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Development, Biomass and Nitrogen Allocation, and Seed-Yield Components in Faba Bean (*Vicia faba*)

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Abstract

Europe has a great demand in plant-protein for food and feed. A high amount of the demand is met by soy import from South America. Europe and Norway aim to become self-sufficient producers in plant-proteins. One of these plant-proteins is faba bean (*Vicia faba*). Faba bean is rich in proteins (~30 %) and has several environmental benefits by being added to the cropping system: By fixing atmospheric N symbiotically, it can contribute to the reduction of synthetic nitrogen fertilizer, and by being used as a break crop in cereal production, it can reduce the risk of diseases, and improve soil structure and fertility. However, the production increase of faba bean in Norway has been slow. Several reasons have been mentioned, such as the yield instability, the right selection of varieties, and the lack of information about the crops development and yield requirements under Norwegian conditions.

This thesis has the aim to contribute information on how different faba bean varieties performed in their development and yield production in Norway in the growing season 2021. The thesis focuses on four topics concerning faba beans: Firstly, phenology, which observes the influence of temperature on the varieties and how temperature affects the yield, as well as observing differences between early and late varieties. Regarding the registration of the development, the two scales BBCH and GRDC are tested and compared. The second topic has its focus on biomass and looks at the question how and where biomass is allocated and how much is accumulated. Nitrogen is at the centre of the third topic, and answers the questions in which plant organs N is mostly allocated and accumulated and how it changes during plant development. The last topic focuses on the seed-yield components, and looks at which extent the variety has an effect on the yield-components and how the previous topics influence the yield.

To research these topics two types of experiments, one greenhouse and one field experiment, were conducted at the Norwegian University of Life Sciences (NMBU) in Ås. In the greenhouse experiment the effect of temperature was tested on the varieties Lielplatones, Vertigo and Sampo. The results showed that at higher temperatures, varieties required more thermal time for emergence until pod maturity. Vertigo was seemingly the most affected by temperature. It was also seen that higher temperature resulted in less yield (lower number of seeds and pods).

The field experiment revealed phenological difference between early (Sampo, Vire, and Louhi) and late (Birgit, Daisy, and Vertigo) varieties. The early varieties needed to sum up less thermal time to reach flowering than the late varieties, and they also reached maturity early by requiring less thermal time. Vertigo started flowering earlier than the other two late varieties but matured

at the same time as them. By using the BBCH development scale a more detailed variation between varieties was seen, whereas the GRDC scale gave an overview of earliness. Chlorophyll content, biomass-, and nitrogen accumulation was higher in the late varieties. Over 80 % of N in the varieties derived from the symbiosis with rhizobium bacteria. There were only minimal differences in protein content between varieties, yet it was observed that Sampo had the highest protein content with around 34 %. A seed-yield component analysis showed that thousand seed weight (TSW) and biomass correlated strongly with the seed dry weight (DW). The late varieties had the highest seed yield and of all varieties Vertigo had the highest yield. Sowing density seems to play a significant role in determining biomass and seed yield and needs to be investigated further. The results contribute to new knowledge about physiological aspects related to plant development and yield formation in faba bean when grown under Norwegian climate.

Additional keywords: *Faba bean (Vicia faba), phenology, biomass, nitrogen, seed yield*

Abbreviations

ANOVA	Analysis of variance
BBCH	Growth stages of mono- and dicotyledonous plants
BNF	Biological nitrogen fixation
	Carbon Nitrogen Sulphur; quantitative determination of these elements, method from J.-B. Dumas
CNS	
DAS	Days after sowing
DW	Dry weight
FW	Fresh weight
GRDC	Grain Research & Development Corporation
GS	Growth stage
LAI	Leaf area index
LAR	Leaf area ratio
LWR	Leaf weight ratio
n.s.	No significance
Ndfa	Dinitrogen fixed from the atmosphere
NIBIO	Norsk institutt for bioøkonomi
PC	Principal component
PCA	Principal component analysis
pr	Per
SE	Standard error
SLA	Specific leaf area
TSW/ TCW	Tousand seed weight/ thouand corn weight

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1. Introduction

Legumes are the second-largest group of food and feed crops globally (Ferguson et al., 2010). Even though legumes display a very important role in our food system, their production within Europe has been decreasing over time (Rossi et al., 2016). Two reasons for this are mentioned: The advantage of producing cereals, which are easy adaptable to many different environments. And second, importing low priced soy for food and feed (Abrahamsen et al., 2019). In recent years however, there has been a changing view on the agriculture in the EU (Billen et al., 2021). There is a shared focus on reducing the negative side-effects of the continuous increase in crop productivity, which puts high pressure on the environment. The focus is being redirected to a more sustainable agriculture that targets environmental issues while simultaneously providing enough food to an increasing population (Billen et al., 2021).

The aim is to make legumes a lasting option in crop systems in Europe. To make that possible the crops have to produce stable yields and rapidly adapt to a changing climate. Norway in connection with other countries in Europe is striving to become self-sufficient in the production of plant proteins (Abrahamsen et al., 2019; Abrahamsen & Waalen, 2020; Billen et al., 2021; Waalen et al., 2019; Watson et al., 2017), yet to achieve this goal it requires more knowledge about legumes, for instance which legume species are suitable for Norwegian conditions and in depth which varieties fit, with regard to different variables. The interest to grow protein crops on cereal areas in Norway is increasing. Instead of making new areas available for legume production, legumes could be added to crop rotations with cereals, that would also reduce the pressure on monoculture production of cereals. (Abrahamsen & Waalen, 2020). An example of a legume that can be produced in Norway is faba bean. Faba bean (*Vicia faba*) is a grain legume, which is rich in proteins and has a positive influence on sustainable agriculture (Duc et al., 2015; Vasconcelos et al., 2020). If it is added to cropping systems it reduces the dependence on N fertilization through its abilities to fix N₂ symbiotically, it also increases biodiversity by being added in crop rotations and by being an important source for pollinators during flowering (Alharbi & Adhikari, 2020; Torres et al., 2012).

Faba bean production in Norway has been scarce (Abrahamsen & Waalen, 2020). The seasons 2018 and 2019 have been causing faba bean instability due to heat and drought stress. This shows that to produce faba bean with continuous stable yield, the varieties need to be better adapted (Abrahamsen & Waalen, 2020). In recent years new trials of faba bean production has been done, to achieve this goal.

In this thesis several novel varieties are being evaluated, that have come on the market in recent years and can be potentially be used in Norway as well. The selection of variety can be divided into different characteristics, depending on the objectives of the cultivation purposes (Prins & Cuijpers, 2020). The choice can be divided into spring or autumn-sown varieties, in addition to early and late maturing varieties, varieties that are rich or poor in tannins, as well as vicine and convicine content (Prins & Cuijpers, 2020). The climate is the most influential factor for the choice of winter or spring faba bean (Jensen et al., 2010). Spring varieties are mostly used in Northern Europe, since they are sensitive to heat and drought (Jensen et al., 2010). They are dependent on precipitation early in the summer to reach high yields (Jensen et al., 2010). In Norway spring-sown faba bean varieties are used.

This thesis concentrates its variety choice on the earliness in pod maturity. The following varieties have been used in the experiments presented in the thesis. The early varieties all derived from Finland. Sampo has been available on the Norwegian market since 2019, with an area share of 15 % (Abrahamsen & Waalen, 2020). Sampo has small seeds, which makes sowing, germination and drying after harvest easier. In addition it has a relatively high protein content and can be used as feed and food crop. Louhi has been new on the market. It is very productive in different soil types and environments, however it has a high content of vicine and convicine, two antinutrients (Pärssinen, 2014). Vire has a lower protein content, however it is high yielding and has a low vicine and convicine content (Pärssinen, 2014). Lielplatones is an old cultivar from Latvia, that has the pod maturity between early and late varieties (Bodner et al., 2018). The late varieties that were selected are all originating from Germany. Vertigo, has been on the Norwegian market as the main variety since 2016 (Abrahamsen & Waalen, 2020). It was grown on 40 % of the area in Norway in 2019 (Abrahamsen & Waalen, 2020). The newer variety Daisy, is a late developing variety in Norway. It has a high seed yield with an additional high protein content (Petersen, 2019b). The last variety tested was Birgit, which is robust and is especially good to use in organic farming (Petersen, 2019a).

Recording plant development is an important task to analyse the correlations between developmental stages and different factors, such as temperature, precipitation, etc. In Europe the BBCH (Growth stages of mono- and dicotyledonous plants) scale is regularly used for faba bean as well as other crops. It is developed after the principles of the Zadok's scale, which developed for the registration of cereal development stages. BBCH was developed in cooperation between several German research centres (Meier, 1997). Another scale that

recently get in use is the GRDC scale (Grain Research & Development Corporation), which was developed for faba bean varieties grown in different areas of Australia and (GRDC, 2017).

2. Objectives

This thesis will concentrate on the objectives of:

1. Phenology

- Analyse the influence of temperature on faba bean varieties
- Describe differences in development between early and late maturing varieties
- Comparison of the two development scales BBCH and GRDC

2. Biomass

- Describe the changes in accumulation and allocation of biomass to plant organs during development

3. Nitrogen

- Describe the changes in accumulation and allocation of nitrogen into plant organs during development

4. Seed-yield components

- Describe in which extent the yield components vary between varieties
- Observe the influence of phenology, biomass allocation and accumulation, and nitrogen allocation and accumulation on the yield

3. Agronomical importance and utilization of grain legumes

The human population is increasing drastically and will rise from the current 7 billion people to 9.3 billion people by the end of 2050 (UNDP, 2010). This, in addition to the global climate change that we are facing, has a direct impact on the agricultural production (Watson et al., 2017). To be able to feed everyone, farmers have to produce the same amount of crop or even more on the same or less cultivation area, which is due to changes in land-use for infrastructure, industry and living area (Billen et al., 2021; Sanz-Sáez et al., 2012). For farmers that often means to use more synthetic fertilizers to increase the crop yield, yet it is causing higher losses of nutrients to the environment, especially nitrogen and phosphorous, thereby negatively affecting the ecosystems and as an end result effect the climate even further through greenhouse gas emission (Billen et al., 2021). Through the increasing wealth of industrial countries, is the consumption of animal products rising, especially meat, fish, and dairy products are primary elements of the diet (Godfray et al., 2010). The consequence has been that a majority of arable land has been used for grain-feed and additionally a high amount of protein-rich feed has been imported from South America, due to low prices of soy (Watson et al., 2017). The most prominent result has been the large scale destruction of rain forest areas in South America, however it has also caused a high loss of crop diversity in Europe, since Europe has made itself dependent on the protein-crop from abroad and now primarily grows cereals for food and feed instead (Huyghe, 2003). In recent years there has been a greater understanding in Europe of the impact of agriculture on ecosystems and climate (Billen et al., 2021). Countries around Europe agree on changing the perspective from only increasing productivity of agricultural products to finding solutions on how to reduce the pressure on the environment while simultaneously producing enough crops to feed a growing population (Billen et al., 2021).

One of the solutions European countries are looking at is to regain self-sufficiency in protein-crops like legumes. Even though legumes are the second-largest group of food and feed crop, after cereals, globally (Ferguson et al., 2010), there has been a visible decline of grain legumes acreages over the last decades (Rossi et al., 2016). Legumes have a variety of positive impacts and can play a key role in a more sustainable agriculture. Due to its symbiosis with rhizobia bacteria in the soil, legumes are able to fix atmospheric nitrogen, which makes them less dependent on nitrogen fertilizer (Heichel & Barnes, 1984). The nitrogen that has been fixed can be used when the legumes are ploughed into the soil as a green manure or when the roots and shed leaves rot in the soil after harvest and ploughed under then, this is especially profitable if legumes are cultivated in rotation with monocultures (Heichel & Barnes, 1984). Soybean is doubtlessly very rich in proteins as a food and feed crop, nevertheless there are also many other

plant species in the family of legumes that are rich in proteins. One of the most common ones are faba beans (*Vicia faba*) (Watson et al., 2017).

Norway wants to: reduce nitrogen run-off, be self-sufficient in livestock feeding with local produced protein-based crops, increase the appeal for meat substitutes in the Norwegian population (Cusworth et al., 2021; Frøseth, 2009). Grain legumes have only been grown in small amounts in Norway, due to competition with low-priced soybean imports from South America (Frøseth, 2009). Norway has a relatively small land area used for agriculture, which is due to widespread mountain areas not suitable for most crop production. Only three percent of the land mass can be used for agriculture and the majority of agriculture is dominated by livestock farms. Therefore, Norway's agricultural income derives mostly from livestock rather than from crop production (Abrahamsen et al., 2019) (Kildahl, 2020; Rognstad & Steinset, 2008). Most agricultural areas are located in the west and south-east and around the fjords, especially the area the lake Mjøsa is an important production area. Due to warm water streams coming from the ocean into the fjords, it is possible to have livestock and crop production even in the northern areas of Norway (Abrahamsen et al., 2019). The country is dependent on pastures and animal production to utilize most of the areas (Abrahamsen et al., 2019). The agricultural areas that can be used for crop production are mainly used for cereal production, those are also the areas that are suitable for grain legumes (Abrahamsen et al., 2019).

As many European countries, Norway has a problem with monoculture, with only cereals being changed between years (Abrahamsen et al., 2019; Waalen et al., 2019). Therefore farmers are starting to add legumes to break the monoculture of cereals. By incorporating the leftover faba bean plant material after harvest, farmers can have a sustainable organic N fertilizer (Abrahamsen et al., 2019). As for now faba bean are not well adapted to the Norwegian climate, the most promising option is to use early varieties even though they are lower yielding than late varieties. Early varieties mature earlier, which is a necessity in areas where there is a longer time-span for the temperature to increase in spring and the temperature decreases fast again in early fall (Abrahamsen et al., 2019; Waalen et al., 2019). The two most important faba bean varieties on the Norwegian market is the Finish variety Kontu as well as the Danish variety Columbo (Abrahamsen & Waalen, 2020). New varieties have to be tested in Norway, since Columbo is no longer available on the EU market and one can see a trend that new varieties are replacing Kontu as well (Abrahamsen & Waalen, 2020). Still the most important varieties for the Norwegian market have been the early maturing ones from Finland (Abrahamsen & Waalen, 2020).

There has been increased awareness in the population about the environmental consequences of meat-based diets in addition to its health concerns and the effect on animal welfare (Cusworth et al., 2021). The consumers are shifting their view to a more plant-based diet (Gonera & Milford, 2018). However, Norway started only in 2017 to bring plant-based products on the market (Alcorta et al., 2021). Norway aims to reduce its greenhouse gas emissions by 40 % before 2030, to meet the Paris Agreement targets (Gonera & Milford, 2018). A proposed solution is to reduce the consumption of red meat by adding legumes as meat substitutes (Alcorta et al., 2021). Legumes have a higher protein content than cereals. In Norway there is no other crop that contains more protein than faba beans (~30 %), which is why it is a good option for a plant-based diet (Øverland, 2021). Yet the majority of the produced pea and faba bean in Norway has been used in the feed industry (Abrahamsen & Waalen, 2020). To be able to use the crop as food, it requires that the faba bean varieties have traits that are important for food quality, as for example the chemical composition and an uniform size of the seeds (Abrahamsen & Waalen, 2020).

4. Faba bean (*Vicia faba*)

Faba bean (*Vicia faba*), also known as horse and broad bean, is one of the most important food and feed legumes in the world. It derived from the Mediterranean and Middle East regions where it is highly used. However, the major producers of faba bean can be found in China, Ethiopia, Australia, UK and Germany. Between 1980 and 2014 the global acreage decreased from around 3.7 to 2.1 mio. ha (Karkanis et al., 2018). Faba bean has a high protein content ranging from 24-30 % as well as a high carbohydrate content around 51-68 %, which both depend on the genotype. (Alharbi & Adhikari, 2020)

Faba bean belongs to the family of Fabaceae (also known as Leguminosae and Papilionaceae) in the genus *Vicia* (Torres et al., 2012) and is a cool-season legume (Etemadi et al., 2019). There are different ways to classify the faba bean types. The most common one used nowadays is classifying into the seed size. The subtype *minor* has a seed size to up to 0.6 g, *equina*, has flattened seeds, which are between 0.6 and 0.9 g, and the last subtype is *major*, which can weigh up to 2 g. The different forms can easily cross, which is why the genetic diversity derives only from geographical isolation, rather than from botanical traits. Thereby no wild forms have been found. (Torres et al., 2012)

The right selection of variety is important for a significant establishment of the crop. To accomplish that it requires knowledge about environment, disease tolerance, the crops purpose and market value (Etemadi et al., 2019). The classification in seed size, needs to be considered when choosing the production purpose of faba bean (Etemadi et al., 2019). While the small seeded varieties are used as animal feed or cover crop, are the big seeded varieties used for the human nutrition in a dried or fresh form (Crépon et al., 2010). In Australia, Europe and Canada modern cultivars are used, in some other countries traditional landraces are still in use and are grown as single crop or in mixture with modern cultivars (Etemadi et al., 2019). Current varieties that could be high yielding and more adaptable to a fast changing environment are being evaluated (Etemadi et al., 2019).

Faba bean can play a significant role in sustainable agricultural systems (Karkanis et al., 2018), by reducing the use of synthetic nitrogen fertilizer, reducing the CO₂ emissions, improving soil structure and fertility as well as increasing yield of the following crop in a rotation (Etemadi et al., 2019). To make *V. faba* a lasting and positive option in cropping system several factors in cultivation need to be considered for an optimal performance of faba bean.

Faba beans can nearly grow in any soil, however they require a well drained soil (Øverland, 2020). Even though they have a long taproot they are susceptible to drought due to little amount of fine roots, which is why it is important to avoid soils that easily dry out (Øverland, 2020). The soil should have a pH ranging from 6.5 to 9.0 (Jensen et al., 2010; Karkanis et al., 2018; Øverland, 2020). While in the Mediterranean countries faba bean is sown in winter sometimes as early as end of summer with the goal to harvest earlier, faba bean in the Nordic countries need to be sown end of winter in spring to reduce the risk of frost damage (Karkanis et al., 2018). Sowing can start when the temperature of the sowing depth has reached around 5°C (Øverland, 2020). In Norway the right sowing time is an important factor, not necessarily for the yield but mostly to reach pod maturity before the end of the growing season (Øverland, 2020). Late varieties should be sown around April to May, while the early varieties can be sown until the 10th of May (Øverland, 2020). The seeding rate is influenced by the TSW (thousand seed weight) (Karkanis et al., 2018) and is dependent on the further usage of the crop, since it not only has an impact on the final yield but also on the extent of the nitrogen fixation and the use as cover crop (Etemadi et al., 2019). The crop needs to produce around 4000 to 4500 kg m² of biomass to contribute sufficient N for the following crop (Parr et al., 2011). There are different information about the sowing density of faba bean, in Norway a seeding density of around 60 plants per m² has been recommended, however some varieties such as Sampo and

Louhi require 80 plants per m². Faba bean and legumes in general are self-sufficient in N. However there have been some discussion if the effective fixation and productivity increases with adding “starter N” to the sown legume (Etemadi et al., 2019). Thereby N limitation in the beginning of the growing season can be reduced. Faba bean has shown that it not necessarily loses its efficiency in fixation with limitations in N, and does therefore not require “starter N” (Etemadi et al., 2019).

V. faba added to crop rotation has usually the function as a break crop in the monoculture production of cereals (Øverland, 2020; Zou et al., 2015). It can also be added to vegetable crop rotations, which is usually happening in Mediterranean regions. There faba bean is used as a pre-crop prior to crops from either the Cucurbitaceae or Solanaceae families (Etemadi et al., 2019). There should be a growing break of faba bean on the same field of 7-8 years to reduce the risk of diseases (Øverland, 2020).

Weed infestation in faba bean can impact the yield greatly by sometimes reducing the yield to 50 % (Frenda et al., 2013; Karkanis et al., 2018). It is important to reduce weed pressure directly from the start. Weed removal between 25 to 75 DAS (days after sowing) have been recommended (Karkanis et al., 2018). The infection with diseases and insects can be another problem causing yield losses. Fungal diseases such as Ascochyta blight (*Ascochyta fabae* Speg.), chocolate spot (*Botrytis fabae* Sard. and *Botrytis cinerea* Pers.), and rust (*Uromyces viciae-fabae* (Pers.) J. Schröt) are especially present in wet conditions and can cause a yield up to 30 % (Karkanis et al., 2018; Øverland, 2020). The risk of infection with Ascochyta blight can be reduced not only with fungicide application, but also through changes in management such as crop rotation, resistant variety usage and late sowing (Karkanis et al., 2018; Stoddard et al., 2010). Frequent application of fungicide has shown to be effective in the treatment against rust and chocolate spot in addition to intercropping with cereals and low planting density (Karkanis et al., 2018). Resistant varieties have also been detected for chocolate spot and rust (Karkanis et al., 2018). The pea leaf weevil (*Sitona lineatus* L.) as well as the broad bean weevil (*Bruchus rufimanus* Boh.) are the main insects infecting *V. faba* (Evensen et al., 2016). The larvae insects cause reduced N fixing abilities, since they feed from the nodules, it also feeds of the leaf edges and the pods and seeds as an adult insect (Karkanis et al., 2018; Øverland, 2020).

The seeds of *V. faba* are great energy sources, which have high amounts of fibre, lysine rich proteins, minerals, vitamins, as well as antioxidants (Crépon et al., 2010). They are either consumed in fresh or dry form. The harvest for the fresh consumed seeds is in the pod filling

stage, however prior to the drying stage (Karkanis et al., 2018). For dry seed production the proper harvesting time is critical (Karkanis et al., 2016). The moisture content is required to be around 14-15% (Karkanis et al., 2018). Even though *V. faba* has many favourable valuable compounds such as high amounts of essential amino acids (Koivunen et al., 2016), the seeds can contain various antinutrients (saponins, tannins, lectins) (Karkanis et al., 2018). The most important antinutrients in faba bean are the level of vicine and convicine (v-c) (Karkanis et al., 2018). These two compounds are stored in the cotyledons and if present in faba bean it can cause favism in people who have a genetic inherited glucose-6-phosphate dehydrogenase deficiency (Khazaei et al., 2019; Luzzatto & Arese, 2018). The v-c level can be reduced through processing prior to cooking, like soaking seeds in water (Khazaei et al., 2019). However there have also been discovered varieties with low v-c content, which has been an aim in recent breeding projects (Khazaei et al., 2019). The seed size is an important factor influencing the use on the market. While *major* types can be used as food for fresh and dry consumption, the minor and *equinoa* types are used in feed (Crépon et al., 2010).

4.1 Phenology

Phenology records the crops seasonal developmental events (Koch et al., 2007). Examples for phenological stages are: leaf unfolding, flowering, fruit ripening, and senescence (Koch et al., 2007). The length of the growing season is defined by the timing of growth and senescence (Cleland et al., 2007). Timing the shift from the vegetative stage to the reproductive stage is important for an optimal seed production (Cleland et al., 2007). Observing the crops phenology is necessary to connect the development of the crop with the prevalent growing conditions (Bodner et al., 2018), since the physical environment is interacting with the development, growth, and the production of the crop (Zabawi & Dennett, 2010). For example, temperature as a factor alone and also together with photoperiod has a great influence on the plants development, and can change how long either the vegetative or the reproductive stage lasts (Cleland et al., 2007). The phenology is therefore directly providing information on the species productivity (Cleland et al., 2007).

Faba beans are annual crops that can either be spring-sown or autumn-sown depending on its location. In north and central Europe, winters are usually long and cold with shorter warm periods, which is why the crop has to be sown in spring as early as possible, to harvest before the weather conditions are not suitable anymore (Bodner et al., 2018; Torres et al., 2012). Therefore, to achieve a high yield, looking at the crops phenology and physiological traits is a

necessity, especially in regards to the climate change that we are facing. (Alharbi & Adhikari, 2020)

The most sensitive phenological stages, which are important for yield build-up, are between flowering and pod development (Bodner et al., 2018; Duc, 1997; Mwanamwenge et al., 1999). Temperatures ranges from 20 C to 25 C are optimal for flowering (Patrick & Stoddard, 2010). Temperatures higher than optimal can lead to abortion, as well as decreased number of pods and seeds (Bodner et al., 2018).

