Zemědělská fakulta
Faculty of Agriculture
Jihočeská univerzita v Českých Budějovicích
University of South Bohemia in České Budějovice

UNIVERSITY OF SOUTH BOHEMIA IN ČESKÉ BUDĚJOVICE

FACULTY OF AGRICULTURE

Department of Agroecosystems

Master's thesis

Influence of wheat varietal mixtures on yield stability, grain quality and flour rheological properties.

Author: Mousa Zoghali

Supervisor: doc. Ing. P. Konvalina, Ph.D.

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Declaration

I declare that I am the author of this graduation thesis and that I used only sources and literature displayed in the list of references in its preparation.

In České Budějovice on ....23-4-2021.... ....

Signature

Abstrakt

V rámci diplomové práce byly hodnoceny čtyři odrůdy pšenice seté (Butterfly, Illusion, Lorien, Vanessa) v režimu ekologického zemědělství. Byly vysety v několika variantách, z nichž byla jedna kontrolní (samostatně zaseté odrůdy). Dále se jednalo o směsi dvou odrůd, kdy byl porost založen smíchaným osivem v poměru 50 % (Butterfly + Lorien, Butterfly + Vanessa, Illusion + Lorien, Illusion + Vanessa). Třetí varianta zahrnovala stejnou směs odrůd, ale byly zasety vždy samostatně obřádek (row by row). Pokus byl založen ve třech opakováních. Analýzy byly prováděny ze směsných vzorků v celkovém počtu dvanáct.

Výsledky ukázaly, že pekařská jakost byla vyšší u samostatně pěstovaných odrůd, ale s minimálními rozdíly. Obsah bílkovin byl vyšší v případě výsevu row by row. Nejlepší pěkařskou jakost měla samostatně pěstovaná odrůdy Butterfly a varianty, kde byla jednou z komponent směsí. Odrůda Vanessa měla nejnižší obsah bílkovin, ale ve směsi s odrůdou Butterfly došlo k jejich zvýšení na 9,39 %. Z hodnocených agronomicky významných znaků byla ovlivněna výška rostliny při setí odrůd ve směsi. Statisticky průkazné rozdíly byly zaznamenány pouze v případě počtu klasů před sklizní (směs odrůd setá row by row). To bylo způsobeno patrně zvýšením produktivního odnožování v důsledku konkurence mezi odrůdami, kdy odrůdy měly více prostoru pro svůj rozvoj než při setí smíchaného osiva. V ostatních variantách byly minimální rozdíly.

Klíčová slova: Pekařská kvalita, Obsah bílkovin, Odnožovací kapacita, Odrůdy pšenice.

Abstract

In this experiment there were four seeded varieties of wheat (Butterfly, Illusion, Lorien, Vanessa) to three groups of sowing, the first group contained of the pure seeded varieties (pure Butterfly, pure Illusion, pure Lorien, pure Vanessa), the second group contained of a mixture of varieties that were sown in a narrow lines (Butterfly + Lorien, Butterfly + Vanessa, Illusion + Lorien, Illussion + Vanessa), and the third group contained also of the same mixture of varieties, but they were seeded in a broad lines, each variety was divided in three replications, these three replications of each variety were mixed to form in the end 12 samples.

The results showed that the baking quality was better in case of the single growing variety with low differences. The protein content was higher in case row by row seeding. The highest baking quality reached in the single variety “Butterfly” and its mixtures. The variety “Vanessa” had the lowest protein content, but it was increased to 9,39 % in the mixture with the variety “Butterfly”. The stalk length was influenced by the variety or there was an influence of the mixture of two varieties. Statistically different results were only in case of the number of spikes before harvest. The tillering capacity of wheat has made an intenser tillering and the number of spikes was a little bit higher in the case of row by row seeding. In other variants there were low differences.

Keywords: Baking quality, Protein content, Tillering capacity, Wheat varieties.

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Content

1 Introduction

Wheat (*Triticum spp.*) is one of the most important and widely grown food crops with more than 25000 different cultivars (Sapone et al., 2012). Its cultivation was started around 10000 years ago during Neolithic Revolution, first series of agricultural revolutions. Due to its wide adaptability to diverse climatic conditions and multiple end-uses along with dynamic nature of genomes and polyploidy character, it has become a crop of financial and nutritional importance (Dubcovsky and Dvorak, 2007).

The trend of production and consumption of wheat is increasing world-wide. According to FAOSTAT, global wheat production is expected to reach a new record of 780 million tonnes in 2021. Nowadays people are more concerned about quality which is forcing processors to use wheat with specific quality attributes. Grain size, protein content and its composition as well as, starch content are important variables that determine wheat quality. And these characteristics depend on cultivar, growing conditions and other environmental factors and the interaction between cultivar and environment (Panozzo and Eagles, 2000).

It is the best of the cereal foods and provides more nourishment for humans than any other food source. Wheat is a major diet component because of the wheat plant's agronomic adaptability, ease of grain storage and ease of converting grain into flour for making edible, palatable, interesting and satisfying foods. Doughs produced from bread wheat flour differ from those made from other cereals in their unique viscoelastic properties (Orth and Shellenberger, 1988). Wheat is the most important source of carbohydrate in a majority of countries. Wheat starch is easily digested, as is most wheat protein. Wheat contains minerals, vitamins and fats (lipids), and with a small amount of animal or legume protein added is highly nutritious. A predominately wheat-based diet is higher in fibre than a meat-based diet (Johansson et al., 2005).

The evaluation of flour and dough characteristics can be conducted using some rheological devices like Farinograph, Extensograph, Alveograph. Therefore, the other quick methods are needed to test the suitability of flours in terms of baking quality (Koksel et al., 2009). A new rheological device – Mixolab has been developed to be able to describe the dough consistency during heating and cooling period for rapid assessment of the wheat quality. This machine measures both flour protein and starch characteristics and provides information about protein weakening, starch gelatinisation, enzyme activity, and gel strength in a single test (Dubat, 2010). In addition, evaluation the wheat baking quality, hydrocolloid effects on the thermo-mechanical properties of wheat, effect of antioxidants on dough mixing properties, etc.

(Kahraman et al., 2008; Ozturk et al., 2008; Abdel-Samie et al., 2010). The effects of antioxidants on dough mixing properties (Abdel-Samie et al., 2010). The search for new varieties with better adaptability to climatic conditions with desirable agronomic and quality performance is one of primary request, which can contribute to the crops diversification, diverse food production and local market development. The bread making quality of wheats can be evaluated by various quality tests. One of the most reliable methods is the direct baking test, this method is time and labour consuming, and it is difficult to use for commercial aims. Indirect methods, more rapid and simple, such as wet and dry gluten content, sedimentation, rheological methods for the evaluation of dough and gluten strength, have been widely adopted, but it does not always differentiate wheats of medium-strong quality (Dhaka et al., 2012). The obtained test results explain both – the protein as well as starch characteristics – pasting behaviour of flour. In addition to the importance of the protein component for the quality of the final product, more recent studies have emphasized the importance of the starch component (Torbica et al., 2016). However, there are not sufficient data to use Mixolab parameters for quality assessment of non-traditional cereals (Grobelnik Mlakar et al., 2014).

Quality for bread wheat is mainly determined based on falling number (FN), protein content and gluten quality. FN is a method aimed at determining the sprout damage and α-amylase activity in wheat grains which determines the flour quality for bread making (Wang et al., 2008). The protein concentration and composition are found to affect the quality of baked products (Johansson, 2002) which in addition are determined genetically and also affected due to environmental conditions (Johansson et al., 2001). As sprout damage is highly dependent on rainfall, the predicted climate change can be more challenging for wheat producing areas that will have increased precipitation during the period of wheat maturation and harvest. Carbohydrates, protein, amino acids, lipids and minerals are the major components in wheat grains that affect nutritional value and end-use quality. Among them protein is an important constituent that determines bread making quality (Pomeranz, 1987; Shewry, 2009). Protein content in fully matured wheat grain varies from 10-20 % (Shewry et al., 1994) and normally from 10-15 % in western Europe. If we assume 10 % protein content in wheat grain, it produced 66.3 x 109 kg total protein and 9,4 kg protein/capita/annum (assuming 7 billion world population) in the year 2012 (Balyan et al., 2013)

1.1 Aims and working hypotheses

This thesis focuses on the influence of wheat varietal mixtures on yield stability, grain quality and flour rheological properties.

The aim of this study was to improve wheat yields and includes further mixing of wheat varieties through crossing, interspecific and intergeneric hybridization, biotechnology techniques, wheat mixtures. The growth of wheat mixtures will lead to yield stability, then it will lead to baking quality stability, and the rheological properties of flour will be positively influenced by the growing wheat in mixtures.

