MSc Thesis Report

Performance of two potato planting structures within a diverse cropping system in the Netherlands

Zewen Yang

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Supervisors: **Dr. Dirk van Apeldoorn** Farming System Ecology Group, WUR

Dr. Johannes Scholberg Farming System Ecology Group, WUR

Farming Systems Ecology Group

Wageningen University, Wageningen

The Netherlands

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Student name: Zewen Yang

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Supervisors: Dr. Dirk van Apeldoorn

Dr. Johannes Scholberg

Examiner: Dr. Egbert Lantinga

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Farming Systems Ecology Group,

Wageningen University, The Netherlands

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Preface

I was struggling for a long time to select an appropriate research topic before I started my current research in Farming Systems Ecology chair group at Wageningen University because I did not know what would be the optimum crop for me to study and which aspect would be a good research topic. Fortunately, I was introduced by Johannes Scholberg to Dirk van Apeldoorn and got fascinated by this project due to organic diverse crops for studying. After learning about this system, I immediately decided to study the organic potato production components within these systems because I found it is a very interesting and innovative research topic which focused on the use of potato variety mixtures.

This was the first year of the research program and a lot of knowledge was lacking. Thus, the aim of this study was to investigate the differences between the two planting structure systems and the effects of different structure systems on their system functioning within this organic diverse cropping system in the Netherlands. I was delighted in the process of this research because I felt I was so lucky to get involved in an international group of students in this research. I am very appreciating my supervisors, Dirk van Apeldoorn and Johannes Scholberg, as they taught me how to do a good research and how to be a positive thinker with their patience and professional expertise. In addition, I must say that I would not have finished my experiment without the help of the international student research group (Steffen Dahlke, Giuseppe Scandone, Shuang Xie and Hashmatullah Hotak) and Dine Volker's assistance; thank you all very much for your help and support. At the same time, thanks to Hennie Halm for helping me with the analysis of nutrient content in the lab and thanks also to other working staff in the physiology lab for their technique support. Last but not least, I would like to be grateful to my parents and friends, who are supportive and reassured through the whole process of my thesis research. From this research, I do not only gain knowledge but also friendships. I am grateful to all of these people, who were involved in this project.

As the initial motivation for me to come to Wageningen University, I hope I could contribute my effort and work to the development of organic agriculture and make our life better. By providing the data I collected and the conclusions I got, I wish it will be beneficial to the following year's research in this project and can contribute to the design and development of an innovative organic potato production system in the future.

Abstract

A study was carried out at the Droevendaal organic farm in Wageningen to investigate the differences of two planting structure systems (potato variety mixture and non-mixture) and the effects of each system on crop performance. Overall, potato variety mixture system performed better than potato variety non-mixture system. The potato variety mixture system presented a higher average plant height and higher canopy size, which resulted in a higher yield compared to potato variety non-mixture. Nitrate leaching was greater in potato variety non-mixture than in the potato variety mixture due to early haulm killing of potato variety non-mixture. However, the soil organic matter content in potato variety non-mixture was significantly higher than potato variety mixture. The insect population of the mixture system was larger and the number of insect species in mixture system were more diverse as well in a sharp contrast with potato variety non-mixture. In terms of late blight, we found that the potato variety mixture system had a significant lower *Phytophthora* infection ratio compared to potato variety non-mixture system.

In terms of spatial pattern within potato variety mixture system, the effect of rows on potato performance was significant regarding plant height, canopy size and tuber yield in the potato variety mixture system and this was probably due to the differences in soil compaction among rows. Effects of rows on soil nutrients and *Phytophthora* infection ratio were not consistent in this research. Blocks appear to have less impact on crop performance, but they did affect the *Phytophthora* infection ratio because the wind direction and dispersal within the field which affected the number of infected potato plants of each block differently.

Finally, there were significant differences among cultivars in the potato variety mixture system as well. Cultivar Connect showed the most dominate growth in the potato variety mixture system due to its larger canopy, higher plant, more root biomass and substantial higher yield. Canopy size and root depth distribution are the specific traits that should be paid attention in potato variety mixture breeding program due to their roles played in the plant growth period. Increasing cultivar combination ability and controlling initial seed growth vigour are important for the following years' research and they are also crucial for the production of crop variety mixture system.

Keywords: diversity, organic potato, potato variety mixtures, position effects, tuber yield, plant characteristics, soil nutrient, late blight

1. Introduction

1.1. Scientific background

During the last half century, conventional cash crop production relied on artificial fertiliser and pesticides to improve crop productivity (Davis *et al*., 2012). Intensified crop management involving improved germplasm (GMOs), excessive use of fertiliser, pesticides and fungicides, production of more than one time per year on the same piece of land and irrigation has increased overall yields per unit time and land in the last half century (Cassman, 1999). Intensive agricultural production systems have been characterised by a high degree of specialisation, narrow crop rotation, high external inputs of chemical fertilisers and biocides (Oomen et al., 1998). However, some of these agricultural activities caused a series of serious environment and soil problems such as eutrophication, soil degradation, environment pollution and food safety problems (Loehr, 1977). Therefore, one of the key challenges of the 21st century in agriculture is finding ways to balance crop productivity and environmental health (Davis et al., 2012). One of the promising ways is to increase biodiversity within cropping systems in time and space (i.e. intercropping, crop variety mixtures, crop rotation etc.). Biodiversity plays an important role in agroecosystems by providing ecological services, including recycling of nutrients, regulating local hydrological processes, suppressing undesirable organisms and detoxification of noxious chemicals (Altieri, 1999). Based on ecological and agronomic theory, one critical aspect to enhance biodiversity in agroecosystems is to intensify the mixing crops in space and time.

Mixed cropping refers to a cropping system in which more than one crop (or more than one variety) is cultivated simultaneously on a farming area in a cropping period, irrespective of their spatial arrangement (Francis, 1986; Jolayemi & Olaomi, 1995). Mixed cropping is one of the traditional farming practices, which was applied by farmers for centuries and it was also the first types of organized agriculture (Francis, 1986; Gliessman, 1985). The maize – beans – squash pattern which derived from Central America since ancient time is one of examples for mixed cropping and the combination of these three crops are so-called ‘milpas’ (Postma & Lynch, 2012). Crop variety mixtures also have been used to a much larger extent than is commonly assumed. Examples include winter wheat variety mixtures in Russia, barley-oat mixtures in German in 1980s, and rice variety mixtures in China (Mundt, 2002).

Currently, various patterns of mixed cropping such as intercropping and crop variety mixture can be found throughout the world and the most diversity is found in the tropics, especially in which small scale farmers operate intensively on a limited land area. However, mixed cropping system should not be only limited in intensive crop production on a small scale by farmers; it also has a great potential for large scale production systems in temperate climates. The potential advantages of mixed cropping systems could change the current situation and create new opportunities for future European rural development to contribute towards the increased sustainability and biodiversity of agriculture as well as preserving landscapes (Eichhorn et al., 2006).

The benefits of mixed cropping can be summarized by four advantages: first of all, an increasing crop yield and a relatively higher productivity is one of characteristics for mixed cropping system; secondly, mixed cropping systems can control pests and diseases much better than monoculture; and thirdly, the mixed cropping system has the potential to enhance ecological services to yield human well-being; last but not least, mixed cropping system also could maintain a greater economic profitability (Malézieux *et al.*, 2009). Therefore, mixed cropping system is worthwhile to be suggested for achieving sustainable productivity (Lenné & Smithson, 1994).

Due to limited research on mixed cropping system, a multitude of problems are needed to be solved for mixed cropping systems in order to be successful in the future. As pointed out by Malézieux (2009), species and variety diversity of ecological communities contribute to stability, but there is little data showing how those properties work within agro-ecological systems. Thus, further exploration on the effects of stability in multispecies-based agro ecosystems is required. One of the concepts for multispecies and multi-varieties is to increase the genetic diversity in order to control diseases and pests, but it really depends on cultivars or species combination ability. Therefore, designing good variety mixtures is needed and finding out optimum cultivar mixtures that will contribute to obtaining a higher yield. The mixture components should be relevant and functional rather than randomly choosing.

Potatoes are one of the most important crops in the world and are consumed everyday like other stable food crop such as rice, wheat, banana and cassava (Walker *et al.*, 1999). Most of these potatoes are cultivated in monocultures. In 2012, 365 million tons of potato was produced in the world (FAOSTAT, 2012). As an important staple food crop in the Netherlands, 15 million tons potatoes was produced in 2014 and with an average yield of 52 tons per hectare (CBS, 2014). Despite huge progress in the scientific understanding of potato productions, there remain two challenges of organic potato production: disease and nutrient management. Due to the climate in summer in the Netherlands (high relative humidity and mild temperatures), late blight (*Phytophthora infestans*) is a serious disease for organic potato production (Haverkort *et al.*, 2008). Generally, organic agriculture produces lower yield compared to conventional potato production (Varis *et al.*, 1996) because of limiting nutrient, disease and pest. To contribute to an improved understanding of these organic potato production problems, this research investigated system functioning of potato variety mixture systems.

1.2. Research objectives and questions

The aim of this study is to investigate if the cropping and planting structure systems affects the system functioning. More specifically, the effects of strip cropping and potato variety mixtures are investigated on potato productivity, plant growth, nutrient utilisation, insects and disease tolerance. These results are described in a discussion on the necessity of breeding for crop mixtures.

The research questions are based on three questions:

1) What are the differences between potato variety mixtures and non-mixture in an organic diverse cropping system on three theoretical productivity levels:

   A. Yield defining factors

   a. Plant characteristic and growth (i.e. plant height, canopy size, leaf chlorophyll content, fresh and dry weight of potato above-ground biomass, root depth distribution)

   B. Yield limiting factors

   b. Soil nutrient availability and plant nutrient uptake (i.e. soil mineral nitrogen, soil total nitrogen, and total nitrogen content of potato tuber)

   C. Yield reducing factors

   c. Pest (i.e. species and population)

   d. Disease (i.e. species, infected plants, with special reference to late blight)

2) Does the performance of the variety mixture system differ by location in the diverse cropping system?
3) What specific traits of potato should be paid attention for organic potato breeding in potato variety mixtures system?

1.3. Structure of this thesis

The first chapter provides the overall context, research questions and the outlines of thesis structure. In chapter 2, details on experimental design, crop management, crop characteristics of each cultivar, measurement method and statistical analysis are described. The effects of planting structures (potato variety mixture and potato variety non-mixture) and positions on crop performance are described in chapter 3. The results consist of four parts: potato productivity, yield defining factors, yield limited factors and yield reducing factors; they will be illustrated and discussed in this chapter. These are followed by a conclusion on these factors. In chapter 4, the effects of cultivars on crop performance are being discussed. Conclusions and recommendations are provided for the following years' research in terms of potato variety mixture system.

2. Materials and methods

2.1. Experimental site

The experimental site is located at the organic experimental farm of Droevendaal (51°59'33"N, 5°39'34"E), a facility of Wageningen University in Wageningen, The Netherlands. The mean annual air temperature and precipitation are 11°C and 829 mm, respectively. The soil type is classified as sandy soil. Droevendaal farm now manages 50 hectares of SKAL certified experimental fields for agriculture and horticulture with a wide crop rotation, including grass, clover, cereals and potatoes etc. Standard tillage has been used in the examined field for many years. The rotation sequence of this field in last few years were white clover, potato, triticale, wheat and cover crop mixture including rapeseed, rye and black radish in winter time. From 2007 to 2010, the grass-clover was sown in the field and after that, potato, triticale and wheat was cultivated. During the winter time, rapeseed, rye and black radish were cultivated in the field. In 2014, this field was planted with 3 m wide strips of grass-clover, potatoes, maize, wheat with fababean, wheat with lupine, sunflower with soybean, flower strip and mustard, which were treated as a diverse organic cropping system.

Fig.1 The layout of the experiment in the field

2.2. Experimental design and crop management

This thesis is part of the systems experiment “the more diverse, the better?” which studies the spatial and temporal effects of crop diversification in agro-ecological system. Within this experiment, multiple crops are studied, which consist of wheat, maize, potato and in combination with other crops, such as fababean, lupine sunflower and soybean. There were eight strips in the field, which included three blocks (24m x 80m) and the field was randomly block designed (Appendix I). Every block consisted of two treatments and four plots (3m x 20m): two crop variety mixtures and two crop variety non-mixtures. In addition, there were 5 meters long potato buffer fields on each side of a plot in order to avoid border effects, so the actual measurement zone was 3m x 10m large, which was used only for non-destructive measurements. Overall, the experiment included six replications for each treatment.

