Impact of management intensity on the plant diversity and soil carbon of grasslands in different agro-climatic regions of Sweden

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Department of Landscape Architecture, Planning and Management
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Abstract

Biodiversity loss is a global issue and within the EU and Sweden, funding is available for practices that can enhance biodiversity. Semi-natural grasslands, either cut or grazed, have proven to host a wide range of biodiversity that can serve important eco-system functions. While dependent on human management through cutting or animal husbandry, it is reported that intensive management can be detrimental to biodiversity. Over- or under grazing, fertilizing, and soil disturbance (ploughing) can damage key species in grasslands.

Land management also affects soil carbon. Globally, soil carbon stocks are being depleted due to unsustainable land conversion and land management. For example, 90% of semi-natural grasslands in Sweden have been converted to arable land or production forest within the last century. Generally, grasslands, natural and semi-natural, are reported to sequester and stock significant amounts of atmospheric carbon, making them a valuable resource in mitigating carbon dioxide levels in the atmosphere. Fertilizing grasslands can boost the carbon sequestration, but at the same time can be negative for biodiversity.

The aim of this thesis was to better understand how varying management intensity affects plant diversity and soil carbon under different climate conditions. This research is based on data gathered on different locations in the south and north of Sweden, which may show differences in the results, since the role of climatic differences in these properties is well known.
The results showed that extensively managed fields did host a wider plant diversity in the fields in the south of Sweden compared to more intensively managed fields, but not significantly so in the north. The fields in the north did however host more species on average than the fields in the south. No significant difference between neither management intensity nor climatic differences was seen with soil carbon, perhaps because of the fields closeness to *steady-state*.

It is discussed whether there may be a trade-off between managing grasslands for carbon sequestration and biodiversity, and that it may be more reasonable to manage the remaining semi-natural grasslands in Sweden and Europe for optimal biodiversity, rather than carbon sequestration. Carbon sequestration may be better targeted at converting carbon depleted arable lands to grasslands which likely already holds nutrients in the soil.

Keywords: plant species richness, pasture, meadow, soil C, subarctic, humid continental, oceanic, ploughing, fertilization, temperate, continental

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Abbreviations

EU	European Union
EXT	Extensive management
INT	Intensive management
LI	Less Intensive management
N	Nitrogen
P	Phosphorus
SOC	Soil Organic Carbon
SOM	Soil Organic Matter
%C	Percent carbon in soil

1. Introduction

The European Union (EU) has clear goals of increasing sustainable agricultural practices in order to stop biodiversity loss (European Commission 2021). The Common Agricultural Policy (CAP) proposed for 2021-2027 (currently postponed to 2023) includes aims of greener agricultural practices, and within the animal husbandry sector specifically mentions diverse grasslands with low-intensity management as a way to increase biodiversity of both flora and fauna. Crop rotations with leguminous crops are also mentioned as a practice that may be implemented to contribute to achieving the sustainability goals of the EU. For the purpose of "carbon farming" (i.e., sequestering and maintaining carbon in the soil and plant cover), establishing and maintaining permanent grasslands, as well as a vast use of these, is suggested as a method (European Commission 2021).

In Sweden, environmental aid can be applied for through the Swedish Board of Agriculture by farmers maintaining pastures and ley (both defined as farmland that is not arable and is maintained with grazing and/or mowing, and mowing, respectively) (Jordbruksverket 2020). On the pasture or lay which the farmer receives environmental aid, no pesticides, fertilization, tillage, sowing, or other management that could potentially harm natural or cultural values of the land, is allowed. The incentive for the funding is explained by the potential of the grasslands to maintain biodiversity which benefits pollination and decreases pests, as well as maintaining cultural and natural landscapes (Jordbruksverket 2020).

There are many types of grasslands, determined by the way they are managed, their age and stability (Allen et al. 2011). Such varieties include annual-, permanent-, and temporary grasslands. Leys, meadows, and semi-natural grasslands are further specification taking into account the origin of the species composition and the management intensity. Definitions of grasslands terminology can be found in Allen et al. (2011). Grasslands occur both naturally in some parts of the world (UCMP 2019), and semi-naturally defined as areas kept from growing into its natural succession of forest by management of mowing and/or grazing (Lemaire 2011; Tälle 2018; Bengtsson et al. 2019). Semi-natural grasslands host some of the widest plant diversity in Europe, and are the habitat of different animals, both relevant for the eco-system functions for crop production. Grassland vegetation depends mainly on low nutrients and light conditions, therefore management towards low nutrient content and the avoidance of dominant species shading the less-dominant species, sustains and maintain the habitat. Grazing and/or mowing with the subsequent removal of biomass provoked a low retention of nutrients in the soil, and the grazing or cutting keeps the vegetation short and favors species adapted for this type of management.

Since the advent of modern agriculture, traditional management of grasslands has largely been abandoned, with for example 90% of grasslands in Sweden having been converted to arable land or forest, leading to a decline of the habitat hosting a wide range of species and eco-system services such as natural pollination (Eriksson et al. 2002; Tälle 2018; Bengtsson et al. 2019).

Semi-natural grasslands in Sweden are typically mowed or grazed, or a combination of both. Mowing is typically done once a year, at the end of summer, or twice a year, at the beginning and end of summer, or less frequently, such as every other year (Tälle 2018 and sources therein). Furthermore, grazing has the added aspect of animals selectively choosing their food and trampling the plants. This often makes grazed grasslands more heterogenous than mowed grasslands, where all plants are treated more equally by the machinery. Pastures or hay-production grasslands resulting from ploughing and sowing of non-native grass species for the purpose of high yield, are known as *improved grasslands* (Bengtsson et al. 2019). They are often fertilized and intensely managed. The global increase in meat consumption is being met by an increase in production of fodder on arable land or on improved grasslands (Naylor et al. 2006).

However, intensification of grasslands for the purpose of high yields has a negative impact on biodiversity. Biodiversity loss is the decrease of biodiversity within a species, an ecosystem, an area, or globally on earth (Rafferty 2019). Although often associated with the extinction of a species, i.e., species richness, biodiversity loss has affects beyond that. A decline in a population of a single species can have effects on the ecosystem functions of an area, thus perpetuating further decline of the species (e.g., harder to find mates) or other species in one way or another dependent on that species. Natural biodiversity loss happens both regularly, due for example to seasonal changes, and sporadically due to natural disturbances. However, these losses are generally overcome quickly compared to biodiversity loss caused by humans. Habitat loss, invasive species, overexploitation, pollution and climate change are identified as key drivers of the mass extinction of biodiversity seen globally today. Habitat loss, typically through conversion of wetland, grassland and forests to urban or agricultural land is seen as perhaps the main cause for the biodiversity loss (Rafferty 2019).

Semi-natural grasslands, although dependent on human management through the practice of mowing or husbandry of grazing herbivores is, as mentioned above, reported to host a wide biodiversity. Conserving these habitats can help mitigate the decline of biodiversity (Heinsoo et al. 2020). Additionally, all types of grasslands maintain and sequester carbon in great amounts, thanks to deep roots and continuous growth and thus make up an important carbon stock globally (Conant et al. 2017; Mctavish et al. 2021).

Global carbon stocks in the soil are greater than atmospheric and live plant material carbon combined (Jackson et al. 2017; Stockholms Universitet 2017). The largest contributor to soil organic carbon (SOC) is the degradation of below-ground plant material, contributing five times more to SOC than above-ground plant litter. Soil organic carbon is lost to the atmosphere when soils are managed unsustainably, such as by intense tilling and bare fallow. The thawing of permafrost due to climate warming is another contributor to lost SOC, but researchers suggest that if soils under human management (70% of SOC is stored in soils under human management) is improved, the loss of carbon dioxide to the atmosphere from thawing permafrost can be mitigated. Less tillage, extensive grazing all year round, addition of plant litter or compost, and the planting or utilizing of perennials are suggested as management techniques to achieve this (Jackson et al. 2017; Stockholms Universitet 2017).

The EIP-AGRI (Agricultural European Innovation Partnership) focus group "Grazing for Carbon" was a panel of experts put together to discuss the knowledge and experience about the relationship between grazing and soil carbon (EIP-AGRI 2018). In the final report of the project, several needs for further research were identified, one of them being the need for greater understanding about regional differences in optimizing plant species mixtures in grasslands, in order to benefit biodiversity and promote carbon sequestration.

An ongoing European research project – Bioinvent – has made research on grasslands in different locations in Europe, where one aspect of interest was to analyze differences between grasslands in favorable and less-favorable agro-climatic regions. The favorability of the regions could depend on temperature, precipitation, altitude, or other factors. In Sweden, the favorable location was in the south, and the less favorable in the north, separated by a geographical distance of about 1200 km.[1] The average temperature has a difference of 5°C, and the precipitation is higher in the south (Table 1).

Table 1. The annual mean temperature (Temp), precipitation (Rain), mean minimum (Min.Temp) and maximum (Max.Temp) temperature and altitude (meters above sea level) for each agro-climatic region included in the sampling for the Bioinvent survey. Abbreviations F = favorable, and LF = less favorable.

	F	**LF**
Temp (°C)	8	3
Rain (mm)	703	602
Min.Temp (°C)	-3	-11
Max.Temp (°C)	20	19
Altitude (masl)	99	112

1.1. Aim and research question

1.1.1. Aim

The aim of this thesis is to compare data on plant diversity and soil carbon in grasslands within two different agro-climatic regions of Sweden, and with

[1] Information shared by Ana Barreiro, SLU.

different management intensities. It may be a contribution to the knowledge about the impact of regional variation in grassland management in order to improve soil quality and biodiversity, both goals of the EU and Sweden.