The leaf expansion and the continuous development of the canopy in addition to the photosynthetic capacity are interacting and thereby influencing the yield. An increase in temperature can increase the number of leaves, yet leaf area and growth rate and at the end the grain yield is decreasing.(Alharbi & Adhikari, 2020)

4.1.1 Morphological and physiological description

Vicia faba has an erect growth. The stems are usually hollow and square-formed (Smither-Kopperl, 2019). They either have one single stem or they have the ability to develop branches, that arise from the leaflets between internodes. The number and shape of leaves can vary depending on the variety or landrace. The leaves do not have tendrils as for instance peas have. The dark spot in addition to an extrafloral nectary is a particularity for *V. faba* species (Torres et al., 2012). The colour of the flowers can vary between white, brown, and/or purple. The black melanin spot is mostly situated on the wings (Duc, 1997). As a member of the Papilionoidea, the flowers can be distinct by a papilionate shape. The flowers arise in clusters from the axils of the leaves (Smither-Kopperl, 2019). They have one ovule and ten stamens. One of the stamens is free, whereas the filaments of the remaining nine are assembled in a sheath surrounding the ovary. At the bottom of the corolla tube, the nectaries are developed. Usually the racemes are short, yet the number of flowers can vary depending on the cultivar. It can vary from one-two to ten-twelve, the *minor* cultivars can even have more flowers. However, the average number lays around four-six flowers on a raceme (Torres et al., 2012). At anthesis the flowers can reach two to three centimetres (Duc, 1997).

The pods are cylindrical shaped with the outside surface that is silky and green, whereas the inside is white wool-coated (Smither-Kopperl, 2019). Genetic and environmental factors are influencing the number of pods that are produced (Torres et al., 2012). One to four pods are produced in average on each node. *V. faba* is known to have a poor flower-pod ratio, which

means that the plants use a lot of energy and assimilates to produce flowers however the plants are not capable to produce enough assimilate for the same amount of pods (Duc et al., 2015; Torres et al., 2012). That's is why many of the flowers that are being produced, do not develop further to pods. According to Rowlands (1960) only 24 % of the ovules develop into maturing seeds. Most of the seeds that develop can be found on the lower nodes (Rowlands, 1960).

Harvesting and processing method can be dependent on the cultivar. If the pods are dehiscent, they are usually used as vegetables, since they will open during harvesting. Like some varieties of pea and common bean, indehiscent faba bean cultivars can be harvested as whole. Depending on the subspecies (*minor*, *equina* or *major*), the length of the pod can vary. The pod length can vary between two to three cm to 30-40 cm and in some major cultivars the pod length can reach 50 cm (Torres et al., 2012). Even the structure of the pods is different between the subspecies (Duc, 1997). The pods of the *minor* and *equina* are shorter and erect, whereas the pods of the major are long and hang (Duc, 1997). The pods of the *major* subspecies can have 8-12 ovules, whereas the *minor* subspecies only contain three to four ovules (Alharbi & Adhikari, 2020).

A single seed weight averages around 0.2 to 2 g, however larger seeds have a higher protein content with more competitive seedling (Alharbi & Adhikari, 2020). In areas with warm climates, the upper layer of the soil dries out easily, therefore large seeded varieties have an advantage. Larger seeds can be sown deeper into the soil where moisture is still available and can easily emerge from there (Alharbi & Adhikari, 2020). Seed size is a very important component with a high heritability, since it is correlated positively with the yield (Duc, 1997). The colour of the seeds can be differentiated as well. The colours ranges from yellow, green to black or violet, with some seeds having additional brown spots or stripes surrounding the hilum (Duc, 1997). The hilum however can be either black or clear (Duc, 1997).

Vicia faba has a strong taproot, such as many legumes have (e.g. alfalfa), however different than alfalfa the rooting system is more shallow with various lateral roots (Smither-Kopperl, 2019). Breeders have developed cultivars with a deeper rooting system, which are proven to be more drought tolerant (Zhao et al., 2018). Through the symbiosis with the soil bacteria *Rhizobium leguminosarum* *bv. viciae*, the plants develop nodules on the roots, which are actively fixing nitrogen from the air (Jensen et al., 2010). Some faba bean cultivars also form a symbiosis with mycorrhiza (Köpke & Nemecek, 2010).

4.1.2 Growth and Development

Vicia faba has an indeterminate growth habit, which means that the different development stages do not happen in a chronological order, but simultaneously. However, through artificial mutagenesis a determinate growth habit has been achieved by backcrossing the *ti* allele which is responsible for the growth habit. (Torres et al., 2012)

V. faba is usually grown in temperate to semi-arid climates, meaning that most of what is known about the growth habit and development of faba bean derive from these areas, which might not be applicable for the production of faba bean under Nordic climates (López-Bellido et al., 2005). In general the development of plants is specifically controlled by temperature as well as moisture and photoperiod (Alharbi & Adhikari, 2020). Emergence can usually be starts around 200-300 degree-days, followed by 830-1000 degree days for the start of flowering, with a base temperature of 0° C (Alharbi & Adhikari, 2020). The growth of one node per degree days of faba beans, can differ in regards to the number of degree days, the genotype, or the other environmental conditions (Alharbi & Adhikari, 2020). The genotypes can be very sensitive to variations in photo-thermal system, there by affecting the time spend to reach each phenotypic stage (Etemadi et al., 2019).

A low germination rate can be caused by a reduced water availability in the soil, thereby a greater conductivity of the available water is needed and as a result the germination rate declines (López-Bellido et al., 2005). After emergence another challenge that can occur is the decreasing spacing between plants, which leads to an enlarged plant competition (López-Bellido et al., 2005). The competition will on the one hand increase the leaf area index (LAI) (López-Bellido et al., 2005).

In the beginning of the vegetative development the accumulation of dry matter correlates with the number of plants (López-Bellido et al., 2005). The length of the vegetative development as well as the weather can affect the numbers of stems (López-Bellido et al., 2005). The number of branches can also be influenced by the growth type, for instance varieties with a determinate growth habit produces more branches, however some of them can also be infertile (Alharbi & Adhikari, 2020). Most of the germplasm deriving from Denmark and Finland have one single stem, due to the fact that the main stem usually produces a higher number of pods (Alharbi & Adhikari, 2020). During the vegetative growth, the most limiting factors are the competition between plants for solar radiation, nutrient and water supply (López-Bellido et al., 2005).

The leaf area regulates the capacity of how much light can be intercepted and the extend of transpiration (Baldissera et al., 2014). The growth of leaf area is influenced by day and night

temperatures, as well as availability of water and the plant density (López-Bellido et al., 2005). The highest leaf area index (LAI) during the begin of flowering until pod filling (López-Bellido et al., 2005). Afterwards the LAI values decrease (López-Bellido et al., 2005). In a study from López-Bellido et al. in 2005 they discovered that the LAI reaches its maximum of sometimes greater than 4 at around 220 to 230 days after sowing (DAS).

The summation of dry matter is slow in the beginning and is increasing over time (Pilbeam et al., 1989). It accelerates until start of pod filling. Due to senescence and shedding of leaves, the dry matter will decrease again (López-Bellido et al., 2005). In addition to dry matter in leaves, the plant will go through net dry matter losses in stems and pods, since the assimilates are being relocated into the seeds (Stützel & Aufhammer, 1992).

The reproductive stage regards after flowering the production of pods and the seeds as well as the period of grain filling (Aguilera Díaz, 1987). Compared to the vegetative development, is flowering, pod filling and maturity prolonged (Adisarwanto & Knight, 1997). Genotype is impacting the number of seeds per pod the most, which it is the most constant trait (Bond et al., 1985). On the other hand the number of seeds per pod between nodes can vary more (López-Bellido et al., 2005). After flowering it can take between 25 to 35 days for the accumulation of biomass in the seeds (Adler & Müntz, 1983).

4.2 Biomass

Carbohydrates are given in the form of sugars, starch and cellulose, making it the main component of the biomass (Alharbi & Adhikari, 2020). The biomass has a direct impact on the seed yield, which is why an effective distribution to the sinks, important for the overall grain yield is important (Alharbi & Adhikari, 2020; Musallam et al., 2004).

The principles of translocation in plants is accomplished by a network connecting essential organs. The two pathways known are the xylem and the phloem. In the xylem, nutrients and water are transported from the roots to the shoots, the phloem on the other side is primarily allocating the carbon deriving from the photosynthesis to the roots and reproductive organs like flowers, pods and seeds. These two vascular pathways provide all the necessary assimilates in fitting proportions from source organs to sink organs. However the assimilates are also stored in sink-site, which means in the example of faba bean assimilates are also stored in the pod walls and later distributed further to the seeds. (Atkins & Smith, 2007; Hay & Porter, 2003)

In faba bean, biomass accumulation is related to the grain yield (Köpke & Nemecek, 2010). The accumulation of biomass varies in which developmental stage the plants are in. In general, an increase of biomass (dry matter), after emergence happens, yet the biomass decreases again during the reproductive phase (Pilbeam et al., 1989). During senescence most of the dry matter is lost, as a result of increased respiration (López-Bellido et al., 2005). In the vegetative growth most of the total dry matter derives from the stem and only a small proportion from the leaves (Loss et al., 1998). However after pod filling most of the biomass will be finally distributed to the seeds (López-Bellido et al., 2005). Maximal biomass accumulation is achieved at the start of seed filling, which means that after this the weight of roots, stem and pods will stay constant (Etemadi et al., 2019). Only the leaf weight has been reported to decrease (Singh & Schwartz, 2010). Croser et al. (2003) has observed that compared to other legume, faba beans before the end of flowering accumulate two-thirds of their overall dry matter (DM). Soybean for instance has a higher variation of DM accumulation before flowering that varies between 30 to 67 % and lentils have lower accumulation that stays around 16 to 30 % before flower initiation (Ayaz et al., 2004; Croser et al., 2003). The partitioning of biomass to the different organs is influenced by different factors, such as the crops phenology or the competition between organs. Between different organs there can be a stronger competition than between others. For example, the competition for assimilates between stems and leaves, between pod walls and seeds, but also the competition between flowers on the same raceme, and between the pods on different nodes. (Alharbi & Adhikari, 2020; Etemadi et al., 2018b; Etemadi et al., 2019)

Depending on the plant's phenology, the competition for assimilates in the reproductive stage, can be divided into phases. The first phase is over the entire period of flowering, in which flowers and young pods compete with the vegetative parts. The next phase happens from the middle of the flowering period, at that point pods among themselves within the same inflorescence compete with each other. The severity of competition on a plant, is determining the rate of flower abortion and as a result determining the seed yield. The last phase is during the physiological maturity of the pods. The pods compete with each other depending on their placement on the plant. (Alharbi & Adhikari, 2020) Often the pods that are placed on the end of the raceme, cannot fully develop and will not complete maturity (Alharbi & Adhikari, 2020). How much biomass is partitioned in different development stages can also depend on the growth habit (Eckert et al., 2011). During flowering and pod set, plants with an indeterminate growth produce more biomass than plants with a determinate growth (Eckert et al., 2011).

In case of abiotic (drought, salinity) or biotic stresses, the plant will change its priorities in biomass allocation. Under water or nutrient deficit, more biomass will be allocated into the roots. When ever the plants are experiencing a low light period, the biomass will be allocated to the leaves to increase leaf area and in addition increase the number of leaves. Too low or too high temperatures can affect the loading of carbon through the phloem into the different sinks and in general change the source-to-sink relationship (Lemoine et al., 2013). In chilling and frosty conditions, faba bean accelerates the concentration of free sugars in the leaves (Alharbi & Adhikari, 2020). When nutrient deficiency is occurring, the plant favours the roots, and distributes more biomass there (Lemoine et al., 2013).

4.3 Nitrogen

Adding Faba bean into cropping systems can have many different advantages. Life cycle assessments (LCA) have proven that faba bean or legumes in general have a positive impact on the environment (Köpke & Nemecek, 2010). One of the advantages is the ability of legumes to fix atmospheric nitrogen through the symbiosis with *Rhizobium* bacteria, the process is called biological nitrogen fixation (BNF) (O'Hara, 1998). In comparison, most crops depend on the addition of synthetically produced nitrogen fertilizer into the soil, which causes higher risks of eutrophication and greenhouse gas emissions (Arora et al., 2013). Therefore, by using legumes in crop rotations, as a form of green manure, or in grass mixtures, the fixed nitrogen from the atmosphere can enter the soil (Heichel & Barnes, 1984; Köpke & Nemecek, 2010). Even in the presence of high amounts of nitrogen in the soil, faba bean is able to still fix the majority of nitrogen from the atmosphere, (Hardarson et al., 1991) (Köpke & Nemecek, 2010). Measurements of the nitrogen from the atmosphere (NdfA) have shown that faba bean reached between 60 to 80 % of NdfA values (López-Bellido et al., 2006). However this values are dependent on the amount of nitrogen available in the soil that can be taken up by faba bean (Köpke & Nemecek, 2010).

There is a high variation on the amount of nitrogen fixed by faba bean, since it depends on the growing conditions and the genotype (Köpke & Nemecek, 2010). BNF correlates with grain yield, in addition to the amount of nitrogen bounded in the grain (Hauser, 1992). The amount of fixed N in the plant changes with development. First nodules are produced two weeks after emergence and maximal fixation rate is reached after flowering. After flowering is also the phase when pods and grains are strong sinks for nitrogen and other assimilates (Köpke & Nemecek, 2010; Vinther & Dahlmann-Hansen, 2005). Higher amounts of fixed N can be

achieved by adjusting agronomic practices to establish a good crop growth, thereby a yield increase will be seen as well (Peoples et al., 2009). One way to adjust agronomy, is by choosing the right cultivar (Köpke & Nemecek, 2010).

Crop productivity is highly impacted by N, since it plays a key role in different plant processes, specifically in the photosynthesis in the form of Rubisco and in the protein content of the grains (Bertheloot et al., 2008). N is involved in the accumulation of biomass, they are interacting and determining the crops productivity (Bertheloot et al., 2008). To have an optimal crop productivity and to prevent nitrate leaching it is important to understand N uptake and its distribution in the crop (Gastal & Lemaire, 2002). To improve nitrogen use efficiency (NUE) one needs to decide which processes in the plant can be altered (Gastal & Lemaire, 2002). The accumulation of N content is closely correlated to the biomass production (Bertheloot et al., 2008; Gastal & Lemaire, 2002; Salon et al., 2001). Therefore crop growth is determining N content (Gastal & Lemaire, 2002). During development the N accumulation and distribution is changing. At the beginning of the development a high amount of the N is taken up either through the roots or through photosynthesis, therefore much of N can be present in the leaves (Gastal & Lemaire, 2002). Further in the development, more N is allocated to the stem. With the start of the reproductive phase, pods and seeds have an increased sink strength (Salon et al., 2001). The amount of N that can be accumulated in the seeds depends on the N supply of the crop, in addition to the seeds node position (Salon et al., 2001). Pods on lower nodes potentially receive higher amounts of N than the pods on nodes positioned higher on the plant. Sinclair and De Wit (1976) observed a “self-destructive” behaviour during seed-filling in soybean. They stated that the seeds have a great demand for N, in fact so great that the N that is allocated to the seeds from the vegetative organs is causing a decrease of physiological activity and as an end result the crop is senescent (Sinclair & De Wit, 1976). This has also been observed in pea (Salon et al., 2001).

Faba bean seeds are important protein sources as food and feed (Köpke & Nemecek, 2010). The seeds consist of important and digestible seed components, such as starch, proteins, and fibres (Hauggaard-Nielsen et al., 2011). However there is a high genotypic variation in the protein content of the seeds (Hauggaard-Nielsen et al., 2011). The European varieties have a protein content up to 30 % of the seeds dry weight (DW), which is a standard for the market (Hauggaard-Nielsen et al., 2011). The chronological order of accumulation of nitrogen is from the leaves to the pod walls to the seeds, whereby the seeds have the highest protein content compared to the other two organs (Etemadi et al., 2018a). The accumulation of nitrogen takes

place during the seed-filling period (Bender et al., 2015). Yet how much nitrogen is allocated to the seeds varies between varieties (Etemadi et al., 2018b).

4.4 Seed-yield components

A reason why farmers are sceptical to produce faba beans, is the high yield instability, where the yield can vary from year to year (Alharbi & Adhikari, 2020; Hebblethwaite et al., 1983). Faba bean is known to have low yields, ranging from 0.6-1.0 t/ha worldwide (Torres et al., 2012). Yet, faba bean is the main protein source in many countries, due to limitations in animal derived proteins (Abdalla et al., 2015). Therefore yield stability has become one of the main breeding objectives in recent years (Torres et al., 2012).

The components that are most important to consider for the yield determination in faba bean, are the number of plants per unit area, the number of pods per plant, the number of seeds per pod and the seed weight per plant. The grain yield can be increased, when there is an increase in those components. Some of the components contribute more than others to the seed yield. (Alharbi & Adhikari, 2020)

These components are to see in relationship to each other and independent as single traits, which help to understand the seed yield physiology of legumes (Greven et al., 2004). Even though a change in on seed-yield component will not automatically change the final yield, it has been shown that higher seed yield can be achieved by maximizing all components (Mirshekari et al., 2013). The recorded seed yields across countries have shown high discrepancies, which can be explained by the variations of seed sizes that considerably changes the planting density, the number of seeds, but also the usage of faba bean (Etemadi et al., 2017). Etemadi et al. (2015) reported a fresh pod yield of eight faba bean varieties a variation of seed yield from 2052 to 17038 kg/hm². Coelho (1987) also mentioned a broad variation in seed yield of 4750-10950 kg/hm² (Coelho, 1987; Etemadi et al., 2015). Each yield component can have different degrees of importance in different legumes (Ayaz et al., 2004). For instance in pea and chickpea the most important component is the number of pods per plant (Pandey & Gritton, 1975; Siddique & Sedgley, 1986). The general number of pods per plant is influenced by environment and management practices (Knott, 1987). But the variation in number of pods is dependent on the variety (Ayaz et al., 2004). Usually when there is a low number of pods per plant, it is caused by competition between plants, which is why an optimal seeding density is important (Moot & McNeil, 1995). Nevertheless, the amount of pods per m² increases with a higher plant

population (Ayaz et al., 2004). In different reports it has been stated that variations in yield mostly derived from variations in pods per m² (Ayaz et al., 2004).

There have been some misleading information regarding the seed size determining the final yield, which argues that larger seeds produce a higher yield than smaller seeds (Etemadi et al., 2018a). However by adjusting seeding density for each seed size adequate yields can be achieved (Etemadi et al., 2018b). The seed size is a stable yield component, therefore the number of seeds per area is the most important factor for determining seed yield, it also contribute to higher variations in seed yield than seeds per pod or the average seed weight (Ayaz et al., 2004). In a study from Ayaz et al. (2004) where they tested the variability of yield of four different grain legume species, they concluded that the number of pods and seeds per m² affected the yield the most. This would mean that seed yield could be improved through influencing those two components (Ayaz et al., 2004).

The determination of the yield can be influenced by other factors as well, such as flower induction, the detainment of flowers, as well as the development of the seeds (Patrick & Stoddard, 2010). Another bigger impact factor is the management practice (Ayaz et al., 2004). Different management behaviours, can influence each component, that at the end can increase or decrease the final seed yield (Ayaz et al., 2004).

As a cool-season legume, the faba bean yield is sensitive to high temperatures and drought (López-Bellido et al., 2005). For an optimal yield, early maturing varieties have been proposed (Etemadi et al., 2019). Those have been observed to require less thermal time (GDD) (Etemadi et al., 2018b).

5. Material and Methods

Two different trials were conducted in the period of February 2021 to September 2021. The first experiment was done under controlled conditions in the greenhouse, while the second experiment was part of a field trial. Material and Methods (as well as the results and discussion) will therefore be partly separated into greenhouse and field experiment, some sections will be dealt with together, which have including information for both experiments.

5.1 Plant material

For the greenhouse experiment only three of the varieties were chosen (Table 1). The complete field experiment on the other hand consisted of 24 different varieties from different breeders. Nonetheless this thesis will concentrate on six specifically chosen varieties for the field experiment. Sampo and Vertigo were also chosen in the field, so they could be compared with the same varieties in the greenhouse. Three of the varieties are known to be early developing, the other three are late developing. The varieties on the field are part of a bigger experiment in collaboration with NIBIO and Graminor.

Table 1: Complete overview of the varieties used for greenhouse and field experiment with origin and breeding company.

Variety	Origin/ Breeder	Earliness	Used in
Sampo	Finland/Boreal	Early	Greenhouse- and field experiment
Vertigo	Germany/NPZ	Late	Greenhouse- and field experiment
Lielplatones	Latvia	Between early and late	Greenhouse experiment
Louhi	Finland/Boreal	Early	Field experiment
Vire	Finland/Boreal	Early	Field experiment
Daisy	Germany/P.H. Petersen	Late	Field experiment
Birgit	Germany/P.H. Petersen	Late	Field experiment

5.2 Recording plant development

In the greenhouse experiment the development of each plant was observed and registered using the BBCH scale (Meier, 1997), which is uniformly used for mono- and dicotyledonous plants. The codes in the scale are separated into the main phenological development stages, those can be identified by morphological characteristics (Table 2). In the field experiment the development stage of each plot was registered using not only the BBCH scale (Meier, 1997) but also the GRDC scale (GRDC, 2017), to compare the efficiency and complexity of the two scales (Table 3). The GRDC scale was developed in Australia, it divides the developmental stages of faba bean in five main stages, with each of those having additional sub-stages, which count as growth stages (GRDC, 2017). In the concept of both greenhouse and field experiment the thermal time from sowing to harvest was summed up as well as the days after sowing (DAS)

Table 2: A summary of the BBCH scale for faba bean with code and description (Meier, 1997).

BBCH	Code	Description
0 Germination	00-09	Dry seed to shoot emergence through soil surface
1 Leaf development	10-19	From visible pair of scale leaves to nine or more leaves unfolded
2 Formation of side shoots	20-29	From no side shoots to nine or more side shoots detectable
3 Stem elongation	30-39	Beginning of stem elongation to nine or more visibly extended internodes
5 Inflorescence emergence	50-59	Flower buds present, still enclosed by leaves to first petals visible, many individual flower buds, still enclosed
6 Flowering	60-69	First flowers open to end of flowering
7 Development of fruit	70-79	First pods have reached final length to nearly all pods have reached final length
8 Ripening	80-89	Beginning of ripening: seed green, filling pod cavity to fully ripe: nearly all pods dark, seeds dry and hard
9 Senescence	93-99	Stems begin to darken , plant dead and dry to harvested product

Table 3: A summary of the GRDC scale for faba bean with growth stage and description (GRDC, 2017).

GRDC	Growth stage (GS)	Description
00 Germination and emergence	GS000-GS006	From dry seed to first leaf unfolded
10 Vegetative	GS101-GS10(x)-GS1(n)	First node (first leaf fully unfolded with one pair of leaflets X = node X leaf fully unfolded with more than one pair of leaflets n = any number of nodes on main stem with fully unfolded leaves, according to variety
20 Reproductive	GS201-GS210	From flower buds visible and still green to pods dry and black, seeds dry
30 Pod senescence	GS301-GS310	From 10 % of pods dry and black to all pods dry and black
40 Stem senescence	GS401-GS410	From 10 % of stem brown/black or most stem green to all stem brown/black, all pods dry and black, seed hard

5.3 Greenhouse experiment

The experiment for observing the development and yield performance of faba bean varieties under two different temperature treatments was performed starting in February 2021 until beginning of June the same year (Table 4). The goal of this experiment was, to observe the development of faba bean. It was also used as a practice run with this type of legume, before starting with the field experiment. It was practiced to register the different development stages, and which variables were the most important to analyse at- and post-harvest.

