Long-term effects of liming and phosphorus application on the root growth conditions for spring barley on a sandy soil

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Front-page photo:	by Julie Therese Christensen 12-04-2017. The photo captures two piles of lime on a grass clover field. The lime is ready for application on the fields.

Contents

Preface

With this thesis, I end my two years master studies in Agro-Environmental Management at Aarhus University. The knowledge that I have gained from the past five years at Aarhus University has served as a great foundation for conducting the research relating to this thesis and making me able to produce a master thesis that I find is highly relevant for today's challenges in sustainable agricultural production. Overall, it has been some interesting years studying what I find highly important in this world: *agriculture*. This work was financially supported by the European Union (Horizon 2020, project: SoilCare, Grant no.677407).

I will now take the freedom thank some people for their help and support during the process. First of all a great thanks to my main supervisor Gitte Holton Rubæk for valuable guidance throughout the process – both when I was on track, and when I was facing frustrations and difficulties. Next, thanks to my co-supervisors, Lars Juhl Munkholm and Hanne Lakkenborg Kristensen for great response and useful critique to my work. Furthermore, a thanks to my co-supervisor and statistician, Rodrigo Labouriau, for helping me through the long process of statistical work.

In order to carry out this thesis, assistance with the experimental work was needed. Technicians from Årslev have assisted the research of minirhizotron; a thanks to Astrid Bergman, Lasse Vesterholt and Bjarne Egedal. Thanks to my TAP-supervisor Stig Rasmussen, who also measured penetration resistance. Thanks to Holger Bak and Michael Koppelgaard who conducted the deep soil cores. A special thanks to Ove Edlefsen for helping out, when I visited St. Jyndevad. All the chemical analysis has been carried out by technicians at AU Foulum. Thanks to Lene Skovmose for guidance on the work with the deep soil cores. A special thanks to Musibau Oyeleke Azeez for great teamwork on the deep soil cores.

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Julie Therese Christensen

List of abbreviations

MR = Minirhizotron

CB = Core-break

P = Phosphorus

N = Nitrogen

C = Carbon

TP = Total phosphorus

PR = Penetration resistance

RF = Root frequency

RI = Root intensity

RT = Reaction number

0.0 = Treatment with no liming added, and with no application of P every year

0.1 = Treatment with no liming added, and with application of P every year

4.0 = Treatment with 4 tonnes lime/ ha, and with no application of P every year

4.1 = Treatment with 4 tonnes lime/ ha, and with application of P every year

8.0 = Treatment with 8 tonnes lime/ ha, and with no application of P every year

8.1 = Treatment with 48 tonnes lime/ ha, and with application of P every year

12.0 = Treatment with 12 tonnes lime/ ha, and with no application of P every year

12.1 = Treatment with 12 tonnes lime/ ha, and with application of P every year

0 P = when no phosphorus was applied every year

1 P = when phosphorus was applied every year

Abstract

This study investigated if optimal long-term application of liming and phosphorus can improve the root growth conditions and potentially increase the yield. The treatments consisted of combinations of four different liming levels and two levels of phosphorus application. Soil physical properties (soil bulk density and penetration resistance) and soil chemical properties (total carbon, nitrogen, phosphorus content and pH) were measured in different depths of the soil. Root growth was measured using minirhizotrons and the core-break method. Observations in the minirhizotron were conducted four times with two weeks intervals starting late May 2016. The core-break was performed on 1-meter deep soil cores taken shortly after harvest. There were significant effects of the long-term treatments on the measured soil chemical and physical properties. No effect on the number of roots at 10 cm depth was found by the core-break method. At 25 cm depth more roots were found in the low liming level when no phosphorus has been applied every year, and the opposite was found when phosphorus had been applied every year. The root frequency increased over time, and the high liming levels had the highest root frequency. The grain yield was found to have an optimum at the medium liming level (4-8 tonnes lime/ha). In summary, optimal long-term application of lime and phosphorus can have a positive effect on the root growth conditions and grain yield on sandy soil.

Summary

Liming is commonly used to increase the fertility of acid soils. Liming can have several beneficial effects on the soil fertility, which includes change of nutrient availability, addition of the plant nutrients Ca and Mg. Furthermore, it can be beneficial for the soil structure. This study investigated the effects of long-term application of different liming levels and two levels of phosphorus on the root growth conditions and grain yield.

The study was conducted in the St. Jyndevad Experimental station in the period May 2016 – August 2016. Combinations of four different liming levels (0, 4, 8, and 12 tonnes lime/ha) and two levels of phosphorus (with and without phosphorus application every year) were used in the study. In selected treatments, minirhizotrons were inserted with 30-degree angle into the soil. Filming in the minirhizotrons was conducted four times with two weeks interval from when the spring barley plants were at the growth stage 29 according to the BCCH scale. After harvest, deep soil cores were collected in all treatments. In the deep soil cores, core-break was performed in five different depths sections to determine root growth. Furthermore, chemical analyses and determination of soil bulk density was performed in all of the depth sections in the deep soil cores. Grain yield was recorded from all plots.

Analysis of the data was carried out using R, and statistical differences were detected with a significance level of 5%. Soil chemical properties (content of phosphorus, nitrogen, and carbon and pH), and soil physical properties (soil bulk density and penetration resistance) had been altered due to the long-term different treatments with liming and phosphorus. Soil bulk density was affected by the long-term liming treatment, but differences were only found when no phosphorus had been continuously applied. The penetration resistance was found to be altered due to the long-term treatment at certain depths of the soil, but differences were not detected in all depths of the soil. Some differences in the content of C, N and P between the different treatments in different depths were found as well, but differences were not detected between all treatments. It was found that increased liming levels had resulted in increased soil pH. The effect on soil pH was found in depth. Since soil pH is known to affect the nutrient availability and toxicity of Al, root growth was expected to be different between the treatments.

The root growth development was found to be different between the different treatments. The analysis of the root frequency found using the observations in the minirhizotrons, showed that there were differences in root frequency at all of the times of observation. The two highest liming levels had the highest total root frequency at the last observation. When analyzing the root frequency in different depth sections of the soil, it was found that all of the treatments ended up

having the same root frequency in the top 20 cm of the soil, but differences were found in the root development. In the 30-50 cm depth section differences in develop in root frequency was also detected, and again the two highest liming levels ended up having the highest. The analysis of the core-break experiment revealed no differences between the treatments in estimated number of roots in 10 cm depth, but differences were found in at 25 cm depth. In the treatments with phosphorus application every year, the estimated number of roots was significantly lowest at the low liming treatment (4 tonnes lime/ha).

The grain yield was affected by the treatments as well. The highest yield levels were found when the soil was limed with 4 or 8 tonnes/ha. A decrease was found when the liming level was above or below this level. This study showed that having optimal liming and P management yield levels can increase. The results also indicated that a larger soil volume was used by the plant roots after long term application of lime and P.

1.0 Introduction

In Denmark and in many other regions around the world, it is common to use liming i.e. addition of calcium carbonate to increase the fertility of the soil. Application of lime increases the pH of the soil and thereby changes the availability of many plant nutrients. Other effects of liming include restriction of availability of aluminum, which can be toxic to plants (Haynes and Mokolobate, 2001). Lime can furthermore increase the soil fertility by improving the soil structure (Haynes and Naidu, 1998) and because it can contain the plant nutrients Ca and Mg (Goulding, 2016). It has been shown that long-term practice with lime in different levels, changes the pH and other soil properties in not only the topsoil, but also in deeper parts of the soil (Rubæk et al., 1998). Aye et al. (2016) also find effects of liming application on sub-soil pH. They furthermore find that this effect is dependent on the duration of the practice, and that the subsoil pH is more affected by longer duration with liming application.

The main functions of roots are to supply the crop with water and nutrients. In this matter root growth and root distribution are crucial factors. Generally, root growth varies between both species and varieties of plants, and their response to different conditions may or may not be similar (Forde and Lorenzo, 2001). The characteristics of the soil as well as the type of crop, and the variety of the specific crop, affect the root system. The availability of the nutrients and thus the supply of nutrients affect the architecture of the root system in the soil (Lopez-Bucio et al., 2003, Forde and Lorenzo, 2001). Long-term repeated liming practice can change this availability of the nutrients in depth by the change in pH and by introducing additional calcium ions to the soil. This in turn may affect root growth by potentially creating a larger volume for the roots to take up nutrients from. That root growth can be restricted by an acid subsoil is discussed by Haynes (1984). He finds that effects in depth of applied lime can be important for root growth in such soils. The root growth, especially below top-soil, is different between soil types (Madsen, 1985). Madsen (1985) finds that spring barley roots on sandy soils have lower root intensity than the roots of crops grown on less sandy soils. Andersen (1986) also finds variations in root growth of spring barley due to different soil types, but found opposite her expectations, that the root growth development to be best on sandy soil than on more clay soil.

Studies on the effects of long-term use of different levels of liming and phosphorus (P) applications on root growth can be done in the long-term experiment in St. Jyndevad Experimental Station on liming and P fertilisation. The experiment started in 1942 and it carries the Danish name "*Vekselvirkningen*" which translates to "*the interaction*". Four different levels of liming have consistently been applied to reach a desired soil pH. Half of all the treatments have received P fertilizer every year. The other half have not received any P fertilizer since the beginning of the experiment (Rubæk, 2008).

The long-term different liming and P fertilisation in this experiment have resulted in clear differences in both soil pH, soil P content, crop growth, and nutrient removal with crops. Furthermore, it has been shown in a previous study that also subsoil properties are influenced by the long term treatments (Rubæk, 2008). Existing literature has rarely described the long-term effects of liming on the subsoil, at least to my current knowledge. In agreement, Rubæk et al., (1998) states that the knowledge of the effects of both liming and P in the subsoil on the P binding mechanisms are unknown. These effects of long-term use of liming can be of great importance to the soil fertility when dealing with soils with natural low fertility such as the studied acid sandy soil, because it may create a larger fertile soil volume for roots to explore. If poor soil fertility restricts root and plant growth, it is likely that a larger volume of fertile soil will affect the crop yield positively. Furthermore, deeper roots may also contribute positively to the improvement of soil properties in the deeper layers, because decaying roots will introduce additional organic material. The overall objective of this project is to increase the knowledge on the effects of the long-term treatments with different levels of liming and P application on the root growth conditions by studying root growth in the long-term field experiment at St. Jyndevad. This thesis aims to investigate how the long-term use of high and low levels of P, and different levels of liming affect spring barleys above- and below ground development in a sandy soil under field conditions. Three working hypotheses set the foundation for this thesis:

1.1 Working hypotheses

1) The physical (soil bulk density and penetration) and chemical properties (content of phosphorus, nitrogen, and carbon, and pH) of the soil have been modified in both topsoil and in subsoil by the different long-term liming and P fertilisation treatments.
2) The differences in soil chemical and physical properties induced by the long-term use of phosphorus and liming will affect the soil as a medium for root growth, and potentially create a larger soil volume favourable for roots.
3) The grain yield and below ground root development will be positively correlated unless the crop experiences stress. If the crop suffers from nutrient limitation, the crop will allocate more energy to root growth development. In toxic conditions, the total crop growth will be restricted.

2.0 Background

2.1 Effects of liming on nutrient availability in acid soils

The agricultural practice on the soil can both increase and decrease soil pH. Some important agricultural practices, which can decrease soil pH include the application of ammonium fertilisers and sulphur fertiliser and growing of legumes. Furthermore, acidic rain can also decrease soil pH (Goulding, 2016). A common practice to reverse the trend of decrease in soil pH is the application of agricultural limes. Agricultural limes are often in the form of $CaCO_3$ or sometimes in a form containing magnesium: $Ca{\cdot}MgCO_3$ (Sims, 1996). Adding lime to the soil can change the soil pH due to the addition of carbonate anions, which can react with H^+. When the lime material is added to the soil, it dissolves gradually. The bicarbonate can then remove H^+, and pH will increase. The reaction when lime is applied as $CaCO_3$ is: $CO_3^{2-} + 2\ H^+ \rightarrow CO_2 + H_2O$. Thereby, when lime is added, soil acidity is neutralized (Brady and Weil, 2010).

Rubæk et al., (1998) investigate the effect of lime application on soil pH on an acid coarse sandy soil in the long-term experiment in St. Jyndevad. The study shows that the long-term effect of liming increased the soil pH in depth down to around 70 cm depending on liming treatment (Rubæk et al., 1998). Likewise, a study by Kostic et al. (2015) shows that adding lime to an acid soil increase the pH, while application of P does not alter the pH. The experimental setup in the study consists of different levels of liming and P in rhizoboxes (Kostic et al., 2015).

The availability of plant nutrients is affected by the soil pH, and the availability of the different nutrients is affected differently. The optimal pH for crop growth is considered to be between pH 5.7 to pH 6.5, when considering the availability of all plant nutrient (Haynes and Naidu, 1998). This optimal pH is of cause affected by the nutrient requirements for the specific crop, and which nutrient in the specific conditions that are limiting (Barrow, 2017).

Limitation of plant growth caused by low supply of P and aluminium toxicity are key reasons for liming application on agricultural soils, but P availability as a function of pH in the soil is a complex matter, and reaching the optimal pH for P availability can be tricky. P can form complexes or be bound to particles such as Ca, Fe, and Al in the soil (Hinsinger, 2001). At acid pH, P is fixated by iron and aluminium, which therefore restrict the availability of P. At alkaline pH, P is fixated by calcium. This leaves a window at around pH 7, where P is most available in the soil (Haynes, 1982). But, there are contradicting views on at which pH the availability of P is highest. Barrow (2017) reviews the literature on effects of pH on plant uptake of P, and he finds that the optimum for P availability is at lower pH than generally accepted. Barrow (2017) divides the P availability as a function of pH to be determined by three processes, when the soil pH decreases to a lower level than 6, which includes 1) increase in P uptake, 2) increase in desorption from soil, and 3) increase in sorption to soil, and he states that:

"The relative contribution of the three effects might differ in different circumstances, but in the data available, the first two dominate [P uptake and P desorption] and uptake increases as pH decreases" (Barrow, 2017)

Thereby, Barrow concludes that the effect of change in pH on P availability is determined by the specific conditions in the soil, and that it is difficult to establish a general correlation between P availability and pH.

As pointed out before, and also found by Barrow (2017), the optimal pH for plant growth is dependent on more than one nutrient, and the plant requirements and the specific soil should be taken into consideration when finding the optimal pH for plant growth.

Aluminium in acid soils can be found in toxic levels of the plants, and pH below 5.5 is considered to have a limiting effect on plant growth (Haynes and Mokolobate, 2001). When aluminium appears in toxic levels it can have an inhibition effect on the nutrient uptake and cell division (Haynes, 1982). In a study on liming conducted by P Kostic et al., (2015), they find that the application of lime could eliminate aluminium stress in wheat plants. Furthermore, they find that adding both liming and P to an acid soil increased the level of available P (Kostic et al., 2015).

2.2 Root growth and nutrient availability

One of the primary functions of roots is nutrient uptake from the soil. Therefore, the spatial distribution and availability of plant nutrients influence the root growth (Hodge et al., 2009). How the root growth of the plant is affected by the nutrient availability is dependent on the specific requirements of the type and variety of the crop in the specific environment. Lopez-Bucio et al. (2003) mention the nutrients nitrogen (N), P, iron, and sulphur as nutrients, which are known to be able to alter the root growth as response to the availability of these nutrients. Furthermore, they state that the response of the root growth to the nutrient state of the soil can be divided into growth of the primary meristem, lateral root growth, and growth of root hairs (Lopez-Bucio et al., 2003). In a situation where the plant experiences P deficiency, it is commonly recognised that an adaptation strategy is to increase the formation of root hairs (Lopez-Bucio et al., 2003, Gahoonia et al., 1997). The formation of root hair increases the surface area of the root exposed to the soil volume, and can therefore potentially increase the uptake of limiting nutrients (Lopez-Bucio et al., 2003). In spring barley, Gahoonia et al. (1997) find that roots of different varieties of spring barley respond differently to different levels of P. Between the varieties, a variation in the respond in the length of the root hairs was observed, as well as a different area of P depletion zone for the different varieties.

Gahoonia and Nielsen (1996) study P depletion of soil by different varieties of spring barley. They find significant differences between the varieties, and they argue that the differences must be caused by the root hair formation and by root exudates. Furthermore, they find that the pH in the depletion zone is not altered by the different spring barley varieties. Schjørring (1987) also studies different varieties of spring barley in relation to P. He studies the response of the varieties to P deficiency and finds that there is variations between both the P uptake by the roots and the root length between the different varieties.