2 Literature review

With more than 25 000 different cultivars, wheat (*Triticum spp.*) is one of the most important cereal grain cultivated worldwide. Starchy endosperm storage tissue from wheat grain is used to produce bread, noodles, pasta and wide range of other food products (Tosi et al., 2011).

The cultivation of wheat reaches far back into history. Wheat was one of the first domesticated food crops and for 8 000 years has been the basic staple food of the major civilizations of Europe, West Asia and North Africa (Sapone et al., 2012).

Large amount of the wheat produced is used for making bread, other baked goods, pasta and noodles or bulgar and couscous as in the Middle East and North Africa (Sapone et al., 2012). Today, wheat is grown on more land area than any other commercial crop and continues to be the most important food grain source for humans. Its production leads all crops, including rice, maize and potatoes.

Wheat is grown on more than 240 million ha, larger than for any other crop, it represents the highest percentage 33,85 % of the global production of cereals (FAO, 2018), and world trade is greater than for all other crops combined. The raised bread loaf is possible because the wheat kernel contains gluten, an elastic form of protein that traps minute bubbles of carbon dioxide when fermentation occurs in leavened dough, causing the dough to rise (Hanson et al., 1982).

2.1 Wheat in organic farming

Organic farming in the Czech Republic is becoming increasingly popular with around 10 % of the agricultural land being organic. The Czech Republic is one of the ten countries with the highest shares of organic agricutural land in Europe, in 2018, it represented 12,8 % of the total organic agricultural area in Europe (FiBL-AMI survey, 2020).

Compared to the other cereal species, there is a wide range of common wheat varieties, it was not easy for an organic farmer to find what they were looking for in the past, as there was a lack of information on reaction of the individual varieties to the organic farming system. The situation has improved recently.

Common wheat is the most frequent cereal species in the Czech organic farming. In 2019, the total growing areas were 13 732,41 ha, the total harvest was 27 942,39 tons, and the yield/ha was 3,07 tons. Also, the wheat produces a very high yield in the Czech organic farming and it is higher than the other cereal species, the number of organic farms was 386, and the orgainc production area of wheat reached 9 100,37 ha, it represents approximately 66 % of the total growing areas of wheat (Ministertvo zemědělství, 2019).

2.2 Productivity and quality of wheat in organic farming

Protein content and its composition as well as, starch content and its ability to gelatinize are important variables that determine wheat quality, and these characteristics depend on cultivar, growing conditions and other environmental factors and the interaction between cultivar and environment (Panozzo and Eagles, 2000).

Quality of grains is determined by their ripeness and moisture. The optimal harvest moisture is 14 %. If we harvest wheat too late, gluten content and falling number decrease and the quality of gluten deteriorates. Therefore, food bread wheat and varieties inclining to lodging are harvested in preference to the other wheat varieties (Moudrý et al., 2008).

For bread-making, grain crude protein concentration (CP %), gluten quality, Hagberg falling number (HFN) and specific weight are among the most important quality parameters (Gooding et al., 1997). Wheat is harvested at the beginning of the full ripeness. Organic form of growing has a negative effect on crude protein content and it makes the food and baking quality more difficult to achieve, but it has a positive effect on the nutrition quality (there are more albumins and globulins in the organic plants) (Krejčířová et al., 2007).

Organic wheat has better parameters of the fodder quality than conventional one (it contains more albumins, globulins and essential amino acids) (Petr et al., 2004). The protein concentration and composition are found to affect the quality of baked products (Johansson, 2002) which in addition are determined genetically and also affected due to environmental conditions (Johansson et al., 2001).

Lower baking quality is another factor making the difference between organic and conventional common wheat. It is mostly caused by the fact that protein content and composition of proteins in wheat grain are influenced genetically and environmentally. They are also influenced by agro technology level and intensity of farming (Šíp et al., 2013). Lower baking quality of organic common wheat is caused by low protein content in grain (Krejčířová et al., 2010; Capouchová et al., 2013). The low protein content is provoked by nitrogen shortage in later growing stage (grains are created and they ripe there). Váňová et al. (2008), for instance, show that organic common wheat grains contain 2 % less crude protein in grain dry matter than conventional common wheat ones. Other authors (Prugar, 1994; Krejčířová et al., 2010) have confirmed it (2–3 % less crude protein content in organic common wheat grains). According to Krejčířová et al. (2010), organic winter wheat achieved lower values of Zeleny test than conventional one. It has indicated worse viscoelastic properties of organic wheat gluten proteins and less possibilities of technological and baking processing.

Gluten protein strength depends on storage protein composition, However environmental factors can modify the gluten quality. Temperature along with nitrogen timing affects gluten strength (Johansson et al., 2005). Field studies show that variation in nitrogen application influences these parameters indicating that there is effect of nitrogen fertilizer in gluten strength and bread volume (Johansson et al., 2001). Higher temperature (> 35˚C) during grain filling can enhance synthesis of gliadin reducing glutenin to gliadin ratio and thus result in weaker dough (Blumenthal et al., 1991). Quality characteristics like protein content, wet and dry gluten and rheological properties should be well defined by baking industries. So, many physical and chemical analysis should be performed before the flour is processed (Miralbés, 2003).

In wheat, both the quantity and quality of protein are crucial. The major types of protein can be divided into three categories: simple, conjugated and derived. However, only simple protein is found in wheat plants, consisting of four major types: albumins (soluble in water and dilute buffers), globulins, prolamins, and glutelins. Gluten, the remainder of wheat flour after removing starch, non-starchy polysaccharides, and water-soluble constituents, comprises alcohol-soluble gliadins and alcohol-insoluble glutenins (Shewry, P.R.; Halford, N.G., 2002). Wheat storage proteins have two basic fraction groups: gliadins and glutenins. Glutenins are known as being the larger polymers in nature and are measured as high molecular weight glutenin subunits (HMW-GS) and low molecular weight glutenin subunits (LMW-GS). These are used as protein markers for predicting the quality of bread and identifying wheat varieties (Branlard et al., 2001; Bradová et al., 2012). Rheological traits are important for processing flour in the baking industry. This index is used for predicting dough-processing parameters and the quality of the end product. To investigate flour and dough characteristics, such as elasticity, viscosity, and extensibility, traditional rheological instruments such as farinograph, extensograph, and alveograph can be used. However, with Mixolab II (Chopin Technologies, Paris, France), a new rheological device, researchers are able to measure the physico-chemical behavior of dough during heating and cooling processes (Švec et al., 2015). During five stages in the process, Mixolab parameters are measured as the change of torque when mixing and heating wheat flour and water. They provide information about maximum torque, protein quality, starch characteristics, enzyme activity, and starch retrogradation (Harati et al., 2020).

2.3 Ecological intensification

Selection of suitable varieties for particular land and climatic conditions is the elementary and essential intensification factor influencing organic common wheat growing. The late harvest decreases the gluten content and falling number, the quality of gluten deteriorates as well.

If we sow wheat into wider rows (e.g. 375 mm), we may hoe the crop stand; it has a positive effect and it enhances the baking quality of wheat (Konvalina et al., 2008). According to Capouchová et al. (2011), if we organize the crop stand differently and we sow wheat into wider rows (125 mm – usual width of cereal rows), we increase the crude protein content in winter wheat grains, If we establish wider rows (not 125 mm but 250 mm), we increase the crude protein content by 0.6 % approximately. If we establish even wider rows (not 125 mm but 375 mm), we increase the crude protein content by 1.2 % approximately. It does not have any negative effect on grain yield.

Location, soil type and cultivars influence yield and protein content (Malik et al., 2012). The agronomical practices also influence protein quantity and quality. Wheat of high protein quantity and quality can be produced with reduced tillage, with providing it with soil and climate conditions are suitable, and the nitrogen management is appropriate (Godfrey et al., 2010). Intercropping with legumes is an effective strategy to improve protein quantity and quality in wheat. Pea and clover grass were most effective and recommended. Catch crops and rotations with legumes were also effective, with clover grass often improving the performance (Moudry et al., 2011). Adoption of minimum tillage should be considered on organic systems where weed competition had been controlled by mechanical weeding or diverse crop rotation. Positive effect of minimum tillage on soil fertility could directly affect crop nutrient nutrition through good rooting when initial soil structure was good (Krejčířová et al., 2010).

There are some chances to improve yield without affecting quality or vice-versa through breeding (Barraclough et al., 2010). Mobilization of nitrogen from various plant parts to head can increase grain protein content and reduction in dry weight of plant biomass (stem weight) can increase grain yield. Understanding the genetic base for dry weight build up and nitrogen concentration of various plant parts can be useful for successful breeding of cultivars with high grain yield and high protein content (Malik et al., 2012). Grain yield and protein content are important parameters in wheat production (Groos et al., 2003). Protein content in wheat grain normally decreases with the increase in grain yield (Simmondsn, 1995). Negative correlation between these two traits is considered to be affected genetically (Groos et al., 2003) that is highly heritable. of various plant parts can be useful for successful breeding of cultivars with high grain yield and high protein content (Malik et al., 2012).