The trial for the field experiment took place in the greenhouses at Kirkejordet at NMBU. For the different temperature treatments, a warm room was set up, with a day temperature of 22° C and a night temperature of 16° C. The cold room had a daily temperature of 17° C and 11° C at night. Both rooms had the same photoperiod of 16 h daylight supplied by HPI lamps. Fertilization and watering were done simultaneously with a 50/50 mixture of Yara Kristalon Indigo and Yara Liva Calsinit every third day in winter and every second day closer to summer.

The experimental design in both rooms were a complete randomized design with five replicated pots. In each pot were five plants sown in pots with 3l of peat soil, which were 12 days after sowing (DAS) reduced to three plants per pot to reduce competition between plants (Fig. 1).



Figure 1: *Plant development in the greenhouse experiment. Reduction of plants from five to three plants 12 DAS.*

Table 4: *Treatment plan greenhouse experiment. Each variety was represented under each temperature treatment.*

Variety	Treatment	
	1	2
Sampo	22°C/16°C	17°C/11°C
Vertigo	22°C/16°C	17°C/11°C
Lielplatonas	22°C/16°C	17°C/11°C

Due to continuous stem elongation of all three varieties, the top of each plant had to be cut to reduce the risk of burnt tips caused by lamps. This was done around the beginning of pod development around 69 DAS (Fig. 2).



Figure 1: *Continuous stem elongation, causing burnt tips. Tip cutting around 69 DAS.*

Harvesting happened at two different dates, due to the differences in maturity between warm and cold room - 104 days after sowing in the warm room and 114 days after sowing in the cold room. At harvest most of the stems were still green, however most of the pods were black.

Each plant was harvested and registered separately to see if there were differences between plants from the same variety. The variables registered were: number of pods per node, number of seeds per node, seeds per pod, and the seeds fresh- (FW) and dry weight (DW). The nodes where pods were grown were placed into node groups, which meant that from the first to the tenth node was group A, from 11th to 16th was group B and from 17th and upwards was group C. This strategy helped to get a better overview in which parts of the plant, pods were developed and to later get a specification where assimilates are allocated inside the plant. Seeds were dried after harvest at 60° C for 48 h and then weighed, with the reasoning to calculate water content and thousand-seed-weight. Some variables were calculated based on others (Table 5).

Table 5: *Overview of the mathematical calculations used to analyse the data*

Name	Calculation	Experiment stage
Leaf area index (LAI)	sum of leaf area (cm ²)/ground area (cm ²)	First and second harvest
Specific leaf area (SLA)	Sum of leaf area (cm ²)/leaf weight (g)	First and second harvest
Leaf weight ratio (LWR/LMR)	LWR= Leaf weight/total plant mass	First and second harvest
Lear area ratio (LAR)	LAR= SLA x LWR	First and second harvest
Biomass	leaves dry weight (+ wilted leaves dry weight) + stem dry weight + pod dry weight + of seed dry weight	First to third harvest
Biomass per plant	Biomass/number of plants	First to third harvest
Proportion leaves, wilted leaves, stem, pods and seeds	organ weight x 100/biomass	First to third harvest
Proportion aborted seeds, deformed pods and open pods	number of aborted seeds/number of seeds number of open (or deformed pods)/ number of pods	Second and third harvest
Seeds per pod	pods per pot/ seeds per pot	Pre-experiment
Thousand-seed-weight (TSW/TCW)	TSW= seed weight after drying (g)* 1000/total number of seeds	Pre-experiment and first to third harvest
Water content	(seed fresh weight (g) – seed dry weight (g)*100/seed fresh weight	Pre-experiment and first to third harvest
Nitrogen content of plant	Sum of (N concentration x weight) of all organs	First and second harvest
Protein concentration	% N x 6,25 (conversion factor)	Second and third harvest

5.4 Field experiment

The field experiment was conducted at Vollebekk in Ås, in the growing season 2021. The varieties were sown with a sowing machine at the end of April 2021.

The single plot length was around 7 meter and width around 1.5 m including the space between plots. The distance between sowing rows had 12.5 cm. The distance between neighbouring plots was 35 cm and the sowing depth met 5 to 7 cm. The 48 plots with different faba bean varieties were organized into two replicate blocks and the varieties were randomized inside those blocks. The first and last plot of each row were the field margins and not considered in the registrations.

The growing season started with a dry period (Fig. 3) optimal for sowing. was also little0404 precipitation in August around the last harvest. Soil samples were taken around the field and analysed in February 2022. The soil was crushed and sieved through a 0.08 mm sieve afterwards they were dried between 30 and 40°C for 26 hours. The results showed an average pH of 6.3, a total N of 0.3 %, a potassium level of 201.67 mg/kg and a phosphorous level of 86 mg/kg.

Application of 20 ml/daa Basagran® herbicide, 10 ml/daa Karate® insecticide and the fungicide Signum® in the quantity of 100 g/daa were applied during the growing season. The irrigation was done once in mid-June with around 20 mm/daa. Fertilization was done with 30 kg/daa PK Yara fertilizer at three different time points (one in June and two in July).

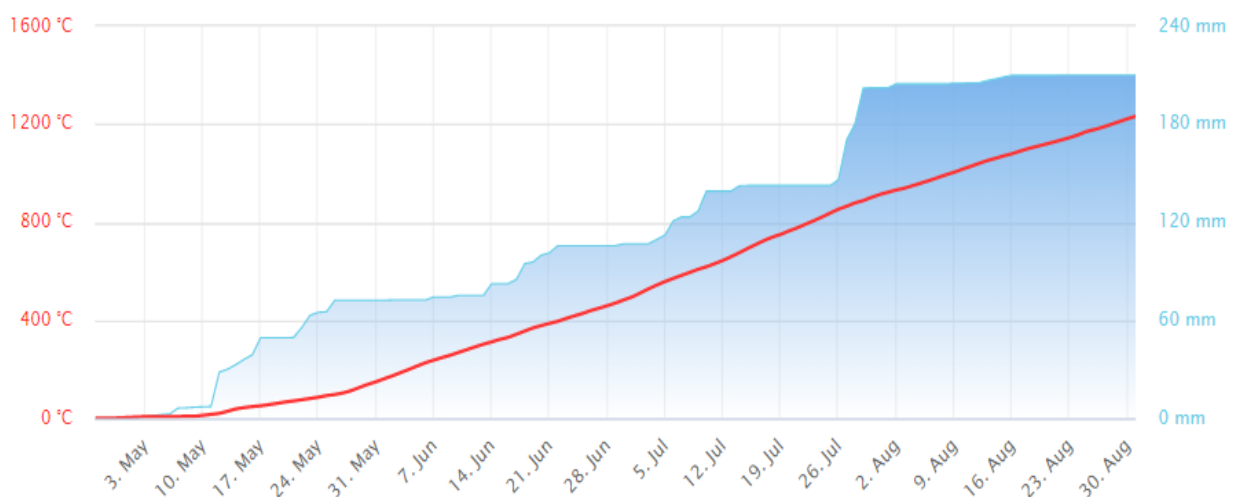


Figure 3: Degree days and precipitation-sum in the growing season 27th of April 2021 to 31th of August 2021. Base temperature 5 °C, number of growth days 123. Blue line is the sum of precipitation and the red line the degree days. (LMT-NIBIO, 2022)

5.4.1 First harvest

Prior to the first harvest, the chlorophyll content ($\mu\text{mol m}^{-2}$) was measured with a SPAD gadget (DUALEX Scientific™, Force-A) on twenty plants across the whole plot (Fig. 4).

The first destructive sampling was done on the 15th of June. A small subplot of 1125cm² close to the edges of the plots were harvested. Since the plants at the edges are often different due to different conditions than in the rest of the plot, those plants were not harvested and not considered in the analysis. At harvest time the varieties were around the principal growth stage of inflorescence emergence. After harvest, the developmental stage was registered on each plant individually using both scales, BBCH and GRDC (Table 2 and 3). Leaves of each plant were counted and height from surface to top was measured. Leaves including petioles were separated from the stem. Leaves that were mostly unfolded were selected for the leaf area meter. Organs were weighed after drying at 60° C for 48 h. The samples were ground to a grain size of around 0.5 mm for the analysis of nitrogen content. The laboratory analysis for the samples from harvest one and two were done by LabTek at NMBU (website: <https://www.nmbu.no/tjenester/laboratorietjenester/labtek/node/40370>) using the CNS-Dumas method. The destructive analysis gave a variation of data. To summarize the data and get more information, calculations were done, resulting in the different variables (Table 5).



Figure 4: *Faba bean field at Vollebekk Ås. 54 DAS. Colour different between plots visible*

5.4.2 Second harvest

Early and late varieties were harvested at a one day difference, due to the extensive workload. The early varieties were harvested a day earlier than the late varieties. At the time of harvest, the varieties were at the phenological stage of pod development (Table 2 and 3).

For this stage, a bigger subplot of 1500 cm² close to the edges of the plots was harvested, with the aim to increase the amount of data. The same registrations and calculations as the first harvest were done (Table 5). In addition pods, seeds, as well as deformed pods and aborted seeds were counted. Aborted seeds were defined as seeds that were very much smaller in comparison to the “healthy” seeds in the same pods and or had turned black. At that point in the development insects and fungi infection, the pea weevil, and the chocolate spot fungi had spread extensively across plots. Therefore a scale was prepared to identify the extent of the infection on leaves, pods and seeds. With zero showing no infection and three being highly infected. Leaves, stems pods and seeds were partitioned into different samples (Fig. 5). Hereby leaves were separated into wilted and not-wilted leaves, since the not-wilted leaves were measured further for the leaf area. After drying at 60° C for 48 h, samples were weighed and ground for the destructive analysis of nitrogen content (CNS-Dumas method), as at the first harvest.



Figure 5: Harvested organs at the second harvest (86 DAS). From left to right: stem, dead leaves, leaves, pods and seeds from the variety Daisy.

5.4.3 Third harvest

At the last harvest early varieties were harvested more than a week before the late varieties (end of August-beginning of September). The early varieties were in the phenological stage of senescence (Table 2 and 3) (Fig. 6), however the infection with the chocolate spot had progressed further, which made it difficult to differentiate between senescent and infected plants. The subplot size chosen for the third harvest was 3000 cm² in each plot. Since now more plants per area were harvested, the number of pods per node were only registered on ten plants that were selected randomly. Number of pods and seeds per plant were registered on all

harvested plants. Most of the leaves had either wilted or been shed from the plant, which is why it was not possible anymore to measure the leaf area. However wilted leaves that were still attached to the stem were considered for weighing. All the other registration were done in the same way as the previous harvests (Table 5), except for the laboratory analysis of N content in the plant organs. For the CNS Dumas analysis, only the N content of the seeds were analysed. The reason was, that at this point the plants across plots and varieties were overly infected by the chocolate spot fungi (*Botrytis fabae*). This infection had severe negative impact on the plants, by for instance enhancing development, increasing proportion of aborted seeds and open pods and potentially have an impact on the distribution of nitrogen into the different organs.



Figure 6: *Plant senescence, shedding leaves 112 DAS in the variety Sampo.*

5.4.4 ^{15}N isotope analysis

A legume that is efficiently nodulated in a soil, free of mineral or organic N, the legume will be dependent on the symbiotic N_2 fixation. In this case the isotopic constitution of the plant would be similar to the atmospheric N_2 . In contrast a non-leguminous plant would have a similar composition of the mineral N in the soil (Unkovich et al., 2008). Natural nitrogen consists of the two stable isotopes ^{14}N and ^{15}N (Peoples et al., 1989). The ^{15}N isotope is lower in the atmosphere than in the soil and for the ^{14}N the opposite (Peoples et al., 1989; Sørheim, 2021; Unkovich et al., 2008). Therefore leguminous plants, that receive all their nitrogen through the fixation of atmospheric N will have a lower content of ^{15}N than the plants taking up N solely through the soil (Peoples et al., 1989; Sørheim, 2021).

To measure the ability of biological nitrogen fixation of the varieties a ^{15}N isotope analysis was prepared at the time of the second harvest. In this experiment above-soil biomass of red dead-nettle (*Lamium purpureum*) was harvested as a reference plant in between rows of each plot of the varieties. The reference plant and the different plant organs - leaves (wilted and not wilted), seeds, stem, pods of the faba bean were dried at 60°C for 48 h and afterwards ground to a sample fineness around 0,5 mm and mixed thoroughly during and after. The results of the N content of the different plant organs from the CNS analysis was used to calculate the sample weight, which is required by the UC Davis Stable Isotope Facility. Due to the nitrogen content varying between the faba bean plant organs the sample weight varied from 2-14 mg . The sample weight of the reference plant was around 3 mg. Samples were sent to the Stable Isotope Facility (SIF) located on the University of California Davis, USA (<https://stableisotopefacility.ucdavis.edu/>, 2021). The University provided us with the $\delta^{15}\text{N}_{\text{AIR}}$ (‰) values. With this values it was possible to calculate the amount of fixed N deriving from the air (%Ndfa) (Eqn. 1).

Equation 1: Calculation of the amount of nitrogen fixed from the air (%Ndfa). $\delta^{15}\text{N}$ is representing the ^{15}N (‰) content of the samples. B is a mean value $\delta^{15}\text{N}$ of shoots of legumes, which is dependent on the N_2 fixation. The B value is often similar on the same legume. For this equation a B value of -0.5 was chosen, which has been recommended by Unkovich (Sørheim, 2021; Unkovich et al., 2008).

$$\%Ndfa = \frac{\delta^{15}\text{N reference plant} - \delta^{15}\text{N faba bean sample}}{\delta^{15}\text{N reference plant} - B} \times 100$$

5.5 Statistical analysis

For the greenhouse and field experiment variance analysis was performed using RStudio (R Core Team, 2020, version: i386-4.0.2).

5.5.1 Greenhouse experiment

Before the analysis, the data collected from single plants were averaged to pot level. Normal distribution of all variables were tested preliminary. Afterwards ANOVA was carried out (Eqn. 2), by testing effect of temperature on different harvest variables. The same test was used to observe the effect of variety. With significant effect of variety it was tested between which varieties there was significant difference. This was possible by comparing means, using Tukey Test. Tukey Test can show where the difference in effect of variable is situated. Lastly, the

interaction of variety and temperature was tested on the variables. When significance was shown, it meant that the varieties were reacting differently to changes in temperature. The significance level was set to 5 % ($p < 0.05$). The ANOVA was carried out with the following model:

Equation 2: *ANOVA model greenhouse experiment. Test effect of temperature and variety and the interaction between temperature and variety on each variable.*

$$\text{Variable} = \text{Temperature} + \text{Variety} + (\text{Temperature} \times \text{Variety}) + \text{Error}$$

5.5.2 Field experiment

Variables had to be calculated (Table 5) from the raw data and for each the normal distribution was tested. For all three harvests it was decided to carry out an ANOVA, hereby replicate and variety were tested to observe an effect on the variables (Eqn. 3). Replicate was used in the model to withdraw the effect of unwanted parameters like hillside. Significance on variety level was then further analysed by comparing the means of varieties through Tukey Test. A principal component analysis (PCA) was done to show the correlation between the original variables and the two principal components. Furthermore, a correlation matrix was designed to observe relationships between variables. This was done by using the Pearson correlation coefficient. Hereby the relationship between two variables can have a value between -1 and 1. The values of -1 and 1 showing negative and positive correlation, respectively, and 0 showing no correlation between the pair of variables. Which means the further a value is from zero the stronger is the relationship or correlation.

Equation 3: *ANOVA model field experiment (used in all three harvests). Test effect of replicate and variety.*

$$\text{Variable} = \text{Replicate} + \text{Variety} + \text{Error}$$

6. Results

As a first task, the normal distribution was tested. The normal distribution using a Q-Q plot was assessed on each variable for both pre-experiment and field experiment (Fig. 7). The normal distribution is determining the model selection. All variables were normal distributed with only a few outliers. The outliers were kept in the analysis and not removed.

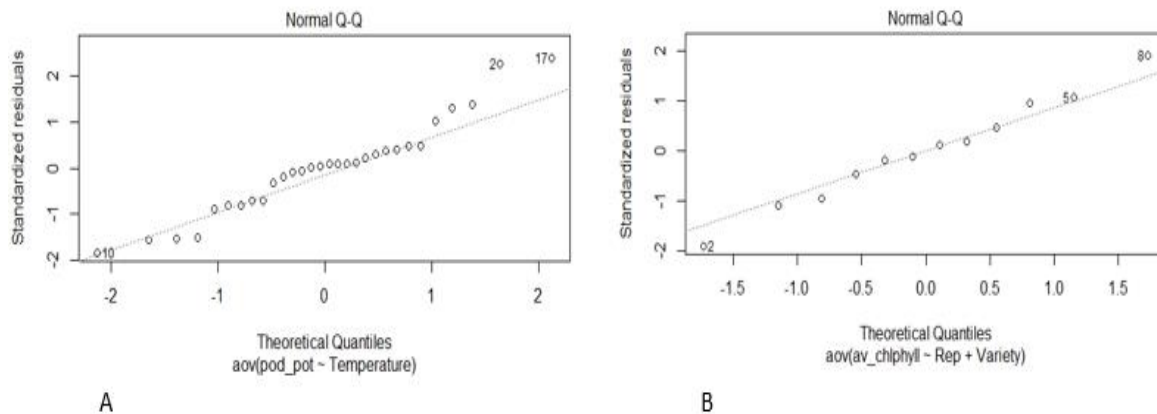


Figure 7: *Q-Q plots (quantile-quantile plot) of two example variables from the greenhouse experiment (A) and the field experiment (B).*

6.1 Greenhouse experiment

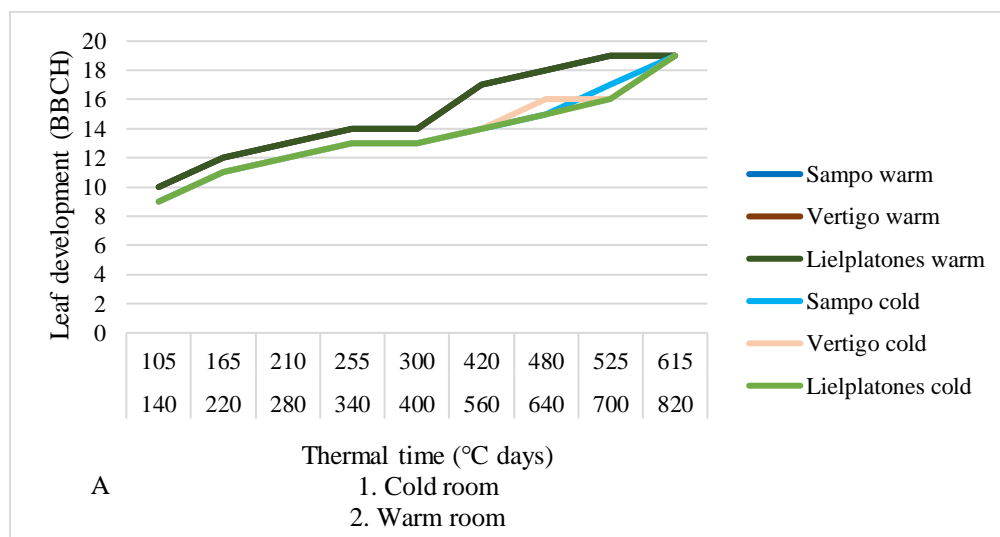
6.1.1 Plant development

The development stages were expressed as a function of thermal time (Fig. 8). Each development stage requires a specific amount of accumulated temperature, to be reached by a plant. The development stages were recorded with help of the BBCH scale (Table 2). The difference in thermal time requirement between cold and warm room at the different main stages were tested on significance. The ANOVA showed no significant effect of thermal time on the developmental stages, $P > 0.5$ level (n.s.).

The varieties under the warm treatment had a higher number of leaves. However, to have a higher number of leaves in the beginning they required more thermal time than when they were under the colder temperature treatment (Fig. 8A). Under the cold treatment the varieties started of with less leaves, yet they reached the last leaf stage as well and as said to reach that stage they required less thermal time. All three varieties in the warm room had the same development. When the varieties were in the colder room some small differences were seen. Vertigo reached the leaf stage 16 at 480°C days, while Sampo and Lielplatones reached it at around 525°C days

with small differences in °C days between each other. Sampo required a little less thermal time to reach leaf stage 18, however the differences are only minor. The recording of stem elongation also showed that the varieties under colder temperature required less thermal time (Fig. 8B). They reached last stem elongation stage (39) at 735°C days whereas under warmer temperature they reached stage 39 at 820°C days. In the warm room there was no difference between varieties in their thermal time requirements for the stem elongation. In the cold room Lielplatones had a minor slower growth than the other two varieties. To reach stage 35 it required minimal more thermal time. From the start of flower emergence more variation between the varieties in the warm was seen (Fig. 8C). At 700°C days it was seen that Sampo had already the first flowers open (BBCH 60) while Vertigo was still at inflorescence emergence (BBCH 50). Vertigo required more thermal time to reach flowering as well as pod maturity in the warm room.

By looking at the thermal time requirement, it can be observed that several of the development stages occur simultaneously (Fig. 8). In the cold room at the leaf stage 11 at 165°C days is also the thermal time requirement of the stem elongation and starting point of the stem elongation 31. Flower emergence requires 480°C days to start, at that point the varieties had first reached stem elongation 34 and leaf stage 16. The simultaneous occurrence of development stages was also seen in the warm room. At 220°C days the varieties were at leaf stage 12 and stem elongation stage 31. Flower emergence required 640°C days at that time the plants were at leaf stage 18 and stem elongation 37.



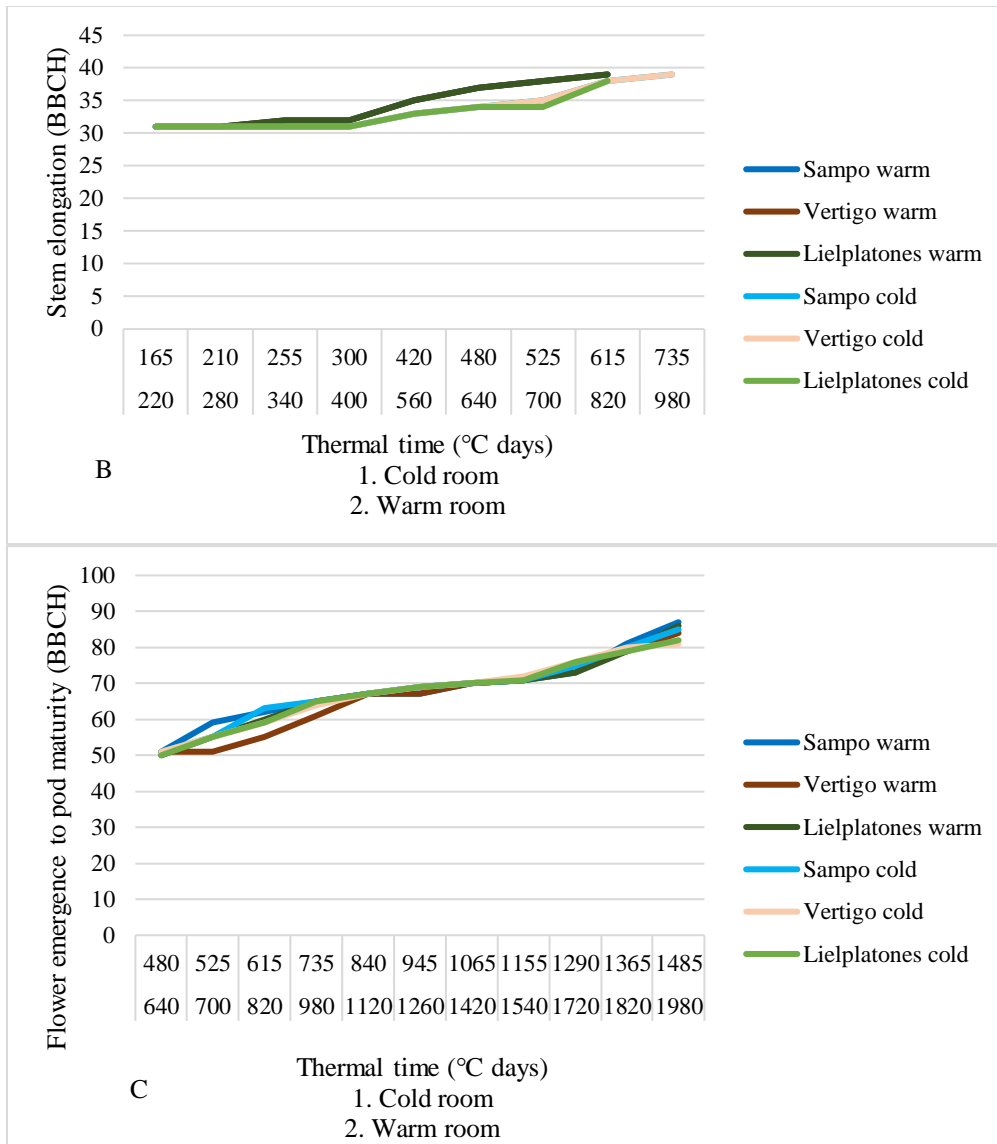


Figure 8: Thermal time requirements for the different development stages (BBCH scale in Table 2): A: the leaf development; B: the stem elongation; C: the flower emergence to pod maturity. The first line on the x-axis presents the degree days (°C days) accumulated in the cold room and the second line of the x-axis presents the °C days accumulated in the warm room.