The basic plant arrangement of the potato crop can be described as follows: the distance between plants was 30 cm and there were approximately 33 plants in each row; the row spacing was 75 cm and each plot consisted of four rows. For potato variety non-mixture system, there was only one cultivar (Raja) sown in the field and four cultivars (Carolus, Connect, Raja, Sarpo Mira) were sown in the potato variety mixture system. The plant sequence of potato variety mixture system was Raja, Carolus, Connect and Sarpo Mira, but the initial plant in each row was different, which aimed to enlarge the distance of same cultivar when considering *Phytophthora* infection. Each cultivar's plant characteristics are described in Table1. Cultivar Carolus and Sarpo Mira are very resistant to *Phytophthora. Infestans* while Raja and Connect are less resistant to *Phytophthora. Infestans.* The weight of seed tuber of Connect was around 170 g while the weight of seed tuber of Raja, Carolus and Sarpo Mira was around 50 g. The tuber colour of Raja and Sarpo Mira is red while the tuber colour of Connect and Carolus is yellow, which make it easy to distinguish each cultivar during harvesting.

Table 1. Basic plant characteristics of four potato cultivars (Raja, Connect, Carolus and Sarpo Mira) and their late blight resistance level

Plant characteristics	**Characteristics of four potato cultivars**			
	Raja	**Connect**	**Carolus**	**Sarpo Mira**
Flower colour	Light purple	Unknown	Violet	Red violet
Maturity	Medium late	Medium late	Medium Early	Late
Skin colour	Red	Yellow	Multi-colour	Red
Fresh colour	Cream Yellow	Yellow	Yellow	White
Tuber shape	Oval	Round Oval	Oval	Long to oval
Tuber size	Medium	Large	Medium	Large
Phytophthora resistance-tuber	High	High	High	Very high
Phytophthora resistance-foliage	Low-medium	High	Very high	Very high

(Source: The European cultivated potato database, Agrico Cooperative, Den hartigh Potato)

Before sowing, solid manure and slurry were applied. A moldboard plough was used in the field. The sowing date of potato was on 20th of May and the potatoes were sown by hand for potato variety mixture system while potato variety non-mixture system was sown by a seeder machine. The seeder machine was driven by a tractor with GPS and potatoes were planted at a depth of 15 cm. During seeding period, three times re-ridging was applied at 2nd week after planting (WAP), 4th WAP and 5th WAP. The Phytophthora started at the end of 7th week after planting and the potato plants in the potato variety non-mixture were mowed at the beginning of 9th week after planting due to agricultural regulations. The harvest date for potato variety non-mixture and mixture was 11th August and 4th September, respectively.

2.3. Field and laboratory analysis

2.3.1. Yield defining factors

2.3.1.1. Plant height

Plant height (from soil surface to the top leaf) was measured at 5^{th}, 7^{th}, 9^{th}, $11^{th}$and 14^{th} week after planting. The central two rows and the edge two rows were all measured to compare the differences among four rows in terms of plant height. A random sampling method was applied by using R to select individual plant and group (four plants are treated as one group due to four cultivars) of each plot in experimental potato field. A total of 8 plants (2 groups) were randomly selected from the first row, central two rows and the fourth row, respectively.

2.3.1.2. Leaf chlorophyll index

Leaf chlorophyll index was measured by a SPAD meter (SPAD 502, Konica Minolta Sensing, Inc. Osaka, Japan) at 5, 7, 9, 11, 14 WAP as well. 3 fully expanded leaves from each plant were chosen to measure leaf chlorophyll index with SPAD meter. We were measuring the leaf chlorophyll index by using the same plants as measuring plant height. The chlorophyll content was expressed by chlorophyll content index (Chang & Robison, 2003).

2.3.1.3. Plant canopy size

Plant canopy size was measured at 7, 9, 11WAP. Samples were taken as the same as plant height. The plant canopy diameter were measured in both vertical and horizontal direction which implies one canopy diameter is parallel to the row and another one is perpendicular to the row (Tumbo *et al*., 2002). The canopy size was calculated by multiplying these two canopy diameters.

2.3.1.4. Fresh and dry weight of above-ground potato biomass

Fresh weight and dry weight of potato plant above-ground biomass were measured at 6, 10, 15 WAP (harvest period). All the samples were randomly selected in the buffer zone of the field by using R. 4 plants of each row were selected in order to exam the differences among four rows and two planting systems. Thus, 196 plants were measured each time for both two systems. Following Garnier *et al.*, (2001), the fresh weight of above-ground biomass including leaves and stems were measured by digital scales before oven-dried at 70°C for at least 2 days. Dry weight of above-ground potato biomass was measured after oven-drying by digital scales as well.

2.3.1.5. Specific gravity

Tuber specific gravity (SG) was calculated by the formula: SG= W air / (W air – W water); where W air is the fresh weight of tuber in the air and W water is the fresh weight of tuber in the water. Around 5 kg of washed tubers were collected and the fresh weigh of tubers in the water and in the air was measured by digital scale, respectively.

2.3.1.6. The root depth distribution

The potato plant rooting depth distribution was measured at 11 WAP. Each plant from four cultivars was sampled in each plot and in total, 48 plants root samples were taken. The root samples were taken from three soil layers and each soil layer was 15 cm depth, from 0-15cm, 15-30cm, and 30-45cm respectively. A hand auger with a 7cm diameter was used to take samples in vertical direction, which is close to the base of the main stem at 7.5cm. Afterwards, potato plant roots were washed from the soil with a fine sieve (0.2-2 mm) and all the organic debris was removed. After washing, we measured the fresh weight directly and dry weight was taken after oven-drying at 60 °C for 48h. Root depth distribution was expressed as dry root biomass per volume (g m^{-3}).

2.3.2. Yield limiting factors

2.3.2.1. Initial soil measurement

Before the initiation of the experiment, the total nitrogen content of soil and manure was estimated. The initial total nitrogen content in the potato field was estimated based on 90 soil samples measurements, with a depth of 0-30 cm for the whole field (See Fig.1).

2.3.2.2. Mineral nitrogen content of soil

Soil mineral nitrogen content for the 0-30cm soil layer was sampled at 6, 10, 15 (Harvest period) WAP. Within one plot, one sample from the first row, central two rows and the fourth row was taken, respectively. The measurement of soil mineral nitrogen content (soil available N-NO_3^- and N-NH_4^+) followed the 0.01 M $CaCl_2$ extraction method (Houba *et al.*, 2000). Samples were dried at 40°C for 48 hours before being extracted in 0.01m $CaCl_2$ at 20°C in a 1:10 (w/v) ratio and analysed by using a segmented-flow system (Auto-analyser II, Technicon).

2.3.2.3. Total nitrogen content of soil and potato tuber

Total nitrogen content of soil and potatoes (tubers) were measured at harvest time (15 WAP). We used the same soil samples from measuring soil mineral nitrogen content and the samples were taken from the first row, central two rows and the fourth row within one plot, respectively. All the soil samples were oven-dried at 40°C for 48 hours before lab analysis. The potato tuber samples were first washed, cutting into small parts and oven-drying at 70°C for 72 hours. After grinding through a 2 mm sieve in a grinding machine, the samples were analysed in the lab. Samples were first digested by a mixture of H_2SO_4 and salicylic acid under the influence of Se as a catalyst(Novozamsky *et al.*, 1983). The digestion was completed by adding concentrated H_2SO_4 at elevated temperature (330°C). Total nitrogen was measured spectrophotometrically with a segmented-flow system – Auto-analyser II, Technicon.

2.3.2.4. Total organic matter content of soil

Total organic matter content of the soil at harvest time (15 WAP) was measured by using the same sample with total nitrogen content. The organic matter in the soil was assessed gravimetrically by dry combustion of organic material in furnace at a temperature of 500-

550°C. The loss in the weight gave an indication of the content of organic matter in the sample(Heiri *et al.*, 2001).

2.3.3. Yield reducing factors

2.3.3.1. Pest

The pest population and species of each plot were measured in the two systems. Pitfall traps were put in the field to study whether there is a difference between two planting systems regarding pest population. Roofed pitfall traps were used. Each pitfall trap was filled with 100 ml preservative, which consists of propylene phenoxetol, propylene glycol and water in a ratio of 1:9:90. A pitfall trap (a diameter of 8.5 cm) with a roof (a diameter of 12.5 cm) was randomly installed in each plot and the location was selected by using R. All the pitfall traps were kept in the field for 48 hours and the collected insects were identified in laboratory by visual observation and microscope.

2.3.3.2. Phytophthora infection ratio

The population of infected plants by *Phytophthora* was measured by visual observation. The whole field was assessed and each four plants were treated as one group. Within one group, the number of infected plants was recorded on every other day in order to study the infection rate between the two different planting systems.

2.4. Statistical analysis

The results were analysed by using R. Shapiro-Wilk normality test and Anderson-Darling normality test were used to test normality of data distribution. Bartlett test and F-test were used to test for the homogeneity of variance. If the data was normally distributed and had equal variance, analysis of variances (ANOVA) and t-test were conducted; an analysis of variances was used for more than two treatments, while a t-test was used for the comparison of two treatments. If the data was neither normally distributed nor unequal variance, non-parametric test such as Wilcoxon Mann-Whitney test and Kruskal-Wallis test was used to assess differences. The Wilcoxon Mann-Whitney test is used for mean comparison of two treatments while Kruskal-Wallis test is used for more than two treatments comparison. The Fisher’s protected LSD-test was used for multiple comparisons in order to investigate whether there is a difference between every two treatments.

Finally, partial least square regression (PLSR) was used for the prediction of the potato yield in order to identify the important indicators for predicting potato yield based on the loadings between potato variety mixture and non-mixture system.

3. The effects of planting structures on potato performance

3.1. Results and Discussion

3.1.1. Potato productivity

The yield of potato variety mixture (3.09 kg m^{-2}) was more than two times higher than the yield of non-mixture system (1.36 kg m^{-2}) which is shown in Table 2 and the difference between the two systems was extremely significant. In terms of the yield of each tuber category, large tuber accounted for the highest yield (1.78 kg m^{-2}) in potato variety mixture, while the yield of medium tuber was the highest (0.81 kg m^{-2}) in potato variety non-mixture. Potato variety mixture system produced significantly higher yield than potato variety non-mixture in terms of large tuber, medium tuber and small tuber.

Table 2. Effects of planting structures (potato variety mixture and non-mixture) and position (block and row) on potato total yield and the yield of each tuber category

Factors	n [a]	Total yield (kg m^{-2})	n	Yield of each tuber category [b] (kg m^{-2})		
				Large	Medium	Small
Treatment						
Mixture	48	3.09	6	1.78	1.21	0.11
Non-mixture	48	1.36	6	0.49	0.81	0.07
P-value		<0.001		0.004	0.007	0.037
Block (Mixture [c])						
Block 1	16	2.86	4	1.56	1.20	0.10
Block 2	16	2.95	4	1.68	1.17	0.10
Block 3	16	3.46	4	2.09	1.25	0.12
P-value		0.187		0.670	0.910	0.164
Row (Mixture)						
Row 1	12	2.94 b [d]		-	-	-
Row 2	12	4.03 a		-	-	-
Row 3	12	1.96 c		-	-	-
Row 4	12	3.42 ab		-	-	-
P-value		<0.001				

[a] n refers to the number of samples

[b] Yield of each tuber category refers to Large (>40 mm), Medium (25-40mm) and Small (<25 mm).

[c] Samples were taken from potato variety mixture for studying the effect of location on yield.

[d] Different letters indicate significant differences according to Fisher's protected LSD-test (P<0.05).

[e] Each cultivar's tuber category was studied in the mixture system, thus the effect of rows was not studied here.

In terms of the influence of block on total yield, no significant differences were found among three blocks, but the data did show a higher yield in block 3 compared to block 1 and block 2.

Regarding each tuber category, there were no significant differences either. Thus, blocks have little effect on potato tuber yield in this study.

However, we found that there were large differences among the four rows. To be precise, row 2 produced the highest yield among the four rows, followed by row 4, row 1 and row 3. Moreover, row 2 (4.03 kg m^{-2}) even yielded more than two times higher potatoes than row 3 (1.96 kg m^{-2}). The difference between row 1 and row 4 was not significant and yields appear to be similar for both row 2 and row 4. It was found that rows has very pronounced effect on potato tuber yield in this study.

