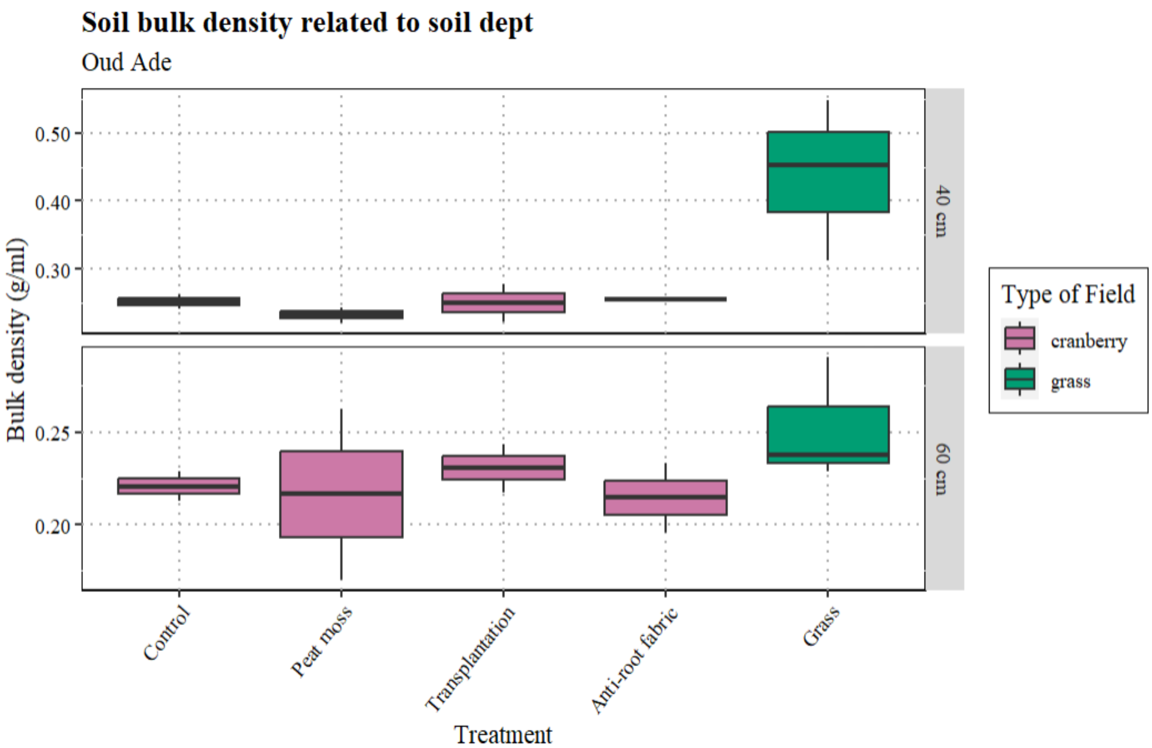

Figure 15: Soil bulk density of the different treatments of cranberry cultivation and the surrounded grassland. Soil dept of the cranberry treatments is relative to the grassland. The absolute dept was 5 cm and 25 cm, because the toplayer of 35 cm was excavated.

Figure 16 shows the bulk density of the different soil depts at the surrounded grassland. The bulk density of the clay soil was between 0.80 g/ml and 0.85 g/ml. The peat soil had a bulk density between 0.20 g/ml and 0.30 g/ml.

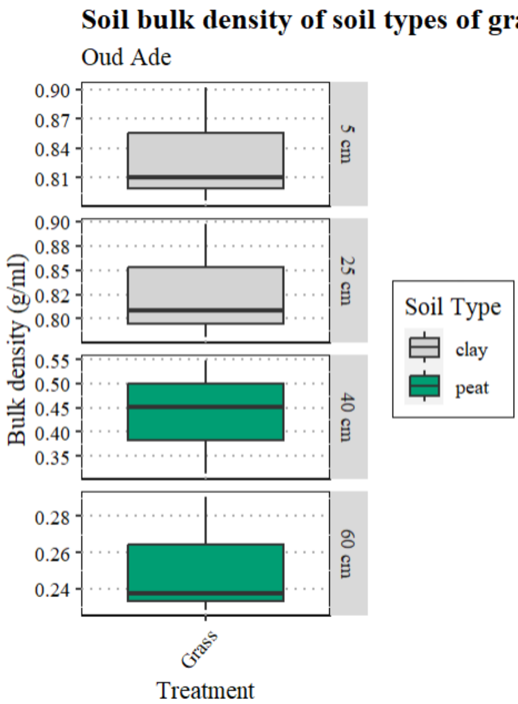

Figure 16: Soil bulk density related to the type of soil and their dept on the surrounded grassland.