1.1.2. Research question

How does management intensity of grasslands affect soil carbon and plant diversity, and do these variables differ between southern and northern Sweden? How can these results contribute to the knowledge of regional differences of grassland management within the EU and Sweden?

1.2. Background

1.2.1. Management effects on plant species richness

Management of grasslands can involve a range of different inputs, such as ploughing, sowing, fertilizing with inorganic and/or organic fertilizers, and varying stocking rates (i.e., grazing animals per unit area).

Fertilization

Plant species diversity is generally considered to be negatively affected by fertilization as fertilizers favors dominant, non-specialized species that outcompete other species, whereas low availability of nutrients favors specialized plants that allow for a broader diversity (Gaujour et al. 2012 and sources therein). Fertilization has a strong negative effect on plant species richness correlated with increasing amount of fertilizer (Müller et al. 2016). Both nitrogen fertilization and even deposition of atmospheric nitrogen, that has increased significantly due to industrialization, pose a threat to biodiversity of both semi-natural and natural habitats globally (Phoenix et al. 2006).

Although management intensity is commonly reported to have an effect on both soil carbon and biodiversity, where fertilization benefits soil carbon sequestration (Conant et al. 2001, 2017), but is negative for biodiversity (Gaujour et al. 2012), this is not always clearly shown. In a study of pastures in northeast United States, plant diversity was measured with relation to management input, soil properties and environment (Tracy & Sanderson 2000). The results from this study showed that only soil phosphorus and percent soil organic matter (%SOM) had significant effects on plant diversity, with soil phosphorus having the more robust effect, inversely related to plant diversity.

Similarly, in a study of alpine hay meadows it was concluded that in the interest of plant diversity, soil phosphorus levels were to remain low (Marini et al. 2007). Phosphorus remains longer in the soil after cessation of it as fertilization, compared to nitrogen, resulting in slower increase in plant diversity after the cessation of P compared to N (Willems & Van Nieuwstadt 1996). A study of 501 grasslands in Europe found that higher levels of soil phosphorus was consistently associated with low plant diversity, noting that soil P deserves more scientific attention when considering plant species richness, similarly to the attention given to negative effects of nitrogen fertilization (Ceulemans et al. 2014). In this study, currently fertilized fields were not included, but rather the varying phosphorus levels in the soil were a result of phosphorus pollution (high levels of soil P as a result of previous excessive fertilizer use). The cumulative and long-lasting nature of P also means that high biodiversity cannot be expected in fields with high P after transforming to an agro-environmental scheme. Previous fertilization with phosphorus may be a cause for the results in the study by Tracy & Sanderson (2000), where fields were previously arable. It has been shown that even 35 years after cessation of fertilizing, plant species richness was still lower compared to unfertilized controls (Heinsoo et al. 2020).

Thus, phosphorus has a negative effect on plant species richness, even long after its stopped use as fertilization. On the other hand, nitrogen fertilization may increase carbon sequestration rate but is detrimental for plant species richness due to outcompeting grassland specialist (Gaujour et al. 2012). Reducing both nitrogen and phosphorus fertilization is proposed as the best way to maintain plant species richness (Ceulemans et al. 2013).

Grazing

Grazing of grasslands by herbivores, such as cattle, goats, sheep, and horses is a method of sustaining grasslands and keeping them from overgrowing with shrubs and trees. Grazing, as opposed to cutting, lets the animals selectively eat what they prefer, but it is important to maintain a grazing pressure that does not lead to over grazing or trampling.

In a study in Switzerland, low-intensity cattle grazing was introduced on previously mown subalpine meadows to analyze how this introduction affected the plant diversity (Fischer & Wipf 2002). The cattle were introduced up to 50 years previous to the study. It was found that even low-intensity grazing had a negative impact on the plant diversity in the meadows. Some suggested explanations for this, as it contradicts similar studies from lower altitudes (Schläpfer et al. 1998), is that the plants in the subalpine meadows may be specialized for the climate and previous management (cutting), and the introduction of grazing cattle affected these plants negatively. An example of such plants are dwarf shrubs of the Ericaceae family, being damaged by cattle. Species better adapted to grazing became more prominent and with greater coverage (Fischer & Wipf 2002).

On the other hand, opposite results were found by (Schläpfer et al. 1998), who found that grazed fields had a slightly higher plant species richness than mowed field, which was attributed to “micropatterns of grazing, trampling and dung deposition”. In this study, 90% of species were the same in the grazed fields as in the mowed, and thus it is concluded that only a small number of species is affected by this management practice. The difference in altitude between the two studies may play a role in the contradicting results, where the first was in subalpine regions (Fischer & Wipf 2002), and the second was at low altitude (Schläpfer et al. 1998).

Another study of alpine plant diversity looked at environmental factors and management intensity in the northwestern alps of Italy. It was found that the highest impact on plant diversity was from environmental factors (elevation and precipitation), and not on management intensity, whereas the conclusions were reversed regarding pastoral value (PV). PV is based on an index of specific quality for each plant species, taking into account "productivity, morphology, structure, palatability and preference by livestock", and then using a formula to give a total pastoral value score from 0-100. The researchers conclude that in order to maximize plant diversity and forage quality, a livestock stocking rate that reduces the negative effect of under- and overgrazing, and instead is in equilibrium with the vegetation carrying capacity must be adopted (Pittarello et al. 2020).

Finally, in a study of remnant and restored grasslands in the Stockholm archipelago, it was shown that cattle grazing supported the dispersal of grassland specialist species from remnant grasslands (i.e., ancient grasslands) into the adjacent restored grasslands (i.e., former arable fields converted to grassland), compared to fields where cattle did not graze. The remnant fields showed the highest benefit of grazing to the grassland's specialist species, compared to restored grasslands where more competitive species often outgrow the specialist. Grazing cattle can also be vectors for seeds and aid the establishment of specialist through disturbance to other species. The small islands of the Stockholm archipelago proved a valuable area to research the dispersal of plant biodiversity in fragmented areas (Kapás et al. 2020).

It seems that the introduction of grazing herbivores must be considered with regard to the previous management methods and regional variations. Grazing can have positive effects and contribute to plant diversity, but it may not always be positive on meadows traditionally mowed, as the trampling can damage certain species.

1.2.2. Management effects on soil carbon

In line with the EIP-AGRI focus group *Grazing for Carbon,* much research shows the potential of permanent grasslands to sequester atmospheric carbon. According to Conant et al. (2001), "grasslands can act as a significant carbon sink with improved management". This was based on worldwide studies of grasslands of different types, where management methods were compared to the soil carbon. Fertilization, improved grazing, conversion from cultivation and native vegetation, sowing of legumes and grasses, introduction of earthworms, and irrigation were studied as management methods. The management methods intended for increasing forage production also increased soil carbon.

In a follow-up study, Conant et al. (2017) looked at several hundreds of more studies since their previous analysis, and found similar results, except that native lands converted to grasslands did not show a significant increase in soil carbon. Irrigation, fertilization, sowing of legumes and grasses, and improved grazing, all showed increased soil C. Converting arable land to grasslands also increased soil C. The rates of accumulation of soil C were between 100-1000 kg per hectare and year.

In a report published by the Swedish Board of Agriculture (Karltun et al. 2010) it is noted that Swedish grasslands are generally reported as sequestering much less carbon than other countries. It is argued that Swedish grasslands, often under management subsidies by the environmental aid mentioned above, are not fertilized, sown, or otherwise managed for the purpose of increasing forage, and thus do not sequester as much carbon as they potentially could. Much of the flora found on the semi-natural grasslands in Sweden is dependent on the management inflicted by grazing cattle, and low nutrients in the soil. Thus, it is noted in the report that there may be a negative relationship between increasing carbon sequestration and maintaining plant biodiversity.

Due to the goal of the EU of increasing carbon sequestration, but also arguing for the benefit of biodiversity on pastures, it is of interest to see how these goals relate to each other.

An important factor for determining the potential for a grassland to sequester and store carbon is how close the field is to *steady-state*. *Steady-state* is the theory of SOC (soil organic carbon) finding equilibrium between how much goes into the system, versus how much goes out of it (through mineralization). The equilibrium is dependent on a constant and unchanged addition of organic matter. Change in addition of organic matter or changed environmental factors such as temperature or moisture can offset the balance (Karltun et al. 2010). In a study of SOC in temperate regions of Australia, researchers showed that crop management techniques such as no-till, stubble retention, and crop rotation performed on arable land previously having been used as pasture could, at best, maintain SOC levels. Tillage had the largest effect on decreasing SOC. Only in crop rotations that incorporated pasture did levels of SOC increase. In fields with SOC far below the assumed *steady-state,* introduction of permanent pastures had the potential to increase SOC with 500-700 kg of carbon per hectare and year (Chan et al. 2011).

According to a study of grasslands and their atmospheric gas exchange (Allard et al. 2007), there may be a trade-off between carbon sequestration and emissions of CH_4 (methane) and N_2O (nitrous oxide). The study, measuring atmospheric gas exchange on a divided field showed that extensive grazing, i.e., no nitrogen fertilizer applied, and with half the stocking density of the intensively managed counterpart, decreased its effect as a GHG sink (greenhouse gas sink) after 1 year. The intensively grazed field (fertilized and grazed to target sward height of 6 cm) continued instead to increase its function as a GHG sink over time. The decrease in soil nitrogen levels in the extensively grazed field after two years may be an explanation for this decrease in carbon sequestration. Thus, as the researchers conclude (Allard et al. 2007), there is support for the hypothesis presented by that managing grasslands for improved carbon sequestration may increase emissions of CH_4 (methane) and N_2O (nitrous oxide). While the CH_4 emissions did not differ between individual animals in the two systems, the fact that there were twice as many animals on the intensively managed fields resulted in a higher CH_4 emission per unit area (Allard et al. 2007).