Protein content in wheat grain can be increased by increasing level of organic nitrogen fertilizer application (Uhlen et al., 2004). But timing of nitrogen application could have different responses depending on the environment i.e. temperature. Split application of nitrogen during stem elongation or at heading can increase protein content in the wheat grain.

Application of post anthesis nitrogen under moderate temperature (24˚C days and 17˚C nights) increased rate of accumulation of protein as well as total protein content in wheat (Dupont et al., 2006). But when grown in same condition under 37˚C days and 28˚C nights (higher temperature) post anthesis nitrogen did not have marked effect on rate of accumulation and total content of protein. However, protein percentage in grains grown at the higher temperature was higher than those grown at the lower temperature. Usually the grain protein percentage increases when the environment conditions like drought and high temperature hinders grain yield to reach its potential (Fowler, 2003). Postpollination application of nitrogen is more effective for gaining wheat with higher protein content as well as higher yield when compared to application of nitrogen by boot stage (Bly and Woodard, 2003).

2.4 Wheat varieties for organic farming

Some specific breeding programmes are ideal for selection of suitable varieties. Such programmes depend on the scientific selection of input materials and selection carried out in the probable soil use conditions (which means low-input or organic farming conditions) (Wolfe et al., 2008). Though the fast development of the organic farming, there are a few varieties which have been bred for the organic farming conditions specifically. In practice, conventionally-bred and tested varieties are grown and they are reproduced in the organic farming conditions. However, such varieties usually do not have many important properties which are required by the organic farming system. They have been bred in order to make their genetic makeup as suitable for the conventional intensive growing as possible (Lammerts van Bueren et al., 2002; Murphy et al., 2007; Konvalina et al., 2011).

Specific requirements are placed on common wheat varieties which are suitable for the organic farming. They are as follows: efficient absorption of nutrients through their root systems (even if there are less nutrients contained in the soil), competitiveness against weeds, resistance to diseases, pests and abiotic stressing factors, stable yield (even in low-input farming system), good quality of production, suitability for organic products and attractiveness to consumers (Wolfe et al., 2008). Moudrý (2006) and Petr et al. (2007) also state that the varieties generating yield through dense crop stands (they make more tillers) are not recommended, as there are worse conditions for growth and more tillers are reduced then in the organic farming. The varieties generating yield through ear productivity are more suitable there. Moudrý et al. (2007) consider 400–450 ears per square metre to be the optimal organic wheat crop stand density.

The organic wheat varieties should be more competitive against weeds than the conventional wheat ones (Oberfoster and Kögelberger., 1996). They should have larger leaves, longer stalks and they should grow faster in the spring. Piorr and Köpke (1985) add that neither semi-dwarf varieties having short stalks nor varieties having tiny grains are suitable for the organic farming.

Conventional varieties have been developed with the aim of combining high productivity and standardized product quality under high-input conditions. Two main areas apparent where organic farming system differs most significantly from conventional farming systems, the soil fertility management, the disease and pest management. The varieties often perform differently in different environments due to genotype-environment interactions, therefore it is important to evaluate characteristics of varieties in conventional as well as in organic farming systems. Currently there is not obtained full answer yet - is the differences between the conventional and the organic growing systems large enough to justify breeding and testing of varieties in both environments. If it is so the main step is the work out the necessity requirements for development organic VCU. Therefore the main problem currently is the lack of information on the relative performance of modern crop species and varieties under organic conditions.

Organic variety trials not intended for national listing, there have been some long term variety testing under organic growing conditions at the Czech University of Live Science. But mostly organic farmers in the Czech Republic have to orientate themselves according to descriptions of variety properties available in the recommended list of conventional varieties or in other information sources. The pertinence of their choice have to be verified in practice (Konvalina et al., 2012). There is a current proposal for a research project to improve the system for organic bread wheat varieties for organic farming (Stehno, Gene Bank, Crop Research Institute, 2012).

Varieties of agricultural crops must pass a test for Value for Cultivation and Use (VCU). This is not the case for other crop groups. Since 1999 a VCU trial series under oganic conditions is run for winter wheat additionally to the normal trial series. Organic VCU-testing may well be a task in the near future in the Czech Republic, but so far there is no possibility to have a variety VCU-tested under organic growing conditions, and there have been no applications so far. (Šafaříková, ÚKZÚZ, 2012). The winter wheat has some VCU challenges like cost of registration due to breeding goals being different to standard traits assessed in registration, important traits are not being evaluated in existing VCU trials, no specific organic VCU in some countries, organic cultivars perform different under conventional testing, and too long straw compared to modern. For the spring wheat, it has too long straw compared to modern as well (Pedersen et al., 2021).

2.5 Use of varietal mixtures

Breeding of wheat varieties for organic farming is a very long and financially demanding process, because of the small Czech market. Therefore, a functional system of certification of conventional varieties should be built in order to increase the efficiency of organic common wheat growing. A lot of modern common wheat varieties are suitable for organic farming system. As an example from Austria shows (a lot of varieties were grown experimentally on organic parcels and tested between 1999 and 2006), such a system can be very efficient and we can run it easily. Since the Austrian experiment was finished and assessed, a system of utility value has been implemented in the organic farming system. Varieties can undergo conventional and organic farming tests there in the Czech Republic, common wheat and barley varieties have been tested officially since 2015 under the auspices of the Central Institute for Supervising and Testing in Agriculture. Preselected varieties are tested in order to be listed and registered as recommended varieties for the Czech organic farming system.

The modern efficient varieties are recommended for organic growing, there are certain risks if we do not know how such modern varieties (which have been bred in order to be grown intensively) react in the organic farming conditions. Grain yield (Murphy et al., 2007) and quality (Wolfe et al., 2008; Capouchová et al., 2013) indicated by protein content and composition of proteins in grain are logical indicators of soil suitability for a certain farming system. Compared to the conventionally grown common wheat variety, the same organically grown common wheat variety produced half yield rate (Mazzoncini et al., 2007). Ingver et al. (2008) show organically grown spring wheat yield to be 34 % lower than conventionally grown one. Results achieved in the Czech Republic have led e.g. to the following study: 10 varieties of winter wheat were tested, and organic varieties achieved 67 % of conventional variety‘s yield (Váňová et al., 2008).

3 Material and methods

The experiments were conducted in the growing season of winter wheat 2019/2020 year, their location was Zvíkov by České Budějovice (48.9758531N, 14.6245594E), the soil of experimental site was loamy soil; altitude of 460 m. Experiment was carried out by using randomized complete block design with three replications. All cultivars were sown on the organic certified research area. Crop rotation belongs to legume family, with common pea (Pisum sativum). The seeding rate was adjusted with a density of 450 germinal grains per m^2. The crop standards were treated in compliance with the European legislation (the European Council Regulation (EC) No. 834/2007, the European Commission Regulation (EC) No. 889/2008).

3.1 Used varieties

The experiment used four varieties of winter wheat (*Triticum aestivum* L.), namely Butterfly, Illusion, Lorien and Vanessa. The varieties and their combinations were seeded in three replications and grown during vegetation period from October/2019 to August/2020 as in the table 1.

Table 1: the seeded varieties and their method of seeding.

Variety	**Method of seeding**
Butterfly	Single
Illusion	Single
Lorien	Single
Vanessa	Single
Butterfly/Lorien	Mixture
Butterfly/Lorien	Row by row
Butterfly/Vanessa	Mixture
Butterfly/Vanessa	Row by row
Illusion/Lorien	Mixture
Illusion/Lorien	Row by row
Illusion/Vanessa	Mixture
Illusion/Vanessa	Row by row

In case of mixture, seeds of the varieties were mixed in share 50 %-50 % before seeding. Row by row – each variety was seeded as single in lines 25 cm wide and the second variety was seeded into interrows.

3.1.1 Nutrition status of experimental field

The nutrition was based on nitrogen provided by the forgoing crop. The basic nutrition analysis of the experimental field was made by Melich 3 method. The results are in the table 2. Whereas nitrogen content was evaluated in the beginning of growing season in the 2020 year. The results of nitrogen content in the soil are in the table 3.

Table 2: Evaluation of nutrient content in the soil (location Zvíkov, 2019).

Location	**Nutrient**			
	P ($mg.kg^{-1}$)	Mg ($mg.kg^{-1}$)	K ($mg.kg^{-1}$)	Ca ($mg.kg^{-1}$)
Zvíkov	56,3	227,2	207,7	1822

Table 3: Evaluation of nitrogen (Nmin) content in the soil (location Zvíkov, 2020).