Abiotic variables

Results of Abiotic variables were used to support the vegetation diversity data. The maps show higher nutrient concentrations close to the ditch. So, the control and anti-root fabric treatment had relatively higher concentrations. The soil moisture and pH varies between the different months (April, May and June). For the figures see the appendix on page 28.

Discussion

The general aim of this study is to investigate the effect of the pioneer stage of cranberry cultivation on environmental processes: vegetation diversity and carbon accumulation. Three aims were introduced, of which two will be discussed more deeply. Firstly, vegetation diversity is discussed, which is followed by the carbon accumulation. Finishing with a review to the general aim of the study.

Vegetation diversity

The aim: (2) 'Can cranberries play a role in restoration of a stable, peat meadow ecosystem that supports peat-specific plant diversity?' was supported by monitoring the vegetation diversity of the pioneer stage of a cranberry cultivation. The results show that, the peat moss treatment is inducing the development of a peat-specific vegetation diversity in the pioneer stage of a cranberry cultivation. Also, the use of plugs instead of P9 would positively affect the vegetation diversity. And, the surrounded grassland didn't provide a lot of seeds to the seedbank yet.

At first, It was found that the plug group of the peat moss treatment had the highest Simpson's biodiversity index. Also, according to the species composition, the peat moss treatment provided niches for some unique, peat-specific species, like: Narrow buckler-fern + broad buckler-fern (*Dryopteris carthusiana x dilatate*) and Common Sundew (*Drosera rotundifolia*). Therefore, it was concluded that transplantation of peat moss introduces a seedbank to the cranberry cultivation in the pioneer stage of succession. While succession continues, this phenomenon may stimulate the development of a stable peat meadow area.

Secondly, the plug groups of every treatment seemed to have a relatively higher Simpson's biodiversity index. This may be explained by the lack of competition on the plug fields. During the pioneer stage, cranberry plugs cover less space than the P9 plants do. Therefore, other vegetation had more opportunity to grow at these fields. This may be a positive effect for biodiversity. However, cranberry plugs may be inhibited or outcompeted by other vegetation at some point.

Furthermore, it was noticed that the surrounded area didn't dispersed a lot of seeds to the cranberry fields yet. Only, Creeping bentgrass *(Agrostis stolonifera*) and Perennial ryegrass (*Lolium perenne*) were overlapping their abundance with the cranberry fields. The input of seeds from the surrounding grassland may increase in the future, because the seed dispersal will increase later during the year. Therefore, it is expected that the number of species derived from the surrounded grassland will increase in the following years.

A last remarkable result, regarding the species composition, was the presence of Asian wavy bittercress (*Cardamine occulata*). This Cardamine species was typically found on the plug groups of all treatments. The plugs originate from the cranberry cultivation at Krimpenerwaard, therefore this might also be an explanation for the origin of Asian wavy bittercress. However, Kaptein (2020) didn't observe this plant species at the cranberry fields at Krimpenerwaard during his study. Therefore, the origin is unlikely to be the cultivation at Krimpenerwaard itself. Probably, the potting soil, used to grow the cuttings, was containing seeds of this Cardamine species. Asian wavy bittercress is identified as an invasive species. They originate from Eastern-Asia and are introduced in Europe after 2000 (Dijkstra, 2001). Currently, this species wasn't occurring in large numbers at the cranberry cultivation at Oud Ade. But, to prevent dominance of invasive species that may disturb the ecosystem, it is suggested to keep an eye on the identity of the species thriving at the cranberry cultivation.

To provide a conclusion for later successional stages of the cranberry cultivation at Oud Ade, monitoring the vegetation diversity in the coming years would be interesting. Whereby, a couple of recommendations can be included. First, activation of peat moss requires wet soil conditions and a lot of patience. Therefore, it is recommended to repeat adding peat moss multiple times to activate their growth. Furthermore, the pH of the soil should be maintained at a constant, acidic level. This will positively affect the growth of both peat moss and cranberry plants. But most important, it will select the vegetation composition. Especially, invasive species are than unable to dominate the ecosystem.