The significant difference of the total yield between potato variety mixture and non-mixture illustrated that different planting structures and cultivars can potentially improve productivity. Different proportion of each tuber category between potato variety mixture and non-mixture could be caused by incidence of *Phytophthora* because the haulm of potato variety non-mixture was cut at the beginning of 9^{th} week after planting, which was prior to the timing of haulm cutting in potato variety mixture system. Even though there was no significant block effect showing in terms of yield, a higher yield of block 3 could still be observed which was probably caused by *Phytophthora* due to the wind direction and the dominant wind direction was from west to east. The differences among rows were likely caused by soil compaction due to tractor trafficking and similar trend was found in neighbour strips as well.

It is clear that variety mixtures improved stability and decreased disease severity, which could result in higher yield compared to monoculture (Smithson & Lenne, 1996). A experiment was conducted in France for three years and researchers found a significantly increasing yield of the susceptible cultivar to late blight (Andrivon *et al.*, 2003). Hence, potato variety mixture has the potential to increase the yield compared with monoculture. However, soil properties could also have a great influence on potato tuber yield. A study on the effect of soil compaction on potato yield found that potato yield and quality were both affected by soil compaction; total yield was even reduced with more than 50% on two tested cultivars (Flocker *et al.*, 1960). The compacted soil could result in decreasing plant vigour which caused delayed plant emergence; and the lower specific gravity of potato tuber in compacted soil was caused by lower soil temperature near the soil surface (Blake *et al.*, 1960). Therefore, soil properties are an important factor causing potato yield and quality differences.

3.1.2. Yield defining factors

3.1.2.1. Plant height

The potato variety mixture system at 5^{th} week after planting showed significantly higher plant height compared to potato variety non-mixture system, but there were no differences at the 7^{th} week after planting between the two systems (Table 3.). During the plant growth period, the average plant height in potato variety mixture system kept a growing trend until the 11^{th} week after planting as shown in Figure 2. In addition, potato plant grew faster at the beginning compared to the growth speed between 9^{th} week after planting and 11^{th} week after planting. In general, the increase in plant height decreased over time which may be related to the shift from vegetative to reproductive growth.

Table 3. Effects of planting structures (potato variety mixture and non-mixture) and position (block and row) on potato plant height

Factors	n [a]	Plant height (cm)				
		5 WAP [b]	7 WAP	9 WAP	11 WAP	14 WAP
Treatment						
Mixture	144	12.7	35.9	49.8	55.0	42.2
Non-mixture [e]	(24/64)	15.9	34.4	-	-	-
P-value		0.013	0.649	-	-	-
Rows (Mixture [c])						
Row 1	48	11.2	39.8 a [d]	55.4 a	60.8 a	48.5
Row 2	16	14.0	42.4 a	59.3 a	62.3 a	57.8
Row 3	32	12.3	27.7 b	37.2 b	42.2 b	34.3
Row 4	48	14.1	35.3 ab	49.4 a	55.4 a	39.8
P-value		0.435	0.0164	<0.001	<0.001	0.135
Block (Mixture)						
Block 1	48	11.5	32.8	47.1	53.1	45.6
Block 2	48	15.2	34.1	49.1	53.7	45.8
Block 3	48	11.5	40.8	53.2	58.4	35.1
P-value		0.086	0.085	0.313	0.152	0.389

[a] n refers to the number of samples; the number in brackets means the number of samples in non-mixture.

[b] WAP = weeks after planting

[c] Samples were taken from potato variety mixture for the study of location effects on plant height.

[d] Different letters indicate significant differences according to Fisher's protected LSD-test (P<0.05).

[e] Potato plants height was measured until the second time due to *P.infestans* seriously happened at the beginning of 9th week after planting and all the plants had to be cut due to agricultural regulation.

Furthermore, we found that there were significant differences among four rows regarding plant height at the 7th, 9th and 11th week after planting; the third row presented a significant lower plant height compared to other three rows in potato variety mixture system. There were no differences among four rows regarding plant height during initial growth.

In terms of the influence of block on plant height, there were no differences among the three blocks during plant growth period. Thus, blocks had little influence on the plant height for potato variety mixture system.

The significant differences of plant height between potato variety mixture and non-mixture system at the beginning of plant growth could be caused by different seed potato vigour of the four cultivars. In fact, the seed potato vigour is determined by seed tuber size and the physiological age of seed potato (Struik & Wiersema, 1999). Because the tuber size of Connect was much larger than other three cultivars and Raja was pre-sprouted earlier than others when planting, all these factors could contribute to the significant differences of plant height. Obviously, the initial growth vigour of potato variety mixture was lower than the growth vigour of potato variety non-mixture through plant height. In addition, the differences among rows regarding plant height illustrated the different soil compaction; the compacted soil brought barriers to have a good development of root system, which further affected the plant height. Similar phenomenon was found in neighbour strips.

Other researchers reported significant differences in plant height among cultivars within years and plant height of earlier cultivars were in general shorter than later cultivars (Deblonde & Ledent, 2001). This finding also could help us to explain the significant difference of plant height because the maturity of four cultivars might differ as shown in Table 1.

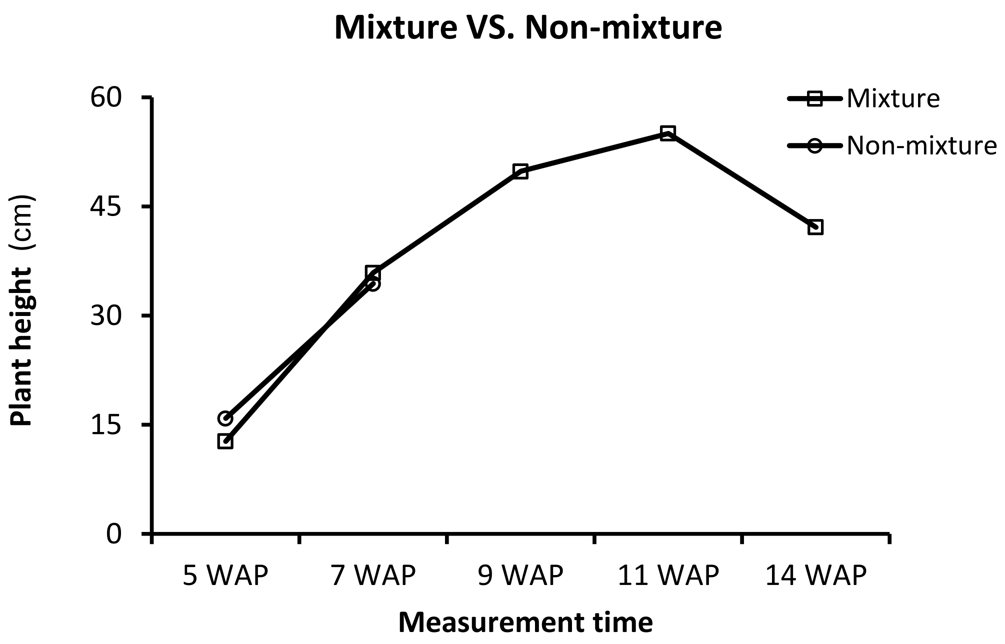

Fig.2. Effect of planting structures (potato variety mixture and non-mixture) on plant height during the potato production period; Plant height in non-mixture system was measured until the second time due to late blight onset.

3.1.2.2. Leaf chlorophyll index

The leaf chlorophyll index did not show a significant difference between potato variety mixture and non-mixture system at 5th week after planting. However, a significant higher leaf chlorophyll index of potato variety mixture was presented at 7th week after planting. In general, the leaf chlorophyll index was decreasing during the period of plant growth within potato variety mixture system. As we can see from Figure 3, the speed of changing leaf chlorophyll index was raising, which meant that the leaf chlorophyll index decreased more quickly at the end of growth period. Meanwhile, the initial leaf chlorophyll index of potato variety non-mixture fell faster than the one in potato variety mixture.

Table 4. Effects of planting structures (potato variety mixture and non-mixture) and position (block and row) on potato plant leaf chlorophyll index

Factors	n [a]	Leaf chlorophyll index				
		5 WAP	7 WAP	9 WAP	11 WAP	14 WAP
Treatment						
Mixture	144	44.4	42.6	36.2	30.8	20.3
Non-mixture [e]	(24/64)	47.5	34.1	-	-	-
P-value		0.298	0.003	-	-	-
Rows (Mixture [c])						
Row 1	36	44.7	41.8 ab [d]	35.5	31.8	20.3
Row 2	36	41.8	36.8 b	36.7	28.3	22.6
Row 3	36	43.9	44.1 a	35.7	29.5	19.4
Row 4	36	45.7	44.3 a	37.0	31.5	20.5
P-value		0.542	<0.001	0.409	0.703	0.957
Block (Mixture)						
Block 1	48	46.2 a	40.8	35.7	31.2	21.2
Block 2	48	48.9 a	45.4	37.2	31.0	21.9
Block 3	48	38.1 b	41.5	35.7	30.1	17.6
P-value		0.007	0.126	0.083	0.475	0.460

[a] n refers to the number of samples; the number in brackets means the number of samples in non-mixture.

[b] WAP = weeks after planting

[c] Samples were taken from potato variety mixture for studying the effect of location on leaf chlorophyll index.

[d] Different letters indicate significant differences according to Fisher's protected LSD-test (P<0.05).

[e] Potato plants height was measured until the second time due to *P.infestans* seriously happened at the beginning of 9th week after planting and all the plants had to be cut due to agricultural regulation.

To be precise, there were no significant differences in terms of leaf chlorophyll index among four rows as was shown in table 4. However, a significant difference was presented at the 7th week after planting and row 2 showed a significant lower leaf chlorophyll index compared to other three rows.

The influence of blocks on leaf chlorophyll index generally was not clear. However, at the first time measurement (5 WAP), leaf chlorophyll index of block 3 was significant lower than block 1 and block 2.

In general, the leaf chlorophyll index showed a downward trend during the period of growth and similar results could be found in a potato research by Canada potato research centre as well (Botha *et al.*, 2006). The difference of leaf chlorophyll index between potato variety mixture and non-mixture system at the 7th week after planting could be caused by late blight because late blight was first observed at the beginning of 7th week after planting. The late blight destroys the chlorophyll structure and therefore chlorophyll content would decrease (James, 1974). Cultivar Raja is less resistant to *Phytophthora* compared to the resistant cultivars (Carolus and Sarpo Mira) in potato variety mixtures; thus the different resistance to late blight on average in both systems may have caused different leaf chlorophyll index at 7th week after planting.

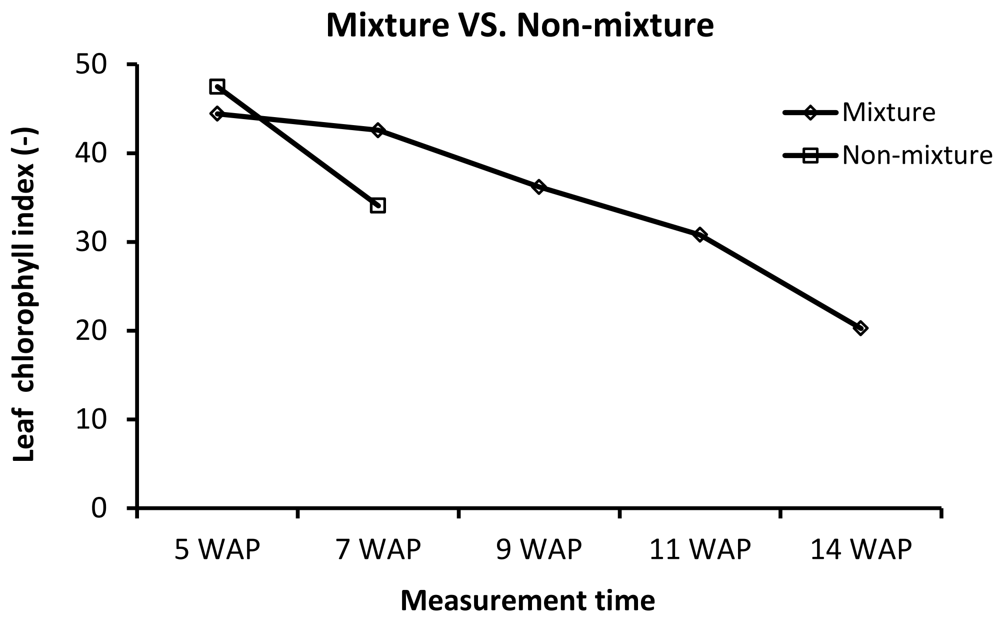

Fig.3. Effect of planting structures (potato variety mixture and non-mixture) on potato leaf chlorophyll index during the potato production period; Leaf chlorophyll index in non-mixture system was measured until the second time due to late blight onset.