Location	**Nitrogen content (11/03/2020)**							
	30-60 cm		0-30 cm		30-60 cm		0-30 cm	
	NH_4^- N	NO_3^- N	NH_4^- N	NO_3^- N	NH_4^- N	NO_3^- N	NH_4^- N	NO_3^- N
Zvíkov	0,48	8,66	1,00	3,51	1,92	34,65	4,02	14,03

3.1.2 Agrotechnology

Agrotechnological operations are in the Table 4. The diseases and pest occurence were monitored. There was only low infection by mildew. From the pests – only sporadic occurence of aphids and flea beetles. The plots were mechanicaly treated by harrowing against weeds two times.

Table 4: Agrotechnological operations (Zvíkov, 2019/2020).

Date	**Operation**
26.09.2019	ploughing
1-5.10.2019	soil preparation – harrowing 2x
07.10.2019	seeding by Hege machine
26.11.2019	harrowing against weeds
18.03.2020	harrowing against weeds
10-11.8.2020	harvest

3.1.3 Climatic conditions during growing season

The vegetation period was wetter in comparison to the previous years, it was also warmer than its normality for a long time. Details are in the Table 5.

Table 5: Climatic data (09/2019 – 08/2020, Zvíkov).

Month	Temperatures (°C)			Precipitation (mm)		
	Mean	Longterm mean	Difference	Sum	Longterm mean	Difference
09/2019	14,2	12,4	+1,8	29,5	50,6	-21,1
10/2019	9,4	7,5	+1,9	33,3	33,9	-0,6
11/2019	5,0	2,4	+2,6	36,2	38,6	-2,4
12/2019	1,9	-1,0	+2,9	31,7	30,2	+1,5
01/2020	0,6	-2,7	+3,3	18,1	26,5	-8,4
02/2020	4,2	-1,1	+5,3	50,3	27,1	+23,2
03/2020	4,4	2,4	+2,0	68,7	34,4	+34,3
04/2020	9,5	6,9	+2,6	18,1	48,6	-30,5
05/2020	11,4	11,9	-0,5	114,0	76,7	+37,3
06/2020	16,3	15,2	+1,1	159,0	99,2	+59,8
07/2020	18,0	16,8	+1,2	72,3	84,6	-12,3
08/2020	19,6	16,1	+3,5	86,0	83,3	+2,7
09/2019-08/2020	9,5	7,2	+2,3	717,2	633,7	+83,5

3.1.4 Varieties evaluation during vegetation period and yield

The length of stems was measured during the vegetation period in each plot at the flowering time of wheat, at the same time the number of spikes was calculated per square meter. After harvest by Wintersteiger experimental harvester the yield was measured and recalculated for the yield in tons per hectare.

3.2 Quality and baking quality analysis

3.2.1 Milling flour

The samples were milled into white flours using a PSY MP 20 (Mezos, Hradec Kralove, Czech Republic) and Quadrumat Junior machine (Brabender, Duisburg, Germany). Flour Mill Test indicates milling properties on small wheat samples. Commercial flour mills can use to this information to adjust mill settings to adjust flour extraction (Tipples, 1980). Small samples of wheat are milled on the machine to produce flour. This flour is used to evaluate properties, such as ash and protein content, and in gluten strength tests, such as the farinograph (Paradiso et al., 2006).

3.2.2 Volume test weight

Test weight is the weight of a measured volume of grain expressed in kilograms per hectolitre ($kg.hl^{-1}$) (Konopka et al., 2004). Test weight is a measure of its quality. High test weight wheat usually has relatively more extractable flour and less bran and is therefore more valuable to the end-user. The minimum test weight for No. 1 grade is 76,65 kilograms per hectolitre and discounts usually are applied when values fall below that value. To reach this standard, kernels must be dense and well filled so that they pack well (Kyptova et al., 2017). Kernel density is effected by the ratio of starch to protein and how tightly the starch granules are woven into the kernel. Factors that impeded starch deposition during grain filling, such as leaf diseases, scab, lodging, drought and high temperatures can reduce kernel density. How well kernels pack is determined largely by their shape. Smooth, rounded kernel pack more tightly than wrinkled ones. Shriveled kernels that are not completely filled will have a low test weight (Schober et al., 2002).

3.2.3 Protein content

The percentage of protein content was determined by the Kjeltec 1002 System (Tecator AB, Hoganas, Sweden), based upon N * 5.7 (in dry matter). Quality for bread wheat is mainly determined based on protein content and gluten quality (Trethowan et al., 2001).

3.2.4 The wet gluten content

The wet gluten test provides information on the quantity and estimates the quality of gluten in wheat or flour samples. Gluten is responsible for the elasticity and extensibility characteristics of flour dough. Wet gluten reflects protein content and is a common flour specification required by end-users in the food industry.

3.2.5 Zeleny test

Sedimentation value was determined using Sedi-tester apparatus in accordance with ICC No. 116/1 (Czech Republic). The sedimentation value according to Zeleny (Zeleny value) describes the degree of sedimentation of flour suspended in a lactic acid solution during a standard time interval and this is taken as a measure of the baking quality. Swelling of the gluten fraction of flour in lactic acid solution affects the rate of sedimentation of a flour suspension. Both a higher gluten content and a better gluten quality give rise to slower sedimentation and higher Zeleny test values. The sedimentation value of flour depends on the wheat protein composition and is mostly correlated to the protein content, the wheat hardness, and the volume of pan and hearth loaves. A stronger correlation between loaf volume and Zeleny sedimentation volume compared to SDS sedimentation volume could be due to the protein content influencing both the volume and Zeleny value (Shewry and Tatham, 2000).

3.2.6 The falling number

The falling number was determined by Perten Falling Number 1310 (Perten Instruments, Hagersten, Sweden) according to AACC 56-81 B and ICC Standard 107/1 (AACC 56-81 B., 2000b; ICC – Standard No. 107/1., 1995). FN is a method aimed at determining the sprout damage and α-amylase activity in wheat grains which determines the flour quality for bread making (Wang. et al., 2008). The level of enzyme activity measured by the Falling Number Test affects product quality. If the falling number is too high, enzymes can be added to the flour in various ways to compensate. If the falling number is too low, enzymes cannot be removed from the flour or wheat, which results in a serious problem that makes the flour unusable.

3.2.7 The dough rheological parameters

Mixolab II. was used to evaluate the baking quality according to the ICC standard method No. 173-ICC 2006 (ICC – Standard No. 173., 2006), which allowed us to evaluate the physical dough properties, such as dough stability or weakening, and starch characteristics in one measurement. The Mixolab measures the consistency of a dough subjected to the dual constraints of mixing and increasing temperatures. It analyzes the quality of protein and the starch using a 50 gram sample of the flour. The Mixolab process has the advantage of being able to measure properties of proteins, starch, and associated enzymes in one test. The greater the decrease in consistency, the lower the protein quality (Schmiele et al., 2017).

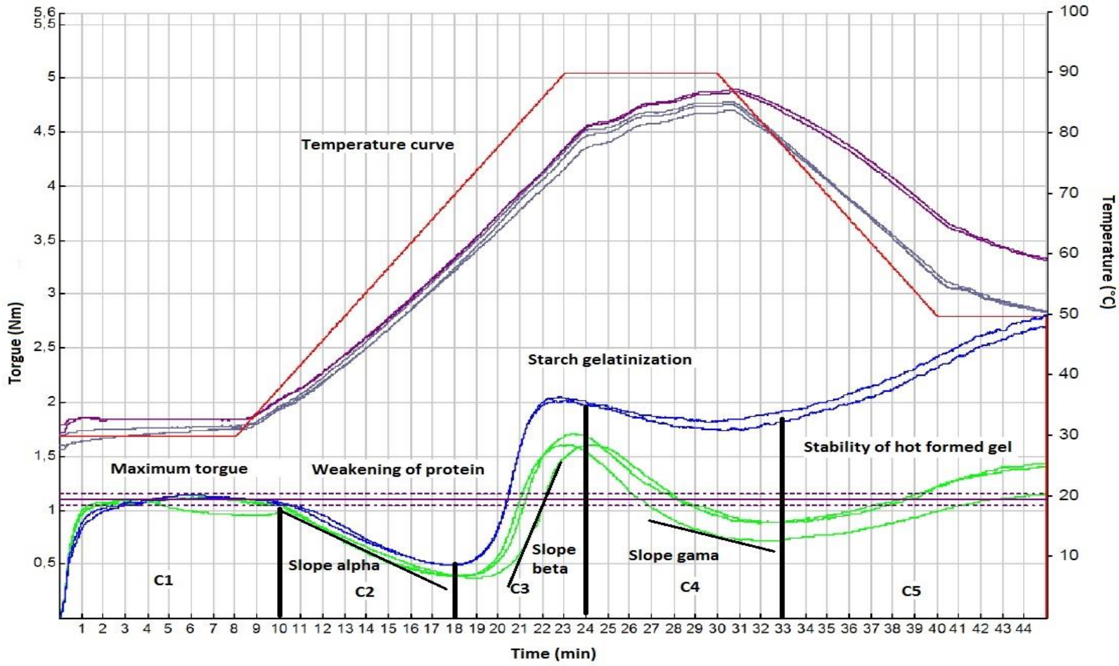

Figure 1: The mixolab II curves of wheat flour (Tran et al., 2020).