Another commonly recognised plant mechanism to increase nutrient availability is the release of root exudates from the roots. A study on this matter has showed that root exudates such as malate and citrate released from the roots can increase the availability of micronutrient in an acid soil. Furthermore, the study shows that the root of the plant is also capable in a situation of an alkaline soil to change the nutrient availability (Jones and Darrah, 1994). In relation to P deficiency, the release of root exudates is a known mechanism, whereby the plant itself can increase the availability of P and thereby potentially the P uptake. In a study done on rape seed oil, it was detected that decreasing the pH in the rhizosphere by the release of root exudates affected the plants ability to deplete the soil for P (Gahoonia and Nielsen, 1992).

As previously stated, one of the primary functions of roots is to supply the plant with nutrients. A study done on root growth in spring wheat in low fertility conditions gave indications on how to study the early nutrient uptake by the roots (Wang et al., 2016). They find that vigorous root growth in spring wheat better indicates early nutrient uptake, than the formation of root hairs. This matter is important, because early uptake of nutrients by the crop can influence on the development of the crop positively and thereby the resulting yield.

2.3 Soil physical properties and root growth

Root growth is affected by the soil properties. Soil of a coarse texture, such as sandy soils, can restrict root growth. Askegaard and Eriksen (2007) investigate root growth of catch crops on the sandy soil in the St. Jyndevad Experimental station, and find that the roots from normally deep-rooted crops were restricted from growing deep on the sandy soil, and they find only few roots below 75 cm. Madsen (1985) investigates root density of spring barley in different soil types and climates. The root density is very different in the top layer between the different soil types, but a tendency to a larger decrease in root density in the more sandy soil. Madsen (1985) finds that roots grow deeper in soil with finer texture than in very sandy soils. Andersen (1986) finds in a study on root growth development of different barley and wheat varieties on different soil types, that there is large variation in the root growth development in the different soil types, and smaller variations between the different varieties grown in similar conditions.

The weather impact on root growth can be significant; e.g. the amount of precipitation during the growth season, but this impact is different for soils with different texture. Madsen (1985) finds that spring barley grown on loamy soil in very wet spring, negatively influences root growth, whereby he finds an increase in root growth of spring barley on sandy soil in wet conditions. Sandy soils are known to have a lower water holding capacity than more loamy soils, and therefore is these observations most likely related to the differences in the water holding capacity. A parameter which can be used to determine the physical conditions for root growth is the concept of Least Limiting Water Range, which is related to the soil structure, is described by Dasilva et al. (1994). This range varies between different soil types and soil structures, and provides information on the water availability for the plant uptake in the different soils (Dasilva et al., 1994). Limits for root growth due to soil physical properties can be found e.g. by measuring soil bulk density and penetration resistance. Under Danish conditions a bulk density above 1.6 g/m^3 is considered to be limiting for root growth (Schjønning et al., 2015). Penetration resistance above the range 1.5-2 MPa is considered critical for root growth (Kadziene et al., 2011).

2.4 Soil organic matter content and root growth

Several beneficial effects for the soil fertility are related to an increase in root depth. Below the topsoil, the carbon (C) content in the soil normally decreases. Maeght et al., (2013) reviews studies of deep roots, and he points out that carbon originating from roots is likely to be stabilized. A higher input of roots in deeper layers can thereby potentially increase the content of C in depth (Maeght et al., 2013). In a review Haynes and Naidu (1998) find that it is likely that the use of lime will result in an increase organic matter content, but they state that the response has not yet been found. Organic matter is known to be beneficial for the soil fertility. The important properties that can be improved by increased content of C, are; soil structure, air permeability, and water holding capacity. Especially the soil structure and the water holding capacity are known to be poor in very sandy soils. So an increased input of root in depth can therefore potentially increase soil fertility of these soils.

2.5 Yield, root growth and nutrient availability

The crops nutrient supply is essential for the yield. The previous mentioned study of Schjørring (1987) with root growth of different spring barley varieties under P deficiency, also provides information on the grain yield for the different spring barley varieties. Schjørring (1987) finds that the varieties, which have the highest P uptake and the longest root length, also have the highest yield. Furthermore, the study shows that generally, the yield levels of all the varieties decreased, when the supply of P decreased.

Travnik et al. (1998) investigate the effect of different combinations of fertilisers and lime on yield. The study finds that the yield levels is highest when both fertilisers and lime have been applied. Furthermore, the study finds that lime in itself is found to increase the yield levels as well, compared to treatments with no supply of either lime or fertilisers, indicating better use of the nutrients.

A study by Conyers et al. (2003) conducted in Australia on the long-term effects of lime application on grain yield levels for different types of cereals, e.g. barley, shows that there is higher yields on the fields that previously had been limed compared to fields, which had not received liming treatments. This study shows that there is a long-term effect of liming, and that the effect was somewhat persistent. This is consistent with the fact, that application of lime is recommended every 5-8th year (Knudsen, 2008).

In the soil, some nutrients are found to be very mobile, whereas others are more or less immobile. Therefore, the architecture of the roots needs to adapt to the crops' need for the immobile nutrient needed. It is important for the crops supply with immobile nutrients that the roots are able to explore a large soil volume. White et al. (2013) have identified the most important traits of crops in order for the plant to acquire immobile nutrients, e.g. P. The identified traits include early vigorous root growth, high degree of root branching, and high root length per mass of soil (White et al., 2013).

2.6 Liming and phosphorus application in Denmark

In Denmark the agricultural advisory service provides the farmers with recommendations for application of both P and liming. The recommendations are based on soil tests on the farmers' fields, and by analysing the results, specific recommendations are given. It is generally recommended to apply lime every 5th to 8th year depending on the pH status of the soils (Knudsen, 2008). There are specific recommendations for the liming application to the soil depending on the soil- and crop type. For very sandy soil it is generally recommended to have a lower soil pH than on more loamy soils (Knudsen, 2008). The pH values that form the basis for these recommendations are the so-called reaction number (Rt), which is pH ($CaCl_2$) + 0.5 (Plantedirektoratet, 1994). Table 2.6.1 divides Rt values from low to high values according to soil type and crop sensitivity. According to this table, spring barley is considered a sensitive crop, and a high Rt for spring barley on a JB 1 soil will be a Rt value over 6.5, and a low Rt value will be considered to be below 5.2. This chart can be used for the farmers/agricultural advisors, when they are done with soil testing, and know the Rt of the different fields on the farm. In addition, for the farmer to use this chart, he or she will also need information on the soil type as well as the crop his or her crop rotation.

Table 2.6.1 *Classification of Rt values for different crops and different soil type. The table has been translated from Danish and adopted from Knudsen (2008).*

Soil type (JB nr.)	**Crop sensitivity to soil Rt**	**Very low**	**Low**	**Medium**	**High**	**Very high**
1-4	Tolerant*	< 5.2	5.2-5.7	5.8-6.1	6.2-6.5	> 6.5
	Medium**	< 5.5	5.5-5.9	6.0-6.3	6.4-6.7	> 6.7
	Sensitive***	< 5.7	5.7-5.9	6.0-6.5	6.6-6.9	> 6.9
5-6	Tolerant	< 5.3	5.3-6.0	6.1-6.5	6.6-6.9	> 6.9
	Medium	< 5.5	5.5-6.2	6.3-6.7	6.8-7.1	> 7.1
	Sensitive	< 5.7	5.7-6.4	6.5-6.9	7.0-7.3	> 7.3
7-9	Tolerant	< 5.3	5.3-6.3	6.4-6.7	6.8-7.2	> 7.2
	Medium	< 5.5	5.5-6.5	6.6-6.9	7.0-7.4	> 7.4
	Sensitive	< 5.7	5.7-6.7	6.8-7.1	7.2-7.6	> 7.6
11	Tolerant	< 4.3	4.3-4.7	4.8-5.2	5.3-5.7	> 5.7
	Medium	< 4.5	4.5-4.9	5.0-5.4	5.5-5.9	> 5.9
	Sensitive	< 4.7	4.7-5.1	5.2-5.6	5.7-6.1	> 6.1

** Tolerant crops are potatoes, rye, oats, and grass. ** Medium crops are winter wheat, winter barley, maize, red- and white clover, rape seed, and field peas. *** Sensitive crops are alfalfa, sugar beet, medick, and spring barley.*

As with liming, recommendations for P application to the fields are also done accordingly to the P status of the fields. P can be applied to the fields as mineral fertiliser or as organic fertiliser. An example of P recommendations can be found in table 2.6.2, which is a table translated and adopted from Rubæk et al. (2005).

Table 2.6.2: *Recommendations for P application according to P status of the soil. The table is translated and adopted from Rubæk et al. (2005).*

P status (mg P/ 100 g soil)	**Level**	**Recommendation**
<1	Very low	Large application of P
1-2	Low	Application of 20-40 % more P than removed by crop
2-4	Medium	Application of P corresponding to the amount removed by crop
4-6	High	Application of 25-50% of amount of P removed by crop
>6	Very high	No application of P

In summary of the table: If the P-state of the fields is at the desired level, the application of P should correspond to the level of P removed by the crops, and if the desired level is more or less, application of P should then be none or be more than the crops remove.

3.0 Materials and methods

3.1 Field site

The experiment was conducted in the long-term field experiment in St. Jyndevad, Denmark. The experimental design of the long-term experiment consists of combinations of different treatments with liming and P. The liming treatments consist of liming levels of 0 kg/ha, 4000 kg/ha, 8000 kg/ha, and 12000 kg/ha for reaching pH of 5.4, 6.2, 6.7 measured in $CaCl_2$, except for the treatments with no liming application (Rubæk, 2008). The P treatments were either with no addition of P or a dose of 15.6 kg P/ha has been applied every year. A single dose of 156 kg P/Ha was applied to half of all treatments at the beginning of the experiment. The treatments have been practiced since 1942 (Rubæk et al., 1998). The soil is a coarse sandy soil (Nielsen and Møberg, 1985). Spring barley is grown in the experiment every year. The field experiment is divided into four different fields designated V and the numbers 1-4. Spring barley is grown in V1 and V2, V3 is set-a side, and V4 is forest. The treatments in V4 have stopped. Further details on the long-term experiment can be found in Rubæk (2008) and in Rubæk et al. (1998). The mean annual temperature is 7.9ºC and mean annual precipitation 870 mm (Møberg and Nielsen, 1986).

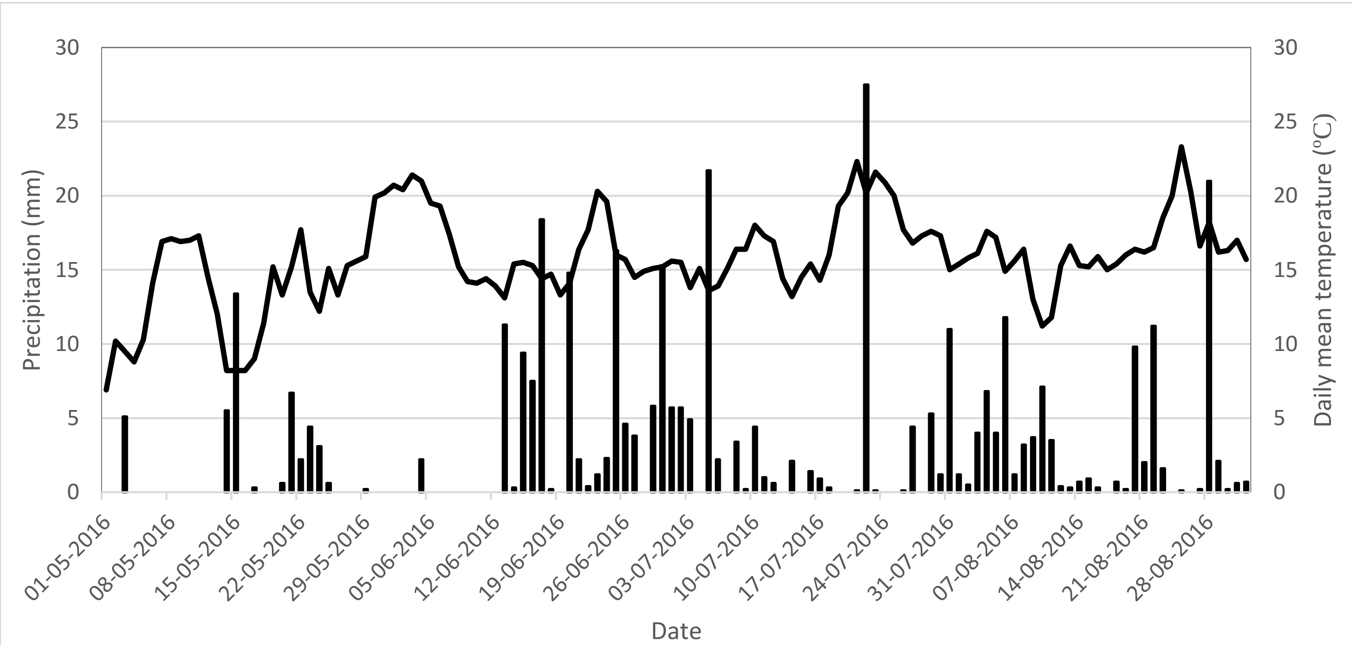

Figure 3.1. *Climate data for research center St. Jyndevad in the period 01.05.2016 – 01.09.2016. The columns show the daily precipitation in mm and the curve show the daily mean temperature in degree Celsius.*

3.2 Experimental design of the present study

This study was carried out from May 2016 to August 2016. All treatments chosen had a start application of 156 kg P/ ha in the beginning of the long-term experiment. Half of the treatments were given 15.6 kg P/ ha per year, and the other half have not been fertilized with P since the first dose. All four liming levels was represented in the study, but some experiments was only carried out in selected treatments. For each of the different treatments the same experiments have been carried out in three replicate plots in the V1 field. A description of the characteristics of the V1 field and the experiment can be found in Appendix 1. Spring barley of the variety KWS Irena was sown in the experiment April 14th 2016. The crops were irrigated with 30 mm on June 2nd and June 8th 2016.

3.2.1 Minirhizotron

Minirhizotrons (MR) were installed on April 26th, 2016 into the following four treatments: 4 tonnes lime with and without P (4.0 & 4.1), and 8 and 12 tonnes lime with phosphorus (8.1 &12.1) (see Appendix 1). Two MR of 1.5 meters length were inserted with a 30 degree angle into approximately the middle of each parcel using a drill, which bored a hole so the MR could be inserted into the soil, and so the top of the tubes aligned with the top of the soil. The installation of MR and the preparation of the MR was done as described by Thorup-Kristensen (2001) with small modifications of the drilling method. Before installation of the MR a 4cm x 4cm grid was drawn on the outside of the glass tubes, and each grid was numbered. The MR was installed shortly after sowing before emergence of the spring barley plants. The roots were filmed four times with two weeks interval (May 17th, May 31st, June 13th and June 28th) beginning when the plants were at growth stage 29 according to the BCCH-scale. For the filming of the roots an Endoskop-camera connected via USB to a computer was used. The videos were played using still photos in the programme Avidemux 2.6.

The root intensity (RI) and root frequency (RF) were found as described by Thorup-Kristensen (2001). The RI was found counting the number of visible roots intersecting the 8 cm cross in each numbered grid. The depth of each numbered grid was found using the angle, which in MR have been installed, and the specific location of the grid on the tube. The RF was calculated as the share of the total number of grids in which roots were observed.

3.2.2 Deep soil cores

Deep soil cores were taken in all the combinations of the different liming levels and the two different phosphorus treatments. For each treatment three 1-meter deep soil cores with a diameter of 9 cm were taken in each of the three replicate plots. The soil-cores were taken on August 22nd six days after harvest of the spring barley using a mechanical soil sampler. The deep soil cores were stored in two half tubes, which were taped together and closed with a lit in each end. The tubes were put into a plastic bag, which was tightly closed using tape. All of the tubes were stored in a fridge until unpacked to perform the next described experiments.

3.2.2.1 Core-break

The core-break experiment was carried out as described by Bennie et al. (1987) and by Wahlstrom et al. (2015), but with some modifications due to the very fragile sandy soil. The soil cores were divided into five different sections using a knife: 0-20 cm, 20-30 cm, 30-50 cm, 50-70 cm and 70-100 cm. The length of the last section varied a bit due to some of the soil had fallen out when the deep soil core was taken. A proximate of the real length of the section was noted down. The soil was split at the middle of each section into two parts gently using a knife. The number of roots was counted on the one intact side.

3.2.2.2 Chemical analysis

From each section of the soil cores, samples for chemical analysis were taken. The C and N content was determined using the Dumas combustion method, where combustion of C and N into CO_2 and N_2 happens at high temperatures and detection of the N_2 and CO_2-gasses on a Vario Max Cube . The pH($CaCl_2$) of the soil was measured in a suspension of soil and 0.01 M $CaCl_2$ with a 1:2.5 weight/volume proportion (Plantedirektoratet, 1994). The soils' total P (TP) was determined colorimetrically after wet destruction in a mixture of concentrated sulfuric acid and perchloric acid heated to 250 degrees for evaporation of perchloric acid, essentially according to Kafkafi (1972).