Abiotic variables

To support the vegetation development of the pioneer stage, abiotic variables have been visualized. In general, the soil of the Northern region of the cultivation contained more nutrients: nitrate & phosphate. Also, the pH seemed higher at Northern region. These results may be explained by the location closely to the ditch. The ditch has relatively high nutrient concentrations and the acidity seems way more alkalic than the cranberry cultivation. These abiotic factors can influence the growth and development of vegetation. In order to monitor the development of vegetation in later successional stages, measuring these abiotic factors is recommended. Also, abiotic factors can serve as an extra explanatory variable, since the design of the cranberry cultivation at Oud Ade may be seen as pseudo replication.

Carbon accumulation at Krimpenerwaard

The aim: (3) 'Does cranberry support the main reason of raising water tables: mitigating climate change and soil subsidence. Are cranberries able to induce the process of carbon accumulation.' is covered by the carbon accumulation results of the successful cranberry cultivation at Krimpenerwaard. Evidence for the activity of the carbon accumulating process was found when analyzing the soil content. However, the elevation measurement contradict the expectation and are questioning this conclusion.

The soil carbon percentage was significantly higher on both sampling depts when cultivating cranberries successfully compared to the grassland and unsuccessful cranberry fields. Also, the bulk density of successful cranberry fields was significantly lower compared to grassland and unsuccessful cranberry fields. Unsuccessful cranberry fields were following the trend of grasslands on both soil depts and for both variables. Calculations of the Soil Organic Carbon in tons/ha weren't possible due to the lack of information about the absolute soil horizon dept. Despite that, low bulk density and high carbon content of the soil already indicate an enhanced carbon accumulating process on with cranberry cultivated fields.

However, the soil surface elevation between 2016 and 2023 showed a decline at Southern region and remains constant at Northern regions of the cultivation. Especially, the successful cranberry field (field 4) seemed to subsidize at a large rate, which conflicts the idea of expanding soils when the accumulation process is activated. An explanation of the subsidizing soils may be the effect of seasonal change on soil structure. Soil moisture and bulk density differs during the seasons. Usually, the moisture percentage is relatively higher and the bulk density lower during winter. During summer, some of the moist will evaporate, therefore the soil becomes more compacted (Franzluebbers, Hons, & Zuberer, 1995). The elevation measurements were performed in August 2016 and February 2023. Therefore, the seasonal change may affect the actual elevation of the soil surface.

For further research, it is suggested to perform an elevation measurement each year at the same month. This will outcompete the seasonal variation. Furthermore, it may be interesting to support the elevation measurements with respirational activity measurements. Elevation measurements are a snapshot of the actual process, while flux chambers monitor the variation of carbon influx and efflux of the soil continuously.

Carbon accumulation Oud Ade

This study also provides a null-measurement of the soil characteristics of the cranberry cultivation at Oud Ade. The bulk density of the cranberry cultivation at Oud Ade already follows the same trend as the bulk density of successful cranberry fields at Krimpenerwaard.

The bulk density of the peat soil of the different cranberry cultivation treatments already follows the same trend as the well-developed cranberry field at Krimpenerwaard. Because, both, Krimpenerwaard and Oud Ade, had a bulk density between 0.20 g/ml and 0.30 g/ml.

A comparison between the peat layers of the grassland and the cranberry treatments was performed at the same horizon (5 & 25 cm dept at the cranberry treatments and 40 & 60 cm dept at the grassland). The only difference was the 35 centimeter thick clay layer, which has been removed at the cranberry cultivation, but is still present at the grassland. The bulk density of clay soil (5 cm dept and 25 cm dept at the grassland) is generally higher than that of peat soil. If the peat layer of grassland and different cranberry treatments were compared in terms of bulk density. The peat layer of the cranberry treatments had a significant lower bulk density than the peat soil of the grassland. This difference may be explained by excavation of the top layer composed of clay, or the absence of heavy machines and cattle on the soil. A heavy top soil exerts a persistent compression on the deeper soil layers, whereas machines and cattle generate a fluctuating compression on the soil. The effect of heavy machines can be seen at deeper layers, than the effect of trampling (Hamza & Anderson, 2005). To provide an explanation for this difference in bulk density, further investigation is required.