The parameters evaluated at Mixolab curve:

- Time for C1: The time evolution of the dough. The stronger the flour, the longer the time evolution (time to reach C1);
- C2: Attenuation of protein due to mechanical work and temperature;
- C3: The gelling starch;
- C4: The stability of the hot gel;

- C5: Measured starch retrogradation in the cooling phase;
- Amplitude - the elasticity of the dough. The higher the value, the more flexible the flour;
- Stability - the resistance against kneaded dough. The longer the duration, the stronger the flour;
- Slope α (C1-C2): Attenuation rate of protein in warming;
- Slope β (C3-C4): Speed starch gelatinization;
- Slope γ (C5-C4): The rate of enzymatic degradation.

In the first stage, hydration of the flour compounds occurs at 30 °C together with the stretching and alignment of the proteins, which leads to formation of the viscoelastic structure. An increase in the torque was observed during this stage until it reached the maximum value (1.10 Nm). The torque decreased to a minimum value in the second stage, which was attributed to the weakening of the protein network for mechanical shear stress and protein destabilization. (Rosell et al., 2007; Ferrer et al., 2006). The third stage demonstrates an increased temperature and gelatinization of starch. The granules absorb the water available in the medium and they swell, so the viscosity increases. In the fourth stage, the amylase activity and the physical breakdown of the granules are associated with a reduction in the viscosity. A decrease in the temperature resulted in an increase in torque, which is referred to as setback and corresponds to the gelation process. The last stage is related to retrogradation. Ferrer et al., (2006). Temperature regime in Mixolab was as follows: 8 min at 30 °C, heating at a rate of 4 °C min−1 for 15 min, holding at 90 °C for 7 min, cooling to 50 °C at a rate of 4 °C min−1 for 10 min, and holding at 50 °C for 5 min (Schmiele et al., 2017).

3.3 Statistical data evaluation

The data were analyzed using the Statistica 12.0 program (StatSoft. Inc., California, USA). The comparison of mean varieties and their division into statistically different categories were conducted using the Tukey’s (HSD) test with p-value < 0.05 considered statistically significant. One way ANOVA was applied for variance analysis (yield, number of spikes, length of plant).

4. Results and discussion

4.1 Evaluation of agronomical important characteristics of wheat varietal mixtures

From table 6, it is possible to see the influence of factors on parameters. The stalk length was influenced by the variety or there was an influence of the mixture of two varieties. The spacing had a low influence. The number of spikes before harvest was influenced by spacing. In this case there was no or a low influence of the variety or its mixture. The yield was more influenced by the variety or its combinations. The spacing had nearly no influence if we take in consideration standard one or row by row. Our result is supported by other authors, thanks for the autoregulation ability of wheat, the spacing has a minimum influence (Konvalina et al., 2014).

Table 6: Evaluation of variety factor and spacing on evaluated characters by analysis of variance (ANOVA), Zvíkov u Českých Budějovic, 2019/2020.

		MS	F	PČ	F	PČ	F
Combination	7	58,1	3,6*	15528	4,6*	7,6	8,5*
Spacing	1	8,2	$0,5^{ns}$	68267	20,0*	3,7	$4,1^{ns}$
Error	27	16,1	-	3406	-	0,9	-
Note: *statistically significance P ≤ 0,05; ^{ns} not significant; MS = mean square; F = test criteria; PČ = average sum of squares.							

More interesting data are in table 6, where the agronomical important characteristics were evaluated as standard spacing/seeding row by row, single variety/mixture of two varieties and varieties combination. There were low differences between the standard spacing and the row by row spacing. Statistically different results were only in case of the number of spikes before harvest. In the case of row by row seeding thanks for the tillering capacity of wheat, there was an intenser tillering and the number of spikes was a little bit higher. In other variants there were low differences.

Interesting results show the evaluation of single varieties and its mixtures. We saw in contradictory results what we expected – lower numbers in the growing mixture. Why it does happen, we cannot say, because we need to repeat the experiment next year. One of hypotheses is the fact of very good climatic conditions on the experimental field in the year 2019-2020. It can support the competition ability of varieties and it results to lower numbers. The hypothesis of growing wheat in the mixtures is based on different expectations – the grown variety in bad conditions is replaced by the second variety (Konvalina et al., 2020).

In the table 7 and figure 2 there are the single data from all the combinations. We can see mainly good yields in case of single grown varieties "Lorien, Butterfly and Vanessa". The lowest yield was in the combinations of variety "Lorien".

Table 7: Evaluation of depending on the spacing of seeding (Standard spacing/ Seeding row by row, Single variety/ Mixture of two varieties; Varieties combination), Zvíkov u Českých Budějovic, 2019/2020.

Factor	**Stalk length (cm)**	**Number of spikes (m^2)**	**Yield ($t.ha^{-1}$)**
Standard spacing / Seeding row by row			
Seeding row by row (25 cm)	92a	463b	7,27a
Standard spacing (12,5 cm)	92a	390a	7,69a
Single variety/ Mixture of two varieties			
Single variety	94a	422a	8,90b
Mixture of two varieties	91a	410a	6,88a
Varieties combination			
Butterfly	95ab	380a	7,54abc
Illusion	90ab	451a	9,44bc
Butterfly+Lorien	96a	347a	6,34a
Butterfly+Vanessa	89ab	455a	7,18ab
Illusion+Lorien	92ab	381a	6,21a
Illusion+Vanessa	88b	458a	7,79abc
Lorien	98a	372a	8,87bc
Vanessa	93ab	487a	9,73c
Note: HSD: honestly significant difference, Means labeled with the same letter within the same column are not significantly different (Tukey's HSD test).			

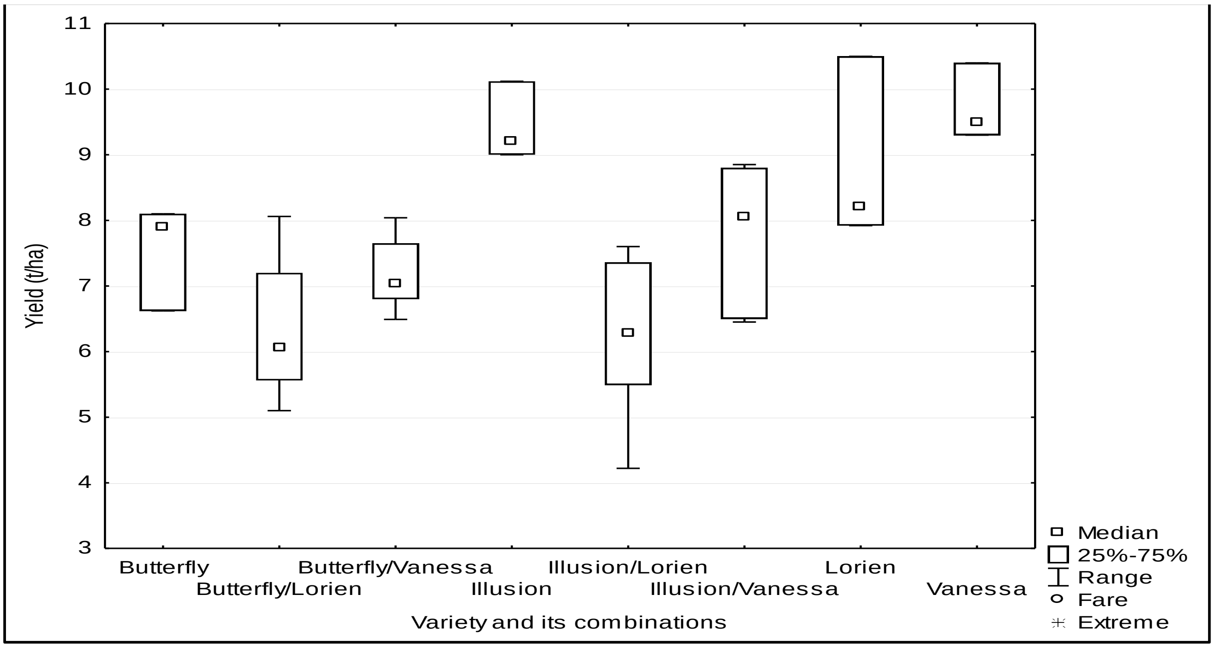

Figure 2: Yield of single seeding varieties and varieties in mixtures, Zvíkov u Českých Budějovic, 2019/2020.