3.1.2.3. Canopy size

Table 5. Effects of planting structures (potato variety mixture and non-mixture) and positions (block and row) on potato plant canopy size

Factors	n [a]	Canopy size (cm^2/plant)		
		7 WAP [b]	9 WAP	11 WAP
Treatment				
Mixture	144	1800	3018	3026
Non-mixture [e]	64	1307	-	-
P-value		0.037	-	-
Rows (Mixture [c])				
Row 1	36	1906	3019 ab [d]	2949
Row 2	36	2028	4375 a	3798
Row 3	36	1235	2142 b	2531
Row 4	36	1995	3150 ab	3176
P-value		0.052	0.017	0.252
Block (Mixture)				
Block 1	48	1480	2956	2920
Block 2	48	1626	2685	2776
Block 3	48	2294	3414	3382
P-value		0.051	0.631	0.838

[a] n refers to the number of samples; the number in brackets means the number of samples in non-mixture.
[b] WAP = weeks after planting
[c] Samples were taken from potato variety mixture for studying the effect of location on canopy size.
[d] Different letters indicate significant differences according to Fisher's protected LSD-test (*P*<0.05).

There was a significant difference on canopy size between potato variety mixture and non-mixture system at the 7th week after planting; the canopy size of potato variety mixture was significantly higher than potato variety non-mixture. To be precise, the canopy size was increasing during the entire plant growth period within potato variety mixture system; however, potato plant canopy grew much faster between 7th week after planting and 9th week after planting than canopy growth during 9th week after planting and 11th week after planting.

The differences of rows on canopy size within potato variety mixture system were significant at 7th week after planting and 9th week after planting. Row 2 presented a significant high canopy size among the four rows, followed by row 1, row 4 and row 3.

In terms of the influence of blocks on canopy size, we did not found significant differences among three blocks; but it was clear to see that blocks 3 showed a larger canopy size compared to block 1 and block 3.

Genotype (cultivar) was one of the factors causing the differences of canopy size between potato variety mixture and non-mixture system. Due to the large canopy size of Connect, the average canopy size of potato variety mixture was higher than canopy size of potato variety non-mixture. Another reason could be caused by *Phytophthora* because late blight started to destroy the field at the beginning of 7th week after planting. In addition, the larger seed tuber size also contributed to larger canopy size at the beginning of plant growth. The different canopy size among four rows illustrated that different soil compactions had influences on nutrient uptake, which was in turn affecting the canopy size. The larger canopy size of block 3 might be caused by *Phytophthora* because the wind direction was from west to east and the block 3 (at east side) got less possibility to be infected by *Phytophthora*. Canopy size is an important parameter which is corresponding to total yield and total dry matter yield of crops depends on the size of leaf canopy, the photosynthesis efficiency and the duration of plant growth (Tekalign & Hammes, 2005).

3.1.2.4. Fresh weight and dry weight of above-ground biomass

Both the fresh weight and dry weight of above-ground biomass of potato did not show a significant difference between potato variety mixture and non-mixtures at the 6th week after planting; however, the fresh weight and dry weight of above-ground biomass of potato variety mixture was higher than potato variety non-mixture. The fresh weight and dry weight of above-ground biomass increased between 6th week after planting and 10th week after planting while it decreased at 15th week after planting in potato variety non-mixture system (Table 6).

Table 6. Effects of planting structures (potato variety mixture and non-mixture) and positions (block and row) on fresh weight and dry weight of potato plant above-ground biomass

Factors	n[a]	Fresh weight			Dry weight		
		6 WAP[b]	10 WAP	15 WAP	6 WAP	10 WAP	15 WAP
Treatment							
Mixture	96	126.5	277.3	170.9	13.9	32.2	24.3
Non-mixture[d]	96	109.6	-	-	12.8	-	-
P-value		0.298	-	-	0.251	-	-
Rows (Mixture[c])							
Row 1	24	118.7	292.1	185.1	12.9	31.3	23.3
Row 2	24	135.3	288.3	194.7	14.7	31.0	28.9
Row 3	24	112.9	259.0	145.4	12.4	31.8	22.0
Row 4	24	141.4	279.3	165.7	15.6	34.5	25.5
P-value		0.895	0.912	0.428	0.887	0.930	0.667
Block (Mixture)							
Block 1	32	111.4	224.7	154.7	12.6	27.6	22.4
Block 2	32	108.5	276.9	155.7	12.8	33.2	21.1
Block 3	32	155.7	330.1	203.0	15.8	35.8	29.6
P-value		0.581	0.224	0.805	0.752	0.221	0.691

[a] n refers to the number of samples

[b] WAP = weeks after planting

[c] Samples were taken from potato variety mixture for studying the effect of location on fresh weight and dry weight of above-ground biomass.

[d] Fresh weight and dry weight of potato plant above-ground biomass was measured until the first time due to *P.infestans* seriously happened at the beginning of 9th week after planting and all the plants had to be cut due to agricultural regulation.

Rows did not show significant differences in terms of fresh weight and dry weight of above-ground biomass in the potato variety mixture system. But row 2 and row 4 had higher fresh weight and dry weight of above-ground biomass compared to other two rows.

Regarding the influence of block on the fresh weight and dry weight of above-ground biomass, there were no significant differences among three blocks in potato variety mixture system. However, block 3 showed higher fresh weight and dry weight of above-ground biomass during the three measurements compared to block 1 and block 2.

The reduction in fresh weight and dry weight of above-ground biomass in potato variety mixture system at 15th week after planting can be caused by *Phytophthora* because the cultivar was not resistant to *Phytophthora;* moreover, the potato plant had reached the tuber maturation stage and vines turned yellow with senescent leaves, which could have caused a reduction in fresh weight and dry weight of above-ground biomass (Robert B. Dwekk, 1993). Smaller canopy size in potato variety non-mixture system is likely to have resulted in reduced light interception and total canopy assimilation rates thereby resulted in lower fresh weight and dry weight of above-ground biomass (Tekalign & Hammes, 2005).

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Table 7. Effects of planting structures (potato variety mixture and non-mixture) and position (block and row) on soil mineral nitrogen (NO_3^- and NH_4^+) , total nitrogen and soil organic matter content

Factors	n[a]	NO_3^- (mg kg^{-1})			NH_4^+ (mg kg^{-1})			Total N %	OM %
		6 WAP[b]	10 WAP	15 WAP	6 WAP	10 WAP	15 WAP	15WAP	15 WAP
Treatment									
Mixture	36	13.92	1.15	0.49	4.97	7.01	6.92	0.16	4.23
Non-mixture	36	14.72	4.04	0.49	5.00	6.88	6.60	0.16	4.42
P-value		0.776	<0.001	0.775	0.704	0.849	0.311	0.874	0.009
Rows (Mixture[c])									
Row 1	12	13.47 a[d]	0.99	0.47	5.00	6.17	7.24	0.16	4.25
Row 2[e]	12	12.34 ab	1.56	0.42	5.31	7.16	6.77	0.16	4.24
Row 4	12	15.95 b	0.91	0.59	4.61	7.71	6.74	0.16	4.19
P-value		0.048	0.366	0.553	0.302	0.241	0.630	0.965	0.815
Blocks (Mixture)									
Block 1	12	14.25	0.99	0.52	4.76	6.44 ab	6.69	0.16	4.19
Block 2	12	14.36	1.39	0.60	4.84	8.38 a	7.40	0.17	4.18
Block 3	12	13.15	1.07	0.36	5.33	6.22 b	6.67	0.15	4.31
P-value		0.622	0.749	0.287	0.412	0.021	0.348	0.737	0.209

[a] n refers to the number of samples

[b] WAP = weeks after planting; OM= organic matter

[c] Samples were taken from potato variety mixture for the study of position effects on soil nutrient.

[d] Different letters indicate significant differences according to Fisher's protected LSD-test (P<0.05).

[e] Soil nutrient measurements took two rows at edges and central two rows as row 1, row 4 and row 2, respectively.

3.1.3. Yield limiting factors

3.1.3.1. Soil nutrient

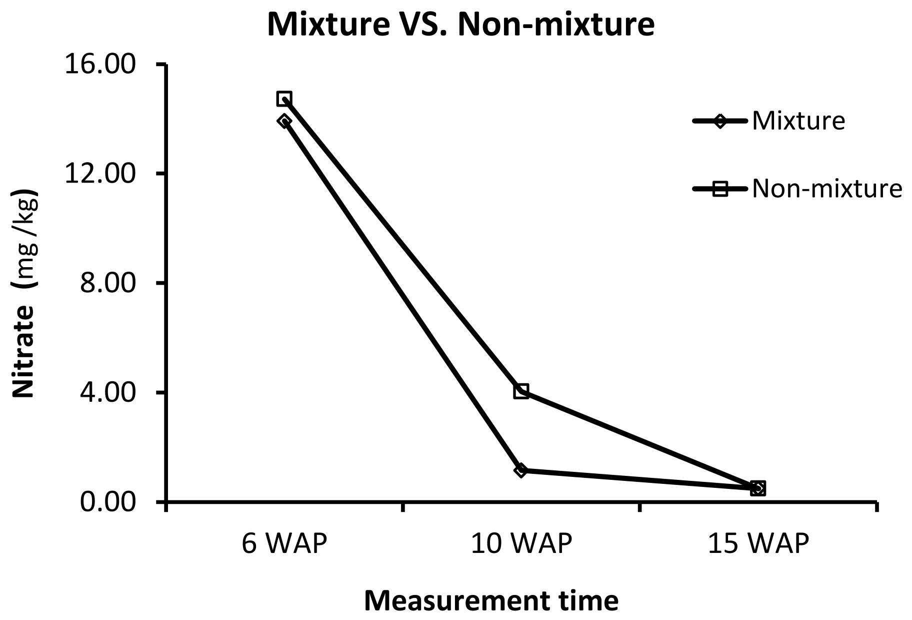

Fig.4. Effect of planting structures (potato variety mixture and non-mixture) on soil nitrate content (NO_3^-) during the potato production period.

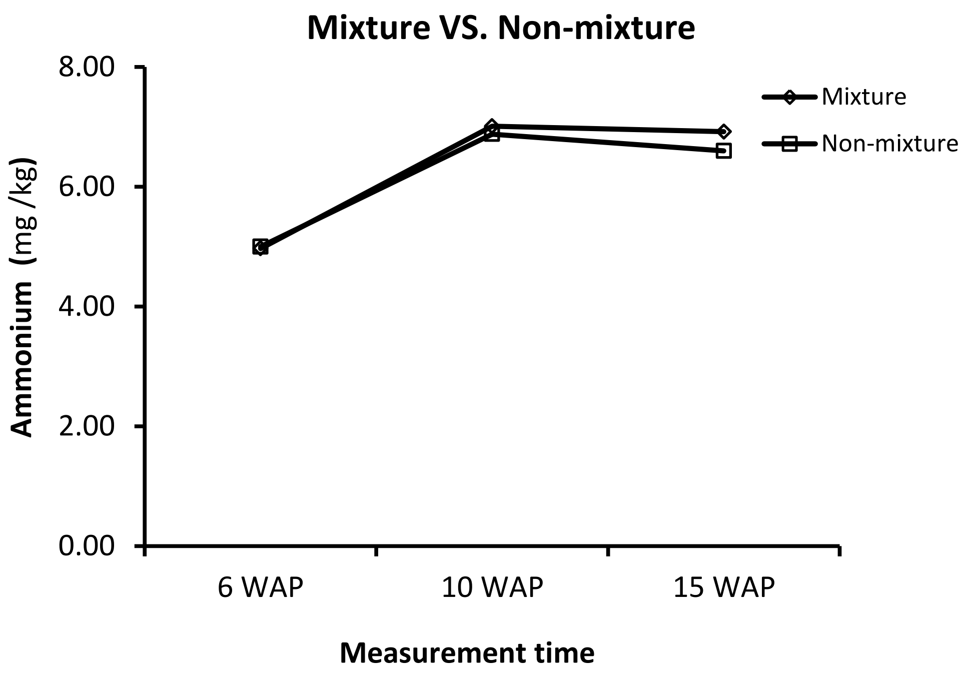

Fig.5. Effect of planting structures (potato variety mixture and non-mixture) on soil ammonium content (NH_4^+) during the potato production period.