6.1.2. Harvest parameters

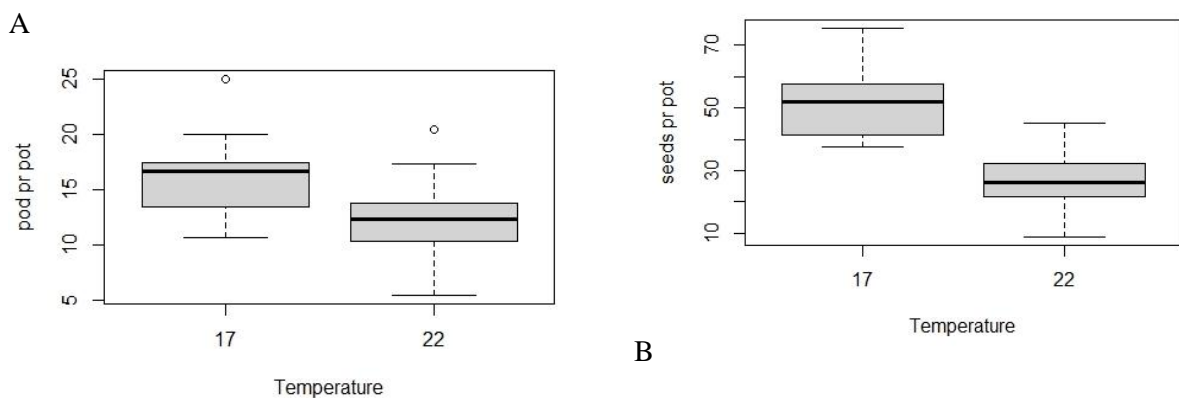
After harvest, the variables were analysed under the effect of temperature, variety and the interaction between those two factors (Table 6), as described in material and methods (5.).

Table 6: Overview of the *F* values and the range of the significant effect. Only variables are shown, where temperature, variety and/or the interaction of both factors had a significant effect. Significance: *, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$, others are marked with *n.s.* (no significance)

	Pods per pot	Seeds per pot	Seeds per pod	FW- seeds per pot	DW- seeds per pot	TSW per pot	Water content
Temperature	10.700 **	49.769 ***	71.561 ***	59.899 ***	29.451 ***	20.246 ***	52.469 ***
Variety	5.321 *	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	67.794 ***	13.316 ***
Interaction	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>	4.964 *	<i>n.s.</i>	<i>n.s.</i>	<i>n.s.</i>

Temperature had a significant effect on all variables (Table 5). The effect of variety on the other hand was rarer and could only be proven on the number of pods, TSW and water content. A significant interaction between temperature and variety was only seen in the FW-seed per pot.

It was observed, that regarding the number of pods, number of seeds per pot, seeds per pod per pot, seed FW, seed DW and water content the varieties performed had higher values under the colder temperature (17°C/11°C), only the TSW was higher in the warm room (Fig. 9).



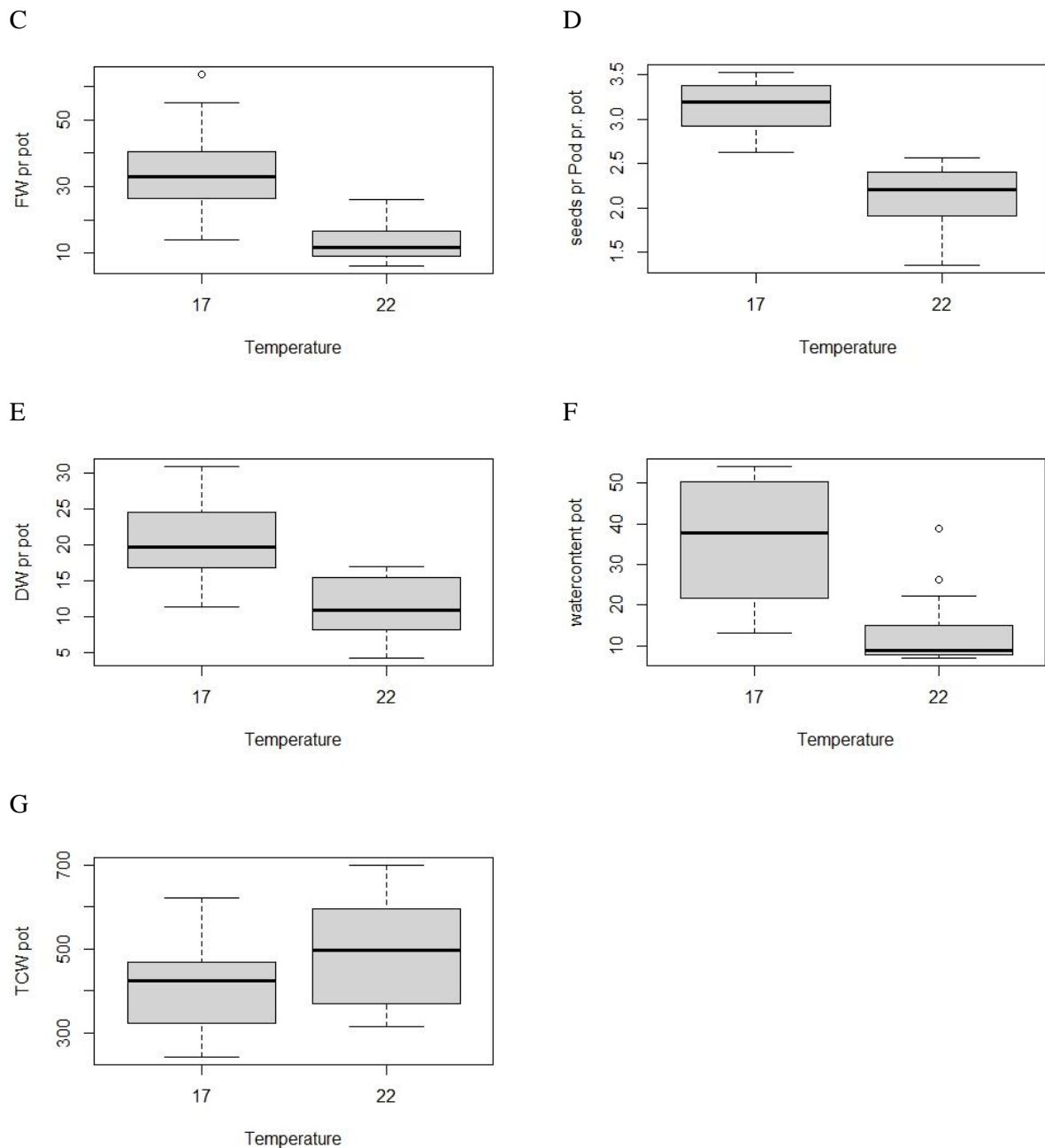


Figure 9: Boxplots of each variable showing the effect of temperature differences. Without regarding effect of variety.

When an effect of variety was tested, a significant effect was only proven on the three variables water content ($P < 0.05$), TSW ($P < 0.001$), and number of pods per pot ($P < 0.05$) (Fig. 10). The seeds of Vertigo had a high water content (~40 %), Lielplatones had a lower water content (~25 %), nonetheless still very high compared to Sampo (~17 %) (Fig. 10A). The significant difference was seen between Vertigo and Sampo ($p < 0.05$). There was no significant difference between Lielplatones and Sampo. The TSW (TCW) showed significant differences between all

three varieties (Fig. 10B). There was a significant difference ($p < 0.001$) between Lielplatones and Sampo and also between Sampo and Vertigo. Again both Vertigo and Lielplatones had a high TSW, the significance is less ($p < 0.01$), however Vertigo has the highest TSW compared to both other varieties. Sampo had a higher number of pods than the other two varieties (Fig. 10C). The difference in number of pods between Lielplatones and Sampo is not significant, the same with Lielplatones and Vertigo. There was only a significant difference between Sampo and Vertigo ($p < 0.05$).

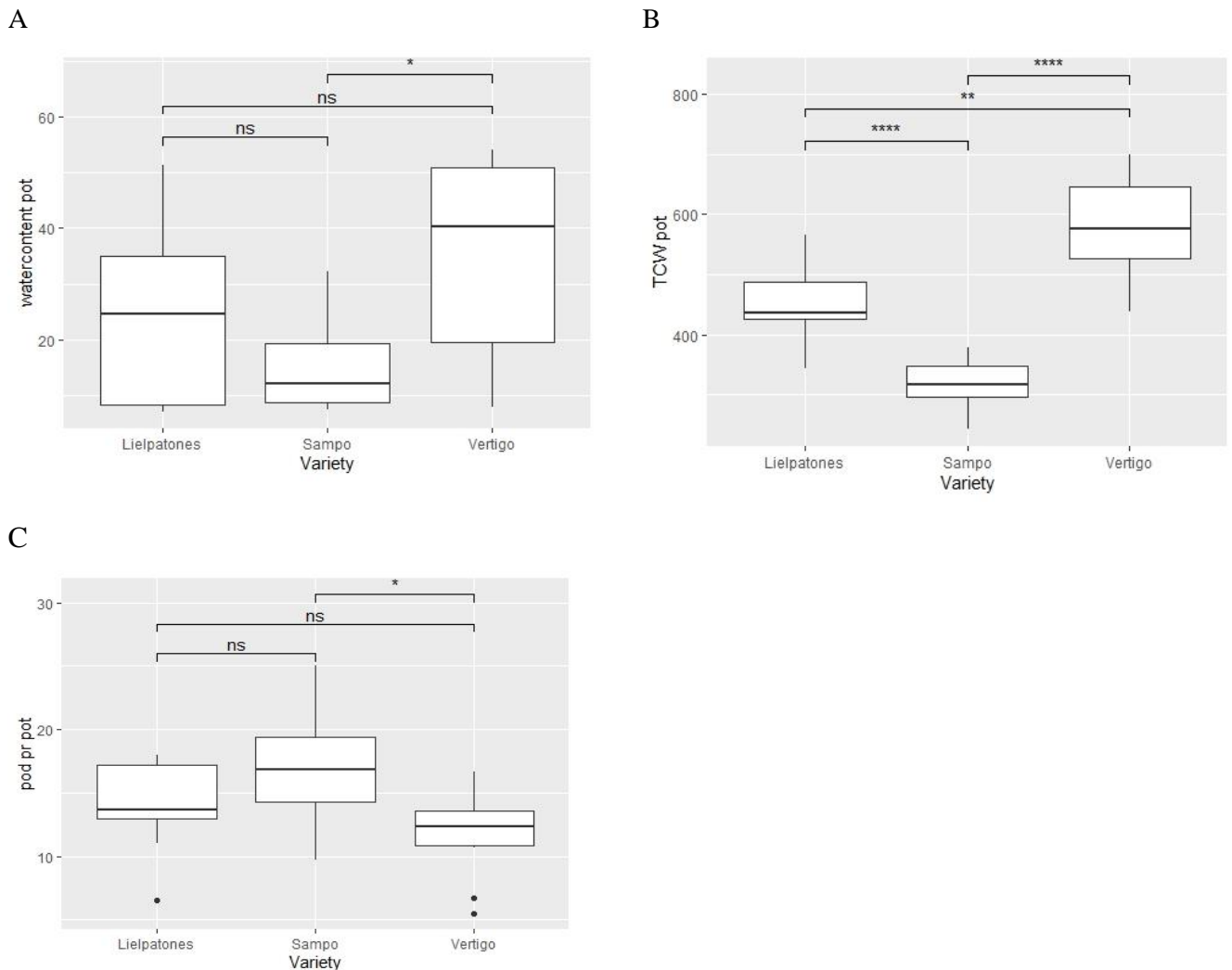


Figure 10: Boxplots display significant effect of Variety on the three variables water content (A), TSW (B) and pod per pot (C).

6.2 Field experiment

6.2.1 Plant development

The emergence rate of each plot was recorded 20, 22 and then again 26 DAS (Table 7). The early varieties Louhi, Sampo and Vire were earlier in emergence. However, on the last day of

registration most varieties had between 90 to 95 % emerged seedlings, only Vertigo (70 %) and Birgit (80 %) were lower.

Table 7: Emergence rate of all 12 observed plots on three different time points. Emergence rate is shown in percent (%).

Plot	Variety	18. May	20. May	25. May
1101	Louhi	60	90	90
1103	Daisy	50	70	90
1105	Sampo	50	90	95
1107	Vire	60	90	90
1202	Birgit	30	50	95
1203	Vertigo	10	30	70
1407	Birgit	50	80	80
1506	Daisy	60	80	95
1601	Vertigo	30	80	95
1603	Vire	80	95	95
1605	Louhi	80	95	95
1608	Sampo	70	95	99

In the field experiment two development scales were tested, to analyse which scale could be most efficient and detailed to use. Detailed meaning how well the scales capture the main development stages to specific days or thermal time.

With the one-on-one comparison of the scales (Fig. 11), broad differences between the scales were observed. While the GRDC only shows two lines, one for early varieties and one for late varieties, shows the BBCH scale more variation between not only early and late varieties but also variation in the development of each variety. In the beginning Vertigo reaches the development stages of leaf development at the same speed as the early varieties, but is then slowed down and has a similar agility as the other two late varieties starting from the end of flowering. The early varieties Louhi, Sampo and Vire have an end of development earlier not only because they were harvested earlier than the late varieties, but also because at the last two recording days, the varieties had already senesced and been diseased. In this way recording the right development stage was not possible anymore. The GRDC scale shows more straight lines, according to that scale the varieties have a more continuous agility of development. In addition,

both early and late varieties are in the same development stage for several days without any changes, yet at around 104 DAS for the early varieties and around 118 days for the late varieties there is an increase in development. BBCH on the other hand has a strong increase after 42 days for early and after 49 DAS for late varieties from leaf development to flowering. The BBCH scale also shows more changes and variation in development.

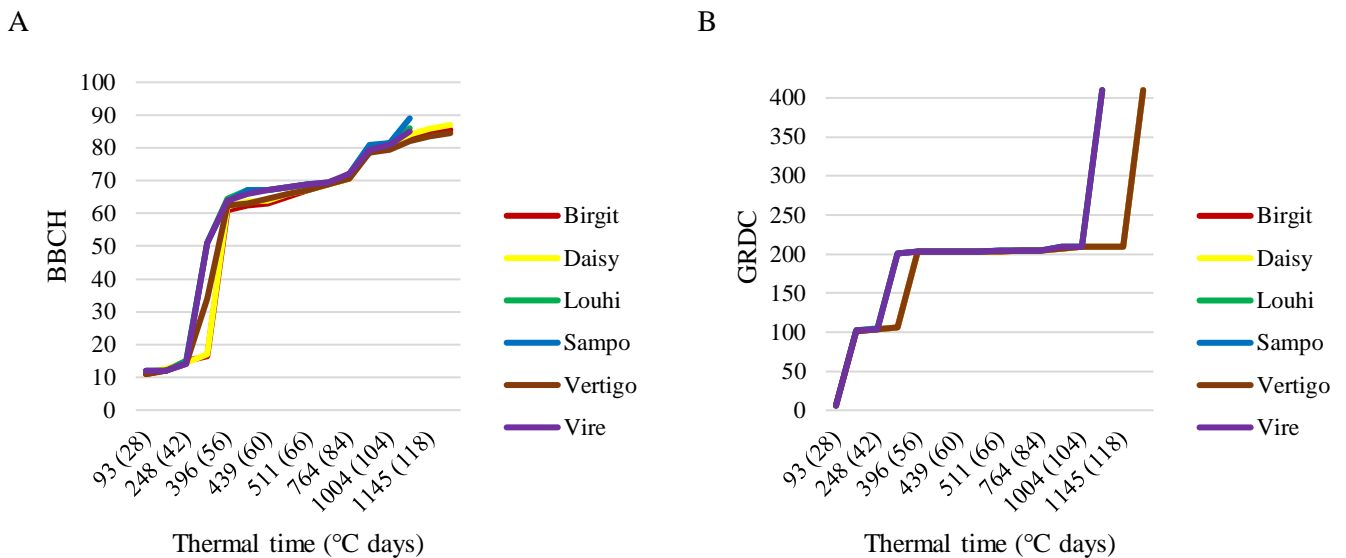


Figure 11: Comparison between BBCH (A) and GRDC (B) development scale under thermal time and the days after sowing in brackets (DAS). Average values of the two replicates

The observations described between the development stages regarding the days after sowing corresponds to the observations made about the thermal time (Fig. 11). The early varieties had the same thermal time requirements. Birgit and Daisy from the late varieties had the same thermal time requirements to reach a new development stage. Vertigo on the other hand had the same thermal time needs as the early varieties in the beginning of the main stage of leaf development. However as development continued, the variety had its own thermal time requirements until flowering, where the thermal time requirements were equal to the other two late varieties (Fig. 11A). The GRDC scale did not show this differences between single varieties, it only showed the broad differences between early and late varieties in thermal time requirements (Fig. 11B).

6.2.2. First harvest

Most of the variables that were tested at the first harvest did not show any significant effect of variety, which were GRDC stage, height, biomass, SLA, LWR, LAR and LAI. Only three of

the variables showed effect of variety; BBCH stage, biomass per plant and average chlorophyll content (Table 8).

Table 8: Overview of the *F* values and the range of the significant effect for the first harvest. Only variables are shown, where replicate and/or, variety had a significant effect. Significance: *, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$, others are marked with n.s.(no significance)

	BBCH	Biomass per plant	Average chlorophyll
Replicate	15.0 *	n.s.	n.s.
Variety	136.7 ***	5.880 *	68.539 ***

There was a significant difference in development (Table 8), using the BBCH scale ($p < 0.001$), but there was no significant difference using the Australian development scale GRDC. In addition, a significant difference between replicates ($p < 0.05$) could be noted. The production of biomass per plant is significantly different between varieties ($p < 0.05$), however the overall biomass production from the harvested plot was not significantly diverse. In the beginning of the experiment when the canopy fully covered the ground, colour differences between plots could be observed. Some plots had a darker green colour. Therefore the chlorophyll content was measured on various plants in the total plot and it was proven that the colouring was significantly contrasting between varieties. The varieties Birgit, Daisy and Vertigo had a higher chlorophyll content and were seen as darker green coloured plots in the field (Fig. 12B). The early varieties on the other hand, had a lower chlorophyll content, and were seen with a lighter green colour.

At the time of the first harvest, Louhi, Sampo and Vire were already at the end of inflorescence emergence (stage 59 after the BBCH scale), whereas Birgit, Daisy and Vertigo were still at the beginning of inflorescence emergence (stage 51 after BBCH scale) (Fig. 12A). As described, the late varieties had a distinctly higher chlorophyll content than the early varieties ($\sim 12 \mu\text{mol m}^{-2}$), yet Vertigo had a lower chlorophyll content ($\sim 11 \mu\text{mol m}^{-2}$) than the other two late varieties (Fig. 12B). Louhi had the lowest chlorophyll content ($\sim 6 \mu\text{mol m}^{-2}$) in comparison to all varieties. In the variable biomass per plant (Fig. 12C), Birgit had the highest share of biomass per plant (2.6 g), followed by Vertigo (2.5 g). Sampo and Vire (~ 1.4 g) had the lowest portion of biomass per plant.

A

B

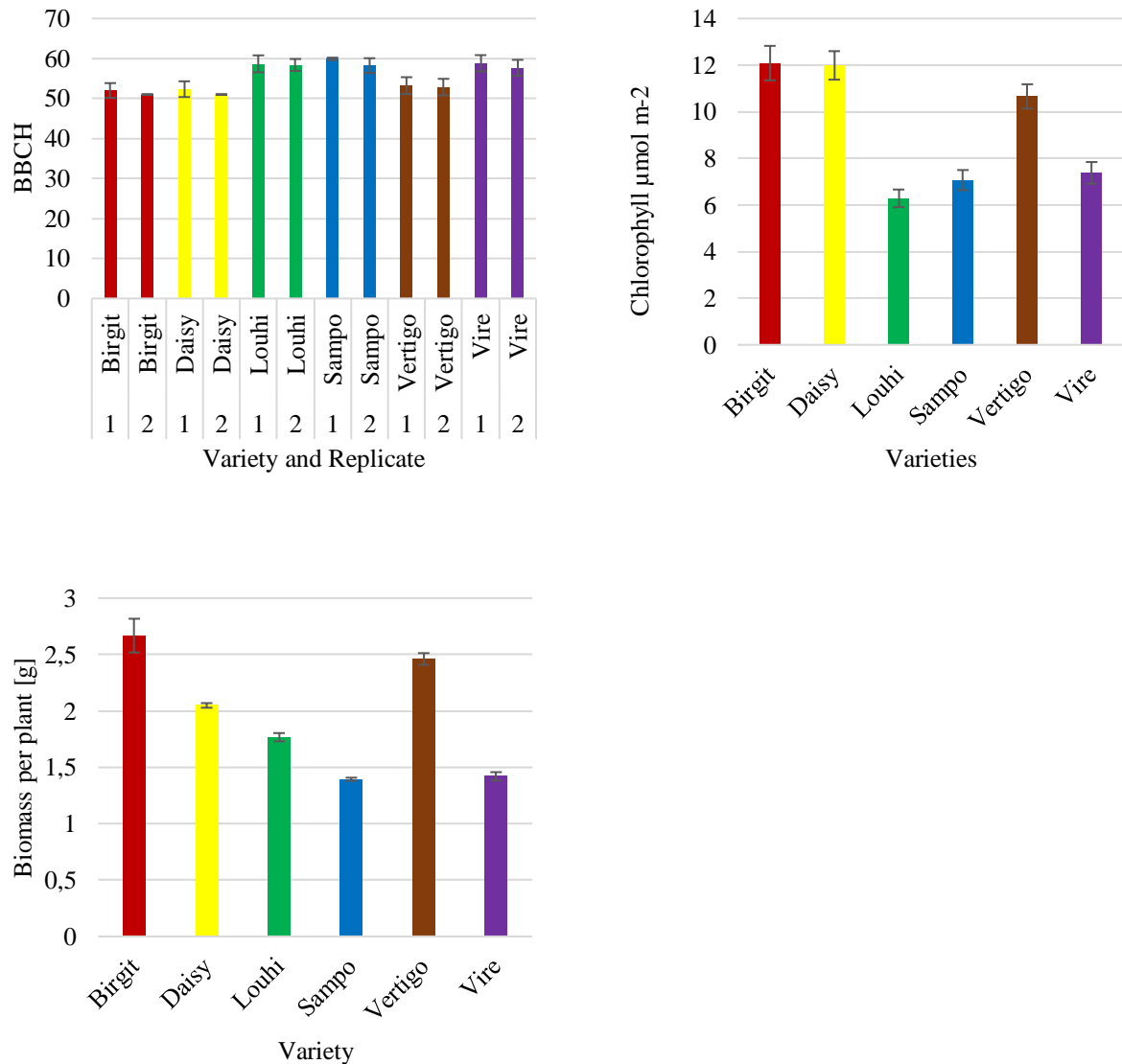


Figure 12: The variables that show significant effect of variety from the first harvest. Graph A: BBCH scale with variety and replicate as factors. Graph B: The chlorophyll content per variety. Graph C: Biomass per plant for each variety. All bars are presented with standard error (SE)

In the principal component analysis (PCA) of the first harvest 51.51 % of the variation is explained by PC1 (principal component one) and the other variation of 23.92 % is being explained by PC2 (principal component two) (Fig. 12). PC1 differs between early and late varieties. The variables biomass, biomass per plant, LAI, height and the average chlorophyll content cluster together and the late varieties are correlating with these variables. On the contrary, the early varieties are correlating mostly with the BBCH scale and a little less with the GRDC scale. One of the replicates of Sampo seems not to be affected by any of the variables, it is mostly shown as an outlier. LAR, SLA and LWR are not clustering with any

other variable and are additionally not correlating with any variety. LAI on the other hand is clustering with the variables in the negative PC1.