4.2 Evaluation of basic baking quality characteristics of wheat varietal mixtures

The quality parameters were evaluated in three basic steps. The first table 8 shows the influence of mixture and single growing variety. In all cases, the baking quality was better in case of the single growing variety. But the differences were low and not significant statistically.

Table 8: Quality parameters of single and mixture seeding (Mean±SD) evaluated by Tukey HSD test.

Factor	Volume weight ($kg.hl^{-1}$)	Protein content (%)	Wet gluten content (%)	Zeleny test (ml)	Falling number (s)
Mixture	72,39±2,40^{a}	9,36±0,54^{a}	19,65±1,87^{a}	23±6,89^{a}	219±23,86^{a}
Single	72,78±3,19^{a}	9,53±0,70^{a}	20,82±3,18^{a}	26±7,87^{a}	226±32,56^{a}
HSD: honestly significant difference, Means labeled with the same letter within the same column are not significantly different (Tukey's HSD test).					

The second table 9 shows the results of varieties grown as single, in mixture and row by row seeding. From this evaluation, it is possible to see some differences in volume weight (statistically different) growing in the mixture and row by row. The protein content was higher in case row by row seeding. The possible explanation is related to the lowest grain yield in this combination. The relation between yield level and protein content was confirmed in organic farming (Konvalina et al., 2017). The varieties with a high yield have usually a lower protein content, because of something happens in the grain like dilution of protein (Konvalina, et al., 2017).

Table 9: Quality parameters of mixture, row by row and single seeding (Mean±SD) evaluated by Tukey HSD test.

Factor	Volume weight ($kg.hl^{-1}$)	Protein content (%)	Wet gluten content (%)	Zeleny test (ml)	Falling number (s)
mixture	71,20±1,96^{a}	9,36±0,54^{a}	20,00±2,08^{a}	23±6,77^{a}	211±21,59^{a}
row by row	74,35±1,66^{b}	9,70±0,50^{a}	21,63±1,24^{a}	23±7,31^{a}	227±23,95^{a}
single	72,39±3,19^{ab}	9,36±0,70^{a}	19,65±3,18^{a}	26±7,87^{a}	226±32,56^{a}
HSD: honestly significant difference, Means labeled with the same letter within the same column are not significantly different (Tukey's HSD test).					

The last table 10 shows only the variety and its combination independent on the different seeding styles. The highest baking quality reached in the single variety "Butterfly" and its mixtures. The variety “Vanessa” had the lowest protein content, but it was increased to 9,39 % in the mixture with the variety “Butterfly”. The final results were somewhere between these two varieties.

Table 10: Quality parameters of single combinations not take into account style of seeding (Mean±SD) evaluated by Tukey HSD test.

Factor	**Volume weight ($kg.hl^{-1}$)**	**Protein content (%)**	**Wet gluten content (%)**	**Zeleny test (ml)**	**Falling number (s)**
Butterfly	$77,12±1,87^{b}$	$10,28±0,18^{c}$	$22,12±0,40^{ab}$	$38±0,69^{f}$	$279±8,52^{c}$
Butterfly/Lorien	$73,06±0,52^{ab}$	$10,23±0,30^{c}$	$22,47±1,10^{b}$	$33±1,75^{e}$	$228±10,55^{bc}$
Butterfly/Vanessa	$73,09±1,78^{ab}$	$9,39±0,27^{a}$	$19,76±1,87^{a}$	$21±1,15^{ac}$	$247±11,32^{c}$
Illusion	$71,47±2,08^{ab}$	$9,03±0,31^{ab}$	$20,88±0,72^{ab}$	$23±0,80^{ab}$	$200±4,73^{ab}$
Illusion/Lorien	$72,62±1,00^{ab}$	$9,43±0,22^{a}$	$21,33±1,14^{ab}$	$23±0,92^{ab}$	$201±5,31^{a}$
Illusion/Vanessa	$72,33±3,71^{ab}$	$9,07±0,55^{ab}$	$19,69±1,90^{a}$	$15±3,39^{d}$	$201±41,85^{a}$
Lorien	$69,82±1,03^{a}$	$9,55±0,09^{ac}$	$21,09±0,20^{ab}$	$24±0,23^{b}$	$213±8,55^{ab}$
Vanessa	$71,15±0,10^{ab}$	$8,54±0,29^{b}$	$14,48±0,49^{c}$	$19±0,63^{c}$	$214±8,34^{ab}$
HSD: honestly significant difference, Means labeled with the same letter within the same column are not significantly different (Tukey’s HSD test).					

Table *11*: Quality parameters of single combinations (Mean±SD) evaluated by Tukey HSD test.

Combination	**Volume weight (kg.hl^{-1})**	**Protein content (%)**	**Wet gluten content (%)**	**Zeleny test (ml)**	**Falling number (s)**
Butterfly - single	77,12±1,87^{e}	10,28±0,18^{f}	22,13±0,40^{cd}	39±0,69^{h}	279±8,52^{d}
Illusion - single	71,47±2,08^{abc}	9,04±0,31^{bde}	20,89±0,72^{ab}	23±0,80^{ab}	200±4,73^{abd}
Lorien - single	69,82±1,03^{ac}	9,56±0,09^{abc}	21,10±0,20^{abc}	24±0,23^{b}	213±8,55^{ab}
Vanessa - single	71,15±0,10^{abc}	8,55±0,29^{d}	14,49±0,49^{g}	19±0,63^{c}	214±8,34^{ab}
Butterfly/Lorien - mixture	72,65±0,10^{abcd}	10,02±0,01^{cf}	21,55±0,01^{bcd}	32±0,02^{f}	218±0,44^{ac}
Butterfly/Lorien - row by row	73,48±0,37^{abde}	10,43±0,30^{f}	23,40±0,67^{f}	35±0,99^{g}	237±4,10^{c}
Butterfly/Vanessa - mixture	71,49±0,46^{abc}	9,15±0,02^{abe}	18,05±0,04^{e}	22±0,05^{a}	237±3,62^{c}
Butterfly/Vanessa - row by row	74,69±0,14^{bde}	9,64±0,02^{ac}	21,47±0,05^{abcd}	20±0,04^{c}	258±1,54^{e}
Illusion/Lorien - mixture	71,88±0,06^{abcd}	9,63±0,07^{ac}	22,37±0,17^{df}	24±0,18^{b}	205±1,19^{ab}
Illusion/Lorien - row by row	73,37±0,92^{abde}	9,24±0,07^{ab}	20,31±0,15^{a}	22±0,16^{a}	196±3,49^{bd}
Illusion/Vanessa - mixture	68,78±2,87^{c}	8,65±0,14^{de}	18,03±0,29^{e}	14±0,22^{d}	183±12,47^{d}
Illusion/Vanessa - row by row	75,87±2,82^{de}	9,49±0,32^{abc}	21,35±0,72^{abcd}	17±0,56^{e}	219±8,86^{ac}
HSD: honestly significant difference, Means labeled with the same letter within the same column are not significantly different (Tukey's HSD test).					

4.3 Evaluation of basic baking quality characteristics of wheat varietal mixtures

Mixolab II. was used to evaluate baking quality according to the ICC standard method No. 173-ICC 2006 (ICC – Standard No. 173., 2006), which allows to the characterization of physico-chemical dough behavior properties as submitted to a dual mixing and temperature constraints. The C1 torgue is related to dough development time (C1 time) and it is an essential index, which is known as the dough development or the gluten development time. The samples having a longer dough development time are related to having a better gluten quality (Konvalina et al., 2017). In the second stage C2 torgue gives the information about the weakening of proteins due to protein denaturation. The good quality wheat has the C2 value higher than 0.4 Nm (Wiwart et al., 2017). The third stage C3 torgue show the process of starch gelatinization. The fourth stage C4 torque show the resistance of starch against the enzymatic hydrolysis due to amylase agent. In the final stage (C5 torgue), the ability of retrogradation of starch granules during the cooling phase at 58 – 60 ^{0}C was assessed.

Table 12: Mixolab parameters of single and mixture seeding (Mean±SD) evaluated by Tukey HSD test, part I.

Combination	**C1 torgue (Nm)**	**C2 torgue (Nm)**	**C3 torgue (Nm)**	**C4 torgue (Nm)**	**C5 torgue (Nm)**
mixture	1,12±0,02^{a}	0,37±0,04^{a}	1,61±0,13^{a}	0,93±0,19^{a}	1,49±0,20^{a}
Single	1,12±0,04^{a}	0,38±0,03^{a}	1,66±0,18^{a}	1,00±0,26^{a}	1,73±0,40^{b}
HSD: honestly significant difference, Means labeled with the same letter within the same column are not significantly different (Tukey's HSD test).					

Table 13: Mixolab parameters of mixture, row by row and single seeding (Mean±SD) evaluated by Tukey HSD test, part I.