The null-measurement at the cranberry cultivation at Oud Ade can be improved. This study only analyzed soil bulk density due to the lack of proper materials. However, soil bulk density does not provide enough information to state that the carbon accumulating process will follow the same trend as the cultivation at Krimpenerwaard. Therefore, it needs to be supported by measuring the carbon percentage of the soil and the soil surface elevation. So, this null-measurement provides a clear basis for further investigation, but cannot state anything about the activity of the peat forming process at the cranberry cultivation at Oud Ade. However, it is expected that the peat moss treatment of this cultivation is able to induce the process of peat formation, therefore it is important to include the different treatments in further research.

General conclusion

In general, this study contributes to the lack of knowledge of cranberry cultivation at the Dutch peat meadow areas and provides recommendations for further investigation of this crop. Further research is required to provide more evidence for the aims (1,2,3) and investigate the situation of later successional stages. The pioneer stage of a cranberry cultivation at the Dutch peat meadow area was very positive in terms of biodiversity. Especially, the addition of peat moss introduced seeds of peat-specific species to the cranberry cultivation. Therefore, peat moss may be able to induce the development of a stable peat meadow ecosystem, when soil conditions remain optimal for this sensitive species. Unfortunately, this study cannot state anything about the carbon accumulating process of the pioneer stage. However, the soil analysis at the well-developed cultivation at Krimpenerwaard detect an activated peat forming process. Although, the elevation measurements remain inexplicable.

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Appendix

Abiotic variables

Nitrate concentration of the cultivation at Oud Ade is shown in Figure 18. Phosphate concentration of the cranberry cultivation at Oud Ade is shown in Figure 17.

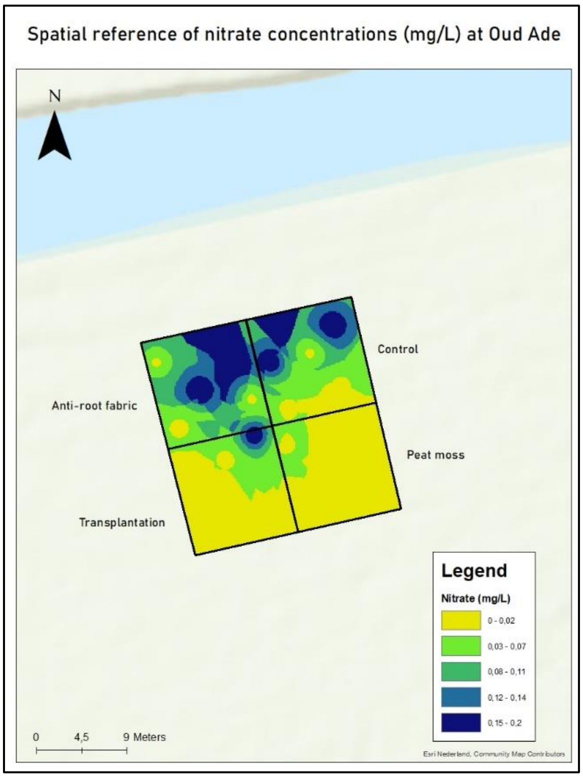

Figure 18: Nitrate concentration in mg/L of the cranberry cultivation at Oud Ade. Measurements of April 2023.