N_2O (nitrous oxide) emissions from fertilizers can be minimized by optimizing the composition and time of application of the fertilizers. Clayton et al. (1997) studied the N_2O flux from different fertilizers applied to grasslands, and found that environmental factors like temperature and precipitation, as well as time of application impacts the flux of different fertilizers. Referring to a combination of an application of urea in spring, and two application of ammonium nitrate in the summer giving a lower emission factor than the two fertilizers used separately, the authors suggest that a varied fertilization scheme, adapted for soil type and crop, as well as climatic conditions, could be employed to minimize N_2O emissions from agricultural lands (Clayton et al. 1997).

In studies looking not at permanent pastures but rather comparing crop production with ley in crop rotations, some interesting data was found. Above-ground plant residue of straw left after a harvest of monocrop cereals has a lower impact on SOC build-up than below ground humification of dead plant material input from ley (Börjesson et al. 2018). Accordingly, root-derived carbon contributed about 2.3 times more to stable SOC than above-ground biomass in a long-term study from the Swedish University of Agriculture in Ultuna (Kätterer et al. 2011). In the study by Börjesson et al. (2018), nitrogen fertilizer did not show an increase in yield of ley, which may be explained by nitrogen-fixing clovers compensating the difference in N-input. Hence, unfertilized leys with nitrogen-fixing clovers also sequestered similar amounts of carbon as fertilized leys in the study. In cereals however, nitrogen fertilization doubled the yield and significantly increased the input of C to the topsoil. The study was conducted in two places in Sweden with different soil profiles; in the loamy soil at Lönnstorp, the *subsoil* sequestered more carbon than the clayey soil at Lanna, which may be due to deeper roots in the loamy soil than in the clayey soil (Börjesson et al. 2018).

In summary, carbon sequestration can be positively affected by nitrogen fertilization and a stocking rate in balance with the vegetation growth. However, climatic differences such as precipitation and temperature, as well as the timing of fertilization also affects the carbon sequestration. With increased nitrogen fertilization, grazing may have to be intensified to keep up with the increased growth, thus leading to higher total methane emissions from the herbivores as well as nitrous-oxide emissions from the fertilizer itself. Conversion from arable land to ley seems to have a significant benefit to carbon stocks in the soil, as roots of ley contribute to carbon stocks far more than above-ground plant residue left after monocultures.

2. Materials and method

2.1. Materials

The material for this thesis was gathered in collaboration with researchers at the Swedish University of Agriculture (SLU) who obtained that data within the European project Bioinvent. Soil samples and plant inventory of grasslands in Sweden was realized during the summer of 2017, at vegetation peak growth in different locations in Skåne (herein after also *South*) and around Umeå (Västerbotten) (herein after also *North*). The data was collected within fields of varying intensity of management: "intensive" (ley in a crop rotation with added nutrients), "less intensive" (permanent grassland with added nutrients), and extensive (permanent grassland without added nutrients). The samples were organized in a gradient, and the researchers classified them to facilitate data interpretation. Thus, in some cases the researchers perception of the fields were taken into account when classifying the fields, and some fields are outliers to the above mentioned classification.

The sometimes-irregular classification becomes evident in the present thesis as the factors of management intensity studied here are not the only factors determining the classification in the original data of the Bioinvent-team. It was decided to use the original classification due to the fact that an interpretation, as well as first-hand information from farmers may contribute more to a realistic interpretation of management intensity than solely a couple of data points (years since ploughing and amount nitrogen fertilization).

2.1.1. Description of data collection

Two regions in Sweden were chosen to represent *favorable* and *less favorable* agroclimatic conditions (as is the purpose of the Bioinvent project). The regions chosen were Skåne (Scania) in the south of Sweden (~55 deg lat.), representing favorable agro-climatic conditions, and around Umeå in the north (~64 deg lat.), representing less-favorable agro-climatic regions. For each region, a total of 36 fields were chosen, of which 12 were classified as *intensely managed,* 12 as *less-intensely managed,* and 12 as *extensively managed*.

The data was collected in the peak growth season of the summer of 2017. In each field, a standardized study unit with four 4 m^2 subplots were established. In the four corners of each subplot, the top 20 cm of soil were gathered with a 2,5 cm diameter auger. The 16 sub-samples were then mixed to obtain a composite sample per field. In the center of each subplot, one soil core for determining soil bulk density was taken as well. Plant species were counted within a 50x50 cm square in the center of each of the four sampling areas per field. Grasses were not individually identified but rather counted as one group.

For the calculation of the soil organic matter, 300 mg of air-dried and milled (<2mm, IKA A10) soil was burned in a muffle kiln (550°C, 3 h, Nabertherm B400) in nickel capsules, and the total soil organic matter (SOM) content was determined by loss on ignition. For measurement of total phosphorus, the remaining ash from the 300 mg of burned soil used for the SOM measurement was digested in 10 ml 13 % HNO_3 (1:4) and analyzed using ICP-OES. Carbon (C) concentrations were measured in air-dried milled soil (<2mm, IKA A10), using a FLASH 2000 elemental analyzer (Thermo Scientific).

The climate data was collected using CHELSA (Climatologies at high resolution for the earth's land surface areas), currently hosted by the Swiss Federal Institute for Forest, Snow and Landscape Research WSL. The data is free to download on their website, and is aimed for researchers (Chelsa Climate 2021).

2.1.2. Classification of management intensity

The researchers of Bioinvent classified the sampling fields according to the management intensity: "intensive" (ley in a crop rotation with added nutrients), "less intensive" (permanent grassland with added nutrients), and extensive (permanent grassland without added nutrients). However, due to other factors noted during the sampling, there were a few outliers in the data, such as three fields regarded as *Extensive* due to very long history of extensive grazing, but that did add some fertilization (organic and inorganic). The information regarding years since ploughing and fertilizer use was realized through a questionnaire handed out to the farmers. Due to varying availability of information to the farmers regarding historic management of the fields, the samples were organized in a gradient, and the researchers classified them to facilitate data interpretation. As the data for this thesis is limited to that gathered by the Bioinvent researchers, their classification will be accepted as reasonable and legitimate.

2.2. Method

2.2.1. Literature study

Relevant studies were mainly gathered through the database *Web of Science*.

Search terms used: plant diversity, plant species richness, biodiversity, carbon sequestration, soil organic carbon, soil carbon, SOC, carbon, ley, grassland, pasture, grazing, meadow, management, management intensity, fertilizer, ploughing, extensive, intensive, phosphorous.

The search results were sorted by relevance, a function within the database, and in some cases, results were refined to studies from Europe or Sweden. In studies citing other research that seemed relevant for my purposes, the study may have been accessed through the link provided within the study or accessed through Google Scholar. Information from the EU or Swedish governmental bodies were found and accessed either through an internet browser, or through their own webpage. In select cases, previous knowledge of publications was used to retrieve those, such as the report *Inlagring av kol i betesmark (Karltun et al. 2010)*.

2.2.2. Limitations

The fields researched by the Bioinvent research team were chosen for being classified as permanent or temporary grasslands. This means that for example ploughed fields currently used for a short period (1-2 years) of ley in a crop rotation were excluded. Thus, the comparisons of this thesis are limited to fields that are not considered arable but yet varying in management intensity for the production of animal fodder.

The number of fields within each degree of management intensity for the north and south respectively was 12, and there was a rather large variation in type of management on these fields. For example, there were some meadows in the north, i.e., fields not grazed by animals, that were classified as *extensively managed,* whereas all fields in the south classified as *extensively managed* were grazed by animals, but yet some were cut in addition to grazing. This practically limits the factors of analysis to the three categories of management intensity (*intensive, less intensive, extensive*).

It became evident through the research that variation in grass species potentially makes up a substantial part of the overall plant species diversity, however, grass species were not counted as individual species but rather as a group. As grasses were present in all fields, the data may state “number of plant species except grass”.

2.2.3. Data analysis

ANOVA

The data was processed in the computer statistical program Minitab.

A One-way ANOVA (analysis of variance) was used within Minitab to test the null hypothesis *all means are equal* against the alternative hypothesis *not all means are equal*. The test assumes equal variance. The significance level was $\alpha = 0.05$. Any result with a *p-value* lower than 0.05 meant the null hypothesis could be rejected. The “pooled” standard deviation was used to calculate the intervals.

The reasonability of using a One-way ANOVA was based on the residual graphs showing a normal distribution.

Within the One-way ANOVA calculation, *Tukey Simultaneous Tests for Differences of Means* (Tukey's method) shows between which groups of variables a difference lies and gives an individual score of significance between the means of specific groups. This is useful in order to show between which management methods there was a significant difference of the given variable, rather than showing an over-all significance based on all data points. For example, if there was a significant difference between *extensive* and *intensive,* but not between *intensive* and *less-intensive*, this would be shown by *extensive* being given a different letter than *intensive* and *less-intensive*.