3.2.2.3 Soil bulk density

The dry soil bulk density in each of the different sections in the soil cores was determined measuring the length of each section and thereby finding the soil volume of the specific section. Only an estimate of the length of the last depth section was noted down due to an uneven surface. The last depth section was therefore not included in the statically analysis of the data. Next, a small soil sample was taken and the weight was measured. The soil was then oven dried at 105 degrees Celsius for one day. The soil was weighed to determine the weight of the soil particles, and thereby determine the soil bulk density.

3.2.3 Penetration resistance

The penetration resistance (PR) was measured on May 10th, 2016 using an automated cone penetrometer similar to the one described by Olsen (1988). The PR was measured for every 10 mm down to a depth of 80 cm with a speed of 30 mm/s.

3.2.4 Yield

The spring barley was harvested on August 18th, 2016. The harvest grain yield in Hkg dry matter per ha was measured for all the different treatments. The netto area of the harvested parcels were 16.5m^2 for the 8.1 and 12.1 treatments, and 25.08m^2 for the 4.0 and 4.1 treatments.

3.3 Statistical analysis

The statistical analysis was conducted in R version 3.3.2. A significance level of $p < 0.05$ was used in all of the analysis. A random variable was added to all models to take out possible correlations due to the experimental design. ANOVA was used to conduct F-test for the model reductions. All models were checked for normality and variance homogeneity. The aim of the statistical analysis was to check if there was an effect and/ or interaction of: treatment (both liming level and P application), liming level, P application, and depth. Furthermore, it was checked whether or not the observed properties for the treatments were statistical different. For further details on the statistical analysis, a comprehensive description of the analysis of the yield data can be found in the Appendix 2. For the analysis of the CB data only the first 3 depth sections was included due to very low abundance of roots in the last two sections. An outlier was removed in the C and N data due to surreal low values. For the bulk density analysis the last depth section was taken out due to the uncertainty about the precision of the method for the last depth section. pH data was found not to be normally distributed, and therefore the Kruskal-Wallis Test for the analysis were performed.

4.0 Results

4.1 Soil bulk density

The long-term liming treatment affected the soil bulk density differently depending on the long-term P treatment. Figure 4.1.1 and 4.1.2 show the mean soil bulk density with and without continuously added P. The curves of the soil bulk densities for the different treatments as a function of depth have been analyzed. When no P had been applied, an effect on the soil bulk density in the different depths of the different liming levels was observed ($p<0.05$). When P continuously had been added, no effect of liming on the soil bulk density could be detected ($p>0.05$). Depth affected the bulk density for all treatments ($p<0.05$), and a significant interaction ($p<0.05$) between liming level and depth both with and without continuously added P was found. The first depth section 0-20 cm had a general lower soil bulk density than in the deeper depth sections. The soil bulk density increased from 0-20 cm depth to 20-30 cm depth. The soil bulk density decreased when 30-50 cm was reached, and it was highest for 50-70 cm depth. In sandy and loamy soils, a bulk density over 1.6 g/m^3 can be considered as critical (Schjønning et al., 2015). Further details on the statistical analysis can be found in Appendix 3 and 4.

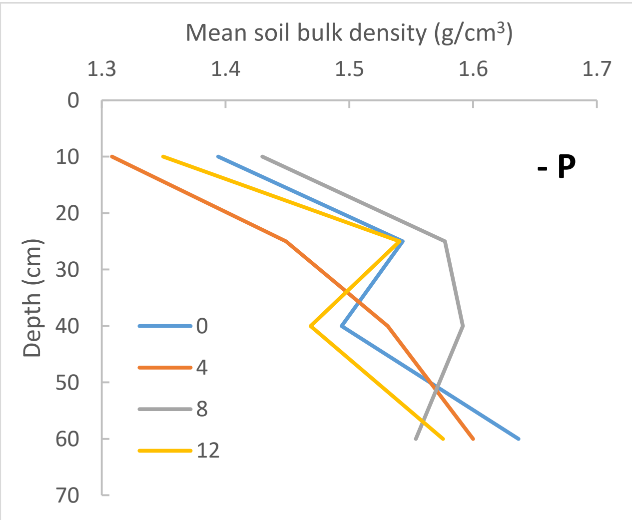

Figure 4.1.1 *Mean soil bulk density (g/cm^3) as a function of depth. Different colors show the different levels of liming (tonnes/ha). No P have been added to these treatments.*

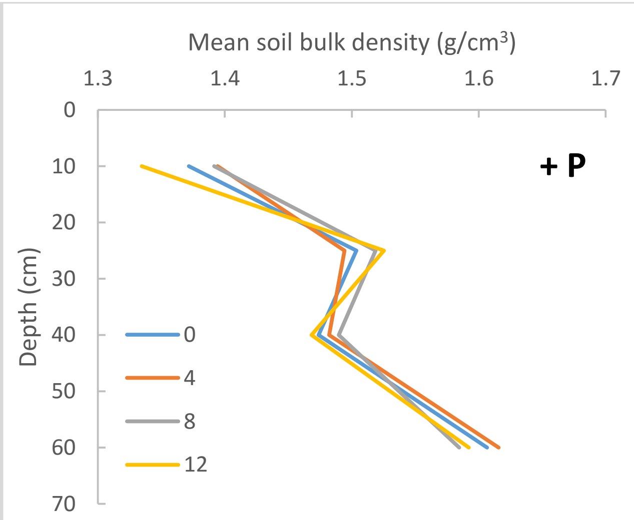

Figure 4.1.2 *Mean soil bulk density (g/cm^3) as a function of depth. Different colors show the different levels of liming (tonnes/ha). P have been continuously added every year to these treatments.*

4.2 Penetration resistance

The PR was measured for all of the eight different treatments. Figure 4.2.1 shows the PR at four selected depths for all of the treatments. The four selected depths were 5, 18, 28, and 40 cm. The chosen depths represent different parts of the soil profile. The 5 cm depth represent the part of the soil, which was harrowed. The 18 cm depth represent the lower part of the plough layer. In 28 cm depth there seemed to be a transition from lower to higher penetration resistance. The last depth chosen, 40 cm, represent the subsoil, where roots also were found. In Appendix 5, a complete overview of the depth can be found. In the Appendix the position of the four selected depths can be found as well. Furthermore, the confidence intervals for the estimated PR can be found in Appendix 6. The PR at 5 cm depth showed no significant ($p>0.05$) differences in PR between the different treatments. Looking at the second selected depth at 18 cm the 0.0 and 0.1 treatments had the highest penetration resistance, and they are found to be statistically different from the 8.0, 8.1, and 12.0 treatments. At the third depth, there was a significant difference between the 0.0 and 0.1 treatments, where the 0.1 treatment had the lowest penetration resistance. No other differences in PR between the treatments was found in this depth. At the last depth, at 40 cm, no differences in PR was detected ($p>0.05$). Generally, the PR increased with depth, but this was not tested statistically. Looking at the graph it can be seen that the PR at 18 cm was in the range 1.5 MPa – 2 MPa, which is considered to be the critical values for restriction of root growth (Kadziene et al., 2011). Furthermore, it can be seen that in the deeper layers, 28 cm and 40 cm the PR is higher than this range. This can also be found in the graph in Appendix 5.

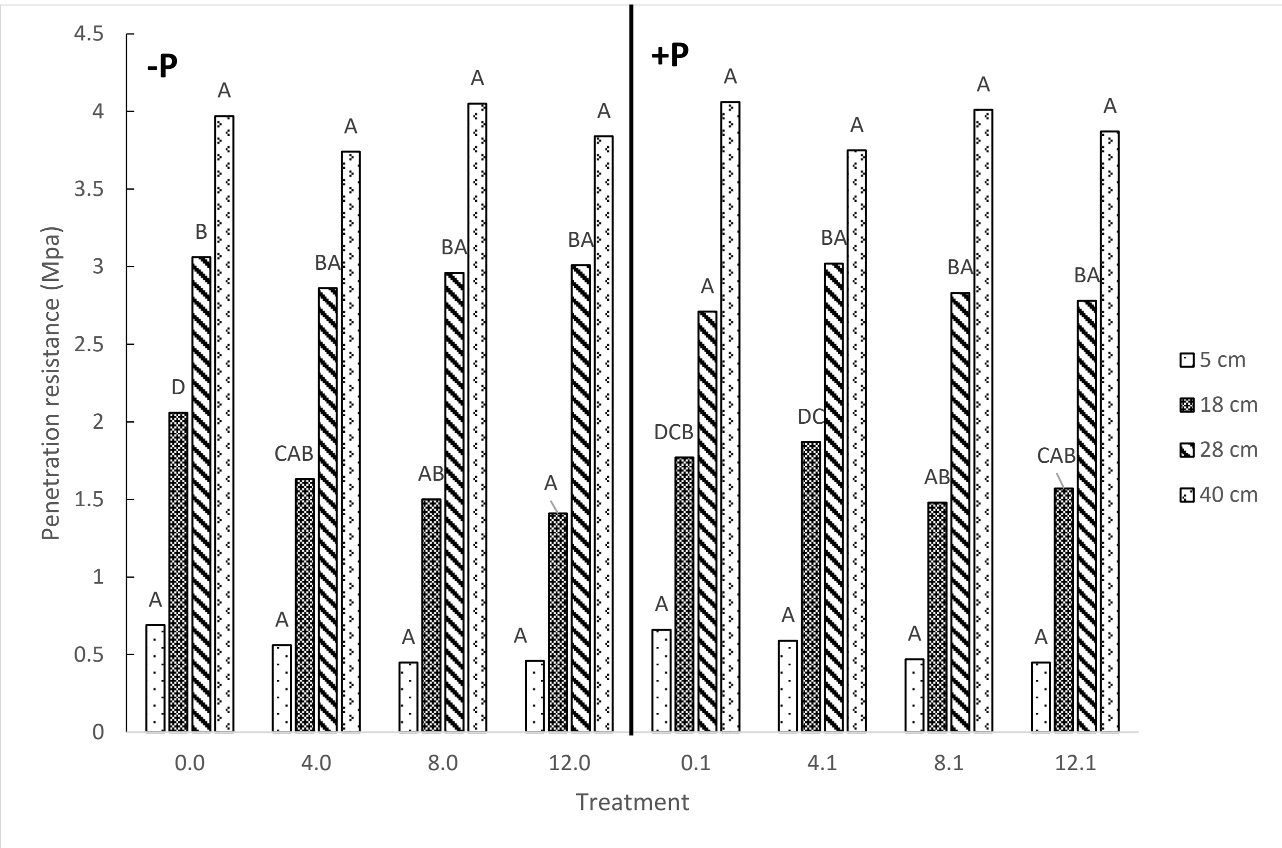

Figure 4.2.1: *Penetration resistance (Mpa) for the eight different treatments in four different depth (5 cm, 18 cm, 28 cm, and 40 cm). Treatment codes are described in list of abbreviations. The letters illustrate whether or not the estimated number of roots for the different treatments is statistical different within the same depth.*

4.3 Chemical analysis

The top 20 cm of the soil from the deep soil core samples was analysed for C, N, TP content and pH. The results are displayed in table 4.3.1 below.

Table 4.3.1 *Chemical properties for the top 20 cm. Estimates of values for C, N, TP content and pH including letters indicating whether or not the treatments are significantly different (p<0.05). Treatment codes are described in list of abbreviations.*

Treatment (P, Liming)	**pH**	**Total P**	**C**	**N**
0.0	3.6 (A)	366 (B)	1.213 (A)	0.0758 (A)
4.0	5.1 (C)	335 (BA)	1.202 (AB)	0.0798 (B)
8.0	6.7 (D)	326 (A)	1.327 (B)	0.0943 (C)
12.0	7.2 (E)	321 (A)	1.345 (B)	0.0907 (C)
0.1	3.8 (B)	448 (C)	1.27 (AB)	0.0831 (B)
4.1	5.3 (C)	425 (C)	1.249 (AB)	0.0822 (AB)
8.1	6.5 (D)	417 (C)	1.318 (B)	0.0897 (C)
12.1	7.1 (E)	424 (C)	1.332 (B)	0.0928 (C)

4.3.1 pH

There was significant effect of the liming on the soil pH ($p<0.05$). Except for the no liming treatments, the soil pH at the same liming levels, both with and without P, was found to be the same. In addition to the analysis of pH for the different treatments in the top 20 cm, analysis of pH in all depth sections was carried out. The result of the analysis is displayed in table 4.3.1.1. Furthermore, figure 4.3.1.1. illustrates soil pH with depth in the different treatments. For the high liming levels pH tended to decrease with depth, whereas the opposite could be observed for the low liming levels. pH within the same level of lime application in depth sections 20-30 cm and 30-50 cm could not be statistically separated. For the depth section, 50-70 cm the same trend was no longer found. In this depth the 12.1 treatment had the significantly highest pH value. Still the 8.0 and 8.1 treatments had the same pH value ($p<0.05$). The 0.0, 0.1, 4.0, and 4.1 treatments could not be statistically separated. For the last depth section, 70-100 cm, once again the 12.1 treatment had a significant higher pH value. As also displayed in the figure, the pH values seemed to become more similar in the last depth, which is also illustrated in the table.

Table 4.3.1.1 *Estimated pH in depth including letters used to determine whether the pH values within the same depth are significantly different from each other. Treatment codes are described in list of abbreviations.*

Treatment (P, Liming)	**0-20 cm**	**20-30 cm**	**30-50 cm**	**50-70 cm**	**70-100 cm**
0.0	3.6 (A)	3.9 (A)	4.4 (A)	4.5 (AB)	4.5 (AB)
4.0	5.1 (C)	5.1 (B)	4.8 (B)	4.8 (ABC)	4.5 (AB)
8.0	6.7 (D)	6.5 (C)	5.8 (C)	5.0 (C)	4.9 (CB)
12.0	7.20 (E)	7.1 (D)	6.3 (D)	5.4 (D)	5.0 (CB)
0.1	3.8 (B)	4.1 (A)	4.4 (A)	4.5 (AB)	4.3 (A)
4.1	5.3 (C)	5.2 (B)	4.9 (B)	4.8 (BC)	4.8 (ABC)
8.1	6.5 (D)	6.7 (C)	5.9 (C)	5.0 (C)	4.8 (CB)
12.1	7.1 (E)	7.0 (D)	6.4 (D)	5.9 (E)	5.4 (D)

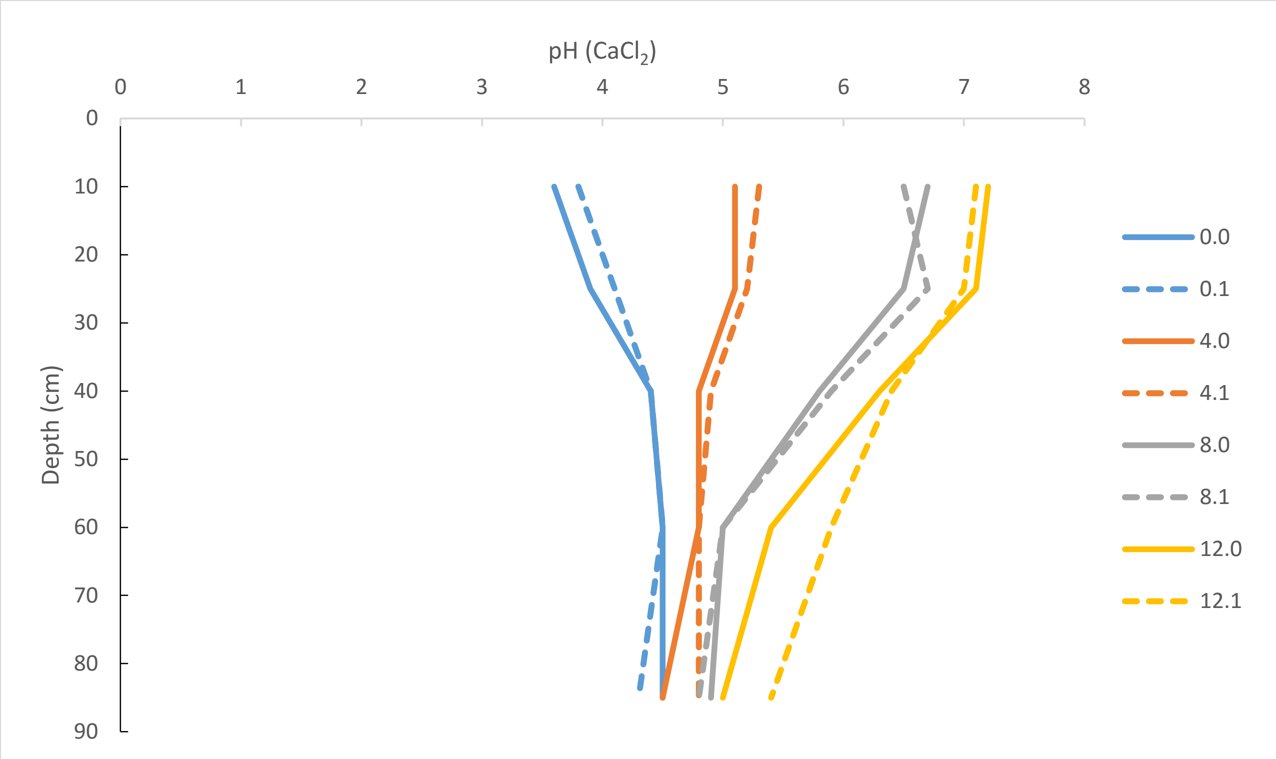

Figure 4.3.1.1. *Estimates for pH in the different treatments in the five depth sections from the deep soil cores. Treatment codes are described in list of abbreviations.*

4.3.2 Total P

TP in the top layer was found to be significantly ($p<0.05$) highest in the treatments, which had continuously been fertilized with P fertilizer. All the treatments with P added every year were significantly ($p<0.05$) different from the treatments with no P added every year. In the treatments, which have not been fertilized with P continuously, a difference between the liming treatments was found as well. It was found that the 0.0 treatment has a significant ($P<0.05$) higher content of TP than the 8.0 and 12.0 treatments.