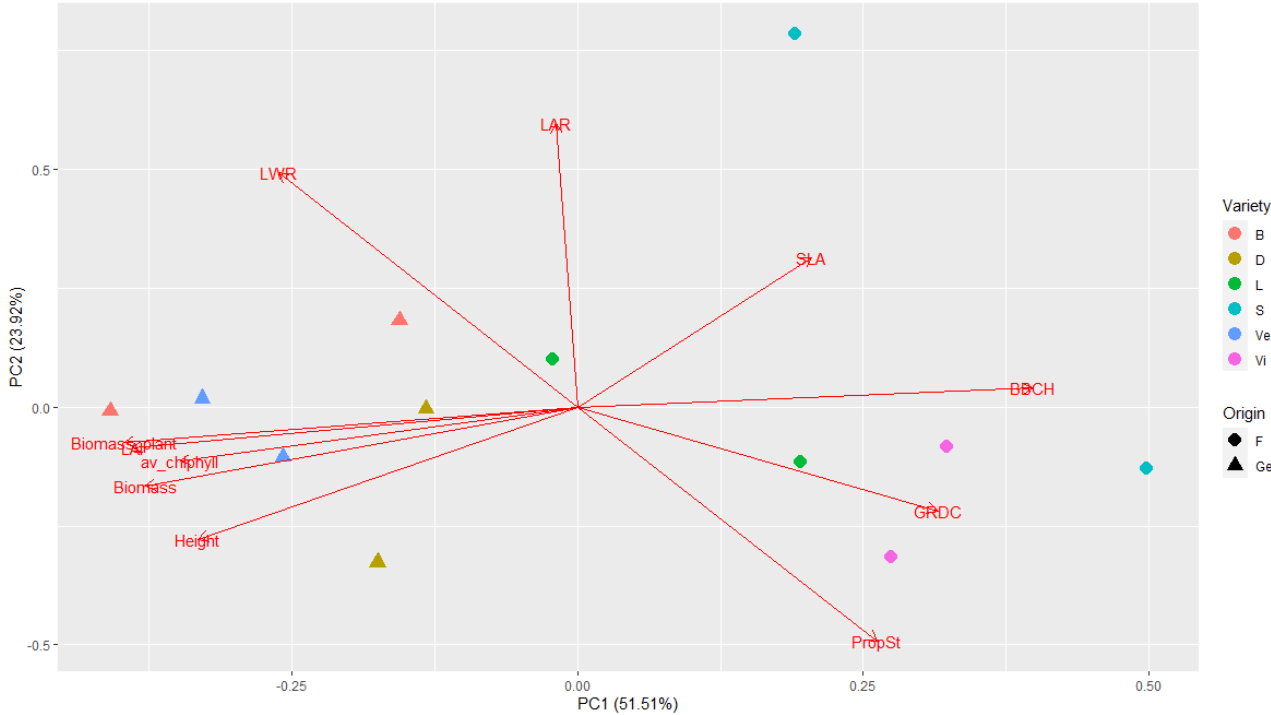


Figure 13: Loading plot of the principal component analysis (PCA) of the first harvest variables. Showing variety and origin as factors. With the variables LWR, LAR, SLA, LAI, BBCH, GRDC, proportion stem (PropSt), height, biomass, biomass per plant (Biomass_plant), average chlorophyll content (av_chlphyll). The early varieties Sampo (S), Louhi (L), Vire (Vi) originate from Finland (F), whereas the late varieties Birgit (B), Daisy (D) and Vertigo (Ve) originate Germany (Ge).

6.2.3 Second harvest

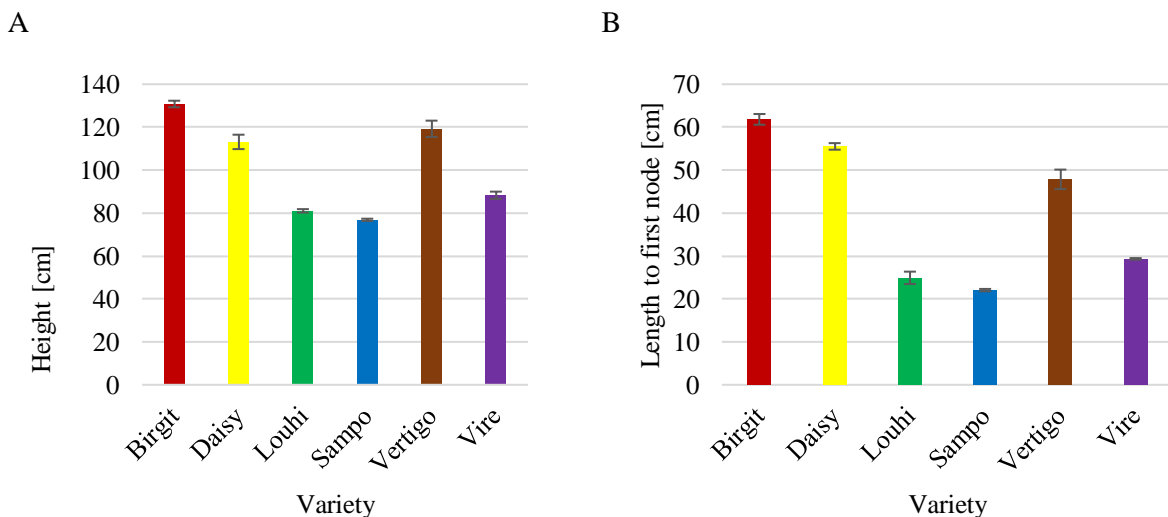
At the time of the second harvest the varieties had reached the fruit (pod) development stage (70-73 BBCH scale). At this stage there was a significant effect of variety on more variables (Table 9). There was no significant effect of replicate. The height differences that were also seen in the field were proven to be significantly different between varieties. Not only the height of the whole plant was significant but also the length from the surface to the first node was different between varieties. Even though some varieties were still at the beginning of pod development, significant differences could already be seen regarding the number of seeds per pod, TSW and the proportions of seeds, pods and deformed pods. At this time of harvest, there were no differences between varieties regarding their development and this time neither BBCH or GRDC could detect any effect of variety. Even though there were significant differences between varieties in TSW, there were however no differences in seed-, FW, DW, or water content. Similarly to the first harvest, LAI, SLA, LWR and LAR are not significantly different between varieties. Variety has no significant effect on the biomass and biomass per plant, although variety had a significant effect on biomass per plant in the first harvest. Important to note is that the number of pods and the number of seeds, which are important for yield formation, do neither have a significant effect as well.

Table 9: Overview of the *F* values and the range of the significant effect for the second harvest. Only variables are shown, where replicate and or, variety had a significant effect. . Significance: *, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$, other are marked with *n.s.* (no significance).

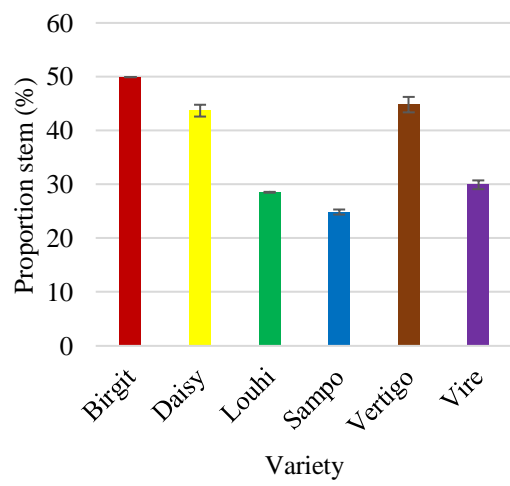
	Height	Length	Proportion	TSW	Seeds	Proportion	Proportion	Proportion
		until	stem		per	deformed	seeds	pods
		first			pod	pods		
		node						
Replicate	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
Variety	6.706*	15.682**	11.306**	5.875*	5.810*	26.229**	10.439*	6.308*

The late varieties were between 20 and 40 cm higher than the early varieties (Fig. 14A). Sampo had the smallest plants (79 cm) and also the shortest distance from the surface to the first node (21 cm) (Fig. 14B). Birgit had in average the highest plants (130 cm) and the longest distance from the surface to the first node (60 cm) in comparison to the other varieties. The biomass proportioned to stem corresponds to the height differences. Birgit, Daisy and Vertigo had comparably more biomass going into the stem (between 45-50 %) than the early varieties

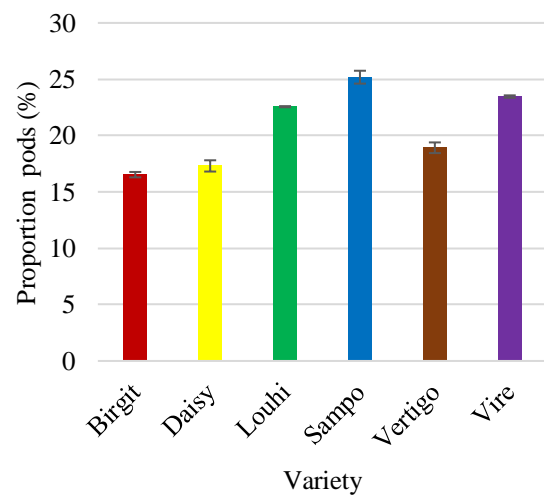
(between 25-30 %) (Fig. 14C). The two variables proportion of seeds and proportion of pods show again a clearer separation of early and late varieties (Fig. 14D and E). Louhi, Sampo and Vire have higher proportions of biomass allocated to seeds (over 25 %) and pods (between 22-25 %) compared to the late varieties (between 10-19 % in the seeds and between 16 % and 19 % in the pods). Even though Birgit, Daisy and Vertigo are large seeded faba bean varieties they have a low proportion of biomass allocated to seeds and pods. The proportion of deformed pods (Fig. 14F) shows a larger span between varieties. The variation seems to be more random. Louhi has the lowest number of deformed pods per plant (0.1 deformed pods per plant), whereas Sampo (0.3 deformed pods per plant) and Vertigo (~0.38 deformed pods per plant) have a large proportion of deformed pods. Birgit and Daisy show a high proportion of deformed pods as well (between 0.25-0.3 deformed pods per plant). Regarding the number of seeds per pod (Fig. 14G), the varieties do not differ widely from each other, compared to the other variables. Moreover there is no differentiation between early and late varieties. Birgit, Daisy, Louhi and Sampo have in average between 2.7 to 2.9 seeds per pod. Vertigo as a late variety (2.4 seeds per pod) and Vire (2.5 seeds per pod) as an early variety have less seeds per pod than the other varieties. The calculation of thousand seed weight (Fig. 14H) detected that Vertigo had a high TSW (249 g/1000 seeds), yet Birgit and Daisy have a low TSW of the late varieties. The two early varieties Louhi (150 g/1000 seeds) and Vire (152 g/1000 seeds) have a higher TSW than Birgit (60 g/1000 seeds) and Daisy (140 g/1000 seeds).



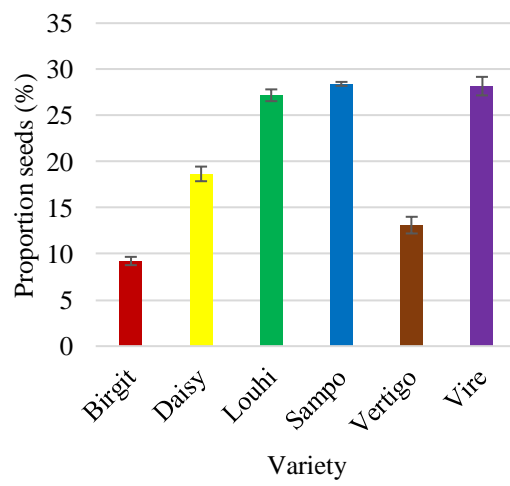
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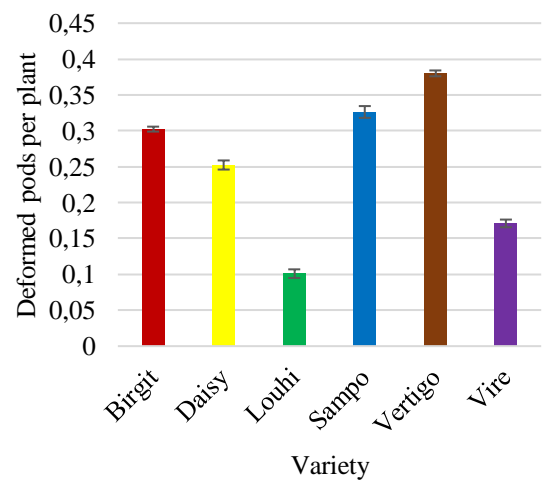
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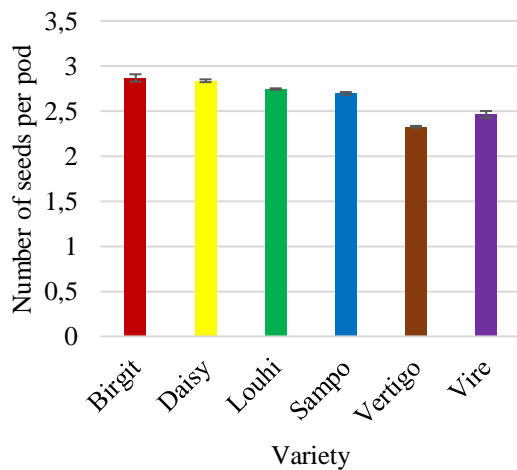
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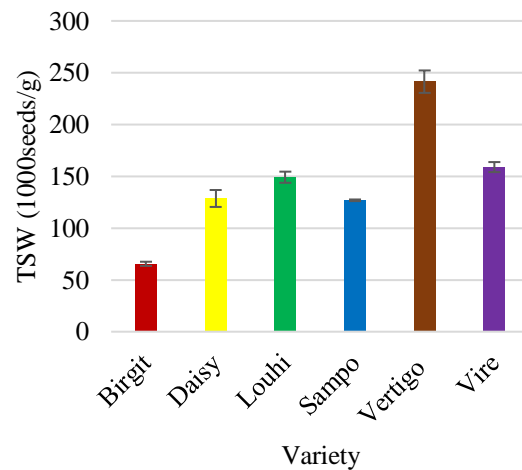


Figure 14: Variables with significant effect of variety from the second harvest averaged by subplot. Graph A: height per variety (cm). Graph B: Length to first node per variety (cm). Graph C: Proportion stem per variety (%). Graph D: Proportion pods (%). Graph E: Proportion seeds (%). Graph F: Number of deformed pods per variety. Graph G: Number of seeds per pod per variety. Graph H: TSW (1000seeds/g) per variety. All bars are presented with SE.

The principal component analysis (PCA) of the second harvest variables has a 48.55 % explanation from PC1 and 19.58 % is explained by PC2. The PCA shows a wider spreading of variables (Fig 15). The early varieties, which are all from Finland, are negatively correlated with the PC1 and the late varieties from Germany are mostly positively correlated with PC1, although one of the replicated of Daisy is negatively correlated to PC1. The variables with proportions of biomass to organs (seeds, pods and aborted seeds) are clustered together positively in the PC2. The proportion of biomass to stem is however clustering together with height and length from surface to first node and the proportion of deformed pods is closer to the LAI, however not completely clustered. The number of pods and the seeds FW are clustered together. Event though BBCH and GRDC are both recording development stage of faba bean, they are in the same direction but not clustering together. The number of seeds per pod are not clustered with any variable and in addition no variety is correlated with the variable, even though the statistical analysis showed a significant effect of variety regarding that variable. Vire is correlated with GRDC and the proportion of seeds, however Louhi seems not be correlated with any of the variables and Sampo is correlated with the seeds DW. The late varieties are not correlating with any of the variables, only Daisy correlated with the proportion of deformed pods.

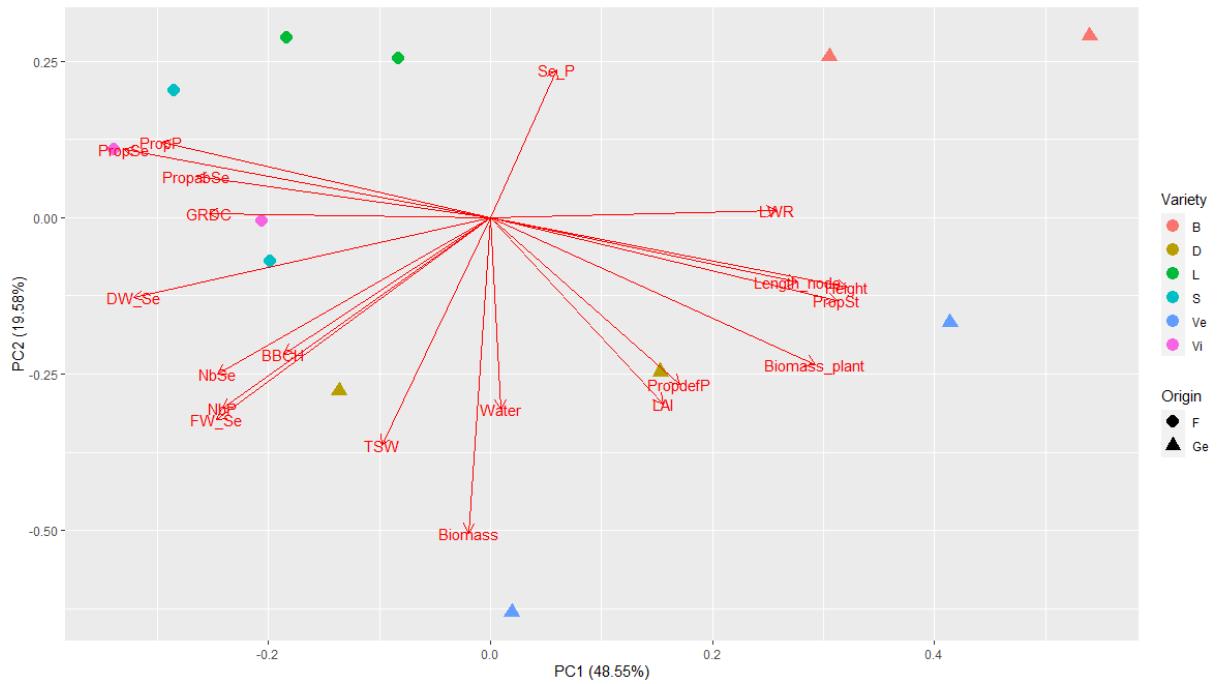


Figure 15: Loading plot of the principal component analysis (PCA) of the second harvest variables. Showing Variety and Origin as factors. With the variables: LAI, LWR, GRDC, BBCH water content (water), thousand seed weight (TSW), biomass, biomass per plant (biomass_plant), seeds fresh weight (FWSe), seeds dry weight (DWSe), proportion dead leaves (PropDL), proportion stem (PropSt), proportion pods (PropP), proportion seeds (PropSe), number of seeds (NbSe), proportion aborted seeds (PropabSe), number of pods (NbP), proportion deformed pods (PropdefP), proportion open pods (PropopenP). The early varieties Sampo (S), Louhi (L), Vire (Vi) originate from Finland (F), whereas the late varieties Birgit (B), Daisy (D) and Vertigo (Ve) originate Germany (Ge).

6.2.4 Third harvest

On the dates of the third and last harvest, most of the plants in the plot were already senesced. At this stage the most important variables concerning seed and biomass yield showed significant differences between varieties (Table 10). There was no significant effect in: seeds water content, biomass proportioned to pods and seeds, number of seeds and pods, as well as the number of aborted seeds and deformed pods, and number of seeds per pod.

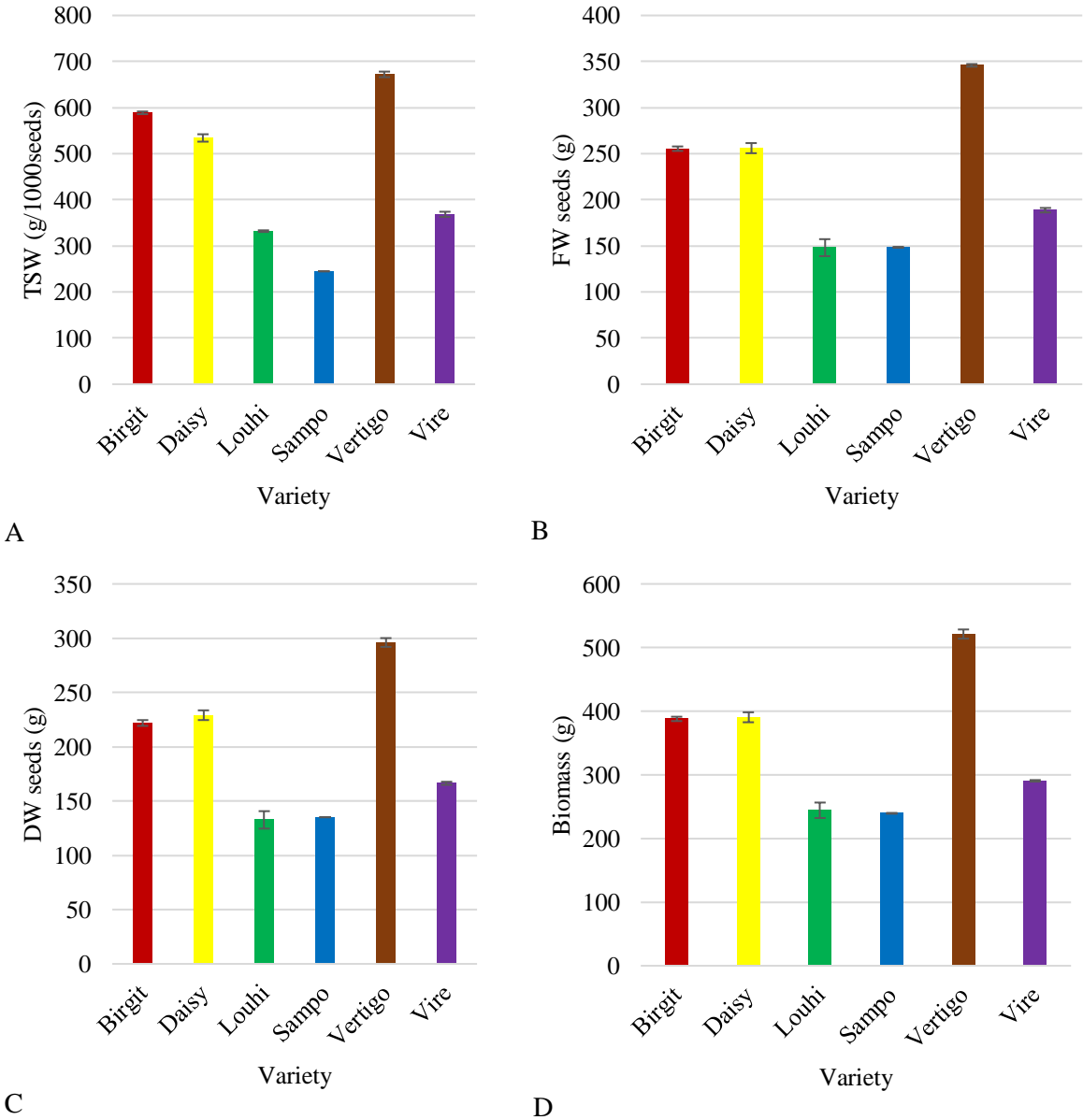
Table 10: Overview of the *F* values and the range of the significant effect for the third harvest. Only variables are shown, where replicate and/or variety had a significant effect. . Significance: *, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$, other are marked with *n.s.* (no significance)

	TSW	Biomass	Biomass per plant	FW seed	DW seed	Proportion dead leaves	Proportio n stem	Pro- portion open pods
Replicate	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
Variety	64.587***	11.72**	5.340*	18.5 54**	11.068 **	5.265*	8.035*	30.878***

What can be observed from all three harvests is that variety had an effect on different variables each time. Only some effects of variety are on the same variables (Table 10): For instance, variety showed effect on biomass per plant in harvest one and three however not in harvest two. At the third harvest the proportion of biomass in the stem and in the dead leaves was significantly different between varieties. Variety had no effect on the proportion of deformed pods as in the second harvest, however there was an effect on the number of open pods. Since it was the last harvest it is important to regard that there were effects of variety on FW and DW seed ($P < 0.01$), which are important variables when it comes to the overall yield potential. In addition to these two variable it was seen that the TSW was strongly affected by variety ($P < 0.001$).