In order to present the data in a clear and understandable fashion, interval plots were used. Interval plots are a function within Minitab that uses individual standard deviation to calculate the intervals. Thus, the individual standard deviation visualized in the graphs is not the same as the "pooled" standard deviation used with in Tukey's method.

Pearson Correlation

Pearson Correlation coefficient is used to determine if there is a correlation between two continuous variables, and whether the relationship is positive or negative, or null (Kent State University Libraries 2021). If the value is close to 1 it is considered a perfect positive correlation, and if it close to -1 it is considered a perfect negative correlation. Interval degrees of correlation are shown in *table 2*.

Table 2. Intervals for strength of correlation from a Pearson Correlation coefficient. r = the sample coefficient.

Correlation degree	**Values interval**
Perfect	± 1
High	$0.5 < \lvert r \rvert$
Moderate	$0.3 < \lvert r \rvert < 0.5$
Low	$0.1 < \lvert r \rvert < 0.3$
None	0

Shannon Weaver Diversity Index

Shannon Weaver Diversity Index is an index that shows the evenness of distribution, and is commonly used to compare diversity in different habitats (Ortiz-Burgos 2016). It can be used to assess the dominance of a single species, indicated as a low score if the dominance is high, or a high score if the distribution of the species within the habitat is more even. Normal values are 1.5 to 3.5, only rarely going upwards of 4.5. The formula used to calculate the index is

$$H' = - \sum (pi \times ln\ pi)$$

where H' is the diversity index, pi is the proportion of each species in the sample, and $ln(pi)$ is the natural logarithm of this proportion.

3. Results

3.1. Management impact on species diversity

Figure 1 shows the relationship between management intensity and the number of species except grasses.

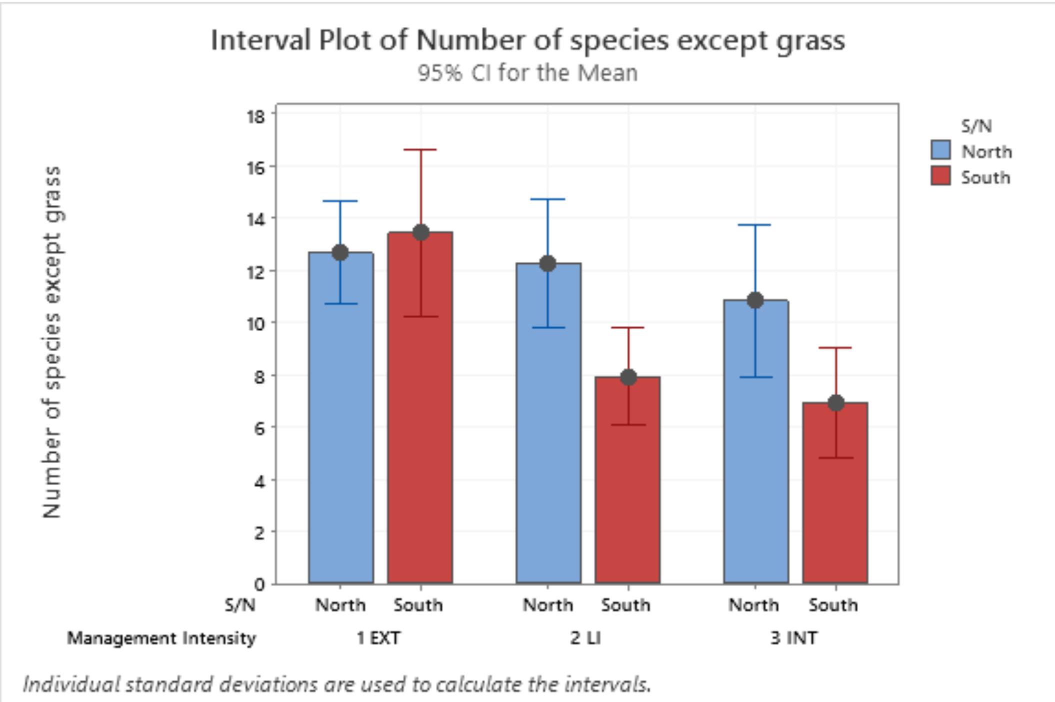

Figure 1. Interval plot of number of species except grass for the three management intensities (1 EXT = extensive; 2 LI = less intensive; 3 INT = intensive) in the North and South.

The results of *Tukey's method* for the number of plant species except grasses in the South, show that extensively managed fields have a significantly higher amount of plant species, than both less-intensive (*P=0.004*) and intensively managed fields (*P=0.001*). There is no significant difference of the number of plant species except grass between the less-intensive and intensively managed fields (*P=0.803*). There is no significant difference of number of plant species except grass between the three management intensities in the North (LI – EXT *P=0.963*; INT – EXT *P=0.488*; INT – LI *P=0.649*).

A Oneway-ANOVA test calculated for each management intensity if there was a significant difference between the mean values of the north and south. There was no significant difference between the means of the extensively managed fields (EXT *P=0.663)*, but there was a significant difference between the means of the less-intensively and intensively managed fields (LI *P=0.005* and INT *P=0.025*), where in both cases the south had a lower number of species.

Figure 2 shows the relationship between management intensity and Shannon Weaver Diversity Index.

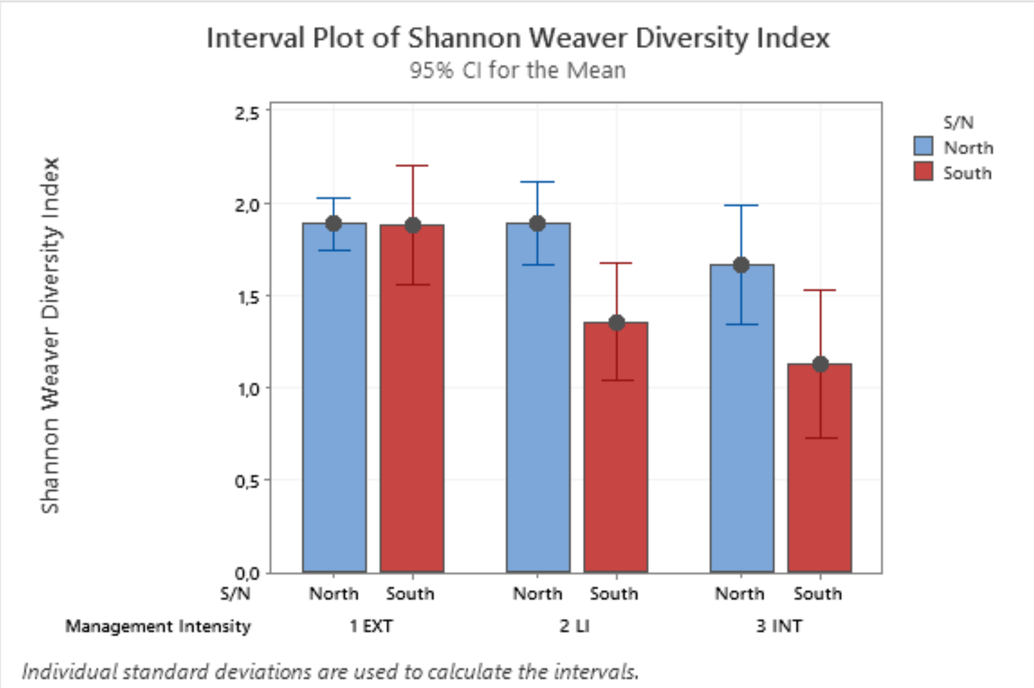

Figure 2. Interval plot of Shannon Weaver Diversity Index for the three management intensities (1 ETX = extensive; 2 LI = less intensive; 3 INT = intensive) in the North and South.

The result of the Shannon Weaver Diversity Index presented in *Figure 2* shows that the extensively managed fields in the South had a significantly higher diversity index score than the intensively managed fields (*P=0.005*) when calculated with *Tukey's method*. The less-intensively managed fields in the South were not significantly different from either the extensively (*P=0.063*) or the intensively (*P=0.569*) managed fields. In the North, the Shannon Weaver Diversity Index score between the three management intensities did not differ significantly (LI – EXT *P=1.000*; INT – EXT *P=0.339*; INT – LI *P=0.334*).

A Oneway-ANOVA test calculated for each management intensity if there was a significant difference between the mean values of the north and south. The pattern of the results was very similar to those of number of species; there was no significant difference between the extensively managed fields (EXT *P=0.971*), but there was a significant difference for both less-intensively and intensively managed fields (LI *P=0.006* and INT *P=0.030*), where in both cases, the south had a lower Shannon Weaver Diversity Index, indication higher dominance of a species.

The correlation between the total amount of soil phosphorus and the number of years since the last ploughing event with the number of species, was significant for the samples from the north (*Figure 3* and *4*), but not for the south.