4.3.3 C

There was a significant ($p<0.05$) effect of treatment on the C content in the soil in the 20 top cm. The extreme treatment with no lime and no P was statistically different from the treatments, which had received the two highest levels of liming (Table 4.3.3.1). In general, the carbon content decreased with depth (Table 4.3.3.1). Some differences between treatments was found at all depth, except for the lowest layer 70-100 cm.

Table 4.3.3.1. *Estimates for carbon content in the different treatments in the five depth sections from the deep soil cores. The letters indicate if the estimates are statically different within the same depth section. Treatment codes are described in list of abbreviations.*

Treatment (P, Liming)	**0-20**	**20-30**	**30-50**	**50-70**	**70-100**
0.0	(1.213) A	(1.101) ABC	0.657 (AB)	0.331 (AB)	0.152 (A)
4.0	1.250 (AB)	1.026 (A)	0.540 (A)	0.273 (A)	0.127 (A)
8.0	1.327 (B)	1.193 (BC)	0.801 (B)	0.429 (B)	0.197 (A)
12.0	1.345 (B)	1.166 (ABC)	0.779 (B)	0.365 (AB)	0.168 (A)
0.1	1.270 (AB)	1.172 (ABC)	0.673 (AB)	0.358 (AB)	0.175 (A)
4.1	1.249 (AB)	1.050 (AB)	0.646 (AB)	0.297 (AB)	0.133 (A)
8.1	1.318 (B)	1.223 (C)	0.769 (B)	0.340 (AB)	0.169 (A)
12.1	1.332 (B)	1.168 (ABC)	0.715 (AB)	0.336 (AB)	0.163 (A)

4.3.4 N

The content of N in the soil was significantly affected by the long-term treatment ($p<0.05$). As displayed in table 4.3.1, the two highest liming levels both with and without P gave a significantly ($p<0.05$) higher content of N in the top 20 cm of the soil than the rest of the treatments. The extreme treatment with no P and no liming gave the significantly lowest amount of N. The extreme treatments content of N could not be statistically ($p>0.05$) separated from the 4.1 treatment.

4.4 Core-break root count

The results from the CB root count can be found in figure 4.4.1. The analysis was only carried out on the first three sections in the deep soil cores due to very low abundance of roots in the last two sections. No statistical differences ($p>0.05$) in the number of roots between the different treatments could be found in the CB in 10 cm depth, but there was a tendency that the number of roots is highest in the high liming treatments. In the CB in 25 cm depth, some differences could be detected between the treatments. The number of roots in the treatment 4.1 was significantly different from number of roots in the treatments where P had been applied every year. The 4.1 treatment was significantly different from 4.0. At 25 cm depth, a decrease in the number of roots could be seen when no P have been added. The opposite could be seen when P have been added continuously every year. The number of roots found in 40 cm depth was also very low, and there was no significant difference between the number of roots in the different treatments in this depth.

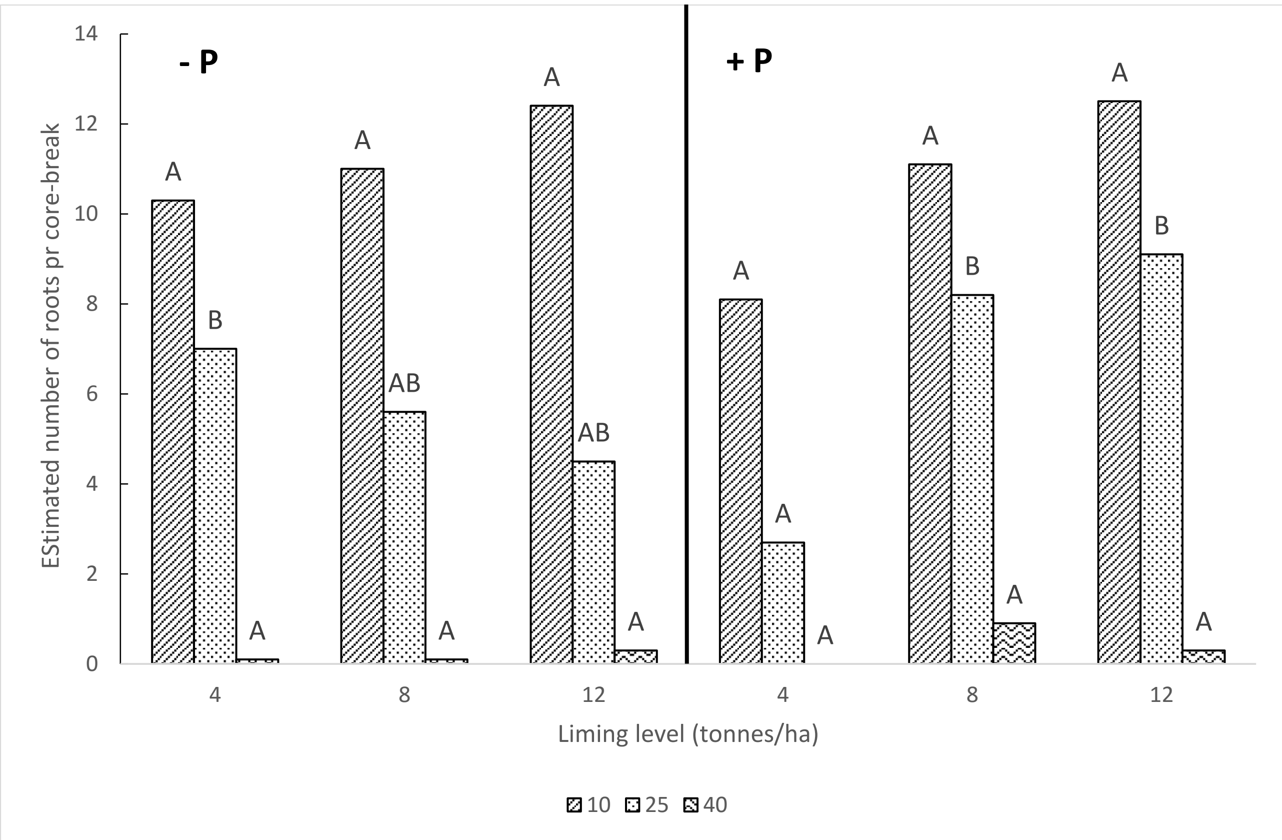

Figure 4.4.1: *The estimated number of roots in the different treatments in the different depth sections in the deep soil core using the method core-break. The surface area of the core-break is 63.6 cm^2. The letters illustrate whether or not the estimated number of roots for the different treatments is statistical different within the same depth. Treatment codes are described in list*

4.5 Minirhizotrons

4.5.1 Root frequency

The RF in the different treatments was determined for each time of observation in the MR. Significant effect of treatment was found ($p<0.05$). Furthermore, a difference in the root growth in time between the different treatments was detected. The growth stage according to the BBCH-scale (Bromand et al., 1995) for the spring barley were registered each time of observation. It was 29 on May 17th, 33 on May 1st, 55 on June 13th, and 69 on June 28th. RF for the total depth of the MR is displayed in figure 4.5.1.1. On the first date of observation, May 17th, the 12.1 treatment was found to have a significant higher RF than the other treatments. Two weeks later, the 4.0 treatment had a significant lower RF than the rest of the treatments. The 12.1 and 8.1 could now not be statistically separated. For the third time of observation the 8.1 and 12.1 could again not be statistically separated, but it seemed that 8.1 now has the highest RF, opposite to the previous time of observation. The 4.0 treatment still had the lowest RF, and it was different from the rest of the treatments. On the last date, the 4.0 and 4.1 treatments was found not to differ from each other and they had a lower RF than the 8.1 and 12.1 treatment. There seemed to be an increase in the RF with time for all of the treatments. This was not tested statistically.

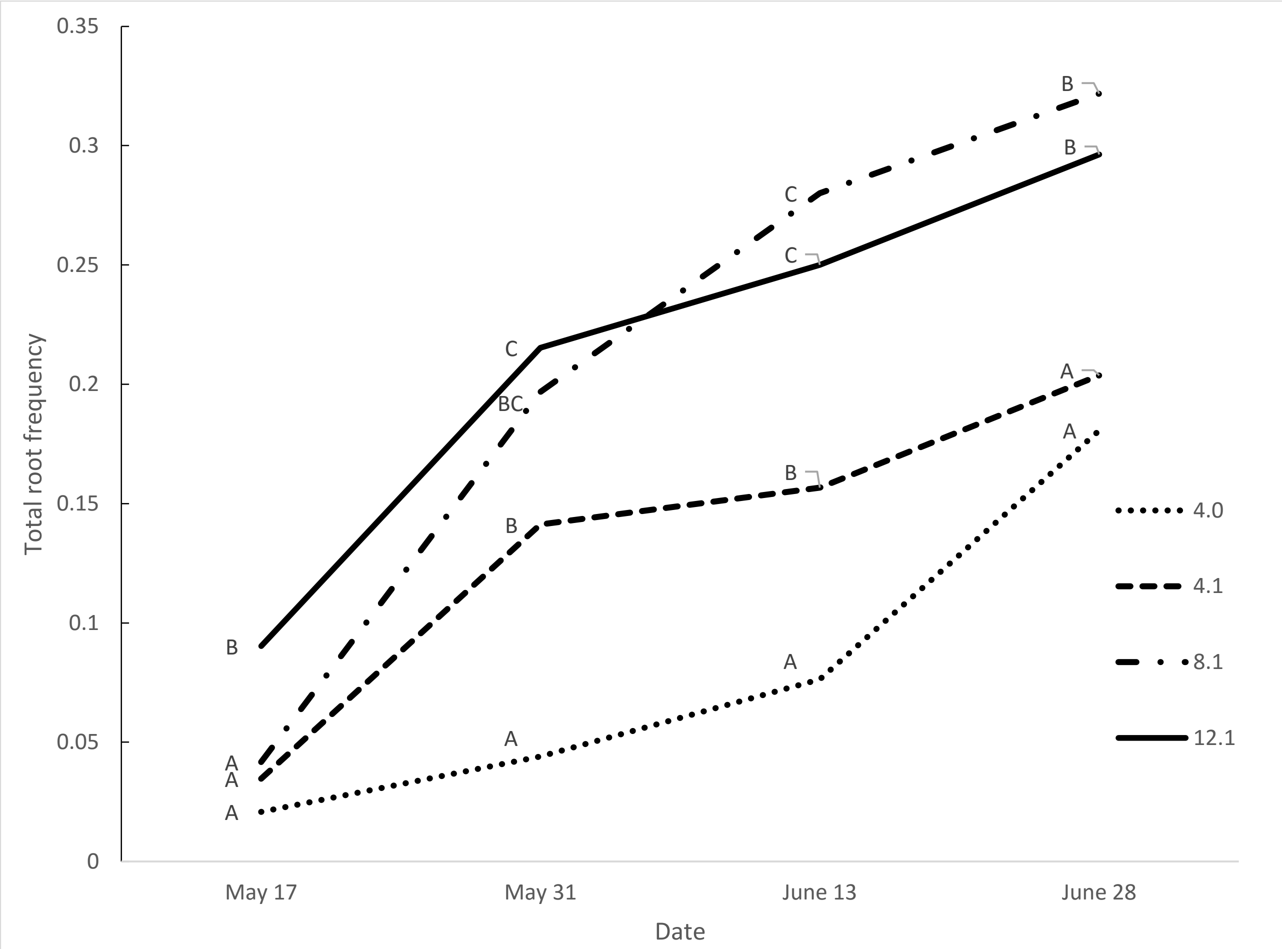

Figure 4.5.1.1: *RF for the four treatments at the four different dates of observations calculated as percentage grids in the MR with appearance of roots. The total length of the MR is 124.71 cm. The letters show which observations within the same date that are significantly different. See Appendix 6 for further details. Treatment codes are described in list of abbreviations.*

The RF for when the soil had been divided into 3 depth sections is displayed in figure 4.5.1.2-4.5.1.4. Figure 4.5.1.2 shows the development in RF for the first depth section (0-20.78 cm). The RF in the top section for the first date of observation shows that the 12.1 and 4.0 was statically different, but otherwise no differences between the treatments could be detected. For the second time of observation, two weeks later, the 4.0 treatment was statistically different from all the other treatments, and this was the same for the third time of observation. At the last observation no differences in the RF for the top layer could be found, which means that the roots statically occupied the same volume of soil in this specific section.

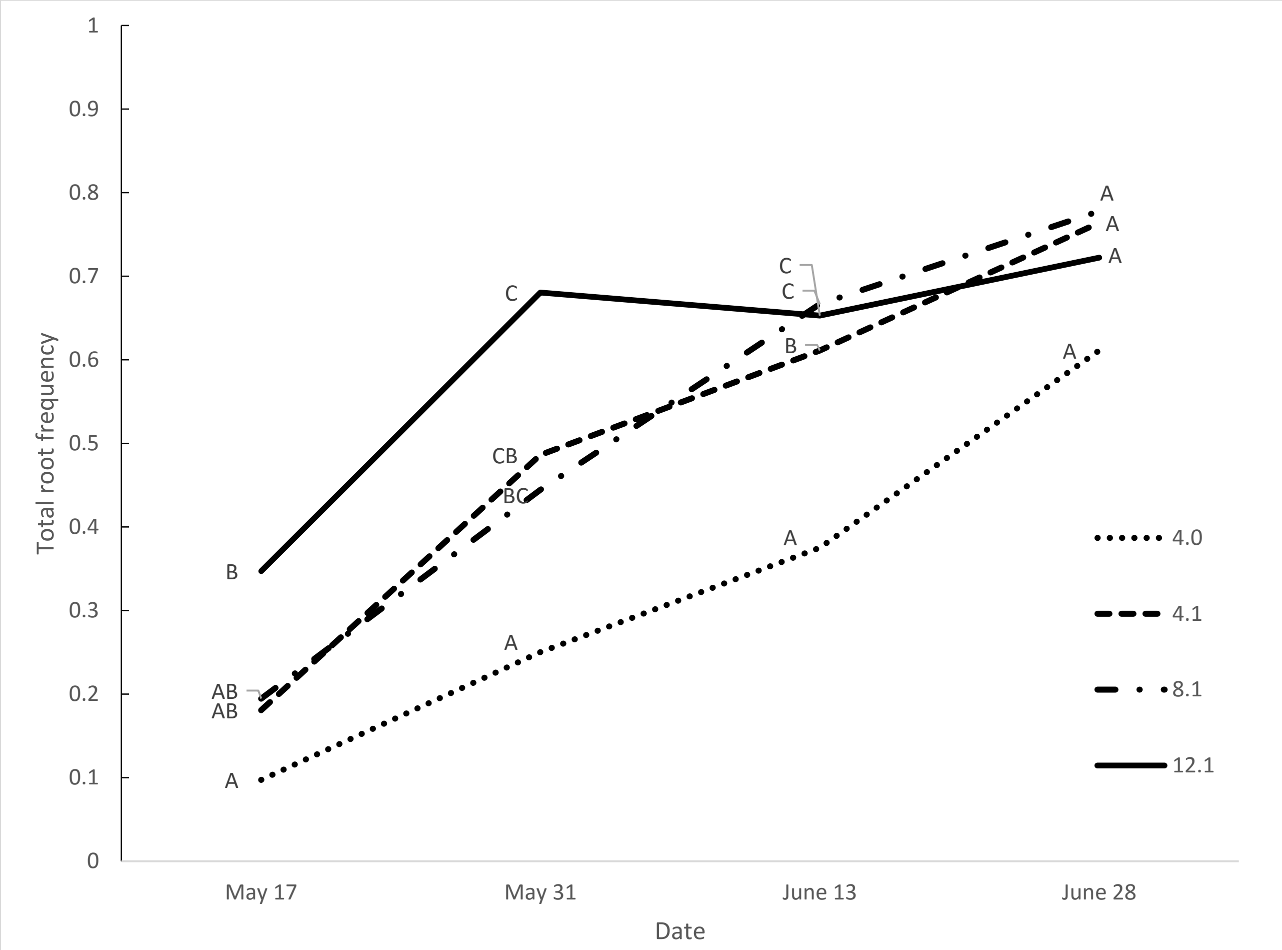

Figure 4.5.1.2: *RF for the depth first depth section (0-20.78 cm). The letters shows which observations within the same date that are significantly different. See appendix 7 for further details. Treatment codes are described in list of abbreviations.*

The RF for the second depth section (20.78-31.18 cm) can be found in figure 4.5.1.3. At this depth at the first date of observation the 12.1 treatment was statistically higher from the rest of the treatments. At the second time of observation, the 8.1 and the 12.1 treatments was found to be the same, and there had been an increase over time in the RF for both of them. The 4.1 treatment had also increased, but it is now significantly different from all of the other treatments. The 4.0 had not increased in RF, and it had statistically a lower RF than the rest of the treatments. On the third time of observation, the pattern was the same as for the second time of observation. On the last date the model used did not detect any statistical differences in RF, but it seemed as if 4.1 and 4.0 was lower than the two other treatments.