Nitrate and ammonium had different during the process of potato plant growth (Table 7). Regarding nitrate (NO_3^-), there was a significant difference at the 10th week after planting between potato variety mixture and non-mixture; the soil nitrate content in potato variety non-mixture (4.04 mg kg^{-1}) was more than three times higher than the one in potato variety mixture (1.15 mg kg^{-1}). The organic matter content in potato variety non-mixture (4.42%) was also significantly higher than the one in potato variety mixture (4.23%) as well. There were no large differences in terms of soil ammonium content and total soil nitrogen content between the two systems. The soil nitrate content showed a downward trend generally during the plant growth period while the soil ammonium content followed an upward trend.

Overall, there were no significant differences in terms of soil nitrate content and soil ammonium content among middle and edge rows except the first measurement of soil nitrate content. It is shown that the soil nitrate content of row 4 (15.95 mg kg^{-1}) was significantly higher than row 1(13.47 mg kg^{-1}) at the 6th week after planting and the differences among rows regarding soil nitrate content did present at the beginning of potato plant growth.

The influence of block on soil nutrient content was not obvious as well. Generally, there was no significant difference among three blocks regarding soil nitrate content and soil ammonium content but the second measurement of soil ammonium content. It is manifest from the Table 7 that the soil ammonium content of block 2 (8.38 mg kg^{-1}) was much higher than block 3(6.22 mg kg^{-1}).

During the peak growth period, tubers started to bulk and plants thus need more nutrients. But the potato plant in potato variety non-mixture was already cut due to severe incidence of late blight at the 9th week after planting and the three cultivars: Carolus, Connect and Sarpo Mira were in tuber bulking stage, which could result in significant differences regarding soil nitrate content between potato variety mixture and non-mixture. In addition, plants have a higher nitrogen efficiency by using nitrate compared to ammonium form (Legaz *et al.*, 1996; Sonneveld & Voogt, 2009) and nitrate usually dominates over ammonium in soils in terms of plant nutrient (Booij *et al.*, 2000). However, nitrate is easily leached compared to ammonium and this could explain why soil nitrate content was much lower than soil ammonium content. The relative lower soil ammonium content at the beginning was related to temperature to some extent because higher temperatures can result in a higher ammonification process (Myers, 1975). Overall, there was more nitrate leaching from the potato variety non-mixture compared to potato variety mixture. The influences of rows on nutrient was not clear owing to sampling design because a difference between central rows and edge rows was expected and we did not measure each row separately. The rows effect may be explored in the following year within this project.

Table 8. Effects of planting structures (potato variety mixture and non-mixture) and position (block and row) on Phytophthora infection ratio

Factors	n [a]	Phytophthora infection ratio [b] (%)						
		7th of July	10th of July	11th of July	14th of July	16th of July	21st of July	23rd of July
Treatment								
Mixture	192	7	9	16	24	29	35	46
Non-mixture [c]	192	20	27	51	80	-	-	-
P-value		<0.001	<0.001	<0.001	<0.001	-	-	-
Rows (Mixture [d])								
Row 1	48	5	5	11	20	26	30	45
Row 2	48	8	11	20	25	34	41	47
Row 3	48	7	9	14	21	28	33	42
Row4	48	8	11	21	30	32	36	50
P-value		0.453	0.163	0.015	0.063	0.232	0.098	0.124
Block (Mixture)								
Block 1	64	8	10	19	26	31	35	42
Block 2	64	9	12	20	25	30	37	46
Block 3	64	4	5	11	21	26	33	50
P-value		0.103	0.092	0.012	0.436	0.395	0.704	0.304

[a] n refers to the number of samples

[b] Phytophthora infection ratio refers to the number of plants get infected divided potato plants rather than infection intensity

[c] Potato plants got infected by *P.infestans* seriously at the beginning of 9th week after planting and all the plants had to be cut due to agricultural regulations.

[d] Samples were taken from potato variety mixture for the study of location effects on *Phytophthora* infection ratio.

Table 9. Effects of planting structures (potato variety mixture and non-mixture) on the species and population of insects

	Insect species and its population in the field [a]									
	Total number	***Araneae***	***Opiliones***	***Ocypus***	***Tachyporus***	***Cetepede***	***Carabidae larvae***	***Harpalus rufipes***	***Amara aenea***	***Colorado bettles***
Treatment										
Mixture	24	17	1	0	1	1	1	1	0	2
Non-mixture	12	6	0	2	0	0	0	2	1	1

[a] The measurement of insect species and its population was at 10th week (23rd July-25th July) and the population of insects was the total number of the treatment.

3.1.4. Yield reducing factors

3.1.4.1. Phytophthora infestans

The *Phytophthora* infection ratio in this research refers to the ratio of infected plants divided by total number of potato plants (Kranz, 1988). As can be seen from the Table 8, there were significant differences between potato variety mixture and non-mixture regarding *Phytophthora* infection ratio. In general, the *Phytophthora* infection ratio in potato variety mixture was much lower than the one in non-mixture in the first four measurements. Before haulm killing of the potato variety non-mixture, the *Phytophthora* infection ratio in potato variety non-mixture (80%) was already more than three times higher than the one in potato variety mixture (20%). Fig. 6 also illustrated that there was a lower spreading speed of potato variety mixture compared to potato variety non-mixture. In addition, the spreading speed of potato variety non-mixture increased after the second measurement.

In general, the effect of rows on the *Phytophthora* infection ratio was not apparent and only the measurement at the third time showed a significant difference among four rows; the *Phytophthora* infection ratio of row 4 (21%) and row 2 (20%) were much higher than the ones of row 1(11%) and row 3 (14%). The same trend can be observed as well during other growth periods.

Blocks generally had limited influences on *Phytophthora* infection ratio but the third time measurement did show a significant difference among three blocks; the *Phytophthora* infection ratio of block 3 (11%) was much lower than the *Phytophthora* infection ratio of block 1 (19%) and block 2 (20%).

The significantly different *Phytophthora* infection ratio between potato variety mixture and non-mixture is in line with the finding by Andrivon et.al (2003) who reported that potato cultivar mixtures reduce disease progress rates and it even delayed disease onset compared to non-mixture system. Pilet et.al (2006) also found that the area under the disease progress curve on susceptible cultivar to late blight was 0 to 20 % less in mixed than in pure plots when no fungicide was applied. Therefore, use of potato variety mixture system may provide a viable alternative to decrease late blight infection to the exclusive use of resistant cultivars (Munk, 2006; Phillips *et al.*, 2005) .

It may be argued that due to the smaller canopy size and smaller plant height of row 1 and row 3, the likeliness of being exposed to *Phytophthora infestans* decreased compared to the other two rows, which could result in a higher *Phytophthora* infection ratio. Late blight dispersed through wind easily and it can be controlled by moisture levels (Henfling, 1987). Furthermore, the wind direction also could cause differences among three blocks regarding *Phytophthora* infection ratio; the wind from west to east reduced the density of *Phytophthora infestans* in the east and block 3 was in the most east part, where showed a lower *Phytophthora* infection ratio.

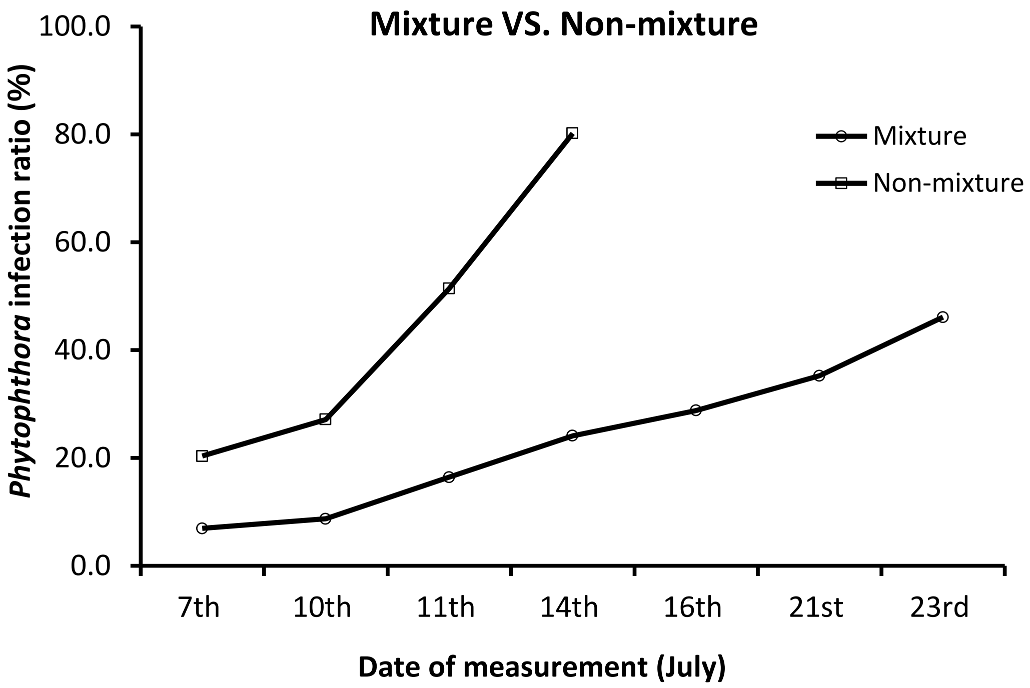

Fig.6. Effect of planting structures (potato variety mixture and non-mixture) on *Phytophthora* infection ratio during the potato production period

3.1.4.2. Pest species and population

The total insects' population in potato variety mixture was two times as large as the one in potato variety non-mixture (Table 9). To be specific, the number of spiders (Araneae) in mixtures system was much higher than non-mixture system. Research showed that spider and carabids can reduce prey population (Greenstone, 1999; Sunderland, 1999). In terms of insects' species, potato variety mixture system had more insect species than non-mixture system as well, which indicated higher insect diversity in potato variety mixture system.

Diversification of resistance is an ecological approach to limit plant diseases and pest expansion through functional diversity (Finckh & Lammerts van Bueren, 2007). Host species may provide a refuge for predators of the pest normally occurring on a second host species (Wolfe, 1985) and this could also happen on cultivars. It may explain why there were more predators in potato variety mixture system.

3.1.5. Partial least square regression analysis

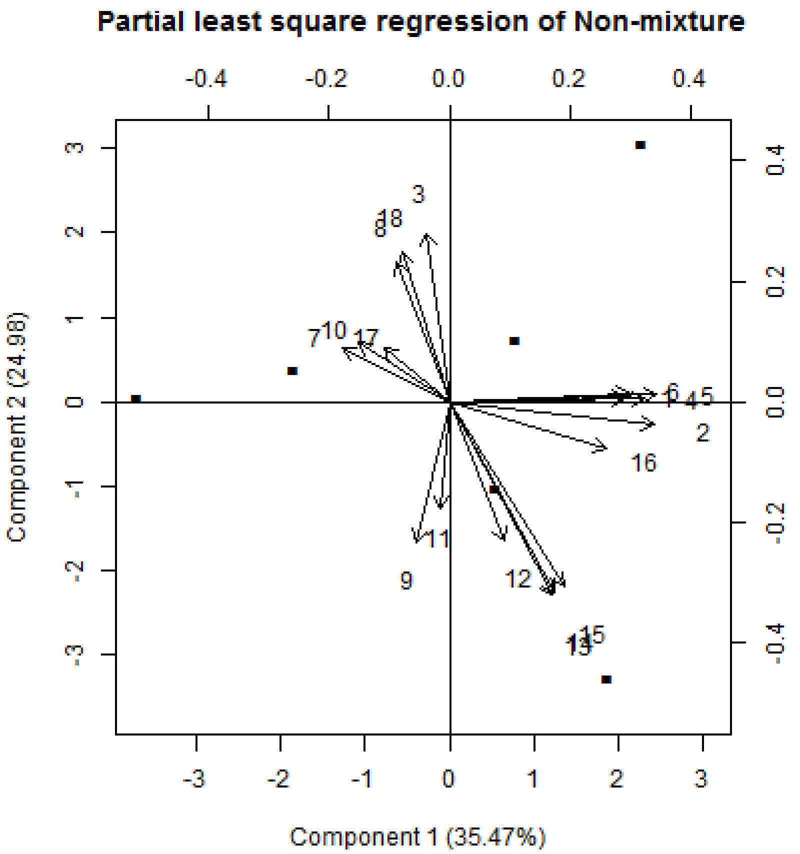

Fig.8. Bi-plot of the partial least square regression analysis of potato variety non-mixture system (X and Y axis represented component 1 and component 2; the loading was shown by coordinate and the arrows represented all the variables, which refer to 1-plant height (1st), 2-plant height (2nd), 3-leaf chlorophyll index (1st), 4-leaf chlorophyll index (2nd), 5-canopy size (1st) ,6-total insect population, 7-NO_3^- (1st) , 8- NO_3^- (2nd), 9- NO_3^- (3rd), 10-NH_4^+ (1st), 11- NH_4^+ (2nd), 12- NH_4^+ (3rd), 13-*Phytophthora* infection ratio (1st), 14-*Phytophthora* infection ratio (2nd), 15- *Phytophthora* infection ratio (3rd), 16-*Phytophthora* infection ratio (4th), 17-total soil nitrogen content, 18- total soil organic matter content)