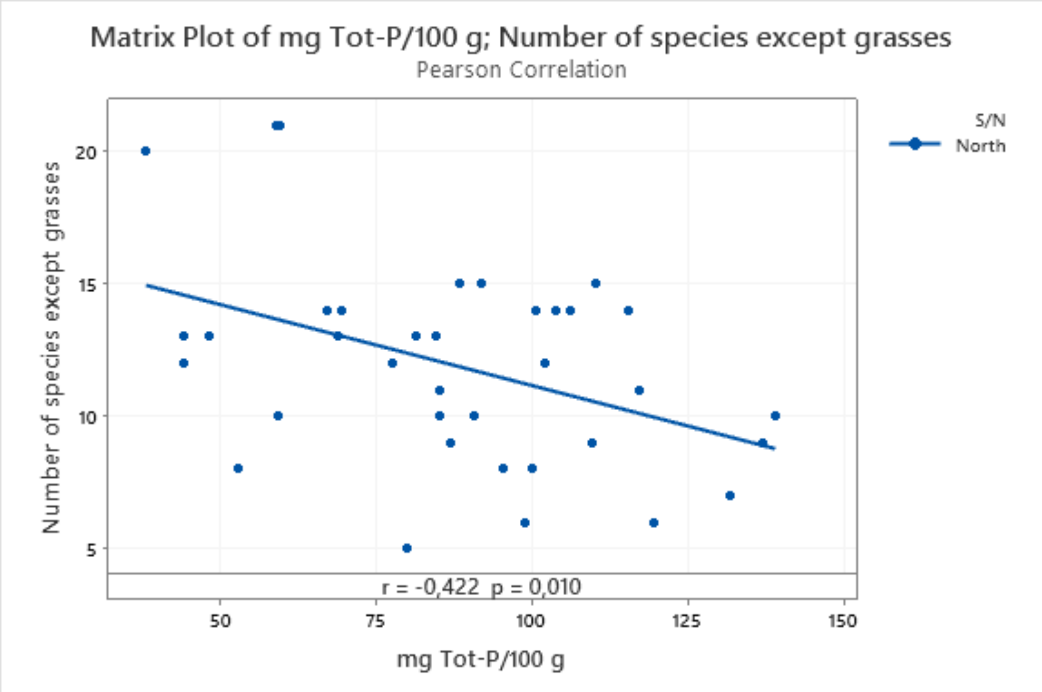

Figure 3. Matrix plot of samples from the North showing the correlation of number of species except grasses and mg of Tot-P/10 0g of soil.

Figure 3 shows that in the north, there is a moderate degree of negative correlation (r = -0.422) between the amount of total phosphorus in the soil and the number of plant species except grasses. No significant correlation was observed in the south (data not shown).

Figure 4 shows a moderate positive correlation of the number of plant species except grasses with the years since ploughing, but only in the north (r = 0.330). The fact that the majority of samples are sub 25 years makes the result less robust.

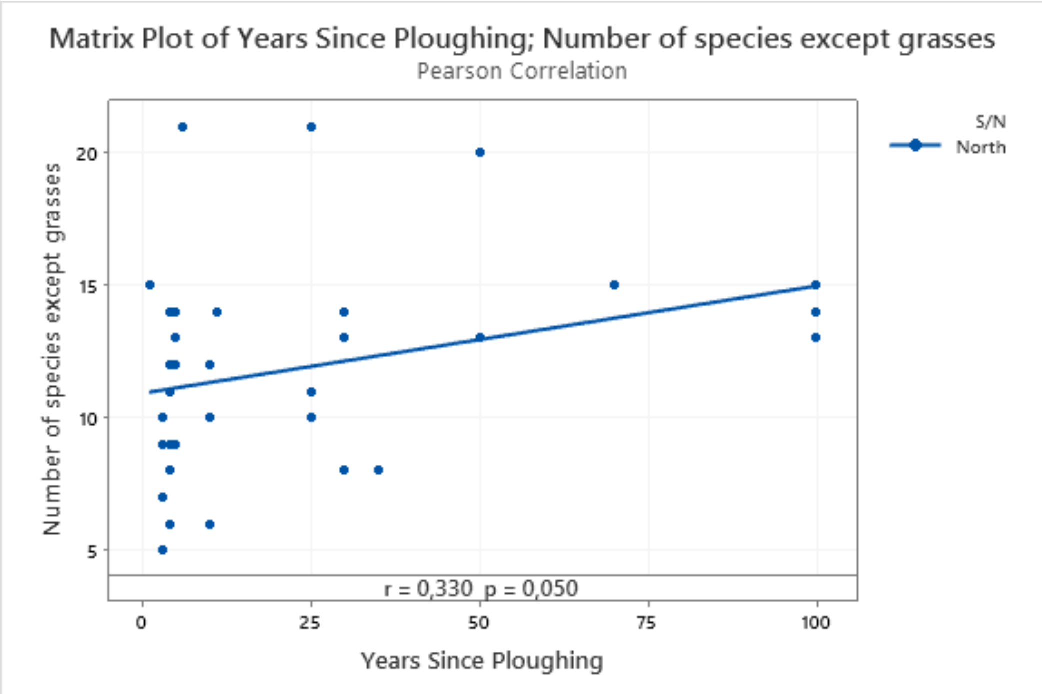

Figure 4. Matrix plot of samples from the North showing the correlation of number of species except grasses and years since ploughing.

Finally, the application of nitrogen as fertilizer did not show a significant effect on plant species richness in neither the south, nor the north (*figure 5*).

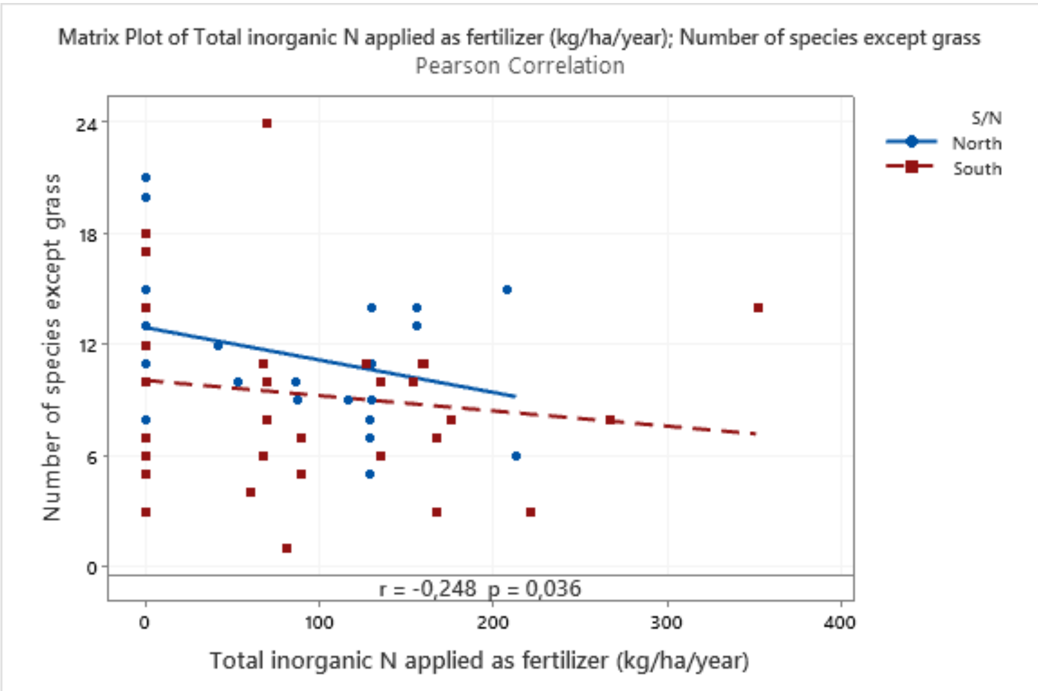

Figure 5. Matrix plot of all samples showing the correlation of number of plant species except grasses and total inorganic N applied as fertilizer.

3.2. Management impact on soil carbon

Figure 6 shows the amount of carbon in the soil under different management intensities.

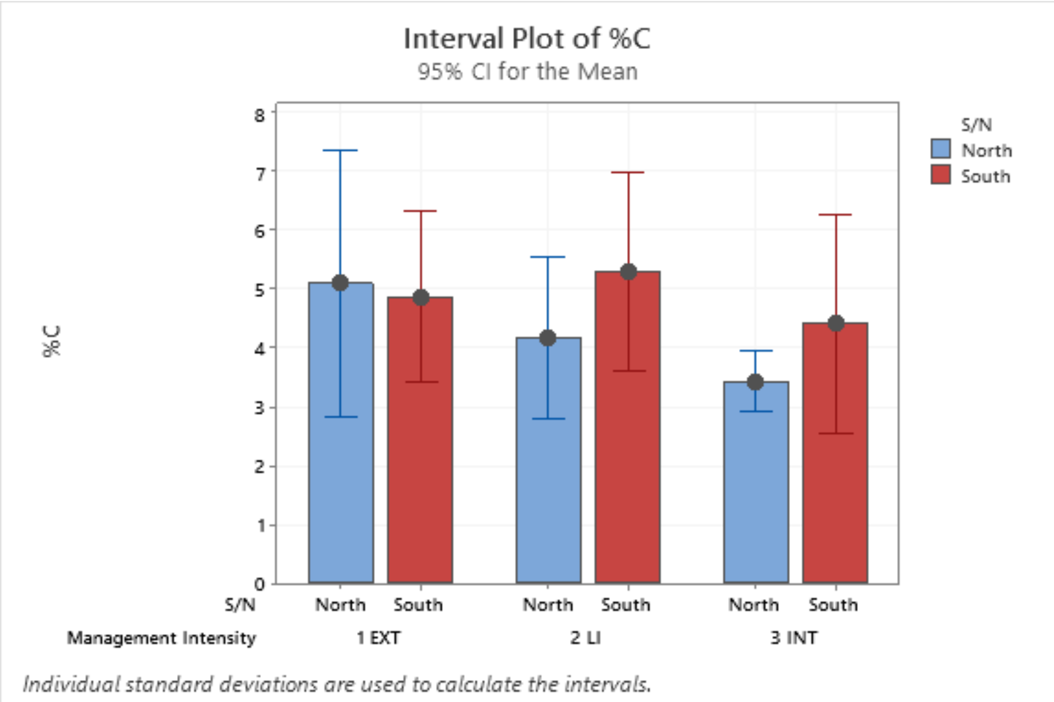

Figure 6. Interval plot of %C (percent soil carbon) for the three management intensities (1 ETX = extensive; 2 LI = less intensive; 3 INT = intensive) in the North and South.