The TSW had changed in proportions, since all varieties had an increased TSW (Fig. 16A). Especially Birgit had a higher TSW (~588 g/1000seeds) than in the second harvest (~65g/1000seeds). Vertigo had still the highest TSW with 672 g/1000seeds from all varieties and Sampo had still the lowest with 245 g/1000seeds. The seeds FW and DW showed similar results. The late varieties had the largest FW (~250-346 g) and DW (~221-296 g), especially Vertigo was highest weighing in seeds FW and DW (Fig 16B and C). The early varieties were on the other hand low in seed- FW (~150-188 g) and DW (~135-166 g). The biomass and thereby the biomass per plant had increased in weight, nevertheless the ranking of the varieties had not changed (Fig 16D and E). Meaning that the early varieties still had a lower total biomass (~244-290 g) and lower biomass per plant (10-13 g) than the late varieties. The late varieties had the total biomass per subplot between 387 to 500 g and biomass per plant around 14-23 g. Vertigo remained the highest in subplot biomass with 500 g and biomass per plant with 23 g. At the third harvest only dry leaves were left and most plants had already shed the largest parts

of their leaves on the field (Fig. 16F). All leaves were considered dead leaves, due to the fact that most leaves were already wilted. No difference between late and early varieties was detectable. Louhi had the highest proportion of dead leaves with around 5 %. Birgit, Vire and Daisy had a low amount of dead leaves which ranged between 2.5 and 3 %. The clear differences between early and late varieties concerning the biomass proportioned to the stem that were seen in the second harvest, had now mostly disappeared (Fig. 16G). The stem proportion had generally decreased from second (between 45-50 %) to third harvest (~22-28 %). For instance Birgit had an average stem proportion of 50 % in the second harvest. At the third harvest it had decreased to around 27 %.



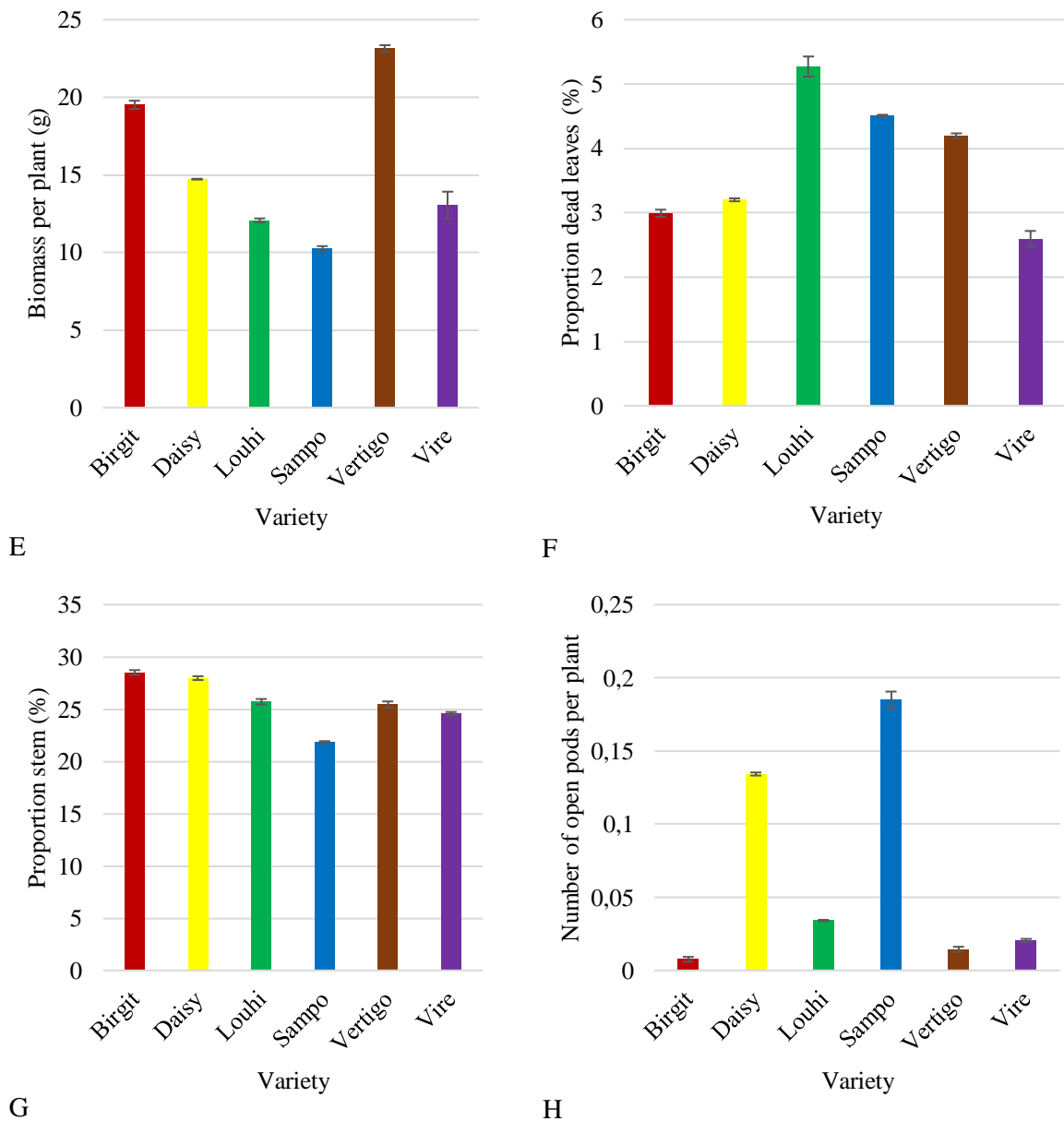


Figure 16: Variables with significant effect of variety from the third harvest averaged by subplot. Graph A: TSW per variety. Graph B: FW seeds (FW yield). Graph C: DW seeds (DW yield). Graph D: Biomass per subplot. Graph E: Biomass per plant. Graph F: Proportion of dead leaves. Graph G: Proportion of biomass to stem. Graph H: Proportion of open pods. All bars are presented with SE.

With the principal component analysis the influence of the variables are influencing the principal components, (Fig. 17) and variables correlating with each other (Table 11). The principal component one (PC1) explains 49.5 % of the variation and principal component two (PC2) the remaining 18.8 %. With the PCA similar variables cluster together. PC1 describes the correlation between varieties. The early varieties (Sampo, Louhi, Vire) cluster together and are positively correlated with PC1. The late varieties (Birgit, Daisy and Vertigo) cluster together

in the negative area of PC1. The early varieties are correlated with number of seeds, proportion of open pods, number of pods, proportion of dead leaves and proportion of deformed pods. The late varieties correlate with seeds DW, biomass per plant, biomass, seeds per pod, TSW, water content.

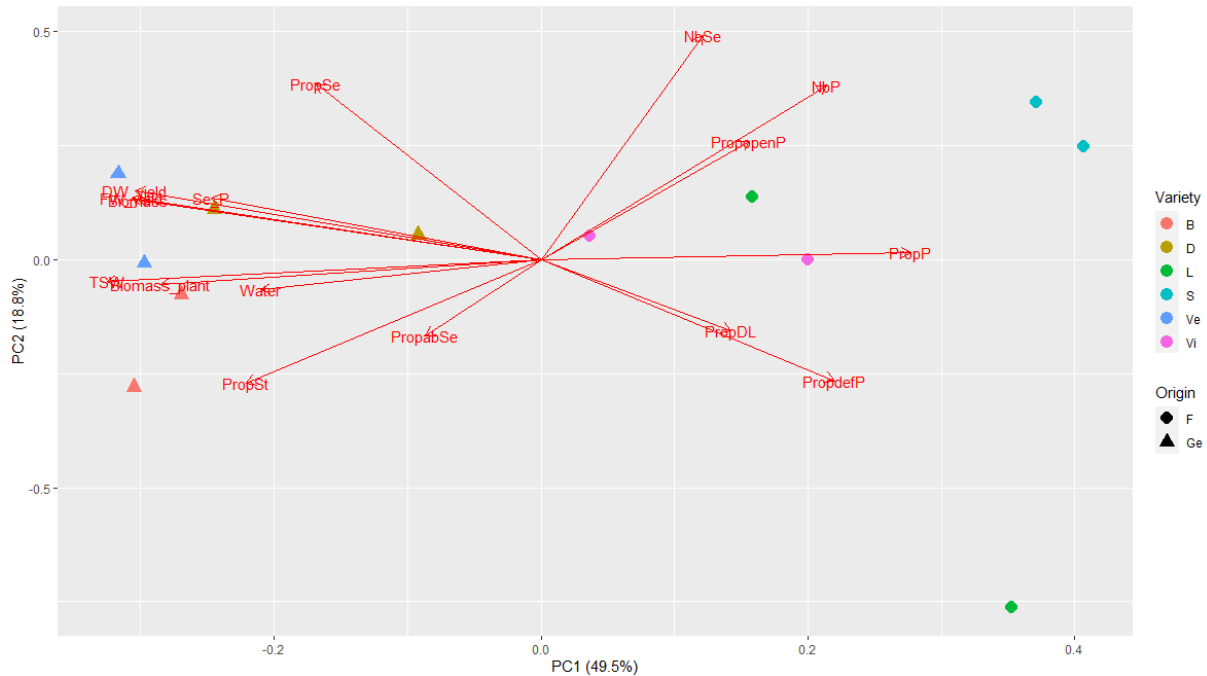


Figure 17: A loading plot of the principle component analysis for the third harvest. Variables: water content (*water*), thousand seed weight (*TSW*), biomass, biomass per plant (*biomass_plant*), seeds fresh weight (*FWSe*), seeds dry weight (*DWSe*), proportion dead leaves (*PropDL*), proportion stem (*PropSt*), proportion pods (*PropP*), proportion seeds (*PropSe*), number of seeds (*NbSe*), proportion aborted seeds (*PropabSe*), number of pods (*NbP*), proportion deformed pods (*PropdefP*), proportion open pods (*PropopenP*). The early varieties *Sampo* (*S*), *Louhi* (*L*), *Vire* (*Vi*) originate from Finland (*F*), whereas the late varieties *Birgit* (*B*), *Daisy* (*D*) and *Vertigo* (*Ve*) originate Germany (*Ge*).

The correlation matrix (Table 11) shows that TSW correlates strongly with: biomass, seed FW and DW ($P < 0.0001$), and biomass per plant ($P < 0.001$). And it correlates in smaller amount with number of pods ($P < 0.01$), water content, proportion of biomass allocated to stem, proportion of deformed pods and number of seeds per pod ($P < 0.05$). The total biomass per subplot correlates strongly with seed FW and DW ($P < 0.0001$), and biomass per plant ($P < 0.001$) and less with the number of pods, proportion of deformed pods and number of seeds per pod ($P < 0.05$). The biomass per plant correlates slightly less with seed FW (0.001) and DW ($P < 0.01$) than the biomass per subplot did.

Table 11: Correlation matrix of the third harvest variables. Variables in order from top to bottom: thousand seed weight (TSW), Water content, biomass per subplot, biomass per plant, seed fresh weight, seed dry weight, proportion dead leaves, proportion stem, proportion pods, proportion seeds, number of seeds, proportion aborted seeds, number of pods, proportion deformed pods, proportion open pods, and seed per pod. Significance: *, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$, $P < 0.0001$ ****. Minus values represent negative correlations.

	TSW	Water	Biomass	Biomass_plant	FW seeds	DW seeds	PropDL	PropSt	PropP	PropSe	NbSe	PropabSe	NbP	PropdefP	PropopenP
TSW															
Water	0.68*														
Biomass	0.91****	0.52													
Biomass_plant	0.89***	0.67*	0.83***												
FW seeds	0.93****	0.60*	0.99****	0.84***											
DW seeds	0.91****	0.51	1.00****	0.81**	0.99****										
PropDL	-0.29	-0.10	-0.30	-0.08	-0.31	-0.33									
PropSt	0.62*	0.12	0.43	0.43	0.40	0.42	-0.31								
PropP	0.31	0.13	0.56	0.49	0.48	0.53	0.10	0.08							
PropSe	0.36	0.24	0.48	0.24	0.52	0.54	-0.66*	0.02	-0.03						
NbSe	-0.43	-0.37	-0.07	-0.37	-0.09	-0.04	-0.08	-0.67*	0.23	0.38					
PropabSe	0.25	0.07	0.07	-0.03	0.08	0.07	-0.57	0.44	-0.35	0.02	-0.37				
NbP	-0.72**	-0.21	-0.69*	-0.81**	-0.64*	-0.66*	-0.01	-0.66*	-0.70*	0.04	0.46	0.02			
PropdefP	-0.59*	-0.56	-0.65*	-0.44	-0.69*	-0.67*	0.45	-0.13	-0.07	-0.67*	-0.20	-0.13	0.09		
PropopenP	-0.48	-0.57	-0.34	-0.51	-0.35	-0.31	0.18	-0.38	-0.18	0.16	0.56	-0.24	0.46	0.30	
Se_P	0.63*	0.14	0.64*	0.57	0.63*	0.67*	-0.37	0.58*	0.22	0.63*	-0.04	0.10	-0.53	-0.44	0.11

6.2.5 Yield component analysis

A yield component analysis was performed for the third harvest, since this harvest represents the potential harvest yield for a farmer (Table 12). For the yield component analysis specific traits were multiplied to receive the final seed yield for each variety. The early varieties Louhi, Sampo and Vire had higher number of pods per plant which was around 7.08 to 8.35, while the late varieties had between 4.97 to 6.42. However, the number of seeds in each pod was in average higher in the late varieties Birgit, Daisy and Vertigo (~3.05-3.30), than in the early ones (~2.68-2.88). The TSW showed that the seeds of the late varieties were bigger than the early varieties. By multiplying the values together for each variety, it was seen that the total seed yield in the late varieties was higher than in the early varieties. The overall highest was Vertigo with 295.18 g per cm², whereas Louhi showed the lowest seed yield with 130.72 g per cm². A statistical analysis of the DW yield (g/cm²) proved that there was a significant difference between varieties (P< 0,05).

Table 12: Yield components in faba bean with result from the third harvest. The results for yield components are averaged per plant for each variety.

Variety	Nb plants per cm ²	Nb pods per plant	Nb of seeds per pod	TSW (g) per seed	DW yield (g/cm ²)
Birgit	20	5.61	3.29	0.59	217.79
Daisy	26.5	4.97	3.30	0.53	230.35
Louhi	20.5	7.21	2.68	0.33	130.72
Sampo	23.5	8.35	2.88	0.25	141.28
Vertigo	22.5	6.42	3.05	0.67	295.18
Vire	25	7.08	2.74	0.37	179.44

6.2.6 Biomass accumulation

The distribution of biomass showed changes in allocation and accumulation to the different organs from the first to third harvest. At the time of the first harvest the highest amount of biomass was allocated to the leaves and less to the stem (Fig. 18A). All three late varieties produced more biomass than the early varieties. Birgit had the highest biomass content with around 2.6 g DW per plant and Sampo and Vire both had the lowest biomass, which was lower than 1.5 g per plant. At the stage of the second harvest a further increase of biomass was seen (Fig. 18B). The late varieties still produced more biomass than the early varieties. Both early and late varieties had started to develop pods, therefore distribution of biomass had moved on to be accumulated in pods and seeds. The proportion of biomass in the leaves had decreased, while the biomass in the stem had increased, this was observed for all varieties. At this point Vertigo had the highest production of biomass with around 15 g per plant, Sampo (11 g) and Vire (around 10 g) had still the lowest biomass production. Early and late varieties showed differences in the accumulation of biomass into the pods and seeds. The early varieties had higher proportions of biomass accumulated into the seeds in comparison to the late varieties. The late varieties on the other hand had a higher proportion of biomass accumulated in the pods rather than the seeds. At the last harvest there was a further increase of total biomass seen in some of the varieties (Fig. 18C). Vertigo had still the highest biomass production with now around 24 g per plant. Sampo on the other hand had a small decrease of 1 g biomass. Most of the biomass was now concentrated in the seeds and nearly no biomass was present in the leaves. In general, late varieties had a higher proportion of biomass distributed to the seeds than the early ones. Vertigo had from all varieties the highest amount of biomass accumulated into the seeds.

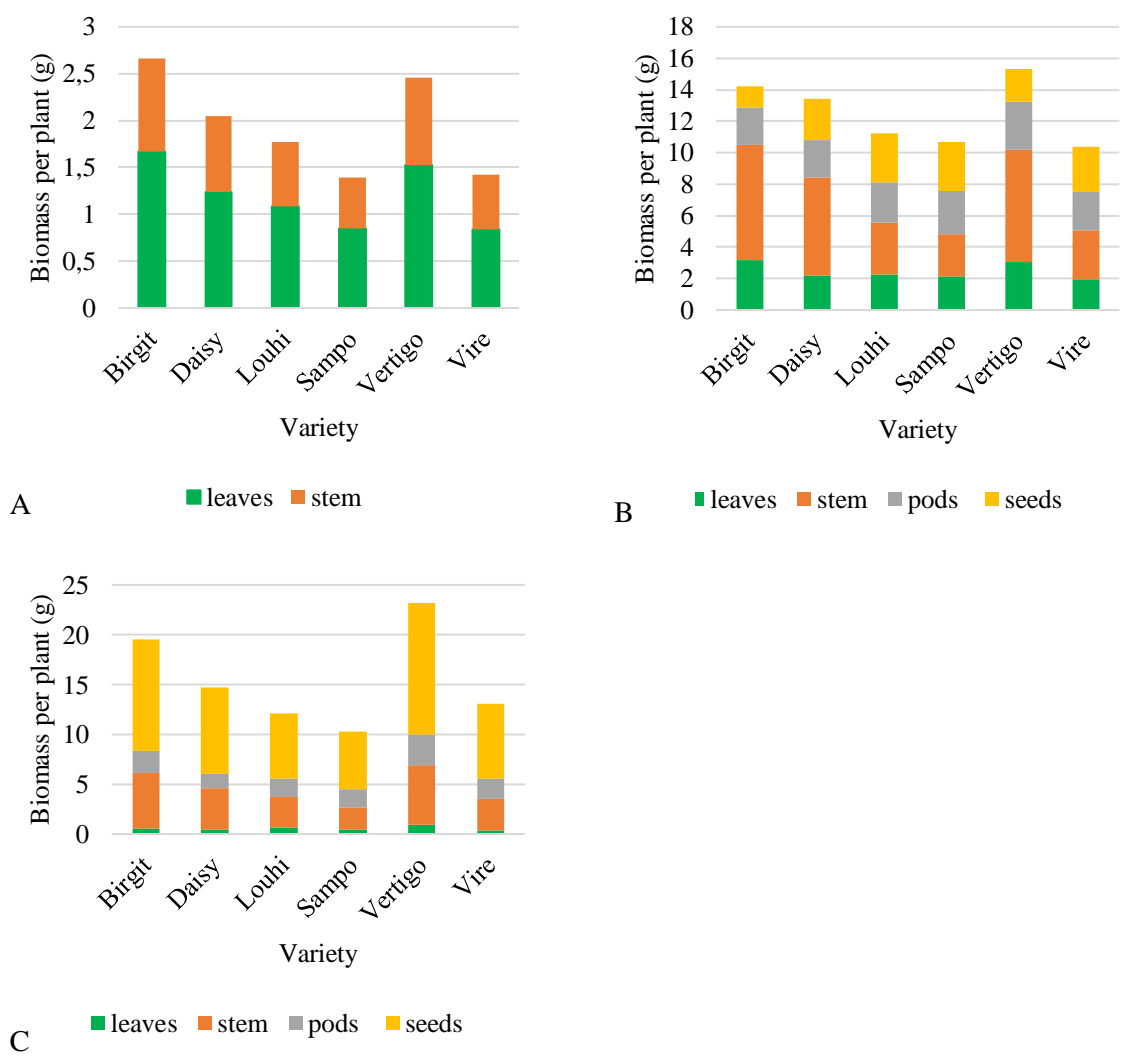


Figure 18: Allocation of biomass to organs per plant of each harvest. Proportion of biomass per plant (g) distributed to the organs from first to third harvest. Graph A: First harvest. Graph B: Second harvest. Graph C: Third harvest.

6.2.7 Nitrogen fixation, nitrogen content, and protein concentration in seeds

The ^{15}N isotope analysis gave the total N_2 fixed present in each organ, which was then further used in calculations to receive the total fixed N_2 of the whole plant. Firstly, it was detected that all varieties had a high percentage of atmospheric N_2 in the plants (Fig. 19). There was no differentiation between early and late varieties all varieties are over 85 %. Daisy had the highest percentage of fixed N_2 in the plants with 90 %.

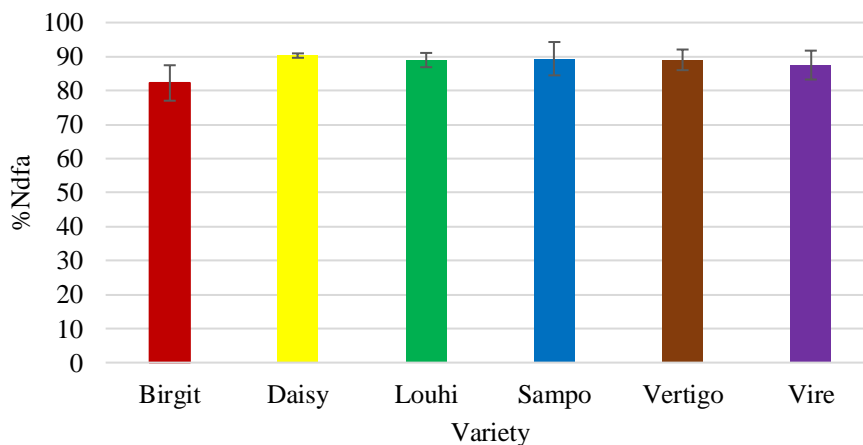


Figure 19: Nitrogen fixation ability of each variety after the ^{15}N isotope analysis. Percentage of N originating from atmospheric N_2 (%Ndffa). Data received from the second harvest. All bars are presented with SE.