Yield prediction formula of potato variety non-mixture system

Y_m=9.95a_1+9.64a_2+6.53a_3+9.87a_4+10.96a_5+11.21a_6-2.11a_7+2.71a_8-6.06a_9-3.60a_{10}-4.27a_{11}-2.45a_{12}-4.32a_{13}-4.13a_{14}-3.13a_{15}+7.17a_{16}+0.02a_{17}+4.99a_{18}

In the potato variety non-mixture system as shown in Figure 7, the first component is dominated by plant height (1st, 2nd), leaf chlorophyll index (2nd), canopy size (1st) and total insects population; the second component are made of leaf chlorophyll index (1st), *Phytophthora* infection ratio (1st, 2nd, 3rd) and total soil organic matter. The plant height, leaf chlorophyll index, canopy size and total soil nutrient content all have a positive effect on total yield while *Phytophthora* infection ratio has a negative influence on total potato yield. In comparison with potato variety mixture system, partial least square regression in potato variety non-mixture seems is more “tractable”. For instance, *Phytophthora* infection ratio was negatively correlated to total potato yield which means late blight is an important indicator for total yield. Furthermore, plant characteristics such as plant height, leaf chlorophyll content ,canopy size and total nutrient content of soil, such as total soil nitrogen content and soil organic matter content were also important indicators for predicting yield.

In general, the yield of potato variety mixture system is dominated by cultivars (genotypes) to a large extent, which are expressed by different plant characteristics and it reduces the risk of losing yield if late blight happens due to the existence of resistant cultivars of late blight. While, the yield of potato variety non-mixture system largely depends on environmental factors such as soil nutrient and the possibility of late blight onset because the only cultivar intrinsically has poor resistance to late blight.

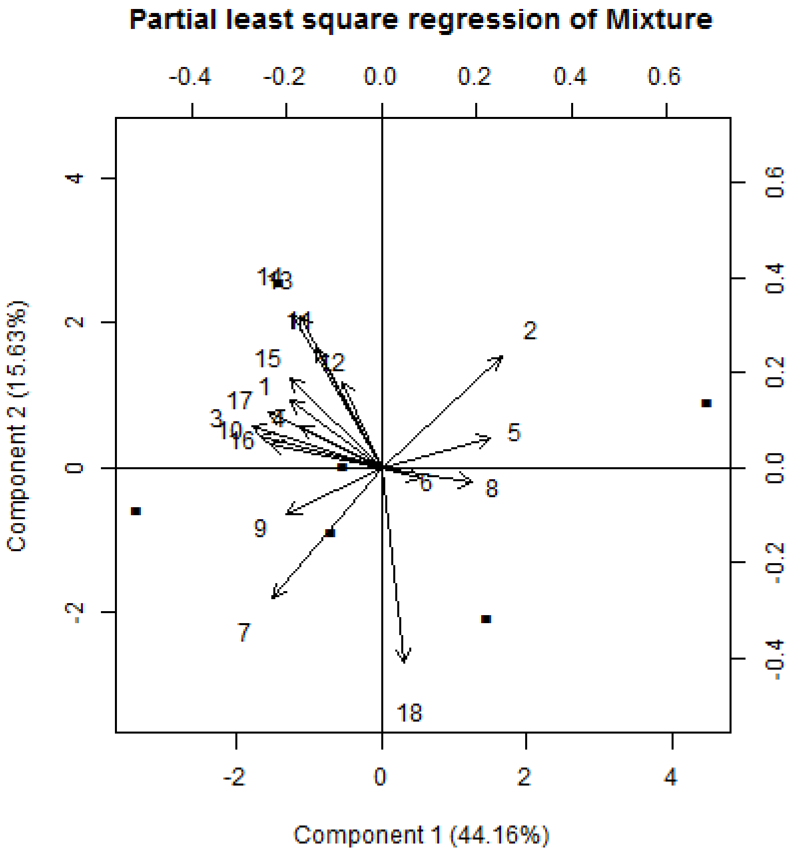

Fig.7. Bi-plot of the partial least square regression analysis of potato variety mixture system (X and Y axis represented component 1 and component 2; the loading was shown by coordinate and the arrows represented all the variables, which refer to 1-plant height (1st), 2-plant height (2nd), 3-leaf chlorophyll index (1st), 4-leaf chlorophyll index (2nd), 5-canopy size (1st) ,6-total insect population, 7-NO_3^- (1st) , 8-NO_3^- (2nd), 9- NO_3^- (3rd), 10-NH_4^+ (1st), 11- NH_4^+ (2nd), 12- NH_4^+ (3rd), 13-*Phytophthora* infection ratio (1st), 14-*Phytophthora* infection ratio (2nd), 15- *Phytophthora* infection ratio (3rd), 16- *Phytophthora* infection ratio (4th), 17-total soil nitrogen content, 18- total soil organic matter content)

Yield prediction formula of potato variety mixture system

Y_m=2.51a_1+30.20a_2-15.11a_3+2.44a_4+18.32a_5+6.73a_6-31.18a_7+12.80a_8-14.35a_9-8.17a_{10}+0.74 a_{11}+2.21a_{12}+4.11a_{13}+4.91a_{14}+0.19a_{15}-11.85a_{16}-5.90a_{17}-20.77a_{18}

In this research, a partial least square regression analysis was conducted in order to figure out what are the main factors determining yield for both planting structures – potato variety mixture and non-mixture. As we can see from the Fig 6, in the potato variety mixture system, the first component consists of plant height (2^{nd}), leaf chlorophyll index (1^{st}), canopy size (1^{st}), soil nitrate content (1^{st}) and total soil nitrogen content; the second component constitutes *Phytophthora* infection ratio (1^{st}, 2^{nd}, 3^{rd}) and total soil organic matter content. Plant height and canopy size both have a positive effect on total potato yield while the leaf chlorophyll index, soil nitrate content (1^{st}), total soil nitrogen content and total soil organic matter content have a negative effect on the yield. The negative effect means the larger of the loading, the lower of the yield is. This counter-intuitive result is caused by mixed stand. For example, the higher of leaf chlorophyll content, the lower yield would be gained because Connect accounted for the largest yield among four cultivars but with lower chlorophyll content compared to Raja and Sarpo Mira, which illustrated that leaf chlorophyll content is not important for cultivar Connect (see chapter 4). Similarly, *Phytophthora* infection ratio has a positive effect on yield which means the more late blight happened, the "more" yield gained and this contradiction may be explained by the fact that the yield in potato variety mixture system were not mainly determined by *Phytophthora* but cultivar because late blight did not damage Connect seriously while it produced the highest yield among four cultivars. Therefore, the plant characteristics such as plant height and canopy size in potato variety mixtures are the dominant indicators for predicting yield rather than *Phytophthora* infection ratio. However, identical relations were found in potato variety non-mixture system, which is discussed in the following paragraph.

3.2. Conclusions

The objective of this research was to investigate if different potato planting structure systems have an influence on system functioning and to explore what are the differences between potato variety mixture and non-mixture within a diverse organic cropping system in the Netherlands. In terms of plant productivity, yield defining factors, yield limiting factors and yield reducing factors, the following conclusions were got.

There was a significant yield difference between potato variety mixture and non-mixture system and the difference of each tuber category was also significant between the two systems. The system yield gap was mainly dominated by highly productive cultivar – Connect rather than by other factors. However, significant yield differences by rows were demonstrated within potato variety mixture and the possibility of this phenomenon could be caused by different soil compaction of each row due to wheel traffic of tractors. Blocks had no significant influences on yield.

Due to the significant yield gap between potato variety mixture and non-mixture, the yield defining factors also presented significant differences. To be more specific, plant height showed significant differences only at the beginning between potato variety mixture and non-mixture. Plant height differed by rows significantly at peak growth period as well. In addition, leaf chlorophyll index at initial growth period did not present differences but the two planting systems had different leaf chlorophyll index significantly at the second time measurement because of late blight. The effect of location (blocks and rows) on leaf chlorophyll content was not significant. Furthermore, a significant difference between the two systems on canopy size was found; at the same time, differences among rows on canopy size were significant at plant peak growth period. In terms of fresh weight and dry weight of above-ground biomass, we found there was no difference at the beginning of plant growth period for both systems and no difference was observed by different rows and blocks as well within potato variety mixture.

Soil nutrients generally did not show significant differences between potato variety mixture and non-mixture, but there was a significant yield gap between the two systems, which means potato variety mixture system has the potential to reach higher nutrient uptake. In other words, there was more nutrient losing in potato variety non-mixture system compared with potato variety mixture.

Regarding pest species and population, potato variety mixture presented higher diversity and larger population than potato variety non-mixture. Another important parameter – *Phytophthora* infection ratio had a large difference between the two planting systems. Potato variety mixture was beneficial to reduce *Phytophthora* infection ratio and decreased the risk of disease onset compared to potato variety non-mixture system. Meanwhile, I found that whether decreased *Phytophthora* infection ratio could result in higher yield depends on other cultivar characteristics and other environmental factors within mixture system because highly productive cultivar could be in a dominant position and be competitive, which could make other cultivar less productive than monoculture.

Potato plant height and canopy size are treated as important indicators to predict potato yield in terms of plant characteristics. *Phytophthora* infection ratio can be useful for predicting yield in potato variety non-mixture rather than mixture on current research, because other factors tend to govern total yield to a larger extend in the complex potato variety mixture system.

4. The effects of cultivars on potato performance

4.1. Results and discussion

4.1.1. Potato productivity and quality

Within the four potato cultivars, Connect produced highest (2.27 kg m^{-2}) total yield and it is significantly higher than other three cultivar based on the multiple comparison which is shown in table 10. In addition, there were no significant differences among Raja (0.33 kg m^{-2}), Carolus (0.27 kg m^{-2}) and Sarpo Mira (0.23 kg m^{-2}) regarding total yield.

Table 10. Effects of cultivars on total potato yield and each potato tuber category

	n [a]	Yield contribution (kg m^{-2})	n	Yield per tuber category [b] (kg m^{-2})		
				Large	Medium	Small
Cultivar [c]						
Carolus	48	0.27 b [d]	6	0.16 b	0.10 b	0.01
Connect	48	2.27 a	6	2.02 a	0.24 a	0.01
Raja	48	0.33 b	6	0.17 b	0.15 b	0.01
Sarpo Mira	48	0.23 b	6	0.08 b	0.13 b	0.02
P-value		<0.001		<0.001	0.002	0.09
Raja (Mixture)	48	0.33	6	0.17	0.15	0.01
Raja (Non-mixture)	48	0.34	6	0.12	0.20	0.02
P-value		0.553		0.127	0.181	0.158

[a] n refers to the number of samples

[b] Yield of each tuber category refers to Large (>40 mm), Medium (25-40mm) and Small (<25 mm).

[c] Samples were taken from potato variety mixture for the study of cultivar differences on yield.

[d] Different letters indicate significant differences according to Fisher's protected LSD-test (*P*<0.05).

In terms of large-sized and medium-size potato tubers, the yield of Connect was still significantly higher than Raja, Carolus and Sarpo Mira while there were no significant differences on small tuber among these four cultivars. In addition, the proportion of each tuber size on different cultivars was also different and Connect had the highest percentage of large tuber compared to other three cultivars.

Raja from potato variety mixture and Raja from potato variety non-mixture produced similar total yield of potato tubers; however, different results can be seen in each tuber category. Precisely, the proportion of large size tuber in potato variety mixture (50%) was higher than the one in potato variety non-mixture (36%), which means potato variety mixture (0.17 kg m^{-2}) produced more large tubers than potato variety non-mixture (0.12 kg m^{-2}).