The percentage of carbon in the soil (%C) had no significant difference between the management intensities, not in the South nor in the North, with the *Tukey's method*. The *P-values* for the North are (LI – EXT *P=0.629*; INT – EXT *P=0.233*; INT – LI *P=0.740*). For the South, the *P-values* are (LI – EXT *P=0.917*; INT – EXT *P=0.907*; INT – LI *P=0.693*).

The means of the north does indicate a trend of lower soil carbon with increased management, although the significance does not show this. The lack of significance when calculated with the Oneway-ANOVA is likely because of the high variance seen in the extensive samples, and to some degree the less-intensive.

A Oneway-ANOVA test calculated for each management intensity if there was a significant difference between the mean values of the north and south. Between none of the management intensities was there a significant difference between the means of the north and south (EXT *P=0.852*, LI *P=0.270*, INT *P=0.274*).

Figure 7 and *8* shows the correlation with soil carbon to the time since ploughing (*figure 7*) and total nitrogen fertilizer added (*figure 8*). The *r*-values and *p*-values shown in the graphs are calculated combining both the north and the south. There was no significant effect of these management strategies (time since ploughing or nitrogen fertilization) on soil carbon, and the same was true for the north and south calculated individually.

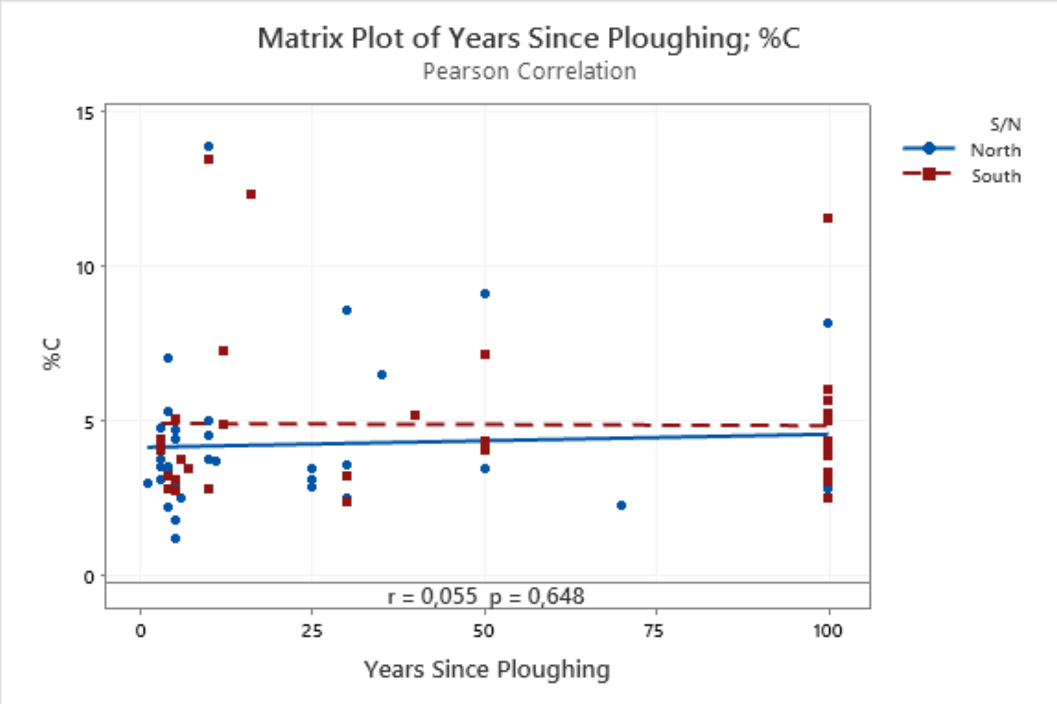

Figure 7. Matrix plot of all samples showing the correlation of %C and years since ploughing.

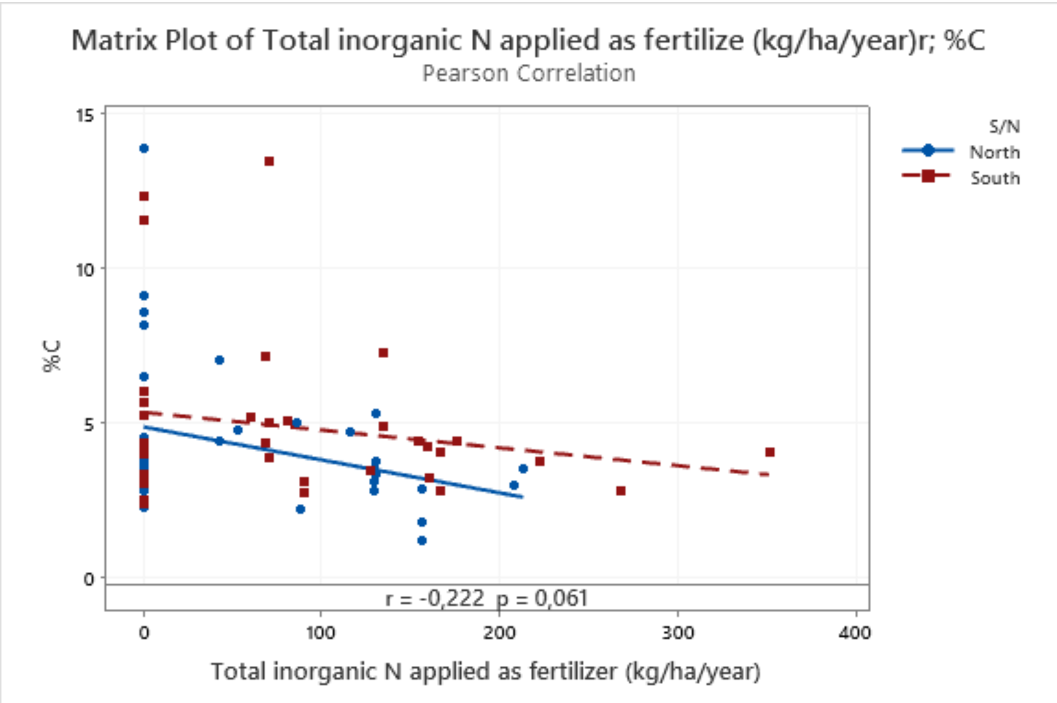

Figure 8. Matrix plot of all samples showing the correlation of %C and Total N added.

3.3. Climatic conditions impact on plant species richness and soil carbon

The following graphs compare annual precipitation and annual mean temperature to soil carbon and plant species richness. All management intensities from both the north and south are included. Precipitation (*figure 9*) and mean temperature (*figure 10*) do not demonstrate a significant correlation with the total soil C.

Annual precipitation was not significantly correlated to plant species richness (*figure 11*). *Figure 12* does show low negative correlation of annual mean temperature to plant species richness, with the north demonstration slightly higher species richness on average.

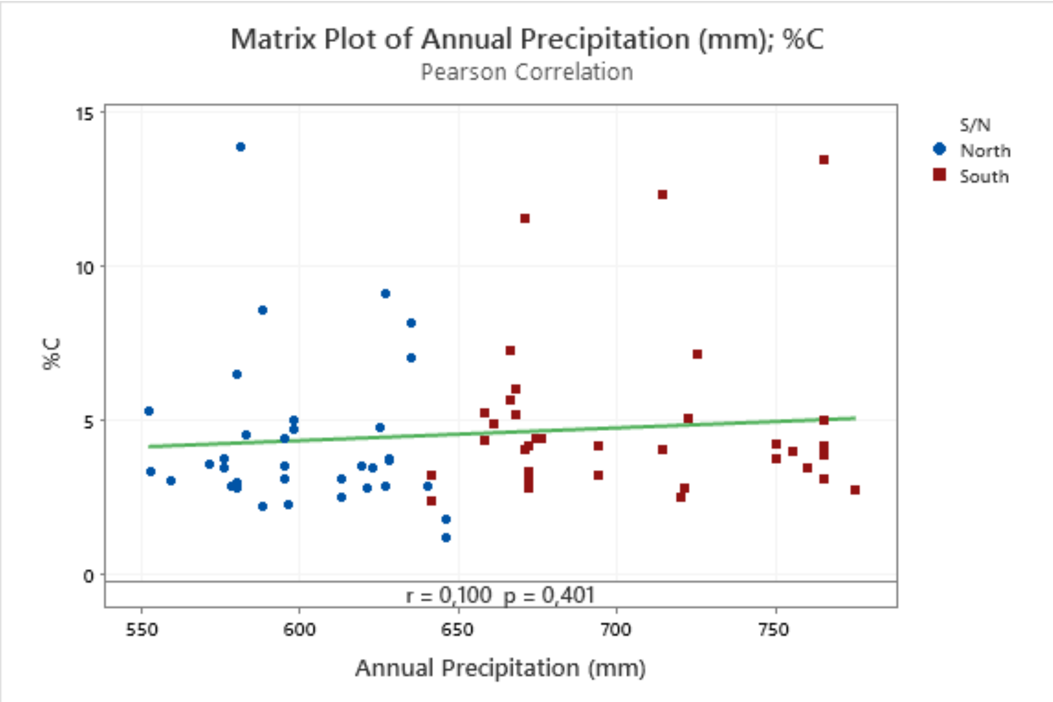

Figure 9. Matrix plot of all sites showing annual precipitation (mm) compared to percent soil carbon.