Combination	**C1 torgue (Nm)**	**C2 torgue (Nm)**	**C3 torgue (Nm)**	**C4 torgue (Nm)**	**C5 torgue (Nm)**
mixture	1,13±0,01^{a}	0,35±0,03^{a}	1,61±0,12^{a}	0,89±0,15^{a}	1,45±0,23^{a}
row by row	1,12±0,02^{a}	0,39±0,05^{a}	1,71±0,13^{a}	0,97±0,21^{a}	1,52±0,15^{a}
single	1,12±0,04^{a}	0,38±0,03^{a}	1,61±0,18^{a}	1,00±0,26^{a}	1,73±0,40^{a}
HSD: honestly significant difference, Means labeled with the same letter within the same column are not significantly different (Tukey's HSD test).					

Table 14: Mixolab parameters of single combinations not take into account style of seeding (Mean±SD) evaluated by Tukey HSD test, part I.

Combination	C1 torgue (Nm)	C2 torgue (Nm)	C3 torgue (Nm)	C4 torgue (Nm)	C5 torgue (Nm)
Butterfly	1,13±0,01^{ab}	0,4±0,02^{ab}	1,55±0,03^{ab}	0,88±0,03^{abcd}	1,43±0,05^{a}
Butterfly/Lorien	1,13±0,01^{ab}	0,35±0,02^{a}	1,67±0,12^{a}	0,90±0,10^{abc}	1,43±0,03^{a}
Butterfly/Vanessa	1,13±0,01^{ab}	0,41±0,05^{b}	1,70±0,07^{a}	1,05±0,02^{bcd}	1,75±0,04^{c}
Illusion	1,09±0,02^{a}	0,34±0,03^{ab}	1,37±0,07^{b}	0,65±0,01^{a}	1,27±0,0^{a}
Illusion/Lorien	1,11±0,02^{a}	0,36±0,00^{ab}	1,59±0,06^{ab}	0,78±0,02^{ab}	1,35±0,02^{a}
Illusion/Vanessa	1,13±0,01^{ab}	0,34±0,08^{ab}	1,68±0,16^{a}	0,98±0,19^{abcd}	1,39±0,30^{a}
Lorien	1,09±0,02^{a}	0,40±0,01^{ab}	1,67±0,02^{ab}	1,25±0,09^{d}	2,15±0,10^{b}
Vanessa	1,17±0,04^{b}	0,37±0,02^{ab}	1,84±0,00^{a}	1,20±0,08^{cd}	2,05±0,12^{b}
HSD: honestly significant difference, Means labeled with the same letter within the same column are not significantly different (Tukey's HSD test).					

Table 15: Quality parameters of single combinations (Mean±SD) evaluated by Tukey HSD test.

Combination	C1 torgue (Nm)	C2 torgue (Nm)	C3 torgue (Nm)	C4 torgue (Nm)	C5 torgue (Nm)
Butterfly - single	1,13±0,01^{abcd}	0,40±0,02^{bd}	1,56±0,03^{ab}	0,88±0,03^{cf}	1,43±0,05^{abd}
Illusion - single	1,09±0,02^{ae}	0,34±0,03^{ac}	1,37±0,07^{g}	0,66±0,01^{a}	1,28±0,00^{ac}
Lorien - single	1,09±0,02^{abe}	0,40±0,01^{be}	1,67±0,02^{cd}	1,26±0,09^{e}	2,15±0,10^{g}
Vanessa - single	1,17±0,04^{d}	0,37±0,02^{ab}	1,85±0,00^{ef}	1,20±0,08^{fg}	2,05±0,12^{g}
Butterfly/Lorien - mixture	1,14±0,00^{bcd}	0,36±0,00^{abc}	1,78±0,01^{def}	0,98±0,01^{df}	1,46±0,01^{bd}
Butterfly/Lorien - row by row	1,12±0,01^{abce}	0,34±0,03^{ac}	1,56±0,02^{abc}	0,81±0,01^{ab}	1,41±0,03^{ab}
Butterfly/Vanessa - mixture	1,12±0,00^{abcde}	0,37±0,01^{ab}	1,65±0,02^{ac}	1,07±0,02^{df}	1,78±0,03^{f}
Butterfly/Vanessa - row by row	1,14±0,00^{cd}	0,46±0,01^{c}	1,76±0,01^{de}	1,04±0,00^{d}	1,72±0,01^{ef}
Illusion/Lorien - mixture	1,13±0,01^{abcde}	0,37±0,00^{abc}	1,54±0,00^{ab}	0,79±0,01^{abc}	1,37±0,01^{abc}
Illusion/Lorien - row by row	1,09±0,01^{e}	0,37±0,00^{ab}	1,65±0,02^{ac}	0,77±0,03^{abc}	1,35±0,02^{abc}
Illusion/Vanessa - mixture	1,13±0,00^{abcde}	0,31±0,03^{c}	1,48±0,05^{b}	0,70±0,06^{ab}	1,19±0,12^{c}
Illusion/Vanessa - row by row	1,13±0,02^{abcd}	0,38±0,03^{ab}	1,88±0,08^{e}	1,27±0,09^{e}	1,60±0,02^{de}

HSD: honestly significant difference, Means labeled with the same letter within the same column are not significantly different (Tukey's HSD test).

The amplitude shows the elasticity of the dough. The higher the value mean the more flexible flour. The most flexible flour was detected in case of variety “Illusion” (Table 19) and generally in case of single variety seeding.

The stability shows the resistance against kneaded dough. The strong the high quality flour have the longer the duration. The highest stability was recorded in the row by row growing (Table 17). The varieties “Butterfly and Illusion” (Table 18) showed the strongest and most resistant glutein against kneading of dough.

The parameter slope α (C1-C2) shows the attenuation rate of protein in warming stage. From our results we can see very low differences (Table 19). It showed all the varieties and its combination to be similar in this parameter.

The parameter slope β (C3-C4) shows the speed of starch gelatinization. From our results we can see more differences between varieties and their seeding. From (Table 17) we can see the more favourable speed of starch gelatinization in case of row by row growing. The mixtures consisting of the variety “Butterfly” had this parameter more favourable too. The best starch speed gelatinization was found in the Butterfly/Lorien - mixture and Butterfly/Vanessa - row by row.

The last parameter slope γ (C5–C4) shows the speed of enzymatic degradation of starch. (Konvalina, et al., 2017). The number more close to zero show more favourable speed of enzymatic degradation of starch. There were low differences between single and mixture seeding. The unfavourable speed of starch degradation showed in Illuson/Lorien - mixture. Generally, the better results of starch degradation were showed in case of single growing varieties.

Table *16*: Quality parameters of single and mixture seeding (Mean±SD) evaluated by Tukey HSD test, part II.

Combination	**Amplitude (Nm)**	**Stability (min)**	**Alfa**	**Beta**	**Gama**
mixture	0,08±0,03[a]	4,51±1,84[a]	-0,08±0,01[a]	0,43±0,08[a]	-0,09±0,03[a]
single	0,09±0,02[a]	5,94±1,00[b]	-0,08±0,01[a]	0,49±0,07[b]	-0,08±0,02[a]
HSD: honestly significant difference, Means labeled with the same letter within the same column are not significantly different (Tukey’s HSD test).					

Table 17: Quality parameters of mixture, row by row and single seeding (Mean±SD) evaluated by Tukey HSD test, part II.

Combination	Amplitude (Nm)	Stability (min)	Alfa	Beta	Gama
mixture	$0{,}10 \pm 0{,}03^{a}$	$4{,}94 \pm 1{,}80^{a}$	$-0{,}08 \pm 0{,}01^{a}$	$0{,}48 \pm 0{,}10^{a}$	$-0{,}08 \pm 0{,}03^{a}$
row by row	$0{,}07 \pm 0{,}02^{a}$	$6{,}94 \pm 1{,}29^{a}$	$-0{,}08 \pm 0{,}01^{a}$	$0{,}50 \pm 0{,}05^{a}$	$-0{,}10 \pm 0{,}01^{a}$
single	$0{,}09 \pm 0{,}02^{ab}$	$4{,}51 \pm 1{,}00^{a}$	$-0{,}08 \pm 0{,}01^{a}$	$0{,}43 \pm 0{,}07^{a}$	$-0{,}08 \pm 0{,}02^{a}$
HSD: honestly significant difference, Means labeled with the same letter within the same column are not significantly different (Tukey's HSD test).					

Table 18: Quality parameters of single combinations not take into account style of seeding (Mean±SD) evaluated by Tukey HSD test, part II.