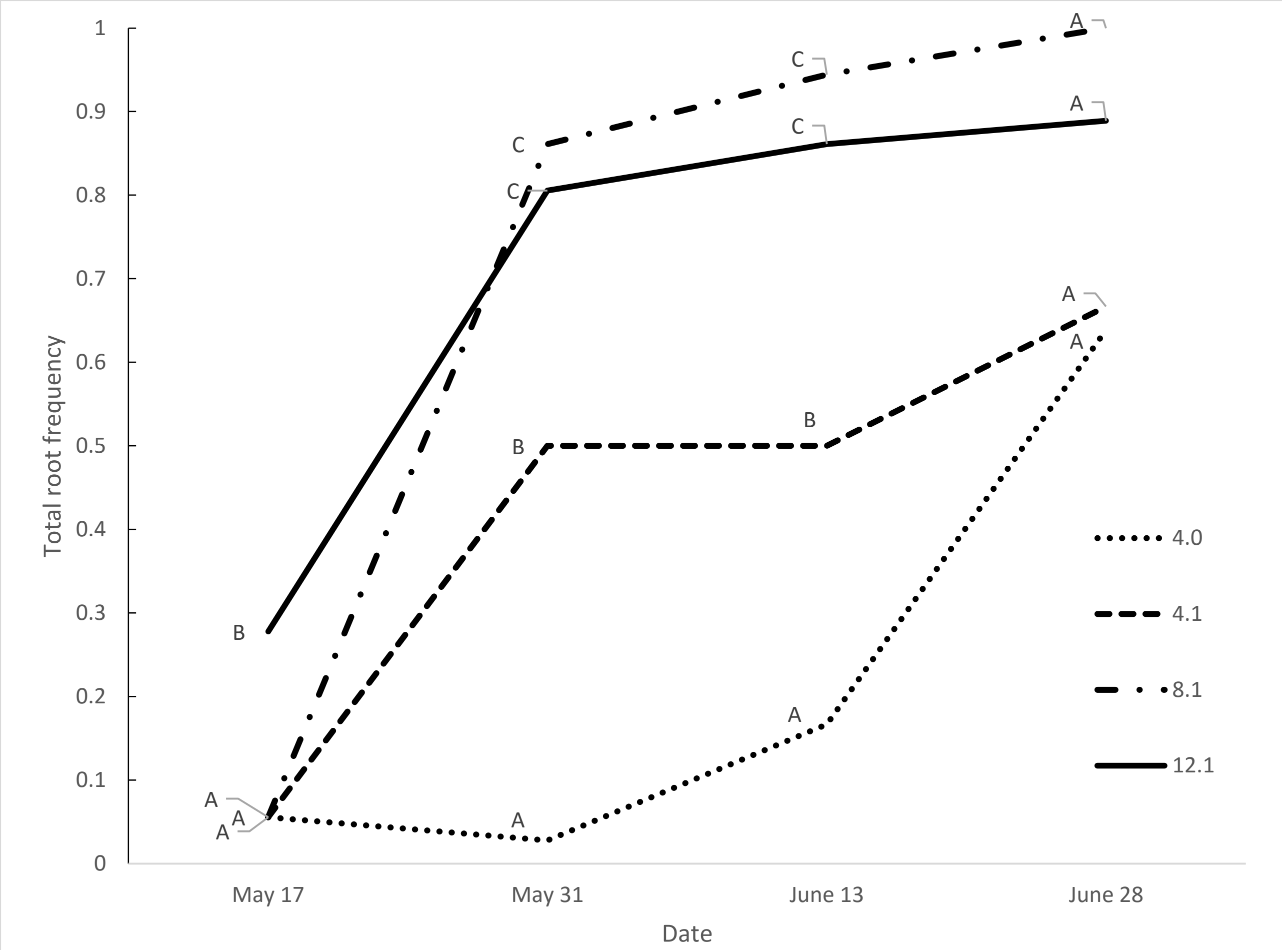

Figure 4.5.1.3: *RF for the second depth section (20.78-31.18 cm). The letters shows which observations within the same date are significantly different. See Appendix 7 for further details. Treatment codes are described in list of abbreviations.*

In figure 4.5.1.4 the RF for the depth third section is displayed (31.18-51.96 cm). At the first time of observation, no roots seemed to have reached this depth section, and therefore all of the treatments was found to be statistically the same. Treatment 8.1 and 12.1 was found to be following each other with increase in RF as the time passes. At the last date the 12.1 and 8.1 had significantly higher RF than the 4.0 and 4.1 treatment.

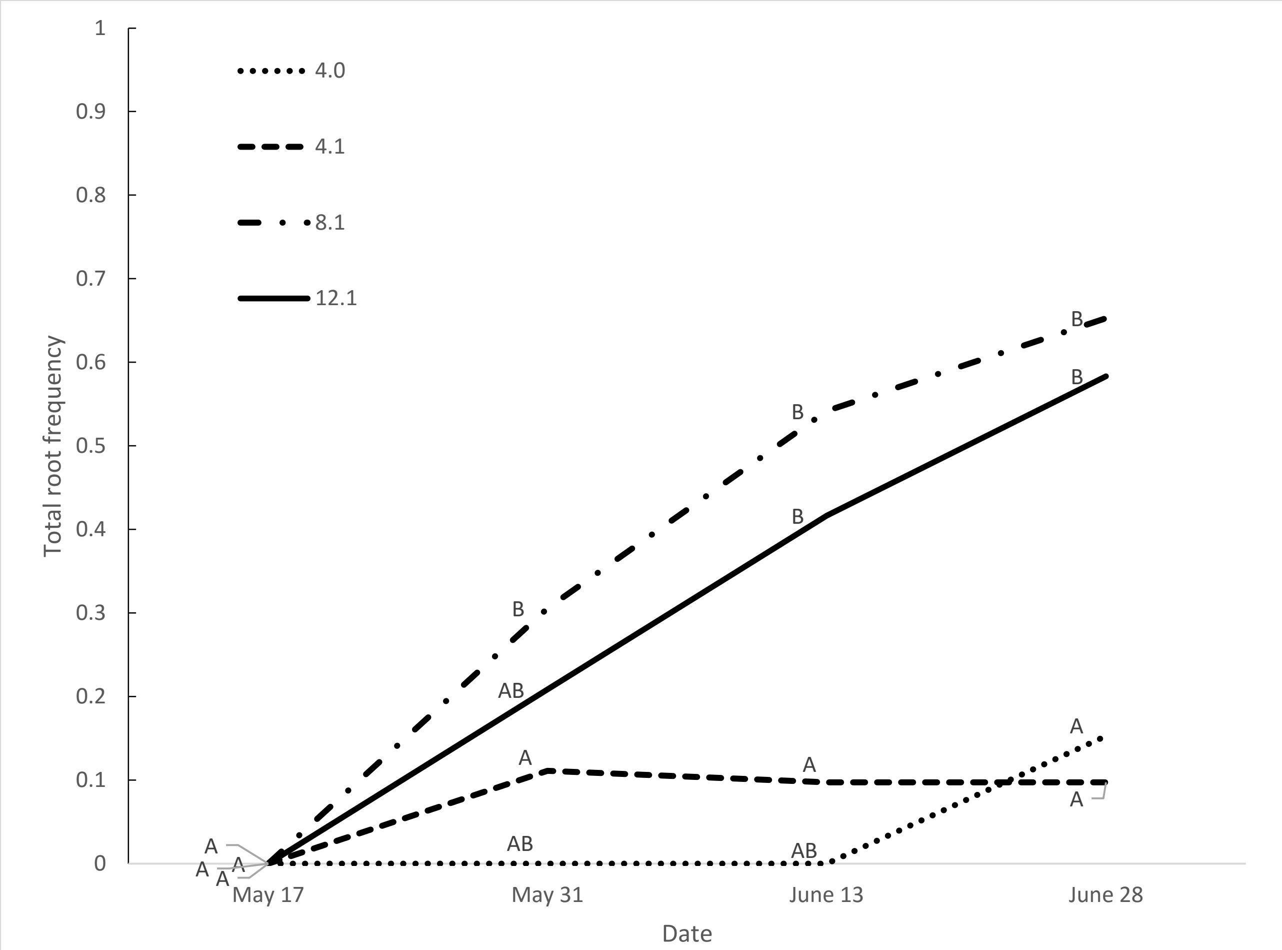

Figure 4.5.1.4: *RF for the third depth section (31.18-51.96 cm). The letters show which observations within the same date that are significantly different. See Appendix 7 for further details. Treatment codes are described in list of abbreviations.*

4.5.2 Root intensity

The RI for five different depth intervals for the different treatment is displayed for each observation time in figures 4.5.2.1-4.5.2.4. The standard error for each observation can be found in Appendix 8. No further statistical analysis was performed on the RI data, because the study focusses more on RF than RI. Therefore, the following can only used to give an indication of the RI with depth. The figures showed that there was increases in RI for each time of observation. For the first time of observation, which was the May 17th, the 4.0 treatment appeared to have the lowest RI, followed by 8.1, 4.1 and 12.1 treatments, respectively. Furthermore, the root intensities seemed to decrease with depth, except for the 12.1 treatments, which appeared to have about same intensity in the second depth interval. On the second date of observation, May 31st, a slightly different pattern in the root intensities could be observed. Again, the 4.0 treatment had the lowest RI, and the RI for this treatment was highest in the top depth interval. The 4.1, 8.1 and 12.1 treatments had around the same RI in the top depth interval. The 4.1 treatments had a decrease in RI, when the top depth interval was compared to the second depth interval. This was contrasting to the 8.1 and 12.1 treatments, which had an increase in RI in the second depth interval. All of the treatments have a decrease in RI from the second to the third depth interval.

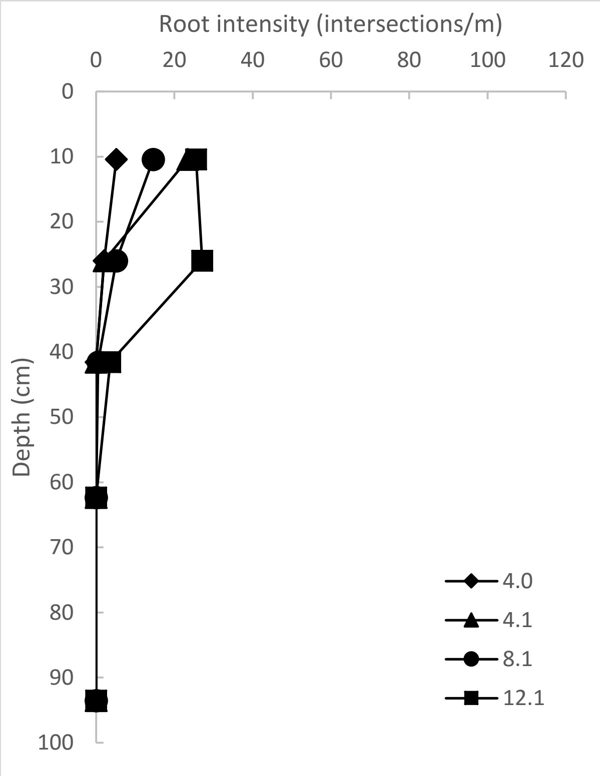

__Figure 4.5.2.1:__ Root intensities for May 17th. RI calculated as intersections pr m-1 for five different depth intervals. The RI for each interval is displayed in the middle of each interval. Treatment codes are described in list of abbreviations.

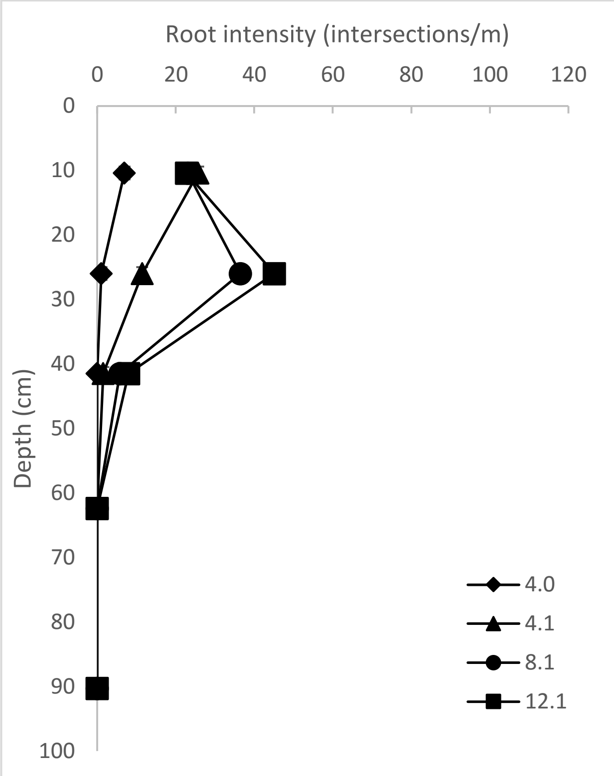

__Figure 4.5.2.2:__ Root intensities for May 31st. RI calculated as intersections pr m-1 for five different depth intervals. The RI for each interval is displayed in the middle of each interval. Treatment codes are described in list of abbreviations.

Figure 4.5.2.3 shows the root intensities for June 13th. Once again, the 4.0 treatment had the lowest RI. As seen before, the other three treatments had around the same RI in the top depth interval. Of the three treatments, it was the 12.1 treatment that has the lowest RI. Yet again, the 12.1 and 8.1 treatment had an increase in RI going to the second depth interval. There the RI for the 4.1 treatment remained about the same for the two first depth intervals. All of the treatments had a decrease in RI from the second to third depth interval.

On the final observation, June 28th, the same pattern as the previous observation was found. The RI for that observation is displayed in figure 4.5.2.5. The RI in the top depth interval for the 4.0 treatment had increased from the last time of observation, and this time the RI in this depth interval was same as for the 12.1 treatment. For the 4.0 and the 4.1 a small decrease in RI was observed from the first to the second depth interval. An increase was found for the 12.1 and the 8.1 treatment between these depth intervals. A decrease in RI was again found for all treatments from second to third depth intervals.