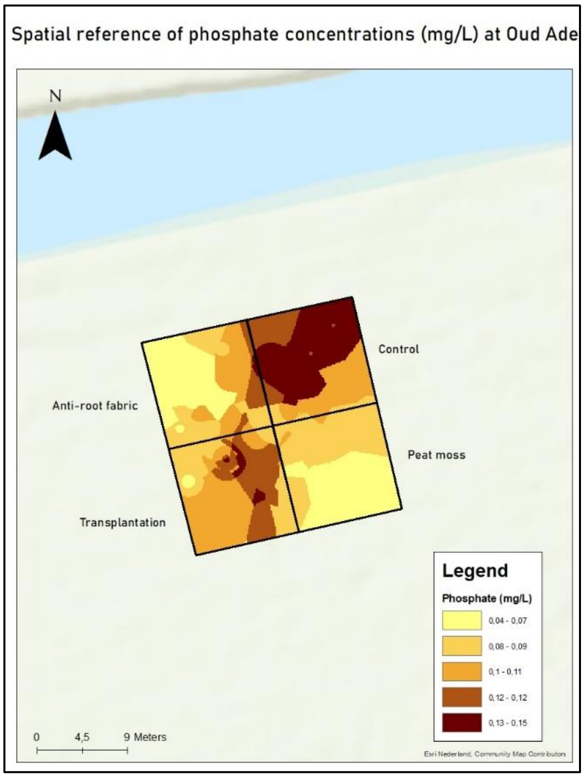

Figure 17: Phosphate concentration in mg/L of the cranberry cultivation at Oud Ade. Measurements of April 2023.

The pH of the cranberry cultivation at Oud Ade were monitored regularly. They are shown in Figure 19 for the months April, May and June 2023.

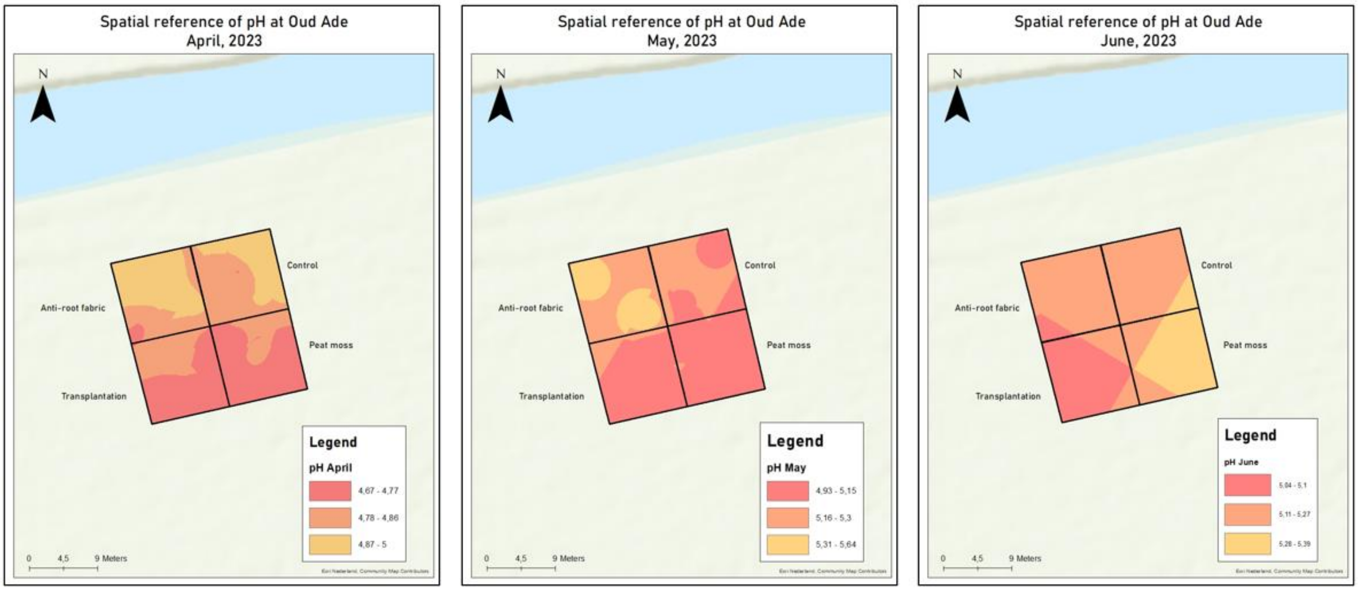

Figure 19: Soil pH of the cranberry cultivation at Oud Ade. Measurements of April, May and June 2023.

The soil moisture percentage of the top layer of the cranberry cultivation at Oud Ade was measured regularly. Figure 20 shows the soil moisture for the months April, May and June 2023.