The CNS Dumas lab analysis resulted in an overview of N concentration in the different organs for each variety (Table 13). At the first harvest only leaves and stem were present, at the second harvest leaves, stem, pods and seeds were present, and at the last harvest only the seeds were analysed. The N concentration was changing during development. The concentration in the leaves was increasing from the first to the second harvest in all varieties. The late varieties had higher N values in the leaves at the second harvest (between 4.1 to 4.5 %). From first to the second harvest were the N concentration in the stems decreasing in all varieties. The nitrogen in the pods was higher in the late (~4.3 %) varieties than in the early varieties (~2.5 %). The N concentration in the seeds showed no clear difference between early and late varieties. From the second to third harvest a decrease in the concentration in the seeds was seen in most varieties, only Sampo and Vire had a slight increase. Sampo had the highest N concentration of all varieties at the last harvest time (~5.45 %), whereas Daisy was seen to have the lowest content (~4.61 %).

Table 13: The N concentration (%) of dry matter in different organs at the different harvest dates. Result derived from CNS-Dumas method. At the first harvest the varieties were in the vegetative stage, therefore only values for leaves and stem. At the third harvest only the seeds were analysed for their N concentration.

Variety	Harvest	Leaves	Stem	Pods	Seeds
Birgit	1	3.9	2.5	-	-
	2	4.4	0.6	3.4	5.2
	3	-	-	-	4.8
Daisy	1	3.2	4.1	-	-
	2	4.1	0.7	3.3	5.0
	3	-	-	-	4.6
Louhi	1	3.3	2.9	-	-
	2	3.4	0.7	2.5	5.0
	3	-	-	-	4.9
Sampo	1	3.6	3.1	-	-
	2	3.7	0.8	2.6	5.2
	3	-	-	-	5.5
Vertigo	1	3.8	2.6	-	-
	2	4.5	0.7	3.4	5.0
	3	-	-	-	4.9
Vire	1	3.4	3.0	-	-
	2	3.5	0.6	2.4	4.5
	3	-	-	-	4.8

Brigit, Vertigo and Daisy had the overall highest total N content per plant (~0.045-0.034 g) at the first harvest (Fig. 20). They not only had a high N content in the leaves (~0.023-0.03 g) but in addition a high content in the stems (~0.009-0.01 g) as well. Sampo (~0.024 g) and Vire (~0.023 g) had the lowest total N content per plant.

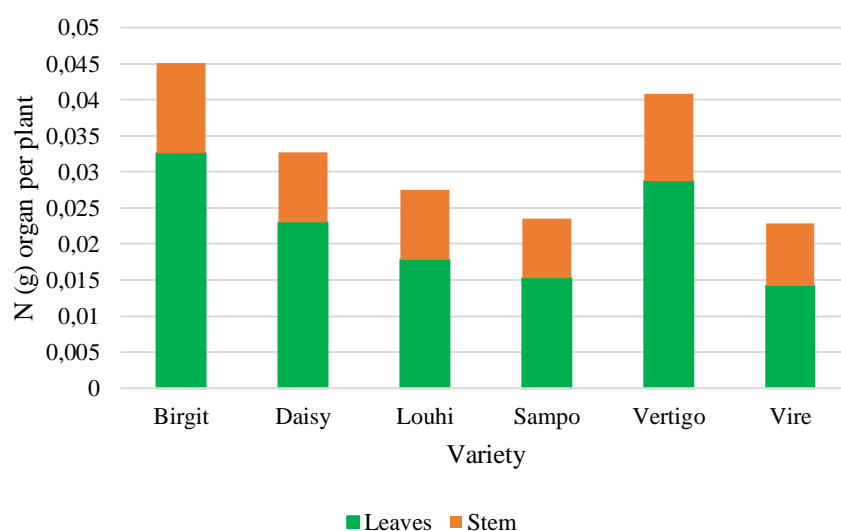


Figure 20: Nitrogen content in organs (leaves and stem) at the first harvest.

At the second harvest, the distribution of N in the organs had changed with increase in plant development (Fig. 21). The total N content had generally increased for all varieties (from 0.02-0.045 g to 0.148-0.19 g). The distribution of N had gone from leaves and stem to pods and seeds. Birgit and Vertigo still had a high N content in the leaves, and less allocation of N into the seeds was seen, in comparison to the early varieties. Birgit had a higher content of N in the pods (0.04 g) rather than in the seeds (~0.034 g). The N content in the stem had drastically decreased in the plants of the early varieties, they had now more N distributed into pods and seeds.

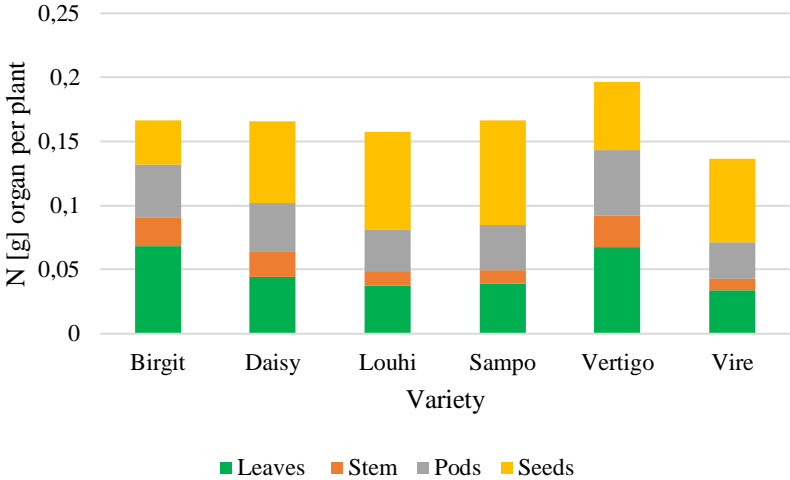


Figure 21: Nitrogen content in organs (leaves, stem, pods and seeds) at the second harvest.

The N content in the seeds increased further until the third harvest (Fig. 22A). The variation of N content between varieties had changed. At this time Vertigo, Birgit, Daisy had the highest N content in seeds (~0.4-0.6 g) compared to the early ones, although the N content in the seeds of the early varieties had increased as well (~0.3-0.4 g). By comparing the N content in the seeds with the biomass proportioned to the seeds side by side, it was seen that even though Vertigo had the highest total N content in the seeds, followed by Birgit, both also had a high proportion of biomass in the seeds (Fig. 22B). Whereas the early varieties had a low N content in the seeds, however, they also had a lower proportion of biomass allocated to the seeds.

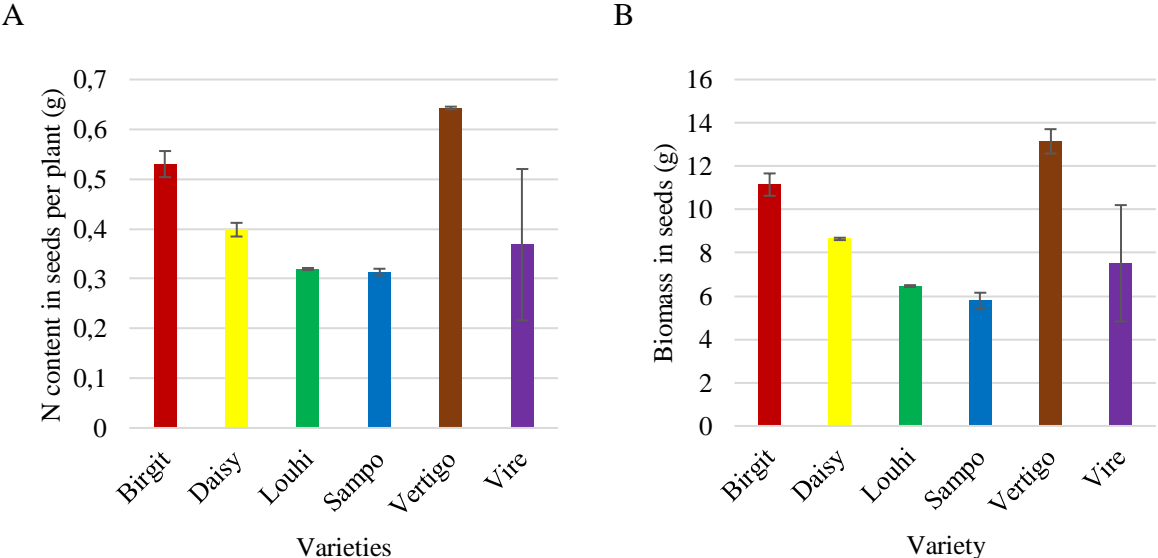


Figure 22: Nitrogen content distributed to seed at the third harvest (A) and biomass proportioned to seeds (B) divided by the number of plants in the subplot. All bars are presented with SE.

With the nitrogen concentration from the CNS Dumas method (Table 13), the protein concentration from the second and third harvest was calculated (Table 5). At the second harvest most of the varieties were close in the concentration, which was around 30 % (Fig 23A). Only Vire was slightly lower with around 27 %. In both, second and third harvest there were only small difference between early and late varieties. The protein concentration decreased in four of the varieties (Birgit, Daisy, Louhi, and Vertigo) until the third harvest (Fig. 23B), only in Sampo and Vire there was a slight increase. Sampo had the highest protein concentration with ~34 %, whereas the other varieties had protein concentrations around 30 %.

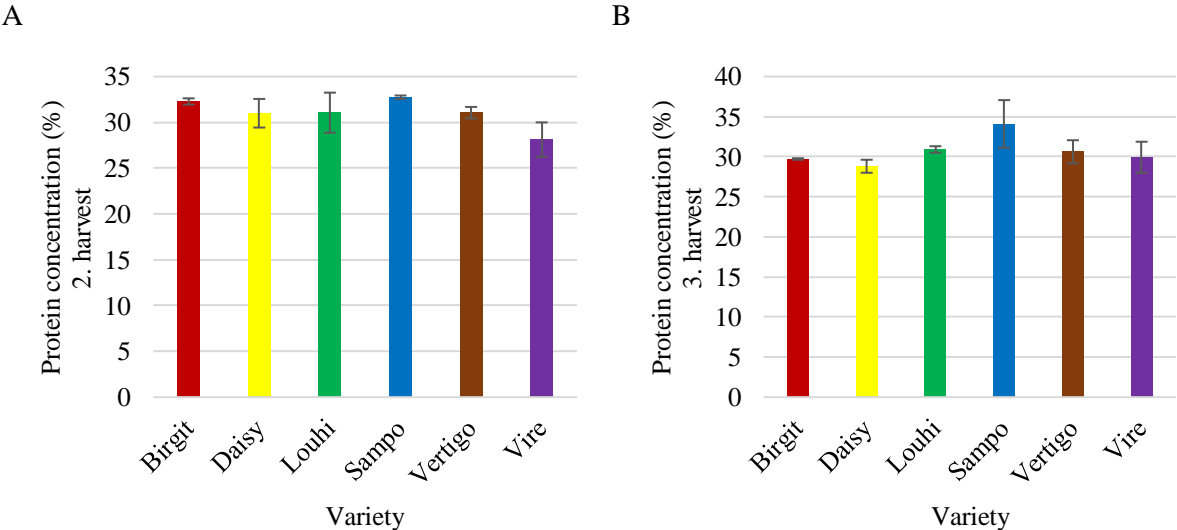


Figure 23: Protein concentration of the seeds at second (A) and third (B) harvest. All bars are presented with SE.

7. Discussion

7.1 Phenology

Yield is influenced by different traits. The phenology of a plant is an important factors to consider in the yield formation. It is therefore important to analyse the underlying traits in the plants phenology (Bodner et al., 2018).

7.1.1 Greenhouse experiment

Legumes go through different main phenological life stages, as for instance germination, emergence and vegetative and generative growth (Etemadi et al., 2018b). These stages and especially the beginning of the flower induction are crucial for the yield determination (Etemadi et al., 2018b). Each species requires a specific heat sum to reach the different development stages and temperature can have a direct effect on how fast the plant will reach those stages (Etemadi et al., 2018b).

The temperature treatments showed that it can affect the development. Further that the warm temperature, in addition to potential drought stress, can impact the development of *V. faba* negatively. Under optimal conditions each variety should require the same thermal time for the main development stages. However, the experiment in the greenhouse showed that the varieties in the warm room required a higher heat sum during the whole development. They especially required more thermal time to reach flowering and anthesis. These stages are also the most sensitive to high temperatures and drought (Bishop et al., 2016). It is important to mention that the temperatures chosen for the rooms in the greenhouse are not considered to be too high or too low for the development in faba beans (Bishop et al., 2016). With that being said the actual temperature in the room possibly could have been higher, especially closer to the summer months, than planned. The rooms used for the experiment are ordinary rooms that are not temperature controlled with modern technology, but rather temperature controlled by opening and closing roof windows, which is why the ventilation perhaps was not appropriate. This can be regarded as an error in the experiment. Since faba bean is sensitive to heat, the maximum temperature is more important than the average temperature.

On occasions of heat stress different responses in different species have been observed (Bishop et al., 2016). For instance, wheat (*Triticum aestivum* L.) has shown accelerate development, specifically in the stage of flower production but heat stress caused pollen infertility (Bishop et al., 2016). Too high temperatures can cause a higher energy (assimilate) requirement for developing seeds and result in a deficit of assimilates (González et al., 2011). The end-result is

a reduced final yield, which agrees with the findings of the greenhouse experiment. In other species such as common bean (*Phaseolous vulgaris* L.) and pea (*Pisum sativum* L.) reduced pod and seed sets, and yield were observed when exposed to temperatures above 30°C (Bishop et al., 2016). For those high temperatures the species required less thermal time to reach maturity, however in this experiment the warm treatment was around 22°C and they required more thermal time to reach maturity. It seemed like Vertigo was potentially most affected by higher temperatures possibly combined with drought stress, since it required more thermal time during flower emergence until flowering (BBCH 50-60) than the other two varieties. A possible explanation is that the varieties in the cold room could have had optimal conditions, therefore their enzyme and metabolism activity was more efficient, since plants under heat stress have a decreased photosynthetic activity, which is why less assimilates can be produced (Wang et al., 2020). The effect of temperature needs to be taken into account when choosing varieties for specific areas. The varieties that are chosen in Trøndelag cannot necessarily be the same as in Viken, due to different requirements in temperature.

7.1.2 Field experiment

To record the plant development in the field two scales were used and compared, the BBCH and GRDC scale. The use of the two scales showed that there are differences on how and to which extent they record the development stages. The GRDC scale only showed differences in development between early and late varieties. Through the GRDC scale it was concluded that the early varieties were early and required less thermal time at the beginning of development during leaf unfolding until pod set and the late varieties were late and required more thermal time at those stages and then again at maturity. However, during the first leaves unfolding and the pod set early and late varieties had the same thermal time requirement.

The BBCH scale could more precisely identify development stages, which facilitated the identification of more differences between the varieties in plant development, not only between early and late varieties. The scale also showed that during flower emergence and flowering the early varieties were earlier, but it also showed that Vertigo is not necessarily a late maturing variety, but in between early and late. Vertigo grown in Canada are usually a medium to early maturing cultivar (Kreutzer Korsvold, 2020), yet in Norway Vertigo is considered a late maturing variety (Strand, 2017). This could be an advantage for choosing Vertigo as a crop if the farmer is uncertain about the environmental conditions of the growing season. Vertigo has already been a popular choice around the Oslo fjord (Abrahamsen & Waalen, 2020).

Both scales showed that pod development and grain filling required the most thermal time.

More detailed information of the development of the varieties was obtained through the use of the BBCH scale. The reason could be that the GRDC scale is developed for varieties grown in Australia. Therefore, the scale might be more detailed for the growing conditions there, but perhaps not applicable to show the differences between varieties for Nordic climates. If one only requires to test the earliness of the varieties, the GRDC can be used as well. It was important to see that the GRDC scale could differentiate early and late varieties in important stages such as flowering and maturity, which is necessary knowledge for farmers to plan the growing season of their crop.

In growing seasons with optimal conditions, such as precipitation after sowing and emergence and warm and dry conditions at pod maturity the late varieties could be possible to use. However, in growing seasons where frost comes early in September, early varieties would be advised to choose. This is because, grain filling requires the most thermal time and in some growing seasons the amount of thermal time cannot be achieved. The field experiment showed that the early varieties required less thermal time and therefore reached pod maturity earlier. It is important to select and breed for varieties that will be able to reach maturity under different climatic conditions.

According to Duc (1997) early flowering and as a result early pod maturity is an important trait for breeding purposes, as this will give possibility to grow winter wheat after faba bean (Øverland, 2020). Under long-day conditions-which are prevailing in Norway flowering can happen earlier than in Mediterranean regions (Bodner et al., 2018). Faba bean has in different studies shown to be sensitive to photoperiod (Patrick & Stoddard, 2010). This is specifically important for the Nordic regions (Bodner et al., 2018). Bodner et al. (2018) showed that higher yield were achieved through prolonged flowering. Yet they stated that the *major* types require higher temperatures for vegetative and generative development and if the temperatures are not corresponding with the requirements of the genotype, they will mature later and cause yield depression (Bodner et al., 2018). This means that photoperiod and temperature need to match for an optimal plant development. The climatic conditions in Norway can vary from season to season, which is why there is a high risk that the later (or *major*) types will not mature in time in some growing seasons.

7.2 Biomass

The production of biomass derives from the photosynthetic activity of developed green leaves (van Bel et al., 2003). The individual leaves start out as sinks and are later when they are

completely unfolded sources for assimilates. That is, if there are no stress factors when the photosynthetic capacity is the highest (Hay & Porter, 2006). Carbohydrates are formed through different process in the photosynthesis, which are further transformed into proteins and lipids. The assimilates are transported through the phloem from the sources to the sinks (Hay & Porter, 2006). The partitioning of these produced assimilates is controlled by the LAI but also by solar radiation, temperature and precipitation (Liu et al., 2019). At the first and second harvest the LAI, SLA, LAR were calculated. These factors are important for the estimation of the canopy's photosynthetic capacity (Amanullah et al., 2007). The LAI is linked to light that can be intercepted by the plant (Bréda, 2008). In the experiments there were no significant differences neither between varieties nor between harvests one and two. There was a visible trend that the LAI of the late varieties was higher in both harvests in comparison to the early varieties. However, the LAI did not give us a significant indication of the photosynthetic activity. LAI is also influenced by sowing date and sowing density, therefore it could be possible to observe changes in LAI regarding those factors in further experiments in the future and observe if the difference between varieties changes by optimizing sowing density (Confalone et al., 2010). The measurement of the chlorophyll content on the other hand showed a significant effect. The chlorophyll content expresses the intensity of the green colour in the leaves and is therefore mostly used to observe nutrient deficiencies (Liu et al., 2019). Since chlorophyll is a pigment in the photosynthesis, it can give direct information of the photosynthetic activity and therefore growth as well (Li et al., 2018). The chlorophyll content was measured at the same day as the first harvest was done. The result showed there were a significant differences between varieties ($P < 0.001$). Graphically it was seen that all three late varieties had higher chlorophyll content than the early varieties. It has been described that the total amount of chlorophyll content in the leaves has a direct impact on the photosynthetic capacity (Li et al., 2018). For this experiment it could be assumed, that since the late varieties had a higher chlorophyll content, they also have a higher photosynthetic capacity. A potential result of the higher chlorophyll content is the higher amount of biomass produced in the later varieties. The late varieties showed a higher content of biomass per plants in the first and third harvest, in addition to a higher total biomass content of the subplot, which shows a potentially a higher photosynthetic capacity. In for example a wheat experiment in Iran it was seen that the chlorophyll content correlated positively with grain yield (Miri, 2009). In case of further experiments with faba bean it is possible to observe the correlation between chlorophyll content and yield in Norway and to focus more on the photosynthetic capacity of faba bean varieties in Norway.

The total biomass per subplot and the biomass per plant is in some harvests significant and in some others no significance was proven. That could be due to different responses in plant density. The sowing machine tries to uniformly distribute the seeds on the field. For this project it was aimed for 60 plants per m², however the distribution in correlation with emergence does not always result in a uniform distribution of seeds. Therefore, the plant density can vary and give different results for biomass in total and biomass per plants. The plant density can affect the accumulation of biomass both positively and negatively (López-Bellido et al., 2005). Often, high biomass production can be achieved with high plant densities, on the other side high plant densities can increase plant competition (Donald, 1963). It is important to have the right balance between optimal biomass production and low plant competition. This should potentially have been a bigger focus in the conducted field trial, since there were wide differences between early and late varieties in their biomass production.

Dry matter production in stems after beginning of pod set can increase, especially with high plant densities, whereas pod weight per plant increases with low densities (Alharbi & Adhikari, 2020). With regard to this field experiment the early varieties had smaller plants, due to less production of biomass than the late varieties. By increasing the sowing densities of varieties with less biomass production (smaller plant) there is a potential to achieve higher yield in those varieties as well.

In the correlation table a high positive correlation between TSW and biomass was observed ($P < 0.001$). The early varieties were also the ones with the lower TSW.

By increasing the plant density in varieties with low TSW, it could be possible to achieve a higher biomass production (Donald, 1963)

Future experiments could be revolving around plant densities and how different varieties react to different densities under Norwegian conditions.

The accumulation of biomass changes with development (Pilbeam et al., 1991), which was seen in this experiment as well. The amount of biomass produced reaches its maximum level until flowering (López-Bellido et al., 2005). In this experiment however the highest amount of biomass per plant was seen at the last harvest, which could be because of the addition of pod and seed weight at the third harvest.

Biomass accumulation is strongly correlated with the formation of seed yield (Köpke & Nemecek, 2010). Therefore to obtain a higher yield in seeds a high accumulation of biomass is suggested (Etemadi et al., 2019). Thus it is important to analyse where biomass is allocated and especially how much is accumulated into the different organs and how this differs between varieties, which is related to the source-sink relationship in the plant. The late varieties had

potentially the higher photosynthetic activity, which could have resulted in a “better” net exporter of assimilates from the leaves (source) to the pods and seeds (sinks) (Hay & Porter, 2003). The level of partitioning is not only changing during development but there is also a constant competition between the sinks (Hay & Porter, 2003). The analysis of competing sinks is however difficult. A strong sink can be defined in its ability to take up assimilates (Hay & Porter, 2003). The reason why the late varieties had more biomass allocated to the seeds could therefore have different reasons. It could mean that their seeds are stronger sinks for assimilates and can better outcompete the other sinks (pods, and stem). It could however also be the result of a higher biomass production in the plants of the late varieties and therefore naturally more biomass can be located to the seeds. This can be supported by the fact that the capacity of a sink is defined by available assimilates (Hay & Porter, 2003). Another potential reason could be the length of the development. The late varieties needed more time to reach maturity. This could mean that they use more time to allocate as much assimilates to the seeds as possible.

A higher biomass content in the late varieties could be achieved by the late varieties developing slower and seemingly grown faster, therefore they can utilize the longer photoperiod and higher temperatures better, to optimise their biomass production (Confalone et al., 2010). During the field trial there was a high infection rate with the chocolate spot fungi. It was observed that the early varieties were more affected by the infection than the late varieties. Biotic stresses can drastically reduce crop productivity. It is possible that the infection reduced the biomass production in the early varieties, meaning that as a stress response the plants accelerated their development through the production of reactive oxygen species (ROS) (Jeandroz & Lamotte, 2017). Through this process it is possible that less seeds per pod were produced, due to the lack of biomass.

In the experiment clear significant height differences between early and late varieties were proven. The late varieties were on average higher. Plant height can be positively correlated with the yield (Link et al., 1999). This is because with longer main stems there is a higher possibility for more nodes producing pods (Bodner et al., 2018). However the high plants were exposed to lodging and this has also been seen in previous research. Therefore the trait is less often added into breeding programs (Bodner et al., 2018).

7.3 Nitrogen fixation, nitrogen content, and protein concentration in seeds

Understanding N uptake and allocation inside the plant has two major advantages: one, the N that is not taken up by the plant is a potential risk for leaching to ground water and second, the N uptake is seen in direct relation to the yield (Gastal & Lemaire, 2002). That means by

understanding the N uptake and distribution in the crop is of ecological and agronomical importance.