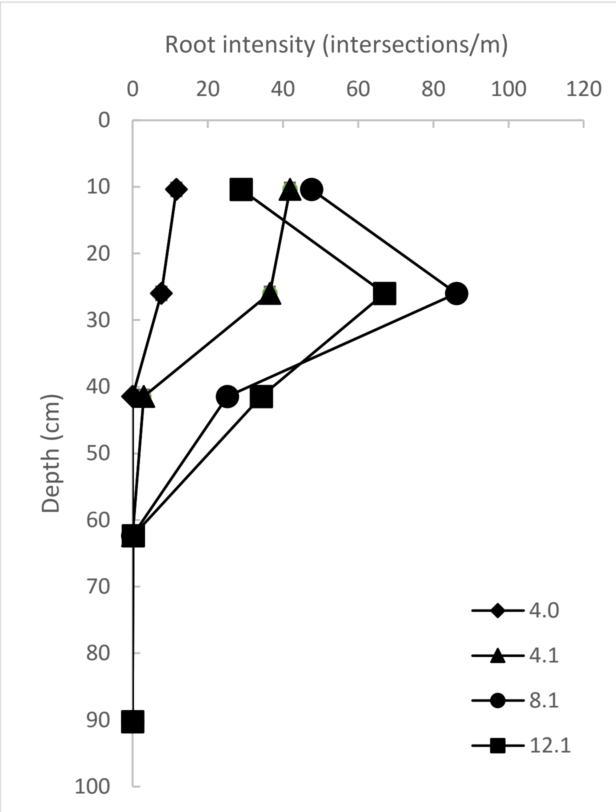

Figure 4.5.2.3: *Root intensities on June 13th. RI calculated as intersections pr m^{-1} for five different depth intervals. The RI for each interval is displayed in the middle of each interval. Treatment codes are described in list of abbreviations.*

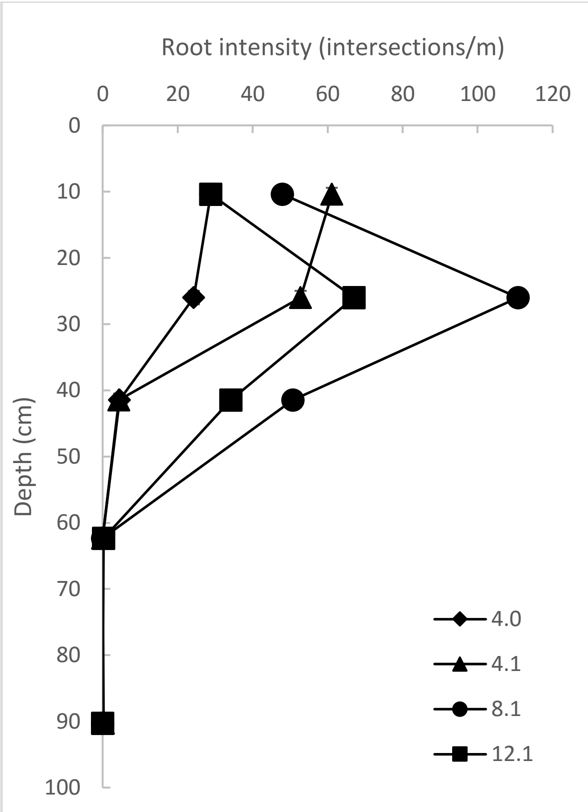

Figure 4.5.2.4: *Root intensities on June 28th. RI calculated as intersections pr m^{-1} for five different depth intervals. The RI for each interval is displayed in the middle of each interval. Treatment codes are described in list of abbreviations.*

4.6 Grain yield

The long-term treatment with liming and P had a significant effect on yield ($P<0.05$). It was found that the treatments with or without continuously P application showed the same response curve to the different long-term liming levels apart from an added constant. Therefore, there was not a significant interaction between liming and P application ($p>0.05$). Continuous application of P every year increased the yield level compared to treatments, which have not received P since the single dose at the beginning of the long-term experiment. Furthermore, an optimal level of long-term treatment with liming for the yield levels in 2016 was identified to be approximately 6.4 tons/ha, and there was not a difference in the theoretical optimal for the treatments with and without continuously added P. Further details on the statistical models used can be found in Appendix 2.

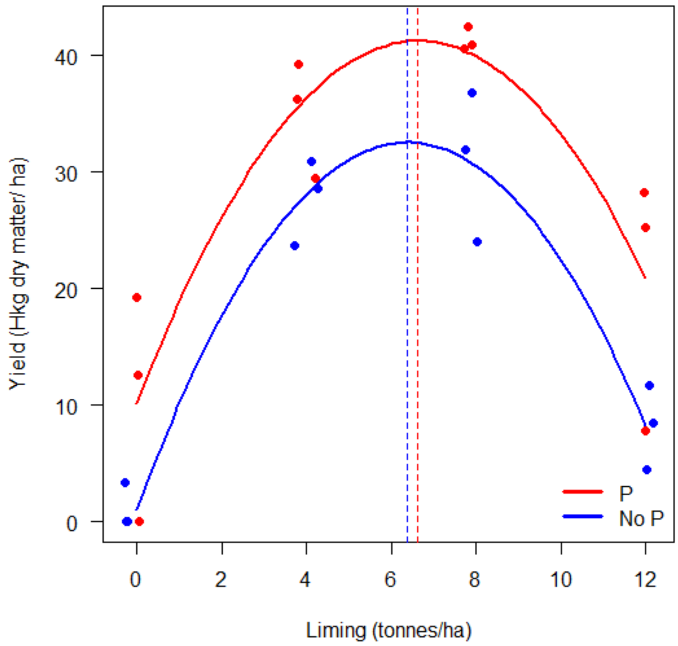

Figure 4.6.1. *Grain yield in Hkg dry matter/ ha as a function of liming level. The red line shows the response curve for when P has been added continuously, and the blue curve shows the response when P have not been added continuously. The two dotted lines indicate the theoretical optimum. The points have been added noise in order to see all of them on the graph. The correct x-values for them are 0, 4, 8, and 12.*

5.0 Discussion

5.1 Soil physical properties

The physical properties of the soil were modified due to the long-term treatments, as expected in the first hypothesis. The physical properties of the soil were affected to a various degree.

5.1.1 Effects on soil bulk density

The soil bulk density changed with depth in all treatments, and the pattern of the curve was found to be the same except for when P has not been applied every year. In the treatments without continuously application of P, the curves of the 4.0 and 8.0 did not have the same shape as the other treatments (see figure 4.1.1). There seem to be no obvious reasons for why these two treatments behaved differently, but it might be created by the long-term differences in the production in these specific treatments. In the last depth section analyzed for soil bulk density (50-70 cm), the critical value of 1.6 g/cm^3 for soil bulk density (Schjønning et al., 2015) was reached.

5.1.2 Effects on penetration resistance

The PR was measured for every 10 mm, and therefore it gives comprehensive knowledge of the development in PR with depth for the different treatments. As the analysis in figure 4.2.1 shows, there was no differences in PR in 5 cm depth, which was likely to be due to the uniform soil mechanical treatments in the top layers of the soil. The results for the root growth both from the CB and from the MR, showed that the roots was concentrated in the top layers, and that differences in root growth could be found in the second (20-30 cm) and third (30-50 cm) depth sections. Assumed, that these differences have been long-term, it is likely that differences in root growth have influenced the PR. Looking at the PR in 18 cm, which is considered to be the lower part of the plough layer, a higher PR could be found for the no liming treatment than the treatments limed, except for the 4.1 treatment. It should be kept in mind, that the MR was only performed in four selected treatments all with liming application, while the no liming treatment was included in the CB analysis. There are therefore no records of the root growths in the no liming treatments. However, grain yield was included, which showed very low levels of grain yield, and low abundance of spring barley roots could be assumed. Haynes and Naidu (1998) suggest that optimal liming and fertilization practice indirectly can improve the soil physical properties, resulting in decreased bulk density due to e.g. increased input of organic matter.

However, as they also state, the effects of liming application on soil physical properties have not been studied thoroughly.

5.2 Soil chemical properties

All of the measured soil chemical properties (pH, TP-, C-, and N- content) were altered due to the long-term treatments with different levels of liming and P, as expected in the first hypothesis. These alterations will be elaborated and discussed in the following sections.

5.2.1 Effects on soil pH

The pH values in the top-layer of the soil had reached higher values than originally aimed at in the design of the field trial in St. Jyndevad. Therefore, the 12 tonnes/ha treatment had more alkaline conditions than aimed at. In accordance with the findings in this study, Rubæk et al., (1998) have previously found increased pH as an effect of liming as deep as 70 cm in another field belonging to the same field experiment. Therefore, when lime is applied on a long-term basis, the alteration of the pH is spread to the deeper parts of the soil than just the plough layer. Findings of Kostic et al. (2015) also show effects of liming on the soil pH, but no interaction with application of P was found in their experiment. In the present study no interactions between liming and P were found in the 20-30 and 30-50 cm depth sections, but in the other depth sections, different pH values with the same liming levels were found depending on P application for some of the liming levels. For example, the 0.0 and 0.1 treatments have a significantly different pH value, with the 0.1 having the highest pH value. The difference might be due to the long-term difference in productivity of the two treatments.

5.2.2 Effects on total phosphorus content

The TP content in the present study was only analyzed for the top 20 cm of the soil. The highest TP content was found, when the treatment had received P every year, and all of the treatments were found to have the same amount of P. In the same experiment, Rubæk et al. (1998) tests the effects of lime or no lime application with P or no P application on the soils TP. Moreover, they find, that the treatments, which have received P every year have the significant highest amount of TP, and they find these treatments to be significantly different from the treatment with no P and no lime application. Furthermore, Rubæk et al. (1998) also investigate the effects of the treatments below the top 20 cm, and found effects on TP in 30-40 cm depth as well, but they find no effects on TP deeper than this depth. In the present study, there was also different TP content within the treatments, which have not received any P fertilization, where the 8 and 12 tonnes lime/ha have a significantly lower TP content than the zero lime treatment. This is likely to be caused by a larger P-removal in the limed treatment, due to higher production. Rubæk et al. (1998) find similar effects on total inorganic P and they explain it with larger P-removal with the crops.

5.2.3 Effects on carbon and nitrogen content

The C-content in the soil was altered by the different treatments, and effects were found as deep as to the 50-70 cm depth section. In the top 20 cm, the extreme treatment with no lime and no P application had the lowest C- content, and could be significantly separated from C-content of the liming treatments with 8 and 12 tonnes lime/ha. The results on grain yield and on root growth development showed clear differences between the different treatments. Therefore, it can be expected that these differences have created a different long-term input of plant residues to the soil, and thereby a different input of organic matter, which can explain the differences in C-content. Haynes and Naidu (1998) also find an increase in organic matter content of the soil when optimal liming and fertilization have been practiced resulting in increased yield levels, and thereby increased organic matter return to the soil. The previously mentioned study of Rubæk et al. (1998) did not show any effects for the treatments included in their study on the C-content in any depths. The N content in this study was found to be highest in the treatments with the two highest liming levels. This can, as the C-content, be related to the higher level of production in these treatments, creating a higher input of plant residues to the soil, since there have not been any differences in the applied N fertilizer in the different treatments.

5.3 Root growth

The second hypothesis states that the previously discussed modifications the in physical and chemical properties of the soil will affect the root growth, and potentially create a larger soil volume favorable for roots. Differences in root growth between the different treatments were found in both root methods used in the study.

5.3.1 Influence of soil physical properties on root growth

Looking at the results from both the CB method and the MR it is clear that the root growth was concentrated in the top layers of the soil, which might indicate restriction of deeper root growth. Restriction of root growth on sandy soil has previously been found in the St. Jyndevad soil by Askegaard and Eriksen (2007). They find that the roots of normally deep-rooted cover crops, (e.g. Chicory) are not found below 75 cm in St. Jyndevad. Comparing the results of the root growth with the measured PR it is clear that the roots fairly fast experience high resistance. As Kadziene et al. (2011) discuss, PR above 1.5-2 MPa will normally restrict root growth. Looking at figure 4.2.1 as well as appendix 5, the critical value for root growth restriction is already met at around 20 cm depth. That the roots are concentrated in the top-layer can also be seen in the RI as well as the results from the CB method.

5.3.2 Discussion of the selected root growth methods

When discussing the results from the two methods used to study root growth, it is important to be aware of the strength and weaknesses of these two methods, even though this is not a study of the two methods. The two methods are discussed by Wahlstrom et al. (2015) who find a larger variation in the results from the MR method than from the CB method. In the present study, it should be noted that the results of the methods can not be directly compared because the time of observation for the two methods were different. The MR study followed the root growth development from early in the plant growth until stem elongation. The CB thereby was conducted after harvest, and when some roots might have degraded already. Small roots and root hairs were not detectable in the used methods. In relation to the effects of the P supply to the plant, these parameters are normally considered to be of great importance (Gahoonia et al., 1997). Wang et al., (2016)'s study on the early nutrient uptake by wheat plants finds that actually the vigorous root growth is a better indicator of this early uptake than root hair. In relation to this study, where the effects of liming was studied, and thereby effects of different nutrient availabilities for the crop, the method of looking at root length, density and frequency does make sense. Especially because the focus of the study was to investigate, if the roots with optimal liming and P application on the long-term basis will increase the use of the soil volume in depth, and thereby potentially increase the fertility of the soil in depth.

5.3.3 Influence of soil chemical properties on root growth

As discussed in the section on soil chemical properties, the soil pH was altered in depth due to the different long-term liming treatments. The pH of the soil affects the nutrient availability. Barrow (2017) suggests that P is mostly available for plant uptake at the pH range 4-6. Taking this into account when discussing the results of the root studies, some explanation to the root growth might be found. Only the 4 tonnes lime/treatments had pH values in the range in the top layers where the roots grow, and in the 4.1 treatment there was a significantly lower number of estimated roots in the second depth than in the 8.1 and 12.1 treatment. It was suggested to be caused by a higher P availability in the 4.1 treatment than in the 8.1 and 12.1 due to higher pH values in these treatments. There was no significant difference in the amount of total P in the treatments. An explaining of the differences in estimated number of roots could be that the P in the soil was more available at the pH level in that liming treatment. This result can be compared to the study of Schjørring (1987) on root length of different varieties of spring barley in P deficiency conditions. Schjørring (1987) finds a correlation between grain yield, root length and P uptake, and the finding of increased root growth in the layer 20-30 indicates a higher uptake of P, due to higher root growth. Furthermore, Schjørring (1987) finds differences between varieties in grain yield, root growth and P uptake. He finds that the all the varieties were affected in the same directions. This indicates that the finding in the present study can be used generally for the crop type spring barley. A weakness in the present study is that the uptake of the different nutrients was not measured, whereby it is difficult to know if the uptake by the crop has varied for the different treatments. Furthermore, there was a significant difference between the number of roots at the second depth section for the two 4 tonnes lime/ha treatments, as well as there was a significant difference in the TP between the two treatments. TP was lowest when P had not been applied continuously. The 4.0 had a significantly higher estimated number of roots, indicating at higher root production in order for the plants to take up P from the 4.0 treatment. No significant difference between the treatments with and with P could be found for the other liming treatments, but it appears that more roots are created at 25 cm depth in treatments with P application every year than in treatments without.

5.3.4 Soil volume favorable for root growth

The results from the MR on RF revealed the development in how roots of the four chosen treatments occupy the soil volume, which is relevant when investigating if a large soil volume becomes favorable for root growth with optimal long-term liming and P treatment. Enlargement of the soil volume favorable for root growth can potentially create a larger and more fertile soil for crop production e.g. due to an increasing return of organic matter to the soil over the years. The RF for the whole depth of the MR revealed differences in RF already at the first time of observation, where the 12.1 treatment had the significantly highest RF. The 8.1 did though catch up with the 12.1 treatments, and at the end it seemed like the 8.1 has a higher RF even though the two treatments are found to be statistically the same. The lower liming level for the 4.1 and 4.0 treatments had a significantly lower RF. These results show that the two highest liming levels had the largest RF, and thereby occupied the largest volume of the soil with roots. When the analysis of the RF is divided into different soil depth sections, even more information on the development of RF is found. The 12.1 treatment has the highest RF in the first depth section at the first time of observation, but at the last time of observation, no differences between the treatments were detected. This was also found in the second depth section for the last time of observation; however, it seemed that the 4.1 and 4.0 had a lower RF. Differences were found in the third depth section. Here, only the 8.1 and 12.1 developed a root system. The development of roots in this section for the 4.1 and 4.0 was very low compared to the 12.1 and 8.1. The long-term application of lime appeared to have enlarged the soil volume in depth with roots, and the total volume of soil with roots was highest for the two highest liming treatments.

5.4 Root growth and grain yield

The third hypothesis states that grain yield and the root growth development will be positively correlated unless the crop experiences stress. In this study, the soil pH is varied, so the crops in the different treatments are expected to experience different nutrient availabilities. At high pH manganese, boron, iron, and P can become less available for plant uptake (Rothwell et al., 2015). At low pH aluminum can be toxic, and as mentioned before, pH below 5.5 can be limit plant growth (Haynes and Mokolobate, 2001, Haynes, 1982). In the soil in St. Jyndevad, the no lime treatment and the 4 tonnes line/ha have pH in the soil below this limit. This is expected to affect the correlation between the root growth development and the grain yield production. In order to investigate this hypothesis, the results from both the CB and the MR will be compared to the yield levels for the specific treatments.

5.4.1 Effects on grain yield production

First of all, the different treatments produced different amounts of yield depending on treatment, and as discussed in the previous section, root growth was also found to be different for the different treatments. It should be noticed that the yield levels was generally lower in the experiment compared to spring barley grown under similar conditions. Expected grain yield levels on a JB 1 sandy soil in Denmark are 43-55 Hkg depending on if the fields are irrigated or not (Eriksen, 2016).

The results from the CB showed only a few significant differences in the estimated number of roots in the different depth sections. However, it seemed that in the treatments with continuous application P, increasing amounts of roots were produced with increasing liming level, both in the first and second depth section. Looking at the corresponding yield levels, the highest yield was produced in the 8.1 treatment, and a slightly lower yield is produced in the 4.1 treatment. There is actually a significant difference in the estimated number of roots between these two treatments, and the 4.1 treatment had produced a lower amount. The 4.1 treatment still had produced a relatively high yield compared to the other treatments. An explanation for this could be the lower soil pH than the treatments with higher liming levels, and thereby a different availability of nutrients. The crop in this treatment may not have had to invest a lot in production of roots in order to take up the necessary nutrients. Another factor possibly influencing the observed differences in root growth between 4.1 and 8.1 treatment is PR. A significant higher PR was found at 18 cm depth for the 4.1 treatment compared to the 8.1 treatment.