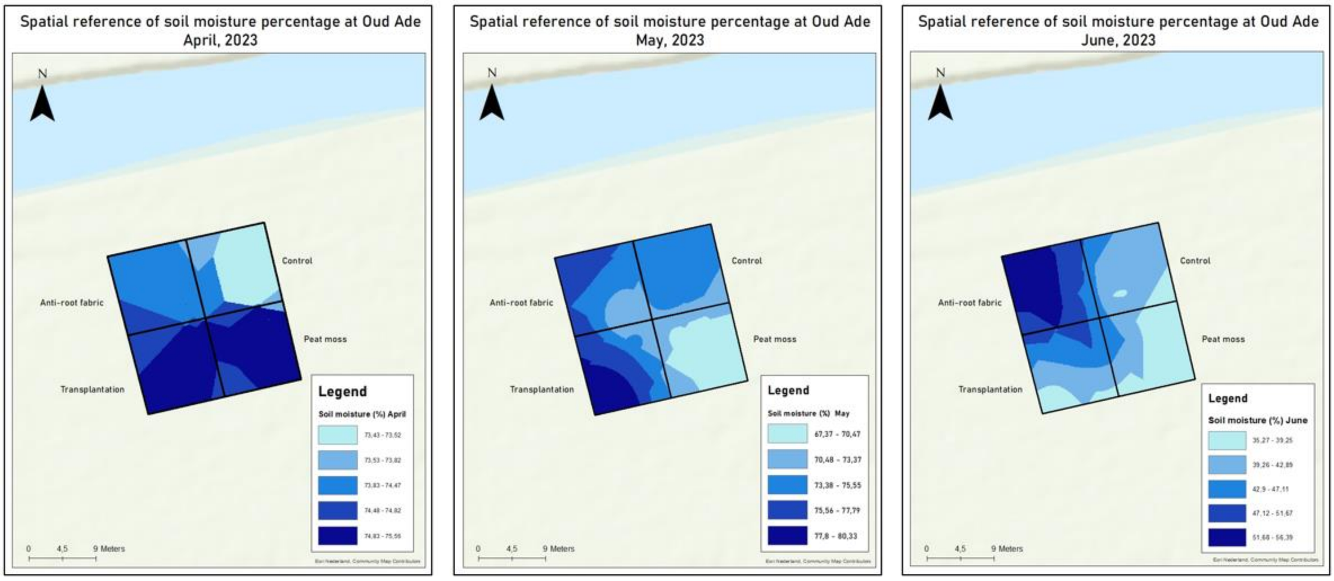

Figure 20: Soil moisture percentage of the cranberry cultivation at Oud Ade. Measurements of April, May and June 2023.

Complete protocols

Protocol: Analyze Soil samples

1. Weight the aluminum container using a scale before putting in the soil.

2. Put the samples in the aluminum container, so that everything fits in. And measure the weight again.

3. Dry the samples by 105 °C for 24 hours.

4. Measure the weights of the samples again and calculate the bulk density and soil moisture.

$$Bulk\ Density = \frac{dry\ mass\ (g)}{volume\ (ml)}$$

$$Soil\ moisture(\%) = \frac{mass\ before\ (g) - dry\ mass\ (g)}{mass\ before\ (g)} \times 100\%$$

5. Take a proportion of the dry samples, for example 10 grams.

6. Measure the weight of a porcelain container, then put the 10 grams of dry soil in it.

7. Measure the weights again, using a scale.

8. Put the samples inside a furnace by 550 °C for 2 hours.

9. Measure the weights of the samples to calculate the organic matter concentration and mineral concentration.

$$SOM(\%) = \frac{dry\ mass\ (g) - residue\ mass(g)}{dry\ mass\ (g)} \times 100\%$$

10. Calculate the proportion of carbon inside the sample by using the conversion factor.

$$Carbon\ (\%) = \frac{SOM(\%)}{1{,}724}$$

Box 2: Protocol for analyzing bulk density soil samples using a furnace.

Colorimetric chemical test: Nitrate and Phosphate

FreshWater Watch

1. Take a sample of water.

2. Rinse the cuvet 2 times with sample water, before filling it until the line displayed on the cuvet.

3. Pull the yellow colored pin out of the tube to create a hole.

4. Remove approximately half of the air volume inside the tube by squeezing it.

5. Keep the tube squeezed while putting him inside the cuvet. The hole downwards.

6. Release the tube, so that the tube sucks up the water inside the cuvet.

7. Shake the tube gently until all powder is dissolved.

8. NO_3: Place the tube at the white paper, and read the values after 3 minutes

   PO_4: Place the tube at the white paper and read the values after 5 minutes.

Box 3: Protocol colorimetric chemical test kits for Nitrate and Phosphate (originating from FreshWater Watch).