As other legumes, *V. faba* has the ability to fix atmospheric N₂ through the symbiosis with rhizobia bacteria (Confalone et al., 2010). Thereby the crop can produce yield without the need of additional synthetic N fertilizer (Mínguez et al., 1993). The potential of fixing atmospheric N₂ is impacted by several factors, crop adaptability, crop management, and plant growth (Costa et al., 1994). In addition each variety can have a different ability to fix N₂. One of the reasons is that the plants need to use a lot of assimilates produced during photosynthesis in the symbiosis (Costa et al., 1994; Franche et al., 2009).

The ¹⁵N isotope analysis after the second harvest showed that all varieties had a very high amount of N₂ fixed from the air, which corresponding to approximately 80 to 90 % Ndfa. These high values match with the analyses made by Sørheim (2021) where she observed a %Ndfa of 80 in Vertigo. Faba bean and soybean are generally known to have very high fixation rates that can range between 75 % and 68 % (Etemadi et al., 2019). Legumes that are able to fix between 50 to 90 % of their N requirements are effective in the N fixations abilities (Etemadi et al., 2019). Therefore it can be assumed that all varieties produced enough assimilates to exchange with the rhizobia symbionts, and have an effective symbiosis (Costa et al., 1994). It could however also mean that the varieties were less effective in taking up N from the soil.

In the growth and development of legumes, photosynthesis and nitrogen fixation are important processes (Vollmann et al., 2011). Chlorophyll content can be an important indicator for photosynthetic activity and it also expresses the nitrogen content in the leaves (Liu et al., 2019). The high chlorophyll content in the leaves could correspond with the higher nitrogen content in the leaves in the late varieties. Since the chlorophyll content can be used as indication for N deficiency (= low chlorophyll content) (Liu et al., 2019), it can express nodulation and N fixing abilities in legumes (Vollmann et al., 2011). For example in soybean it has been detected that around 50 % of the nitrogen content in the leaves is part of Rubisco (Vollmann et al., 2011). In a study carried out by Vollmann et al.(2011), it was shown that the chlorophyll content increased with increase in N supply provided through the fixation during development. In this thesis it is rarely possible to compare the nitrogen content obtained through fixation with the chlorophyll content, since there were as said very similar results between varieties in their amount of N₂ fixed from the atmosphere, and in the chlorophyll content there were more differences. Furthermore the ¹⁵N isotope analysis was done during the second harvest and chlorophyll measurements in the first, therefore it is possible that the difference in chlorophyll content between varieties might have changed until the second harvest.

As the biomass distribution is changing during development, so is the distribution of the nitrogen content. The accumulation of nitrogen is highly correlated with the accumulation of biomass (Gastal & Lemaire, 2002). At the first and second harvest it was seen that the late varieties had a higher N content in the leaves than the early varieties. The leaf N content determines the photosynthetic response of the leaf due to a high proportion of Rubisco that is represented in the N content of the leaf (Gastal & Lemaire, 2002). It is possible that since the N content in the leaves of the late varieties is higher, they have a higher light use efficiency (Gastal & Lemaire, 2002; Hirose & Werger, 1987). As a result it is possible that the late varieties were able to produce more biomass, which also corresponds to the higher chlorophyll content in the leaves (see 7.2). During the increase of biomass the leaf : stem ration in nitrogen is generally decreasing (Gastal & Lemaire, 2002). This is due to the fact that an increased proportion of N, but also biomass is allocated to the reproductive organs. However, the total N content is increasing as well (Gastal & Lemaire, 2002). This was also seen in the field experiment. The increase in grain content is not only dependent on the supply of N but also on the allocation capacity from vegetative organs to the reproductive organs of the plant (Salon et al., 2001). Leaves and stems contribute the most to the N content in the seeds (Salon et al., 2001). Since the late varieties produced more N in the stems and leaves, they were able to mobilise more N to the seeds as well. By comparing N content with biomass side by side, it can be seen that the N content reflects the variation of biomass in a high degree. Birgit, Daisy and Vertigo produced the most biomass and they also have the highest N content. However, on the other hand, the concentration of N is less correspondent to the N content. It can be seen that Sampo has the highest N concentration in its organs from first to third harvest. It is specifically important that Sampo's higher N concentration in the seeds possibly corresponds to its higher protein concentration in the seeds as well.

V. faba has a high protein concentration in the seeds and can therefore easily compete in consumption share with soy, pea or lentils (Sharifi, 2014). The protein concentration of faba bean in Norway can be set around 30 % and is therefore a good source for feed and food (Øverland, 2020).

There have been discussions if the protein concentration and yield are negatively correlated (Skovbjerg et al., 2020). Studies involving pea and chickpea have confirmed such negative correlations, however there have also been studies where the correlations have been absent (Frimpong et al., 2009; Skovbjerg et al., 2020; Stoddard et al., 1993). Skovbjerg et al. (2020) therefore reported negative correlations between protein concentration and yield specific to the

varieties they used in their study. In this thesis the protein concentration varied between 34 % (in Sampo) to 29 % (in Birgit) in the third harvest, however it was detected that the protein concentration in some varieties decreased from the second to the third harvest. At the third harvest specifically the three late varieties had a decreased protein content, whereas the three early varieties had either no change (Louhi) or a small increase (Sampo and Vire).

There are different explanations to consider: One, during maturation the starch content is increasing and thereby diluting the nitrogen content (Skovbjerg et al., 2020). Starch content increases during seed development, whereas the proteins that are synthesized during the beginning of seed development are degraded simultaneously as the embryo grows (Kreutzer Korsvold, 2020; Warsame et al., 2018). Secondly, another factor is the dependence of the protein concentration to the variety (Skovbjerg et al., 2020). In this thesis there were differences in protein concentration between early and late varieties. In Skovbjerg et al. (2020) it was seen that the varieties with low TSW had a higher protein content than the varieties with a higher TSW. This corresponds to our findings of the protein content in the third harvest, where Sampo had a higher protein content even though it had a very low TSW. It could be that there is an indirect negative influence of the TSW. Yet Alharbi and Adhikari (2020) stated that big seeded varieties have a higher protein concentration, however as said the small seeded varieties Sampo, Vire, and Louhi had a higher concentration. It can therefore not be made any final statement what variable indicates a higher protein concentration, since it is influenced by several ones.

7.4 Seed-yield components

The seed yield results from the general growth and development of the crop (Confalone et al., 2010). Together growth and development determine the sink strength and thereby how much biomass is partitioned to the seeds. The seed yield shows a direct relationship to the effectiveness of translocation of assimilates (Etemadi et al., 2018b). The effectiveness of translocation to the different sinks can depend on various factors, like the genotype, abiotic and biotic stresses and growing season (Etemadi et al., 2018b). The seed yield each year reflects the environmental effects on the seed-yield components (Ayaz et al., 2004). Therefore, the following discussion about the seed yield will comprise the traits that have been discussed in the previous section and that are each impacting the final yield. This thesis looked at different traits of faba bean that influence the yield. It is possible that the differences in these traits make some varieties more profitable in cropping systems in Norway than others.

As explained in the section of the phenology discussion (7.1.1) the warm temperature room affected the development of the varieties negatively, further this resulted in negatively affecting the yield components. All varieties produced significantly more pods, seeds, seeds per pod, and seed dry weight in the cold room. The cold room could have had better climatic conditions, where the plants had a slower development and faster growth than in the warm room, meaning they had more time to accumulate assimilates into the seed sinks.

Since the seed yield can be limited by the production of biomass, periods with too high temperature possibly combined with water stress during the main biomass production may have caused low production of pods, and seeds in the warm room.

The climatic conditions in Northern Europe are show-cased by shorter growing seasons, but with possible heat and drought waves (Bodner et al., 2018). There are not that many studies about varieties under Northern European climate conditions (Bodner et al., 2018). Therefore, investigating the influence of different temperature regimes on varieties further can possibly give novel information on how temperatures prevalent in Northern Europe effect varieties and correlate with the yield components.

By looking at the two seed-yield components (TSW and number of pods) that were significantly different between varieties, it was seen that Vertigo had a higher weight than Sampo and Lielplatones. Yet Sampo had a higher number of pods. This differences between Vertigo and Sampo were also observed in the field.

The seed yield is comprised by several yield components, some of which can correlate more with the yield than others (Alan & Geren, 2007). Correlations between components and yield is also indicating a form of yield stabilisation (Alan & Geren, 2007). As described in the theoretical section, the yield in faba bean is determined by the number of plants per m², the number of pods per plant, the number of seeds per pod, and the TSW per seed (Alan & Geren, 2007). To increase seed yield it is necessary to observe which components correlate with the yield more positively than others.

With the correlation matrix (Table 11), it was observed that TSW, biomass and seed FW correlated the most ($P < 0.001$) and biomass per plant ($P < 0.01$) and seeds per pod ($P < 0.5$) but still positively with the seeds DW. The number of pods and deformed pods correlate negatively ($P < -0.5$) with the seeds DW. In the seed-yield component analysis it was seen that the early varieties had a higher number of pods, however the late varieties had a higher number of seeds per pod. It could mean that the early varieties used more biomass to produce pods rather than to allocate the biomass further to the production of seeds. Therefore, it is possible that the number of pods is negatively correlated with the DW of the seeds. The number of seeds is only

slightly correlate with the seed's DW, since it could be more important to have a higher biomass partitioned to the seeds, which was observed with the extent of the significant correlation between biomass and seeds DW. It must be said that the late varieties produced more total biomass and more biomass per plant, therefore they also had more biomass available to be allocated to the seeds. In this experiment TSW correlated very highly with the seed's DW, which could mean that varieties with bigger seeds produce higher yields.

As previously mentioned, the production of biomass shows the efficiency of photosynthesis in the variety, the seed yield on the other side reflects efficiency of translocation of assimilates to sinks (Etemadi et al., 2018b). The correlations that were observed in our experiment correspond with other research (Etemadi et al., 2018b; Loss et al., 1998). Etemadi et al. 2018 stated the same result as was seen in this experiment, that the varieties with the highest biomass production also had the highest seed yield. It was however stated by López-Bellido et al. (2003) that the correlation is not necessarily always present. That means that high biomass production does not always result in the highest seed yield (López-Bellido et al., 2003). An effective sink-source relationship can determine a higher seed-yield as well (Etemadi et al., 2018b), which is mostly determined by the harvest index (Alharbi & Adhikari, 2020). Even though the calculation of the harvest index was not part of this thesis, an indication of the source-sink relationship can also be observed in the biomass partitioned to the different organs as mentioned in 7.2.

The final result in the yield component analysis showed that Vertigo produced the highest yield. The question is what trait does Vertigo have that made it perform best or better than the other varieties?

Vertigo is in some of the development stages (BBCH 20 to 50) between early and late varieties, however flowering and pod maturity require the same thermal time and DAS as the late varieties. In growing seasons with cold temperatures prevailing in spring, Vertigo could be an option. This could be due to a lower requirement of thermal time in the stages of biomass production. The variety could have a quicker start than for instance the other two high yielding varieties Birgit or Daisy, since it was observed to have a higher LAI. To add the observations from the greenhouse experiment, Vertigo was the most affected by potential heat and drought in the phase from flowering to maturity, since at these stages the development slowed down and the plants required more thermal time to mature. Therefore, Vertigo could be better adaptable to colder climates. Even though Vertigo had a higher biomass production with less required thermal time than the other two late varieties Birgit and Daisy, there is still a risk that Vertigo could have high yield losses under drought periods. With climate change also affecting

Nordic countries, Norway will be expected to have a higher risk for drought in the southern regions in the upcoming years (O'Brien et al., 2006). Vertigo should be tested over a longer period to investigate its performance under different Norwegian climatic conditions.

In the first and second harvest, Vertigo does not stand out between the other varieties. Its total biomass production (per subplot) and biomass per plant, as well as the biomass proportioned to the stem is only the second highest after Birgit. Vertigo showed a weak allocation of biomass to the seeds and pods at the time of the second harvest (pod filling stage). The PCA of the second harvest indicated that Vertigo correlated positively with height and biomass proportioned to stem, LAI and other variables. The switch arrives with the third harvest, where Vertigo resulted in having the highest production of total biomass and biomass per plant, moreover it had the highest seed FW and DW. Even though it had the highest N content per plant at the second harvest and the highest N content per seed in the third harvest, its protein content was only average. Thus, the protein content did not decrease as much from second to third harvest as in Birgit and Daisy. The seed-yield component analysis resulted in a low number of plants per cm² and number of pods, however the number of seeds per pod were the third highest and the TSW of Vertigo was the highest of all varieties, which was already the case at the second harvest. The result of Vertigo producing the highest yield, even though many of the variables have lower values in comparison to the other varieties, shows the importance of TSW.

In general the TSW is fundamental to measure the seed size and further directly impacting the seeding management (Boychyn, 2022). Crops with larger seeds have automatically a higher TSW and reversed (Boychyn, 2022). The TSW is indirectly related to several of the seed-yield components and is directly linked to germination and the final yield (Mirić et al., 2007). Other studies by Berhe et al. (1998), Tadesse et al. (2011) and Ulukan et al. (2003) confirm the direct positive effect of TSW on the seed yield. For the evaluation of the seed yield components, as well as phenology, height and protein content, the subspecies *minor* and *major* play a distinctive role (Bodner et al., 2018). Since Vertigo, Birgit and Daisy belong to the *major* and Vire, Sampo and Louhi to the *minor* type, this statement agrees with that the yield produced in this experiment differs between these subspecies. The sowing density should be adjusted to the seed size. As said previously, all varieties were sown uniformly without regard to seed size, which could have caused more difference in yield than necessary. In a growers guide provided by “Norsk Landbruksrådgivning” for Viken (Øverland, 2020), a sowing density of 60 plants per m² is recommended, which was also done in this experiment. However, they also described that the varieties Louhi and Sampo require a higher sowing density of around 80 plants per m². It is

possible that this has added to the lower yields in these two varieties. Even though higher sowing densities have shown to increase lodging, it has also been seen that it increases the yield. Further experiments regarding sowing densities should be done to confirm variety specific densities for an optimal crop performance.

When focusing on the weather conditions in North Europe, the Finnish varieties will in some years potentially be the better option, due to their earliness in maturity. Of the early varieties, the highest yield was seen in Vire and the lowest in Louhi. The yield potentials of Sampo and Louhi were observed in a field trial carried out by Abrahamsen et al. (2018). They observed that Louhi had the highest total yield at different locations in comparison with Sampo and another variety called Kontu. They additionally stated that in a Finnish trial from 2007 to 2014 Louhi had a 10 % higher yield (Abrahamsen & Waalen, 2020; Strand, 2017). Since the field experiment for this thesis were only carried out in one growing season, it cannot be clearly stated which of the variety from the early once should be chosen to grow in case of early frost for example.

In general the results collected in this thesis respond to only one growing season, it is possible that by conducting the same experiments over several growing seasons, different results would be found. Nevertheless, the results of this thesis can be of help to design further experiments with faba bean in the future.

8. Conclusion

Faba bean (*V. faba*) due to its high protein content has a high potential to be integrated into cropping systems in Norway and thereby to reduce the soy imports.

Aim of this thesis was to describe the response of faba bean to the four objectives: Characterisation of phenology, biomass accumulation and allocation, nitrogen accumulation and allocations, and seed-yield components.

The greenhouse experiments gave a good starting overview of the crop itself. It showed the influence of temperature on faba bean and how the different varieties response in their development. The varieties in the warm room required a higher accumulation of heat from emergence until flowering. Additionally, they produced less pods and seeds under the warmer temperature treatment. Separating the effect of temperature between the varieties, it can be assumed that Vertigo reacted more negatively to the warmer temperature than Sampo or Lielplatonas, since it required more thermal time during the whole development.

By using the developmental scales BBCH and GRDC in the field experiment, it was seen that regardless of late and early development the varieties required the same amount of thermal time from emergence to stem elongation. The early varieties required less thermal time to reach flowering than the late varieties. The early varieties reached maturity with less thermal time than the late varieties. With the BBCH scale more variation between varieties was shown. It was observed that Vertigo was between early and late varieties in their timing of flowering, but matured at the same time as the late varieties. With the GRDC scale it was only possible to distinguish between early and late varieties.

The observation of biomass at three different time points in the development showed that the late varieties produced more biomass. They allocated and accumulated more biomass to the leaves and had a higher chlorophyll content already in the first harvest than the early varieties. This could indicate that the late varieties had a better photosynthetic activity. The early varieties had allocated more biomass to the pods and seed than the late varieties at the second harvest. A potential reason could have been that rather than harvesting the varieties at the same stage in the development they were harvested at the same date, the early varieties were already at the stage of allocating more biomass to pods and seeds, rather than being more effective in the allocation. At the last harvest the late varieties had allocated more biomass to the seeds than the early varieties. Potential reasons could be that they are more effective since they produced more biomass to also allocate a greater amount to the seeds. It could also be since the late varieties are bigger seeded, their seeds are stronger sinks and require more biomass.

All varieties contained over 80 % atmospheric N₂, fixed through the symbiosis with rhizobia bacteria. The N content in the organs reflected the variation in biomass and in a smaller degree the N concentration. Sampo had the highest protein content (~34 %). Due to its small seeds, it could be that protein content was less diluted with other assimilates.

The late varieties produced a higher final yield than the early varieties. The seed-yield component analysis indicated that the bigger seeded varieties, which were the late maturing ones, had the higher yield. Since the early varieties had smaller seeds and smaller plants, it could be possible, that by changing plant density, a higher yield of the early varieties could be achieved as well. Vertigo performed best of all varieties. It had the highest seed yield and also the highest production of biomass.

9. References

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Appendix

Table 1A: ANOVA output. Testing effect of the DW yield from the yield component analysis in the third harvest. Significance: *, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$. (R Core Team, 2020)

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Replicate	1	1141	1141	1.325	0.3017
Variety	5	39900	7980	9.268	0.0144 *
Residuals	5	4305	861		

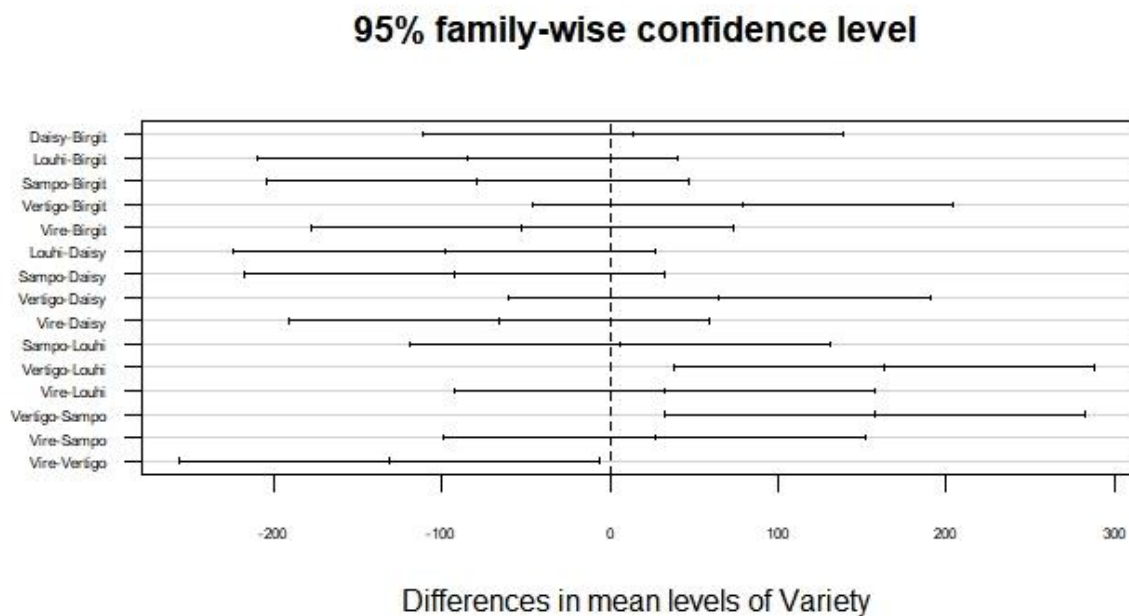


Figure A1: Means and confidence interval of varieties from the third harvest regarding the DW yield (g/cm^2). Lines that are completely under or above the zero mark, represents varieties that are significant different from each other. (R Core Team, 2020)

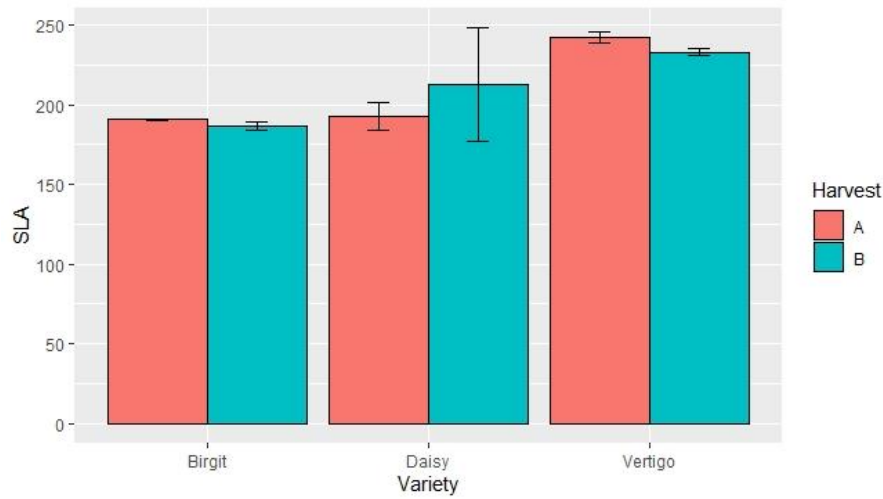


Fig. 2A: Difference in specific leaf area (SLA cm²/g) in the late varieties between first (A) and second (B) harvest. (R Core Team, 2020)

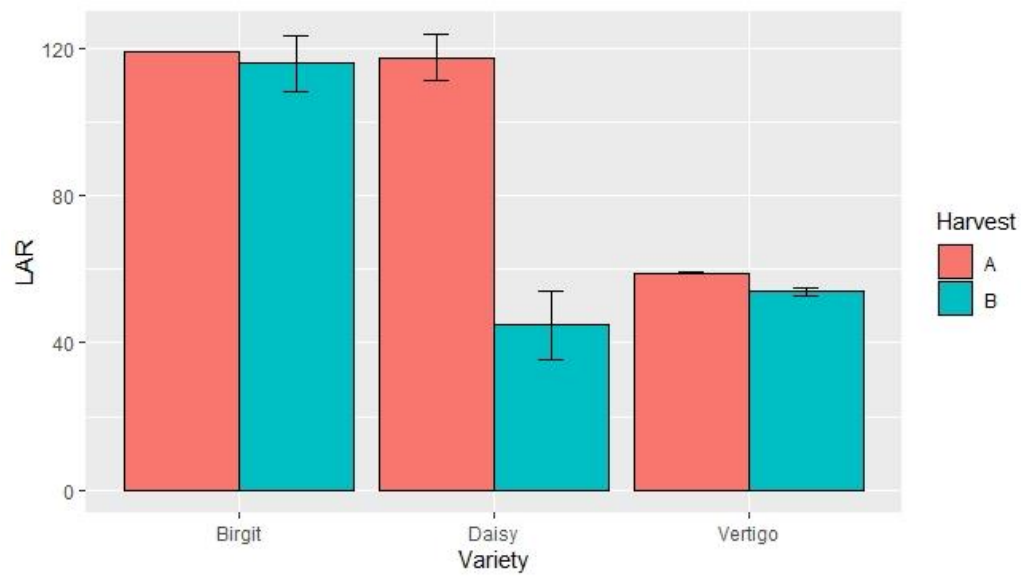


Fig. 3A: Difference in leaf area ratio (LAR cm²/g) in the late varieties between first (A) and second (B) harvest. (R Core Team, 2020)



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