On one hand, the higher yield of Connect is determined by its genotype, which means Connect itself is a productive cultivar. On the other hand, the external factors could also have influence on the total yield, such as the seed tuber size. The seed tuber size of Connect was much larger than other three cultivars when planted and larger seed tuber size means large surface with more sprouts, which resulted in more stems and higher growth vigour at the beginning (Struik & Wiersema, 1999). The more stems and higher growth vigour could be beneficial to a higher yield eventually. In addition, the competition among cultivars could also cause the lower yield and higher yield. Therefore, interactions among cultivars should be paid attention when staring a potato variety mixture system.

Table 11. Effects of cultivars on potato quality

	DM [a] (%)	SG [b]	Yield (kg m^{-2})	Total N (%)	Total C (%)	N uptake (g m^{-2})
Cultivar [c]						
Carolus	24.0 a [d]	1.09 a	0.27 b	1.1	43.5	11.2
Connect	22.8 b	1.08 a	2.28 a	0.8	41.6	73.7
Raja	20.4 c	1.06b	0.33 b	1.4	42.7	14.2
Sarpo Mira	24.7 a	1.09 a	0.23 b	1.0	43.3	8.9
P-value	<0.001	0.005	<0.001	-	-	-

[a] DM= Dry matter content; SG= Specific gravity

[b] Different letters indicate significant differences according to Fisher's protected LSD-test (P<0.05).

[c] Samples were taken from potato variety mixture for the study of cultivar differences of potato quality.

[d] Different letters indicate significant differences according to Fisher's protected LSD-test (P<0.05).

It is surprising to note that there is no significant difference on the yield of the same cultivar in potato variety mixture and non-mixture especially when Raja in potato variety non-mixture was cut early than the Raja in potato variety mixture. Because Raja is not resistant to late blight and it was not productive in this research compared to Connect, the yield difference was not significant at the end. We could assume that Raja in potato variety mixture stopped to grow when the Raja in potato variety non-mixture was cut. However, a research conducted in France in 1993, 1997 and 1998 found late blight severity was significantly lower in a susceptible cultivar growing in rows in potato variety mixture than in non-mixture systems and significant yield also increased for the susceptible cultivar (Andrivon et al., 2003). Thus, whether the yield of susceptible cultivar increased in potato variety mixture may also be related to competitive relations among the different cultivars.

Considering the effects of cultivars on tuber quality, we found there were significant differences on dry matter content of tubers and specific gravity among four cultivars. Sarpo Mira and Carolus have higher dry matter content, followed by Connect and Raja. The specific gravity of Raja was significantly lower than other three cultivar, which is correlated to tuber dry matter content (Wilson & Lindsay, 1969); the specific gravity of potato tubers was also affect by tuber size, tuber dryness, dirt and debris when measuring (Shetty, 2013). Due to different yield and total nitrogen content of the tuber, the total nitrogen uptake showed a greater difference; Connect took up much more nitrogen than Carolus, Raja and Sarpo Mira.

4.1.2. Yield defining factors

4.1.2.1. Plant height

Table 11. Effects of cultivars on plant height

	n [a]	Plant height (cm)				
		5 WAP [b]	7 WAP	9 WAP	11 WAP	14 WAP
Cultivar [d]						
Carolus	36	7.6 c [c]	28.8 b	46.4 b	52.5 b	51.9 b
Connect	36	21.0 a	55.3 a	66.5 a	68.4 a	73.1a
Raja	36	13.3 b	32.6 b	44.8 b	48.4b	-
Sarpo Mira	36	9.0 c	26.9 b	41.7 b	50.8 b	43.6 b
P-value		<0.001	<0.001	<0.001	<0.001	<0.001

[a] n refers to the number of samples

[b] WAP = weeks after planting

[c] Different letters indicate significant differences according to Fisher's protected LSD-test (P<0.05).

[d] Samples were taken from potato variety mixture for studying the differences among cultivars on plant height.

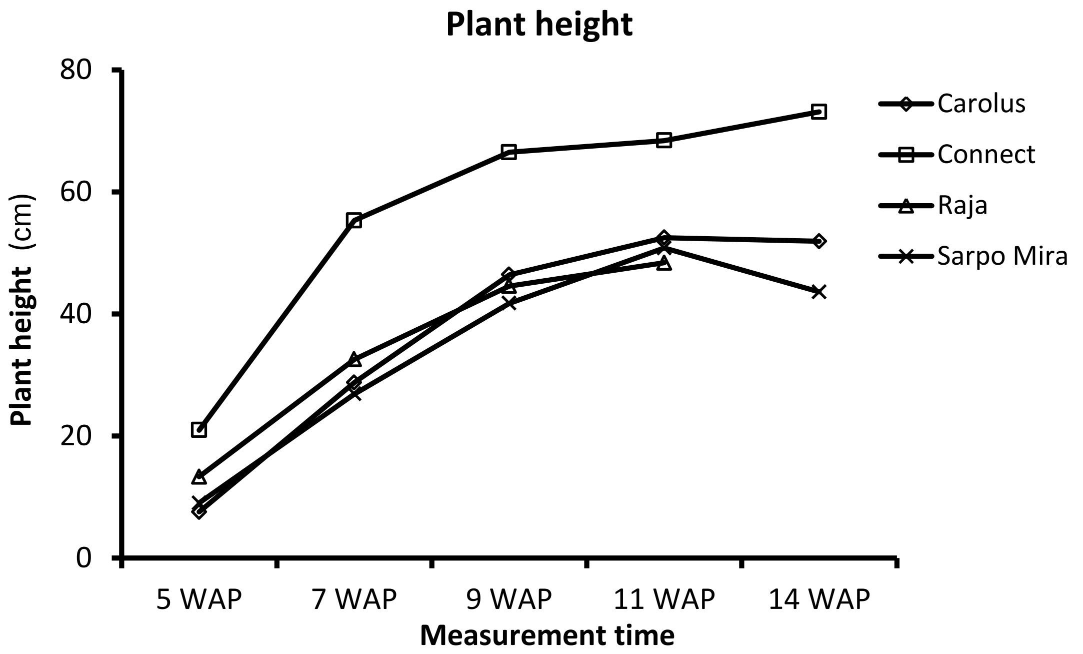

Fig.9. Effect of cultivars on plant height during the potato production period

As what we can see from the Fig.8, Connect already showed a faster growth than the others in terms of plant height on the 5th week after planting; Raja also presented a significant difference on plant growth compared to Carolus and Sarpo Mira. However, at 7th week after planting, only Connect showed a significant difference with other three cultivars regarding plant height and similar results can be found at 9th week after planting 11th week after planting. The plant height of 14th week after planting generally did not increase and some cultivars even became shorter due to *Phytophthora* and old physiological age. But Connect still grew and this is probably because Connect is a medium late cultivar and has higher plant vigour

(Den Hartigh). From the plant height, we found the plant height differed from each other at the beginning which could be related to different growth vigor because the seed potato were from different companies where potato could have different storage conditions and treatments(Hartmans & Van Loon, 1987).

Overall, there was a significant difference among four cultivars regarding plant height and Connect was the highest cultivar in contrast with Raja, Sarpo Mira and Carolus.

4.1.2.2. Leaf chlorophyll index

Table 13. Effects of cultivars on potato plant leaf chlorophyll index

	n [a]	Leaf chlorophyll index (-)				
		5 WAP [b]	**7 WAP**	**9 WAP**	**11 WAP**	**14 WAP**
Cultivar [d]						
Carolus	36	36.0 b [c]	39.3	33.5	27.5 b	25.8 a
Connect	36	46.2 a	41.7	35.8	30.7 b	27.5 a
Raja	36	50.2 a	42.8	36.1	29.6 b	-
Sarpo Mira	36	45.2 a	46.5	39.3	35.4 a	27.6 a
P-value		0.003	0.065	0.050	<0.001	<0.001

[a] n refers to the number of samples

[b] WAP = weeks after planting

[c] Different letters indicate significant differences according to Fisher's protected LSD-test (P<0.05).

[d] Samples were taken from potato variety mixture for studying the differences among cultivars on leaf chlorophyll index.

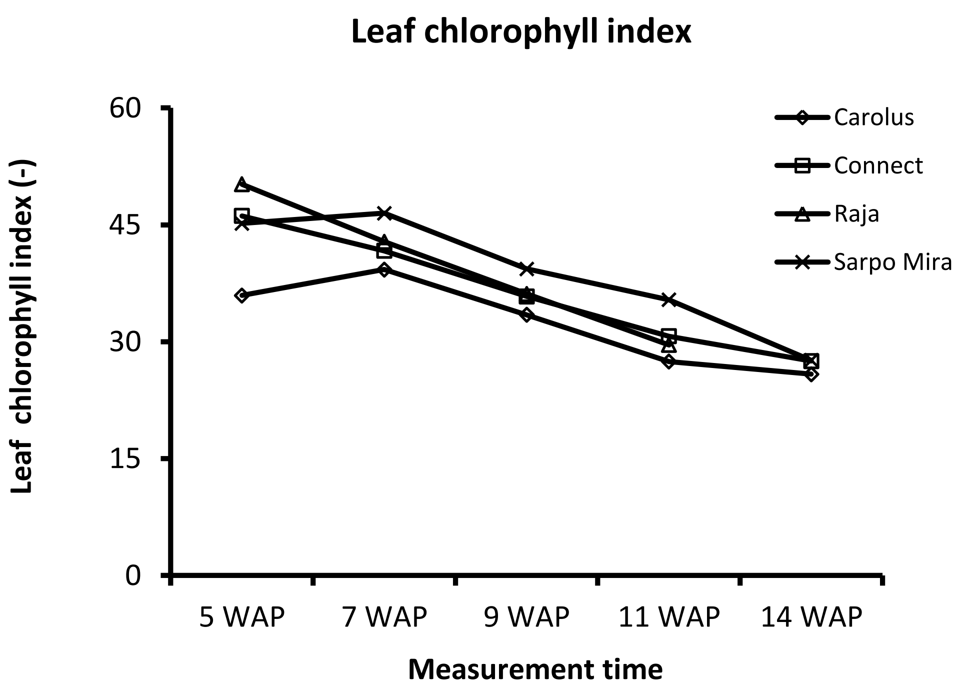

Fig.10. Effect of cultivars on potato leaf chlorophyll content during the potato production period

The potato leaf chlorophyll index showed a significant difference among four cultivars at the beginning of potato growth (5th WAP) and the chlorophyll index of Carolus was significantly lower than other three cultivars. However, there were no significant differences at 7th and 9th week after planting among the four cultivars regarding leaf chlorophyll index. At the 11th week after planting, Sarpo Mira had significantly higher leaf chlorophyll index than other three cultivars. Because of the infection of late blight on Raja, the leaves were completely senesced at the end of growth period. Overall, it is shown that there was a downward trend of leaf chlorophyll index during the process of potato growth.

Leaf chlorophyll content in field-grown potato could be affected by nitrogen supply, genotype and the age of plant (Mauromicale *et al.*, 2006) and the older the plant, the lower the leaf chlorophyll content would be. Because of the genotype and late maturing characteristics, Sarpo Mira had a higher leaf chlorophyll index at 9th and 11th week after planting. In addition, Phytophthora causes a decreasing chlorophyll content, which is found in tomato plants (Zhang *et al.*, 2003). Therefore, late blight could have had an impact on leaf chlorophyll content for susceptible cultivar, such as Raja.

4.1.2.3. Canopy size

Table 14. Effects of cultivars on potato plant canopy size

	n [a]	Canopy size (cm^2/plant)		
		7 WAP [b]	9 WAP	11 WAP
Cultivar				
Carolus	36	840 c [d]	1311 c	1514 b
Connect	36	3473 a	6643 a	7036 a
Raja	36	1606 b	2349 b	1297 b
Sarpo Mira	36	1283 bc	1771 bc	2257 b
P-value		<0.001	<0.001	<0.001

[a] n refers to the number of samples

[b] WAP = weeks after planting

[c] Samples were taken from potato variety mixture for studying the differences among cultivars on the canopy size of potato plants

[d] Different letters indicate significant differences according to Fisher's protected LSD-test (P<0.05).