The 8.1 and 12.1 treatments were found to have the same RF and estimated number of roots in the core-break, but the 12.1 treatment had a lower yield than the 8.1 treatment. The soil pH for the 12.1 treatment was higher than for the 8.1 treatment, which indicates that the nutrients in the 12.1 were less available and the crop therefore had to allocate more resources into the production of roots, and this seemed to have a negative consequence for the production of grain yield. The treatments without application of P followed the same tendencies for response of the grain yield production to the different liming levels, but with a general lower production of grain yield. The crops seemed to produce less roots in the second depth section with increasing liming level, which was opposite the treatments with P application. Since there is a significant different response in the estimated number of roots to the same liming levels at 20 cm with and without P a significant interaction between liming and P was found. The treatments without P application every year did give a general lower yield than in the treatment with yearly P supply. An explanation of the lower production of roots in the second depth could be due to limitation of the crop growth due to the significantly lower levels of P.

The development in RF can also be compared to the production of yield. The 8.1 and 12.1 treatments have the highest RF, and the development in RF for the two treatments were found to be similar, except for a higher RF for the first time of observation for the 12.1 treatment. Where the 8.1 produced the highest yield of all the treatments, the high RF in the 12.1 treatment did not result in a high production of yield, but in a relatively low yield. Comparing the 8.1 to the 4.1 treatment, a lower slightly lower yield was achieved, but a significantly lower RF can be observed. The 4.1 is therefore likely to be more effective in the uptake of nutrients than the treatments with higher liming levels. As found in the previous section, the RF is the same in the top layer, and the differences in RF show in the second and third depth section. The 12.1 and 8.1 explore a larger soil volume in depth with roots, but it appears that it is only the 8.1 treatments, which gain a higher yield level.

Liming the soil too much, clearly influenced the grain yield production negatively, even though the crop still produced a large amount of roots. This shows clear indications of over-liming, and show the negative impacts of applying too much lime to the fields. In a review, Haynes (1982) mentions several cases where liming has resulted in yield reductions. It is clear that the two highest liming levels gave the highest production of roots, and that lower amount of roots could be found at lower liming levels. However, the yield responses to the different treatments show that the 4 and 8 tonnes lime/ha give the highest yield levels. Conyers et al. (2003) also found positive effects of long- term liming practice on the yield levels.
In relation to this study, the 8.1 treatment appeared to be the optimal of the tested treatments, due to the highest production of yield, and most extensive production of roots, which in the long-term will increase the return of organic matter to the soil compared to the 4.1 treatment.

6.0 Conclusion

Long-term treatments with different levels of liming and P application on a sandy soil had a significant effect on the root growth conditions. The long-term treatments had altered both the soils' physical and chemical properties. The different liming levels had altered the soil pH, so increased liming application had resulted in increased soil pH. These effects was observed in depth as well. The root growth development was found to be significantly different depending on the long-term treatment. The differences was mostly found below the top 20 cm, but the early RF was found in the top 20 cm to be highest for the 12.1 treatment. Furthermore, differences in root response to different liming levels in the 20-30 depth depended on the phosphorus application. The largest RF was found for the 8.1 and 12.1 treatments, indicating that high liming application increase the volume of soil with roots. The grain yield levels showed a response both to liming level and to the P application. An optimum for grain yield production was found to be between the 4 tonnes lime/ha and 8 tonnes lime/ha with application of P every year. The lower and higher liming application showed a clear decrease in grain yield. In conclusion, the optimal long-term liming and P application appeared to be able to improve the root growth conditions in depth on the sandy soil, and to have a positive effect on the grain yield level.

7.0 Perspectives

This master thesis have enlightened us on new aspects of long-term effects of liming and P application. That the soil pH increases with increasing levels of liming application was already well known e.g. in Conyers et al. (2003) and Rubæk (2008). Furthermore, it was known that over-liming and under-liming could have negative consequences for the productivity of the soil (Rothwell et al., 2015, Haynes, 1982). However, when looking on how the root growth development as well as grain yield levels respond to the long-term liming and P application provides us with new knowledge of these soil improving factors. To have knowledge on the long-term responses to common agricultural practices such as liming and P fertilizer is highly important when addressing important questions on how to achieve sustainable agricultural production.

Liming application on acid soil like the St. Jyndevad soil must be considered essential in order to have an agricultural production on the soil, since hardly any yield is produced in the no liming treatments. Liming at low levels does produce a relatively high yield compared to the achieved yield levels in the experiment. However, it does not produce an extensive root system, and the return of organic residues to the soil must be considered to be lower than with optimal liming treatment. An effect of liming and P treatment on the soil carbon content was found in this study. The positive effects on the soil fertility due to improved crop growth from optimal lime and P application is also pointed out by Haynes (1984). He also mentions increase in organic matter to be one of the important reasons for this.

Obviously, too high long-term liming levels is not beneficial for the grain yield levels, as the crop is investing a larger share of energy and resources in root growth. Furthermore, application of lime is costly for the farmer, whereby yield reductions induced by this costly treatment is unsustainable. The most yield producing liming levels and P application was found also to have an extensive root system compared to treatments with lower liming levels. However, this extensive root growth was also found for the highest liming level, which must be considered to be over-limed to the yield reduction. It would be interesting to know the long-term effects when liming is withheld for a period, allowing soil pH to decline again. How fast will root growth development and grain yield be responding to postponed liming application?

The experiment provides us with important information on what happens, when a soil is not fertilized with P for a very long time. The differences in the yield levels between the no P treatments and those who have received P fertilizer every year was clear – continuously application of P every year increase the grain yield levels. The root growth development (measured using MR) seemed also to be affected by the P treatment, but the design of the experiment did not investigate all the different liming levels with and without continuously P application. Nevertheless, looking at the results which were generated from the MR on the 4 tonnes lime/ha both with and without P, clearly a poorer root growth development could be found when no P had been applied since the beginning of the long-term experiment.

Taking these findings into boarder perspectives with regards to a long-term sustainable agricultural production, climate change as well as environmental protection can be discussed. Finding an optimal liming and P treatment, which generates a high yield as well as a high production of roots, can be considered important with regards to climate change. As also discussed by Powlson et al. (2011) increased soil organic carbon in the soil from increased yield levels can be a possible climate change mitigation strategy. In the design of the experiment only the application of liming and P is different, the other soil treatments including application of other nutrients is the same. The utilization of other nutrients e.g. nitrogen is dependent on the surrounding growth conditions. Therefore, optimizing liming and P application so a high yield level is achieved must improve the utility of the applied nitrogen fertilizer. In relation to environmental protection, having a high utilization of both P and N is highly important, since these nutrients when lost from the agricultural land, can have a negatively impact water bodies (Kronvang et al., 1993, Kronvang et al., 2009).

Clearly, there are several perspectives in optimizing application of both lime and P. This study have illustrated positive effects on the soil fertility by of optimizing these common agricultural management practices in the long-term, and therefore should these practices not be neglected.

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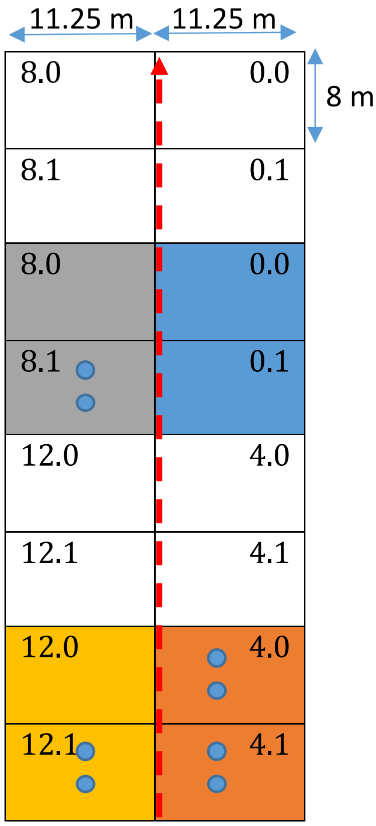
Appendix 1: Description of field design



The long-term field experiment in St. Jyndevad consists of four fields: V1, V2, V3, and V4. The V1 and V2 are grown with spring barley. The design of the experiment is systematic as illustrated in the figure (see treatment codes in the list of abbreviations). The V1 field, where the study was carried out, consists of three replicates of the design illustrated in the figure placed in continuation of each other. The white squares in the figure illustrates treatments, which have not received a single start dose of P in the beginning of the experiment. These treatments was not used in the study. The color coated squares has all received a start dose of 156 kg P/ha in the beginning of the long-term experiment. The size of the brutto parcels is indicated in the figure. The size of the netto parcels for harvest of grain yield can be found section 3.2.4. The MRs was inserted into approximately the middle of the parcels, as shown in the figure (blue circles). The machine, which was used to insert the MR, drove in between the treatments as indicated with the red arrow. The deep soil cores were taken randomly within the netto parcel.

Appendix 2: Analysis of yield data

Description of the experiment and variables.

Yield data have been recorded for eight different treatments with three replicates. The eight treatments consist of combinations of plots with and without continuously applied P, and with four different levels long-term application of liming. The statistical analysis are carried out with a significant level of 0.05. The variable yield is defined as:

Y_{lpr} is the yield of the r^{th} repetition (for r=1,2,3) with the p^{th} level of P (for p=0,1) and the l^{th} level of liming, (with l=0,4,8,12 tons/ha).

It is assumed that the random variables Y_{111} , … , $Y_{12,1,3}$ are independent, normally distributed and have the same variance.

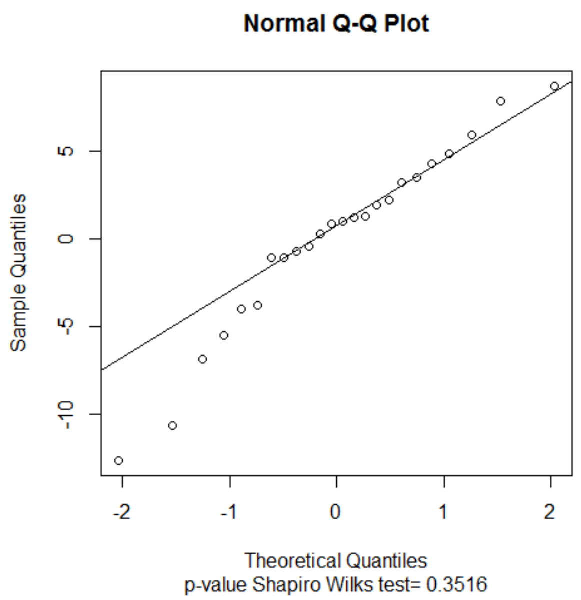
First it is tested whether the observed yields meet the assumptions by fitting a model with an effect modification (or interaction) between the level of liming and the level of P, i.e.

$E\ (Y_{lpr}) = I_{lp},$

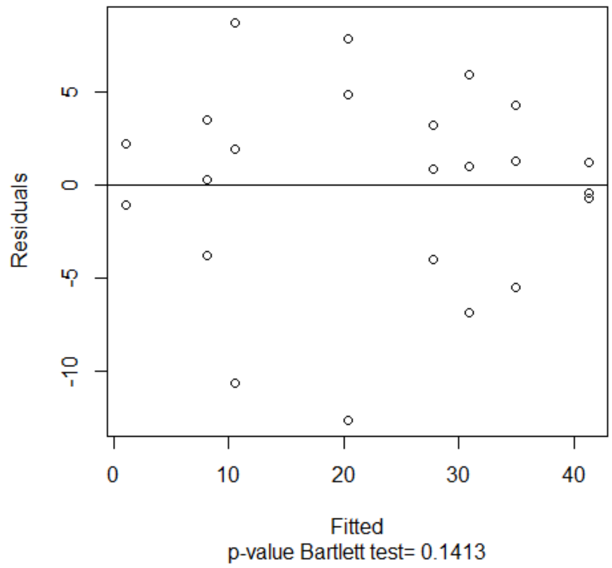
and the residuals of this model (i.e. Y_{lpr}- $E(Y_{lpr})$ is analysed).

The assumptions of normality and variance homogeneity are tested using a Shapiro-Wilks test and the Barlett test.

The Shapiro-Wilks test gives a non-significant p-value of 0.3516, indicating that there are no detectable signs of deviation from the normality. Furthermore, the residuals are checked for normality using a qq-plot. From these tests the model cannot be rejected.



For checking the homogeneity of variance the Barlett test is made, which a non-significant P-value = 0.1413. The model can therefore still not be rejected. Looking at the residuals at the figure below, no systematic errors can be seen.



Next it is checked whether or not the model can be simplified even more. An additive model is created, and defined as:

$E(Y_{lpr}) = L_l + P_p$

The new model is tested against the effect modification model using a F-test. This gives a non-significant p-value larger than 0.05, and therefore it cannot be rejected that the two models are different from each other.

Next the additive model is reduced to a QD model, Quadratic model

$$E(Y_{lpr}) \begin{cases} \alpha 0 + \beta 0 + \gamma 02, p = 0 \\ \alpha 1 + \beta 1 + \gamma 12, p = 1 \end{cases}$$

Where $\alpha 0 + \beta 0 + \gamma 0$ are parameters that define the parabolic response when using p = 0, and $\alpha 1 + \beta 1 + \gamma 1$ are parameters defining the parabolic response when using P = 1. The test gives a non-significant p-value higher than 0.05.

It can now be tested whether or not the additive model can be reduced to the QD model using ANOVA. This test gives a non-significant p-value, and therefore it cannot be rejected that the two models are different from each other.

Next it is tested if the variables $\beta_0 + \alpha l_0^2 = \beta_1 + \alpha l_1^2$. This model is called QD-common:

$$E(Y_{lpr}) = \begin{cases} \alpha 0 + \beta + \gamma 2, p = 0 \\ \alpha 1 + \beta + \gamma 2, p = 1 \end{cases}$$

Where β and γ are parameters that define the parabolic response when using both P = 0 and P = 1.

The QD-model and the QD-common-model is tested using ANOVA. P = 0.798. The test does not show any evidence that the models are different from each other, and can therefore be reduced to the QD-common model.

Next it is tested if $l_0 = l_1$. The model is defined as:

$E(Y_{lpr}) = l + \beta + \alpha l^2$

Testing this model against the QD-common-model gives a significant p-value below 0.05, and therefore the models can be assumed to be different from each other. The model cannot be reduced further than to the QD-common model.

The optimal level of liming is calculated as the maximum of the parabola.

$f(l) = \alpha + \beta l + \gamma l^2$

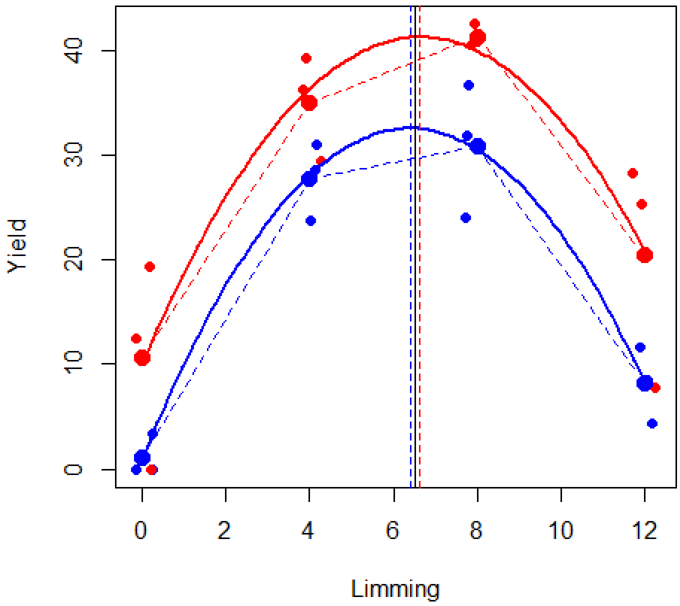
$df(l)/de = f'(l) = \beta + 2\gamma l$

$l = -\beta/(2\gamma)$

The model and the data is illustrated in the figure below.

Conclusions:

- There is an optimal long-term level of liming at 6,4 tons/ha for year 2016.
- The response curve for yield as a response for liming is the same for the two levels of P apart from an added constant.
- Therefore, the optimal liming is not affected by the level of P application.



Appendix 3: Analysis of bulk density data when no P have been added

Description of the experiment and variables.

Bulk density data have been measured for eigth different treatments with 3 replicates. The bulk density have been measured from five different depth in a 1 meter depth soil core. The eigth treatments consist of combinations of plots with and without continuously applied P, and with four different levels long-term application of liming. The statistical analysis is carried out with a significant level of 0.05. The variable bulk density is defined as:

B_{lprd} is the bulk density of the r^{th} repetition (for r=1,2,3) with the l^{th} level of liming, (with l=0,4,8,12 tons/ha) and the d^{th} depth in the soil core (for d=0-20, 20-30, 30-50, 50-70, and 70-100). Due to error in method, the fifth depth have been taken out of all analysis.