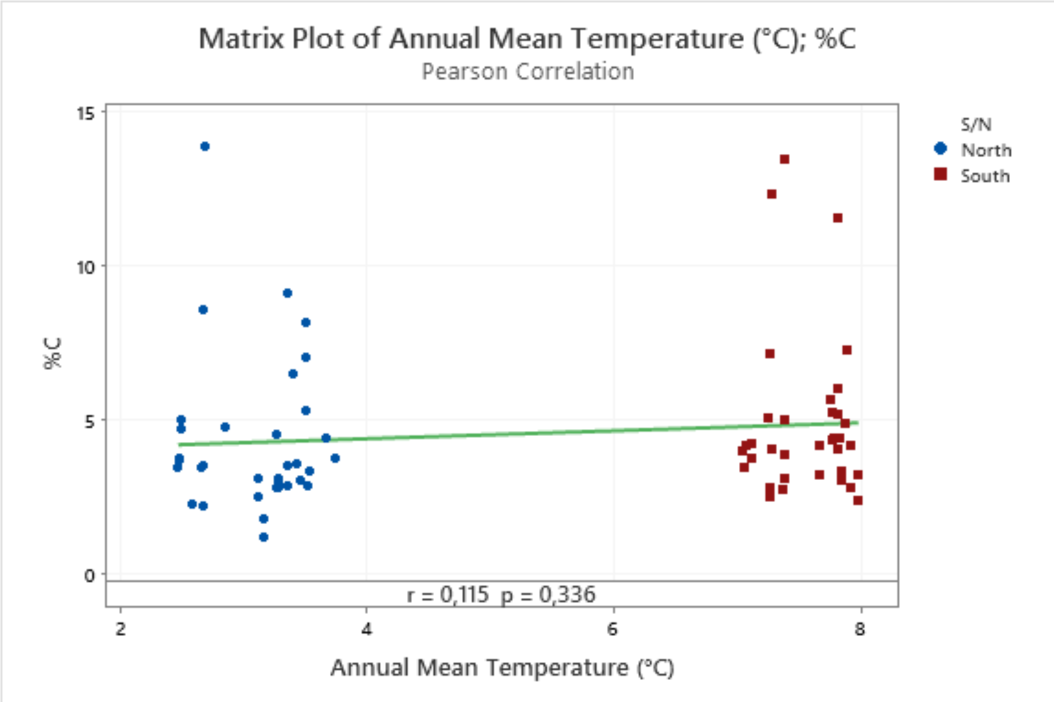

Figure 10. Matrix plot of all sites showing annual mean temperature (°C) compared to percent soil carbon.

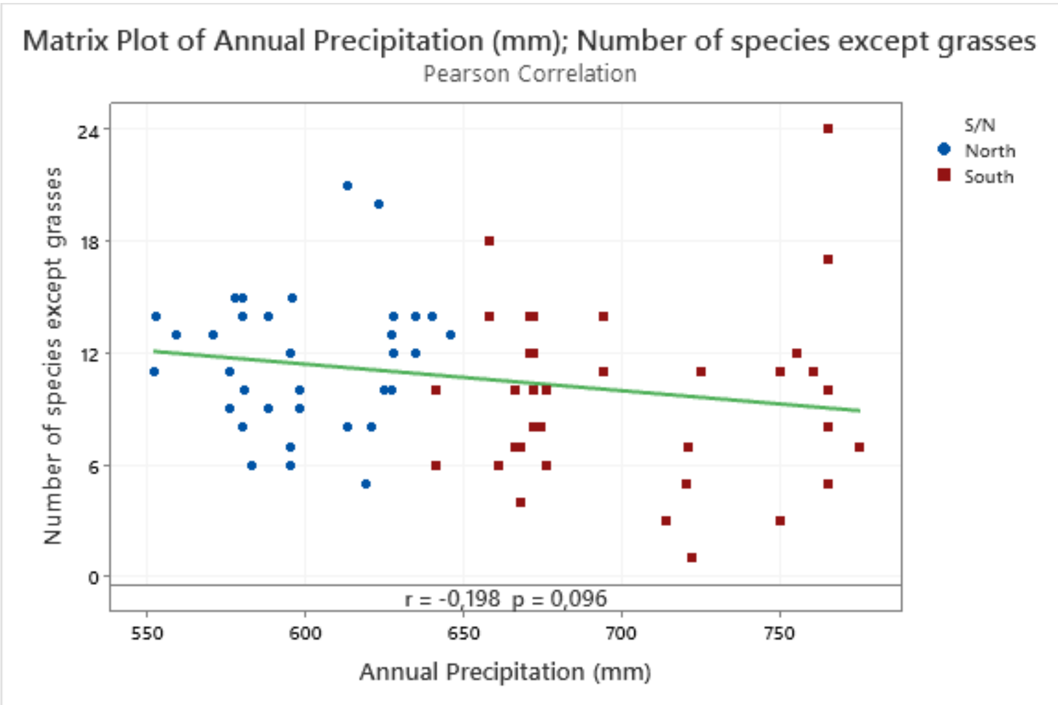

Figure 11. Matrix plot of all sites showing annual precipitation compared to number of species except grasses.

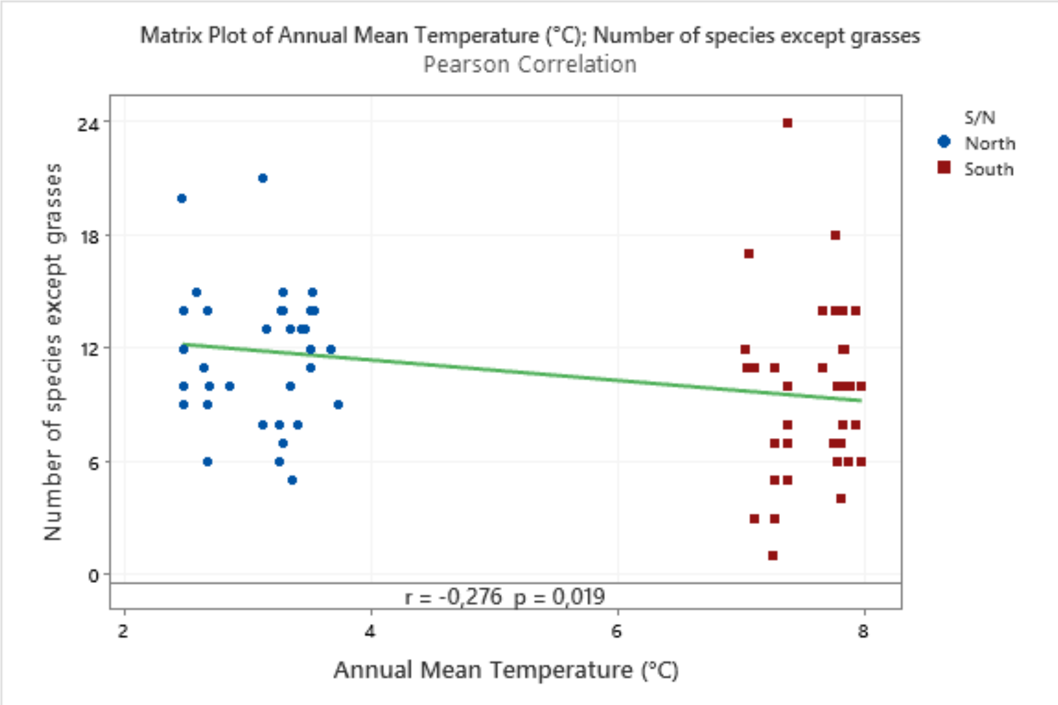

Figure 12. Matrix plot of all sites showing annual mean temperature compared to number of species except grasses.

4. Discussion

4.1. Interpretation of results

Of all the results, significant differences were mainly seen in plant diversity (plant species richness and Shannon Weaver Diversity) between the different management intensities in the south, but also that the north was generally higher than the south (*Figure 1; 2*). In the north, soil phosphorus and time since ploughing each had a moderate degree of correlation to plant species richness, but the same was not seen in the south (*Figure 3; 4*).

A possible reason for the insignificant difference between species richness among the management intensities in the north could be that the less-intensive and intensive fields may be managed comparatively less intensively than the corresponding fields in the south. This could be due to smaller farms in the north, with fewer animals. Fewer animals, smaller machines, and a shorter grazing season due to the climatic differences (and thus less grazing pressure per year) could all be factors explaining the little difference of plant species richness between the management intensities in the north. Another contributing factor could be the generally more mosaic landscape in the north, with more forest and natural landscape surrounding the grasslands, as compared to the south where much of the surrounding land is likely conventionally managed arable land, or otherwise has a previous history of human management. With surrounding conventionally managed arable land, there may be a lack of habitats that harbor plant species, and there may also be spillover of herbicides into the grasslands. Smaller farm sizes supporting more biodiversity than large farms was found in a study in the southeast of Sweden (Belfrage et al. 2005). Observations of the surrounding landscape from the sites were recorded by the Bioinvent-team, and it seems that the south has many more fields with cereal crops surrounding the grasslands, whereas the north is more commonly surrounded by other grasslands, ley, forest and rivers, perhaps indicating a wider range of habitats for biodiversity.