From Table 14, it is found that Connect had the largest canopy size and the canopy size of Connect was significantly higher than the ones of other three cultivars during growth period. In addition, the canopy size of Raja was significantly higher than the one of Carolus as well at the first two measurements. The canopy size of Carolus, Connect and Raja between 7th and 9th week after planting grew faster than the canopy size growth between 9th and 11th week after planting. However, the growth speed of canopy size of Sarpo Mira between 7th and 9th week after planting was the same as the one between 9th and 11th week after planting, which can be seen from Fig. 11.

The branching capacity of cultivars is the major determination of canopy size (Collins, 1977) while other factors also could have an impact on canopy size, such as disease, maturing time. Sarpo Mira was a late maturing cultivar and the growth speed of its canopy size was more stable than other cultivars until the end of measurement. Raja was influenced by late blight seriously and it canopy size started to decline from the 9th week after planting.

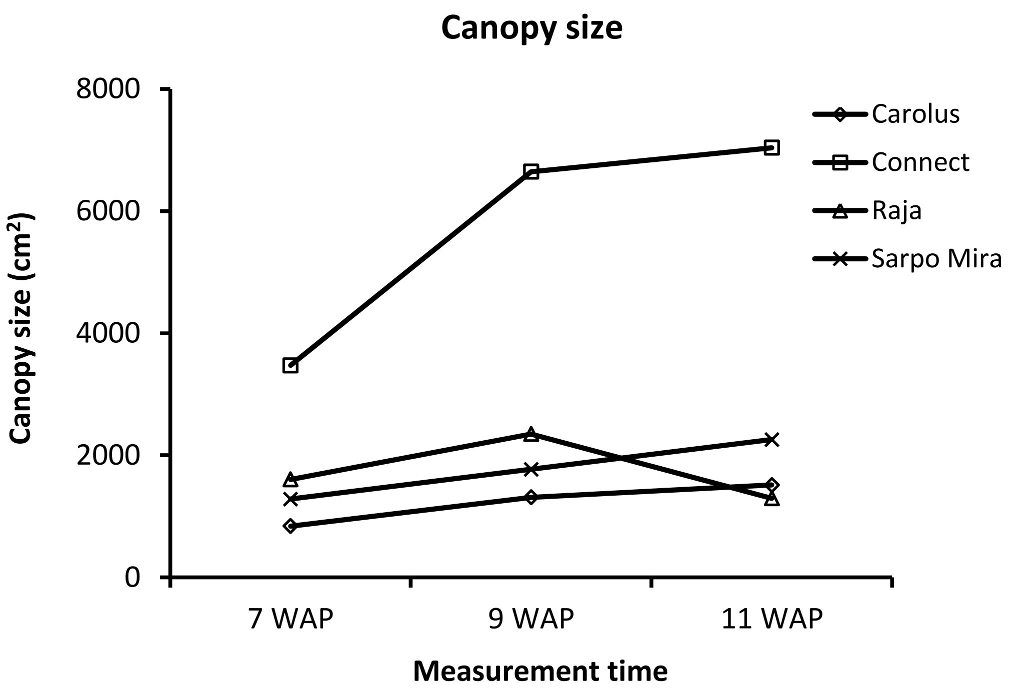

Fig.11. Effect of cultivars on potato canopy size during the potato production period

4.1.2.4. Fresh weight and dry weight of above-ground biomass

Table 15. Effects of cultivars on fresh weight and dry weight of above-ground biomass

	n [a]	Fresh weight of above-ground biomass			Dry weight of above-ground biomass		
		6 WAP [b]	10 WAP	15 WAP	6 WAP	10 WAP	15 WAP
Cultivar [c]							
Carolus	24	78.6 b [d]	134.5 c	58.8 b c (48)	9.5 b	14.9 bc	9.4 bc (48)
Connect	24	253.8 a	531.1 a	497.5 a (48)	26.2 a	69.4 a	69.5 a (48)
Raja	24	71.3 b	107.6 c	16.6 c (48)	8.5 b	12.1 c	2.5 c (48)
Sarpo Mira	24	97.1 b	335.9 b	106.3 b (48)	10.5 b	32.4 b	15.4 b (48)
P-value		<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

[a] n refers to the number of samples, the number in brackets means the number of samples of last time

[b] WAP = weeks after planting

[c] Samples were taken from potato variety mixture for the study of cultivar differences on fresh weight and dry weight of potato plant above-ground biomass.

[d] Different letters indicate significant differences according to Fisher's protected LSD-test (P<0.05).

As can be seen from Table 15, there were significant differences among four cultivars in potato variety mixture regarding fresh weight and dry weight of above-ground biomass. Connect had significantly high fresh weight and dry weight of above-ground biomass compared to Raja, Carolus and Sarpo Mira during the plant growth period. At the 10th week after planting, the fresh and dry weight of Sarpo Mira was higher than Carolus and Raja significantly as well. In addition, the fresh weight and dry weight of all the cultivars at the end decreased compared to the second measurement.

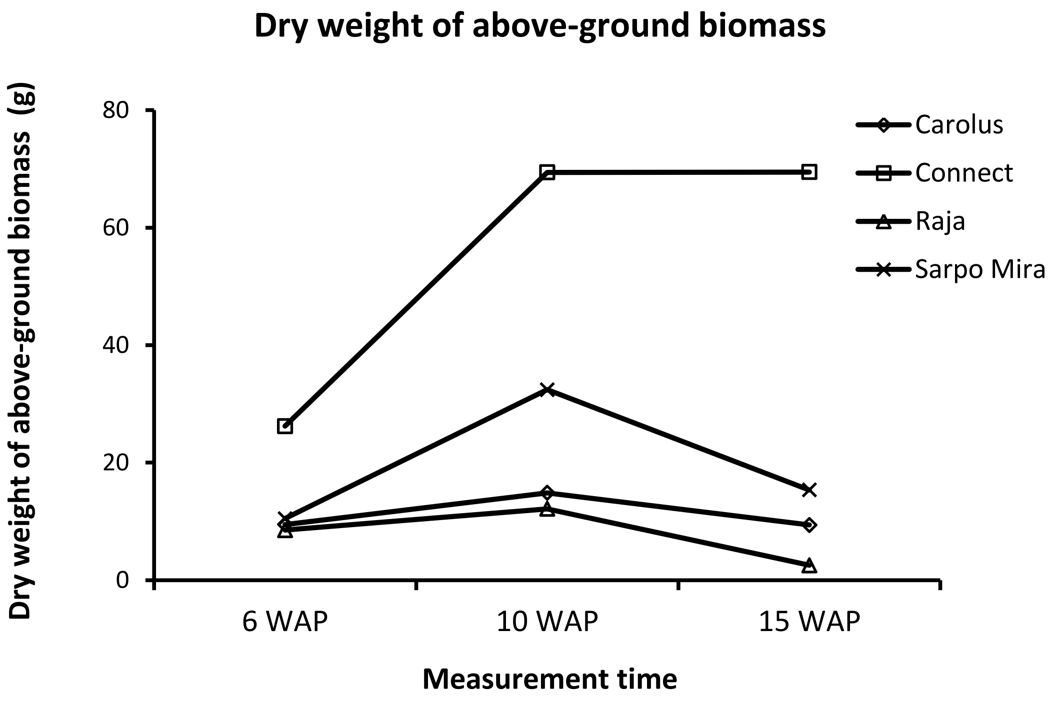

Fig.12. Effect of cultivars on dry weight of potato plant above-ground biomass during the potato production period

From the fresh weight and dry weight of above-ground biomass at the beginning, we found that Connect was already very competitive due to its significantly higher weight. At the beginning of planting, the seed tuber size of Connect was much larger than other cultivars and this caused different growth vigour afterwards, which resulted in a competitive condition of Connect. Furthermore, late blight was a major factor to limit the development of fresh weight and dry weight of above-ground biomass for Raja as well. Fresh weight and dry weight of above-ground biomass are also affected by nutrients and genotypes (Moinuddin *et al.*, 2005).

4.1.2.5. Root depth distribution

Table 16. Effects of cultivars on potato plant root depth distribution

Factors	n [a]	Dry weight of root biomass (g m^{-3})			
		Total	0-15 cm	15-30 cm	30-45 cm
Cultivar [c]					
Carolus	6	445.4 ab [b]	138.5	239.6	67.2
Connect	6	624.3 a	337.2	200.2	86.9
Raja	6	300.1 b	146.2	119.3	34.6
Sarpo Mira	6	630.3 a	271.7	178.9	179.7
P-value		0.027			
Raja (Mixture)		300.1	146.2	119.3	34.6
Raja (Non-mixture)		337.3	190.7	70.5	76.1

[a] n refers to the number of samples

[b] Different letters indicate significant differences according to Fisher's protected LSD-test (P<0.05).
[c] Samples were taken from potato variety mixture for the study of cultivar differences on root depth distribution.

The analysis of root depth distribution showed that there were significant differences among cultivars. To be specific, the total dry weight of root biomass of cultivar Connect and Sarpo Mira were significantly higher than Raja and Carolus. In each soil layer, cultivars performed differently and most of the roots of four cultivars were distributed in the first and second 15 cm soil layer. Between 30cm and 45 cm of the soil, there was little dry weight of root biomass except Sarpo Mira. Carolus had most roots in the second 15 cm soil layer and other three cultivars had the most roots in the first 15 cm soil layer.

The Raja in potato variety mixture produced less root than the Raja in potato variety non-mixture. Raja from the potato variety non-mixture in the first 15 cm produced much more root than the Raja from the potato variety mixture as well. However, there was more root in the second 15 cm soil layer for the Raja in potato mixture compared to the Raja in potato variety non-mixture.

Raja in potato variety mixture might be more competitive than the raja in potato variety non-mixture due to its root distribution. Variety had a major influence on the ultimate depth of rooting (Stalham & Allen, 2001) and the genotypes played an important role in it. Other factors such as soil water content, irrigation, soil compaction all have different impact on root depth distribution(Opena & Porter, 1999).

4.2. Conclusions

The aim of this chapter was to study the cultivar differences within potato variety mixture system and to summarize the challenges and problems which have potential to be improved in the future. Genotypes showed to have a direct influence on plant characteristics and good cultivar combination or appropriate ecological combination ability among cultivars appears to be of importance for potato variety mixture system.

The most important parameter – yield, showed significant differences. In fact, only cultivar Connect produced significantly higher yield than other three cultivars: Raja, Carolus and Sarpo Mira. However, higher yield of Connect coincided with taller plant height, larger canopy size, greater potato above-ground biomass and more root biomass in sharp contrast with other cultivars. Every parameter of plant characteristics is interacted with each other and their performance determines final yield. Overall, the cultivars showed significant differences in terms of yield, plant height, canopy size, fresh and dry weight of above-ground biomass and root depth distribution.

In general, Connect outperformed all other cultivars for most growth processes and thus has the highest yield. However, under the conditions of the current study, it was too competitive and therefore the potato variety system as a whole may still not have functioned optimally. As discussed above, genetic traits and initial tuber size could contribute to its dominant position. Thus, using more uniform seed tuber size and pre-planting conditions across all cultivars appear to be relevant when implementing future studies. In addition, cultivars resistant to late blight played an important role in potato variety mixture since they can reduce the risk of yield reduction associated with late blight to a large extent. However, more detailed studies are needed to examine the processes that govern the interactions among cultivars, which is in order to design ecological combination of different cultivars effectively.

4.3. Recommendations

1. Use of similar seed tuber size and controlling pre-planting storage conditions are suggested to minimize the differences in initial seed vigour, which could result in the dominance of certain cultivars in the potato variety mixture system.
2. The growth duration of cultivars should be the same in order to control plant growth and it is easy for harvest as well. The same growth duration of cultivars can ensure uniform plant growth and reduce the possibility of competitive cultivars.
3. The effects of rows should be studied by measuring each row in the future rather than measuring central two rows together and edge rows together.
4. It is better to test the growth vigour of seed tuber before planting potatoes. Pre-test for growth vigour is essential because it is influenced by storage environment and seed and large differences of growth vigour among cultivars easily cause irregular plant growth.
5. The severity of late blight could be studied rather than only late blight infection ratio on susceptible cultivars, but appropriate methods are needed in order to evaluate its effect on potato variety mixture compared to potato variety non-mixture.
6. The soil structure should be continued to investigate because differences on plant growth of different locations were observed.
7. Planting date should be suitable and the planting date of this research was too late (in the middle of May). It is suggested to plant potato between the middle of April and the start of May; it also depends on the weather and cultivar maturity.

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