First an interaction model is created to test whether there is an effect of liming and depth:

$E\ (B_{ldr}) = I_{ld},$

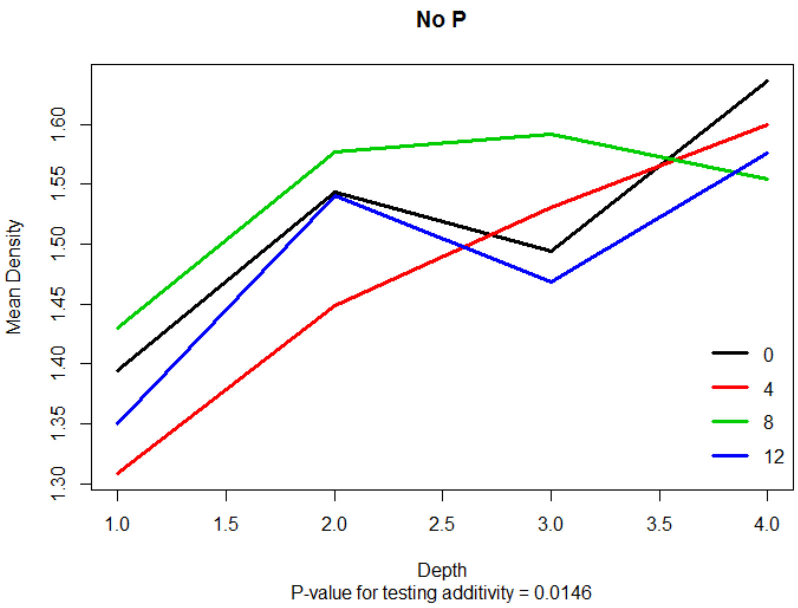
Next the model is reduced to an additive model:

$E(B_{ldr}) = L_l + D_d$

The new model is tested against the effect modification model using a F-test. This gives a significant p-value smaller than 0.05, and there it can be assumed that the two models are different from each other, and therefore the model can be reduced to the additive model.

Conclusion:

There is an effect of depth on the bulk density. Furthermore, there is an effect of the liming level on the bulk density when no P have been added continuously.



Appendix 4: Analysis of bulk density data when P have been added

Description of the experiment and variables.

Bulk density data have been measured for eight different treatments with three replicates. The bulk density have been measured at five different depths in a 1 meter depth soil core. The eight treatments consist of combinations with and without continuously applied P, and with four different levels long-term application of liming. The statistical analysis is carried out with a significant level of 0.05. The variable bulk density is defined as:

B_{lprd} is the bulk density of the r^{th} repetition (for r=1,2,3) with the l^{th} level of liming, (with l=0,4,8,12 tons/ha) and the d^{th} depth in the soil core (for d=0-20, 20-30, 30-50, 50-70, and 70-100). Due to error in method, the fifth depth have been taken out of all analysis.

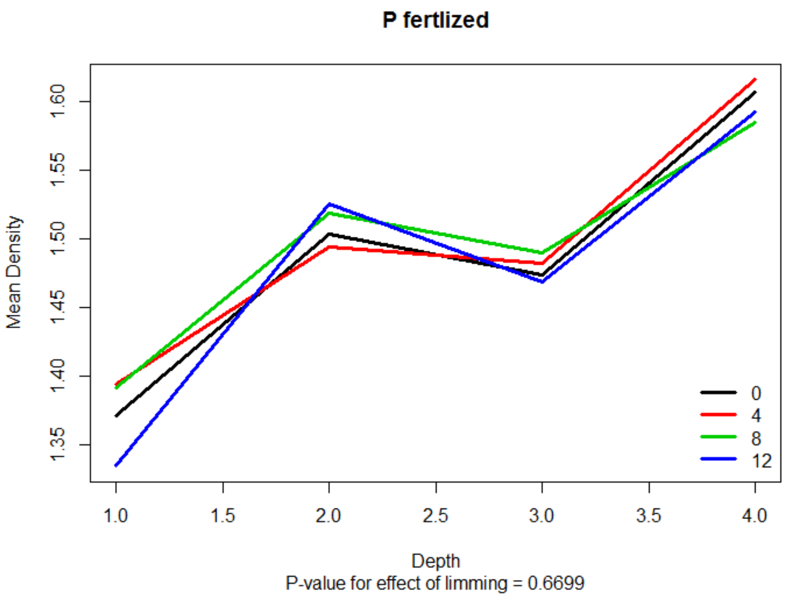
First an interaction model is created to test whether there is an effect of liming and depth:

$E\ (B_{ldr}) = I_{ld,}$

Next the model is reduced to an additive model:

$E(B_{ldr}) = L_l + D_d$

The new model is tested against the effect modification model using a F-test. This gives a non-significant p-value higher than 0.05, and there it cannot be assumed that the two models are different from each other, and therefore the model cannot be reduced to the additive model.



Conclusion:

There is an effect of depth on the bulk density. There is no signs on effects of liming on the bulk density when P have been added.

Appendix 5: Penetration resistance, graph

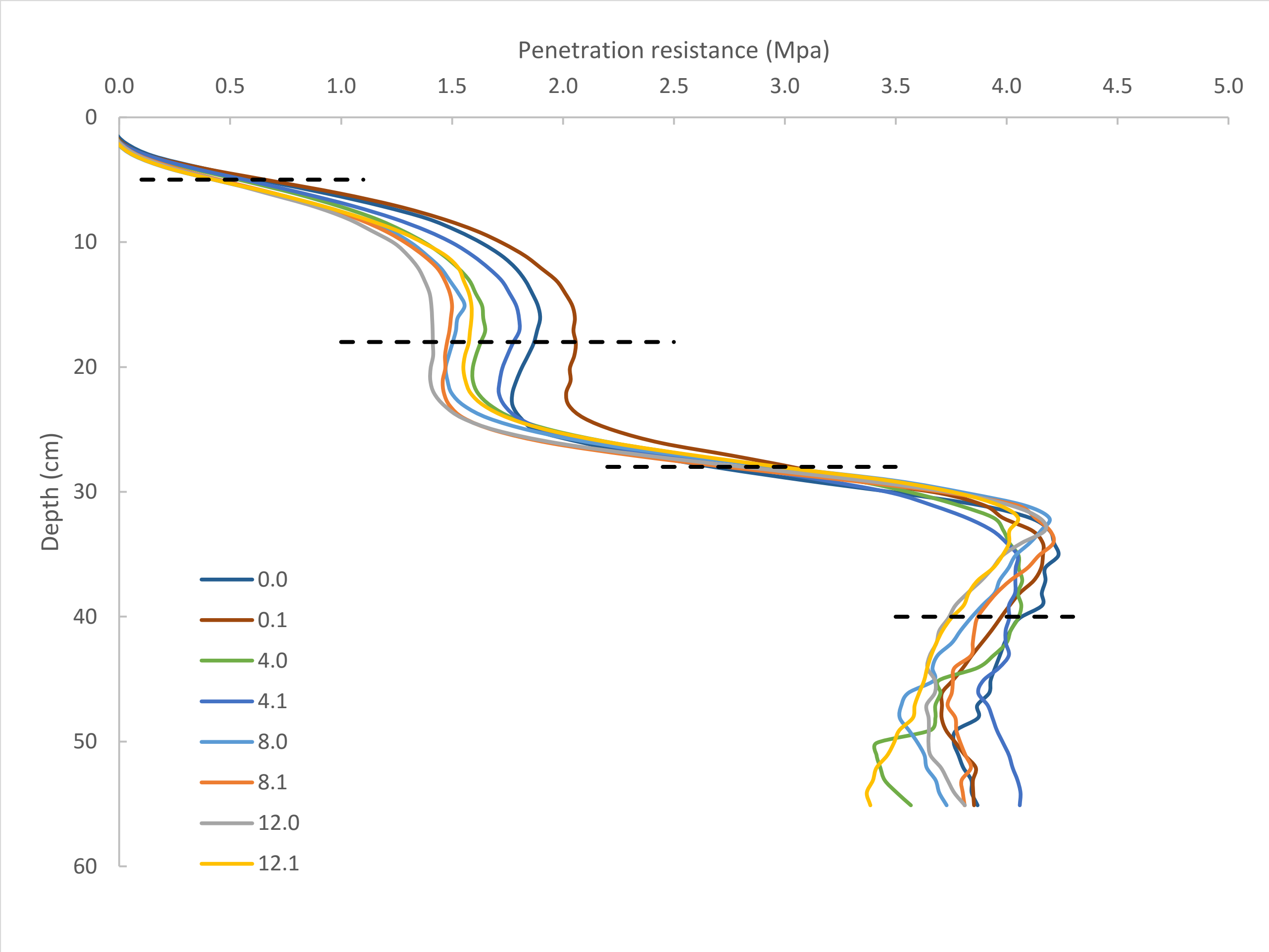


Figure appendix 5.1: *Penetration resistance (Mpa) as a function of depth (cm) for the eight different treatments. Depths at which statistical analysis was performed is marked with a dotted line. Treatment codes are described in list of abbreviations.*

Appendix 6: Penetration resistance, confidence interval

Table appendix 6.1: *Estimate, test of significance and confidence interval for penetration resistance at 5 and 18 cm depth. Letters in the test column show if treatments are significantly different from each other. Treatment codes are described in list of abbreviations.*

P	**Liming**	**5 *cm***			**18 *cm***		
		Estimate	**Test**	**Conf. *Int***	**Estimate**	**Test**	***Conf. Int***
0	0	0.69	A	0.54-0.84	2.06	D	*1.91-2.21*
0	4	0.56	A	0.41-0.71	1.63	CAB	*1.48-1.78*
0	8	0.45	A	0.30-0.60	1.5	AB	*1.35-1.65*
0	12	0.46	A	0.31-0.61	1.41	A	*1.26-1.56*
1	0	0.66	A	0.51-0.81	1.87	DC	*1.72-2.02*
1	4	0.59	A	0.44-0.74	1.77	DCB	*1.62-1.92*
1	8	0.47	A	0.32-0.62	1.48	AB	*1.33-1.63*
1	12	0.45	A	0.3-0.6	1.57	CAB	*1.43-1.72*

Table appendix 6.2: *Estimate, test of significance and confidence interval for penetration resistance at 28 and 40 cm depth. Letters in the test column show if treatments are significantly different from each other. Treatment codes are described in list of abbreviations.*

P	**Liming**	**28 cm**			**40 cm**		
		Estimate	**Test**	**Conf. Int**	**Estimate**	**Test**	**Conf. Int**
0	0	3.06	B	2.91-3.21	3.97	A	3.82-4.12
0	4	2.86	BA	2.81-3.11	3.74	A	3.90-4.20
0	8	2.96	BA	2.86-3.16	4.05	A	3.69-3.99
0	12	3.01	BA	2.71-3.01	3.84	A	3.59-3.89
1	0	2.71	A	2.56-2.86	4.06	A	3.91-4.21
1	4	3.02	BA	2.68-2.98	3.75	A	3.86-4.16
1	8	2.83	BA	2.63-2.93	4.01	A	3.72-4.02
1	12	2.78	BA	2.87-3.17	3.87	A	3.60-3.90

Appendix 7: Minirhizotron RF, confidence interval

Table appendix 7.1: *Total root frequency for the entire length of the minirhizotron*

Treatment	May 17th	May 31st	June 13th	June 28th
4.0	0.021 (0.011-0.040) A	0.044 (0.028-0.068) A	0.076 (0.055-0.106) A	0.181 (0.147-0.220) A
4.1	0.035 (0.021-0.057) A	0.141 (0.111-0.177) B	0.160 (0.128-0.197) B	0.204 (0.168-0.244) A
8.1	0.042 (0.026-0.065) A	0.197 (0.162-0.237) BC	0.280 (0.240-0.324) C	0.322 (0.279-0.367) B
12.1	0.090 (0.067-0.121) B	0.215 (0.179-0.257) C	0.250 (0.211-0.293) C	0.296 (0.255-0.341) B

Table appendix 7.2: *Root frequency for the depth first depth section (0-20.78 cm).*

Treatment	May 17th	May 31st	June 13th	June 28th
4.0	0.097 (0.047-0.190) A	0.250 (0.164-0.362) A	0.375 (0.271-0.492) A	0.611 (0.495-0.716) A
4.1	0.181 (0.108-0.287) AB	0.486 (0.373-0.600) CB	0.611 (0.495-0.716) B	0.764 (0.653-0.848) A
8.1	0.194 (0.119-0.302) AB	0.444 (0.335-0.560) AB	0.667 (0.551-0.766) B	0.778 (0.668-0.859) A
12.1	0.347 (0.247-0.464) B	0.681 (0.565-0.778) C	0.653 (0.537-0.753) B	0.722 (0.608-0.813) A

Table appendix 7.3: *Root frequency for the second depth section (20.78-31.18 cm).*

Treatment	May 17th	May 31st	June 13th	June 28th
4.0	0.056 (0.014-0.197) A	0.028 (0.004-0.173) A	0.167 (0.077-0.325) A	0.639 (0.473-0.777) A
4.1	0.056 (0.014-0.197) A	0.500 (0.342-0.658) B	0.500 (0.342-0.658) B	0.667 (0.500-0.800) A
8.1	0.056 (0.014-0.197) A	0.861 (0.707-0.941) C	0.944 (0.803-0.986) C	1 (0-1) A
12.1	0.278 (0.157-0.444) A	0.806 (0.645-0.904) C	0.861 (0.707-0.941) C	0.889 (0.739-0.958) A

Table appendix 7.4: *Root frequency for the third depth section (31.18-51.96 cm).*

Treatment	May 17th	May 31st	June 13th	June 28th
4.0	0 (0-1) A	0 (0-1) AB	0 (0-1) AB	0.153 (0.087-0.255) A
4.1	0 (0-1) A	0.111 (0.057-0.207) A	0.097 (0.047-0.190) A	0.097 (0.047-0.190) A
8.1	0.028 (0.007-0.104) A	0.306 (0.210-0.421) B	0.542 (0.426-0.653) B	0.653 (0.537-0.753) B
12.1	0.056 (0.021-0.139) A	0.208 (0.130-0.317) AB	0.417 (0.309-0.533) B	0.583 (0.467-0.691) B

Appendix 8: Minirhizotron, root intensity, confidence interval

Table appendix 8.1: *Average root intensity including standard error for the different treatments at the four different dates of observation. The mid of the depth intervals are given in cm.*

May 17th	**4.0**		**4.1**		**8.1**		**12.1**	
Mid of depth interval	**Average**	**Standard error**	**Average**	**Standard error**	**Average**	**Standard error**	**Average**	**Standard error**
10.39	1.74	2.17	7.81	10.88	4.86	2.62	8.51	5.27
25.98	0.69	0.60	0.69	1.20	1.04	1.80	9.03	5.74
41.47	0.00	0.00	0.00	0.00	0.17	0.30	1.22	0.60
62.36	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
90.28	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
May 31st	**4.0**		**4.1**		**8.1**		**12.1**	
Mid of depth interval	**Average**	**Standard error**	**Average**	**Standard error**	**Average**	**Standard error**	**Average**	**Standard error**
10.39	6.94	5.85	25.69	26.05	23.09	3.94	22.74	11.14
25.98	1.04	1.80	11.46	10.05	36.46	5.80	45.14	8.67
41.47	0.00	0.00	1.56	1.38	5.73	6.89	7.99	5.42
62.36	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
90.28	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
June 13th	**4.0**		**4.1**		**8.1**		**12.1**	
Mid of depth interval	**Average**	**Standard error**	**Average**	**Standard error**	**Average**	**Standard error**	**Average**	**Standard error**
10.39	11.63	12.31	41.84	20.69	47.57	25.53	28.82	9.18
25.98	7.64	13.23	36.46	30.78	86.11	48.64	67.01	16.15
41.47	0.00	0.00	2.95	2.67	25.17	4.67	34.20	41.93
62.36	0.00	0.00	0.00	0.00	0.00	0.00	0.17	0.30
90.28	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
June 28th	**4.0**		**4.1**		**8.1**		**12.1**	
Mid of depth interval	**Average**	**Standard error**	**Average**	**Standard error**	**Average**	**Standard error**	**Average**	**Standard error**
10.39	28.82	13.59	61.11	11.43	47.92	14.39	37.33	11.99
25.98	24.31	13.75	52.78	36.42	110.76	26.46	96.18	28.27
41.47	4.51	6.04	4.34	5.06	50.69	46.96	49.65	46.33
62.36	0.00	0.00	0.00	0.00	0.00	0.00	0.35	0.30
90.28	0.00	0.00	0.21	0.36	0.00	0.00	0.07	0.12