The results of how management intensity and climatic differences (precipitation and average temperature) affects soil carbon were not significant in any case. In the literature, fertilizing and avoiding ploughing is often reported to support increasing soil carbon. The results did not demonstrate a correlation between time since ploughing and soil carbon, but this could possibly be explained by all sample fields in this study being relatively long-lasting, even the intensively managed ones. The reports of high carbon sequestration rates are usually shown when converting arable monoculture land to grassland (Chan et al. 2011; Conant et al. 2017), in other words when the field is far from *steady-state*. It is likely that the fields in this thesis are not as carbon-depleted as intensively managed crop fields can be, and thus closer to *steady-state*. Compared for example to the soil organic carbon content reported in trials of fields with ley-rotations or monocultures in Sweden (Börjesson et al. 2018), the carbon content in the fields of this thesis were on average higher, regardless of management intensity. The fields in this thesis having comparatively high carbon content regardless of management intensity, thus being closer to *steady-state,* could explain the lack of significant variations between them.

With all plots included, climatic differences between the north and south of Sweden did not demonstrate significant effects on neither soil carbon nor plant species richness (except a very low negative correlation of annual mean temperature and plant species richness). It has been found that precipitation has a significant positive effect on the number of plant species in grasslands (Pittarello et al. 2020), as well as increasing annual precipitation showing a negative correlation to soil carbon sequestration (Derner & Schuman 2007), neither of which were significant correlations observed in this study. It may be that the sample size in this thesis was too small, and with too much variation between the fields of the north and south to draw any conclusions on whether or not climatic differences between northern and southern Sweden should play a role in determining optimal management to benefit either plant diversity or soil carbon. It may also be the case that the climatic differences between the southern and northern fields in this thesis are not large enough to cause significant differences demonstrated by other studies, such as Pittarello et al. (2020).

4.2. Relation to previous knowledge

The results in this thesis show an increase of plant diversity with extensive management (in the South) that is in line with both other studies (Gaujour et al. 2012; Müller et al. 2016) and with the Swedish Board of Agriculture argument for the implementation of extensively managed grasslands (Jordbruksverket 2020). Interestingly, this trend was not observed the North. Notably, though the north did not show a significant difference between management intensities, it was overall higher in plant diversity (richness and Shannon Weaver Diversity) than the south on the less-intensively and intensively managed fields, and about the same for extensively managed fields.

Although this study did not measure carbon sequestration in the soil, as it is based on a one-time sampling. It could be expected that continuous management that in other studies are said to promote the buildup of carbon in the soil (fertilization and no-till), would show higher amounts of carbon compared to fields with less favorable management for carbon sequestration (Allard et al. 2007; Chan et al. 2011; Conant et al. 2017). In the current study, the less-intensively managed fields were generally not ploughed for at least 10 years but were fertilized. As these practices are associated with carbon sequestration, it may be expected that the less-intensively managed fields would have high amounts of carbon in the soil, but a significant difference were not observed in this study.

There was an inverse correlation between the amount of phosphorus in the soil and the number of plant species, but this was only observed in the North (*figure 3*). A negative correlation between soil phosphorus and plant species richness is in line with other research showing this relationship (Willems & Van Nieuwstadt 1996; Tracy & Sanderson 2000; Marini et al. 2007; Ceulemans et al. 2014). The fields in the north, especially the less-intensive and intensive fields, applied significantly more phosphorus to the soil than the south, due to a higher use of organic fertilizer. In the current study, also the time since ploughing had a moderate correlation with the amount of plant species as seen in *figure 4*, again only in the north. On average, the fields in the north were ploughed more recently than the fields in the south (average 25 and 50 years ago respectively), possibly explaining the correlation being observed only in the north. Recovery of plant species richness from ploughing is reported to take centuries to millennia before returning to the former state of old-growth grasslands (Nerlekar & Veldman 2020). Although this may refer more to natural than semi-natural grasslands, it can still be assumed that ploughing has long lasting negative effects on plant species richness.

Nitrogen fertilization did not demonstrate a significant correlation to soil carbon in the north or south. Fertilization has been shown to increase carbon sequestration (Conant et al. 2001, 2017) and it could be expected that increased carbon sequestration would correlate to higher amounts of soil carbon, but again, fertilized fields did not show higher amounts of carbon than non-fertilized in this study. There was more use of fertilizer in the more intensively managed fields, which were fields that were also ploughed more frequently and perhaps had a higher number of annual species compared to more deep-rooted perennial species on permanent grasslands. Nitrogen fertilization was likely used for above-ground production (i.e., fodder) and not for the intent of raising soil carbon. The management accompanying more nitrogen fertilization may have been counterproductive for soil carbon, thus not showing the expected results. The amount of added nitrogen was generally within the range of other studies showing results of increased carbon with fertilization (Börjesson et al. 2018; Heinsoo et al. 2020).

4.3. Implications

The results of this study demonstrate the variety of management techniques under the umbrella-term *grasslands*, and the implications this variety brings along. The initial testament to this is how the researchers of the Bioinvent-team organized the sample fields according to many factors determining management intensity, where a single factor did not suffice to make the distinction between the intensities. Thus, drawing conclusions on how a single input/factor, such as time since ploughing, nitrogen fertilization, precipitation, or soil phosphorus is not possible. Instead, a broader view of management is necessary to give recommendations or otherwise draw conclusions, but this also requires many more samples than were available to this thesis. More samples resulting in a wide spectrum of each variable could potentially provide more robust results and correlations.

Management rules for semi-natural grasslands subsidized by the Swedish Board of Agriculture include not fertilizing and not ploughing the fields (Jordbruksverket 2020). This is in order to promote biodiversity. In the south, extensively managed fields, generally not fertilized and with many years since last ploughing (if ever) did show a higher species richness than the other management intensities. In the north, the same significance was not seen, but there was however a positive correlation between greater species richness and longer time since ploughing, as well as lower soil phosphorus. Soil phosphorus was not a factor included in the classification of management intensity, as it is not in itself a management, but it could suggest that phosphorus fertilization (through organic fertilizer) ought to be avoided as it inevitably increases soil phosphorus, thus negatively affecting plant diversity.

Although the results do not show a particularly robust or causative relationship between management inputs and plant species richness, it seems safe to say that the Swedish Board of Agriculture ought to keep devoting resources to preserving the little semi-natural grasslands still intact. The time it takes for plant species richness to recover from short times of fertilizer inputs makes it not worth the damage it can make to biodiversity (Ceulemans et al. 2014).

The insignificant difference in soil carbon in relation to management and climate on the grassland in this study may suggest that grasslands in Sweden ought to be managed to optimize biodiversity, whereas reaching goals of carbon sequestration could be better reached by conversion of arable land into well-managed grasslands or ley-rotations (Conant et al. 2017; Börjesson et al. 2018).

4.4. Strengths and limitations

This study, based on data gathered by the Bioinvent research team in 2017, compared the classification of management intensity done by the Bioinvent team with plant diversity and soil carbon. The classification was based on time since ploughing, fertilization, and an interpretation of the historic and current management on the fields. Therefore, this study is not conclusive on whether ploughing, fertilization, grazing pressure or any other management in and of itself has an effect on the two parameters plant diversity and soil carbon. Instead, what can be learned from the results is how a gross amount of input from many sources into a grasslands system may affect the parameters plant species richness and soil carbon.

A shortcoming of the study is that grasses were not counted as individual species but rather as one group, as there may be a diverse range of grasses in the fields, specifically grasses especially suited for low nutrient levels and grazing.

History of how fields have been managed historically was not always certain due to information available to the farmers and in turn, the researchers of the Bioinvent team.

4.5. Recommendations

This study has given an indication that at least in Sweden, management intensity of grasslands does not have a linear relationship to plant diversity or soil carbon. Further research including a broader sample collection may make correlations between specific inputs more robust and could aid in deepening our understanding of how to optimize grassland management for specific purposes, be it benefiting biodiversity, sequestering carbon, or providing optimal fodder for livestock. This study did not analyze carbon sequestration (as a change over time) as it was based on one-time samples. Repeating the sampling over many years could paint a different picture on how management impacts the grasslands. As seen in other studies for example, plant diversity takes many years to recover from phosphorus fertilization (Ceulemans et al. 2014). This could be true for the sample plots in this study, but future research would have to follow this development over many years.

4.6. Final remarks

The EIP-AGRI focus-group "Grazing for Carbon" noted a need for greater understanding about regional differences in optimizing plant species mixtures in grasslands in order to promote both biodiversity and carbon sequestration (EIP-AGRI 2018).

In this study, there was some difference shown in the correlation between management intensity in the south compared to the north, possibly showing that regional difference does make a difference in how management of grasslands should be practiced in order to benefit carbon sequestration and plant species richness. However, irregular management methods even within the management classification makes it difficult to draw conclusions.

As noted in this thesis, there may be a trade-off in the management of grasslands for either promoting carbon sequestration or promoting biodiversity. Increased biomass production, resulting in more above- and below-ground organic matter contributes to sequestering atmospheric carbon, and this increase is achieved mainly through fertilization (although the results of this thesis did not support this). However, grazing pressure also plays a role, and a grazing pressure below the plant productivity will inevitability lead to more plant biomass, and in time even to succession to forest. This will however also lead to dominance of species suited toward low or no grazing pressure, which may outcompete much of the plant species diversity more suited to grazing. In promotion of plant species diversity commonly associated with grasslands, the right grazing pressure has to be adopted in order to not over- or under graze, and added nutrients ought to be avoided.

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