Combination	Amplitude (Nm)	Stability (min)	Alfa	Beta	Gama
Butterfly	$0{,}08 \pm 0{,}00^{ab}$	$5{,}35 \pm 0{,}15^{a}$	$-0{,}07 \pm 0{,}01^{a}$	$0{,}45 \pm 0{,}01$	$-0{,}07 \pm 0{,}00^{ab}$
Butterfly/Lorien	$0{,}07 \pm 0{,}02^{ab}$	$6{,}52 \pm 1{,}24^{a}$	$-0{,}08 \pm 0{,}01^{a}$	$0{,}55 \pm 0{,}05^{c}$	$-0{,}07 \pm 0{,}01^{a}$
Butterfly/Vanessa	$0{,}07 \pm 0{,}03^{ab}$	$5{,}95 \pm 1{,}87^{a}$	$-0{,}07 \pm 0{,}00^{a}$	$0{,}54 \pm 0{,}04^{bc}$	$-0{,}07 \pm 0{,}04^{a}$
Illusion	$0{,}12 \pm 0{,}00^{b}$	$3{,}30 \pm 0{,}10^{a}$	$-0{,}08 \pm 0{,}00^{a}$	$0{,}35 \pm 0{,}08^{a}$	$-0{,}08 \pm 0{,}01^{ab}$
Illusion/Lorien	$0{,}09 \pm 0{,}03^{ab}$	$5{,}57 \pm 1{,}65^{a}$	$-0{,}08 \pm 0{,}00^{a}$	$0{,}43 \pm 0{,}05^{a}$	$-0{,}11 \pm 0{,}01^{b}$
Illusion/Vanessa	$0{,}09 \pm 0{,}03^{ab}$	$5{,}70 \pm 2{,}44^{a}$	$-0{,}08 \pm 0{,}01^{a}$	$0{,}44 \pm 0{,}11^{ab}$	$-0{,}09 \pm 0{,}02^{ab}$
Lorien	$0{,}09 \pm 0{,}01^{ab}$	$5{,}45 \pm 0{,}05^{a}$	$-0{,}08 \pm 0{,}00^{a}$	$0{,}44 \pm 0{,}09^{abc}$	$-0{,}08 \pm 0{,}04^{ab}$
Vanessa	$0{,}06 \pm 0{,}01^{a}$	$3{,}95 \pm 0{,}65^{a}$	$-0{,}07 \pm 0{,}01^{a}$	$0{,}48 \pm 0{,}01^{abc}$	$-0{,}10 \pm 0{,}00^{ab}$
HSD: honestly significant difference, Means labeled with the same letter within the same column are not significantly different (Tukey's HSD test).					

Table 19: Quality parameters of single combinations (Mean±SD) evaluated by Tukey HSD test.

Combination	Amplitude (Nm)	Stability (min)	Alfa	Beta	Gama
Butterfly - single	0,08±0,00[abc]	5,35±0,15[ac]	-0,07±0,01[ab]	0,45±0,01[abcd]	-0,07±0,00[ac]
Illusion - single	0,12±0,00[d]	3,30±0,10[b]	-0,08±0,00[acde]	0,36±0,08[b]	-0,08±0,01[abc]
Lorien - single	0,09±0,01[abcd]	5,45±0,05[ac]	-0,08±0,00[abc]	0,44±0,09[abc]	-0,08±0,04[abc]
Vanessa - single	0,06±0,01[a]	3,95±0,65[ab]	-0,07±0,01[ab]	0,48±0,01[abcd]	-0,10±0,00[ab]
Butterfly/Lorien - mixture	0,06±0,00[ab]	7,65±0,25[d]	-0,09±0,00[d]	0,59±0,03[d]	-0,07±0,01[ac]
Butterfly/Lorien - row by row	0,09±0,03[abcd]	5,40±0,10[ac]	-0,08±0,00[ab]	0,52±0,03[ad]	-0,08±0,01[abc]
Butterfly/Vanessa - mixture	0,10±0,01[bcd]	4,25±0,25[ab]	-0,07±0,00[b]	0,52±0,05[ad]	-0,04±0,01[c]
Butterfly/Vanessa - row by row	0,05±0,01[a]	7,65±0,05[d]	-0,08±0,00[abc]	0,56±0,01[ad]	-0,10±0,02[ab]
Illusion/Lorien - mixture	0,12±0,01[d]	4,55±1,05[ab]	-0,08±0,00[ace]	0,44±0,08[abc]	-0,12±0,01[b]
Illusion/Lorien - row by row	0,07±0,01[ab]	6,60±1,60[cd]	-0,09±0,00[cde]	0,43±0,01[abc]	-0,11±0,00[ab]
Illusion/Vanessa - mixture	0,11±0,02[cd]	3,30±0,80[b]	-0,07±0,00[b]	0,37±0,04[bc]	-0,08±0,01[ab]
Illusion/Vanessa - row by row	0,07±0,01[abc]	8,10±0,10[d]	-0,09±0,01[de]	0,50±0,03[acd]	-0,10±0,00[ab]
HSD: honestly significant difference, Means labeled with the same letter within the same column are not significantly different (Tukey's HSD test).					

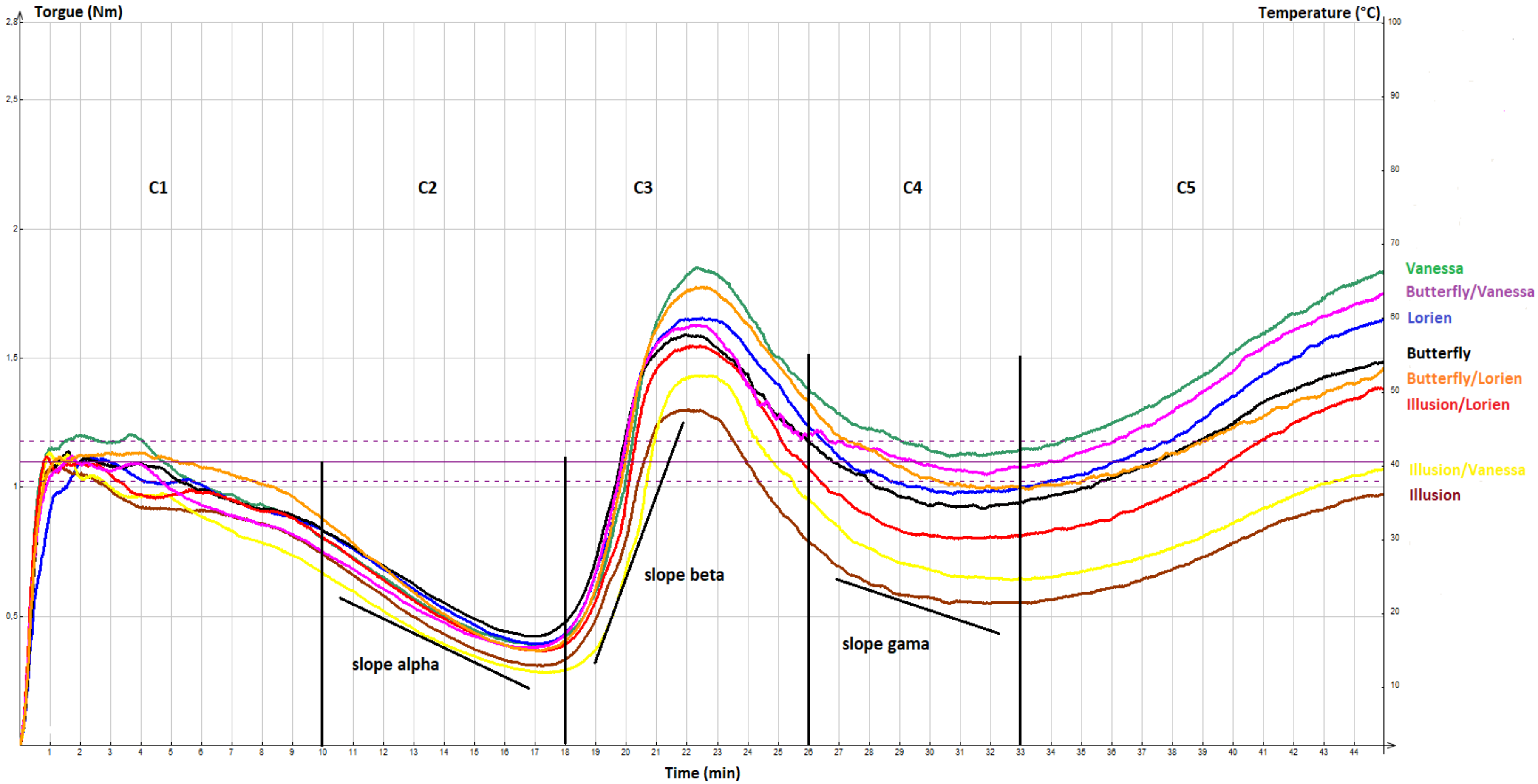

Figure 3: Mixolab curve – single varieties + varieties seeded as mixture.

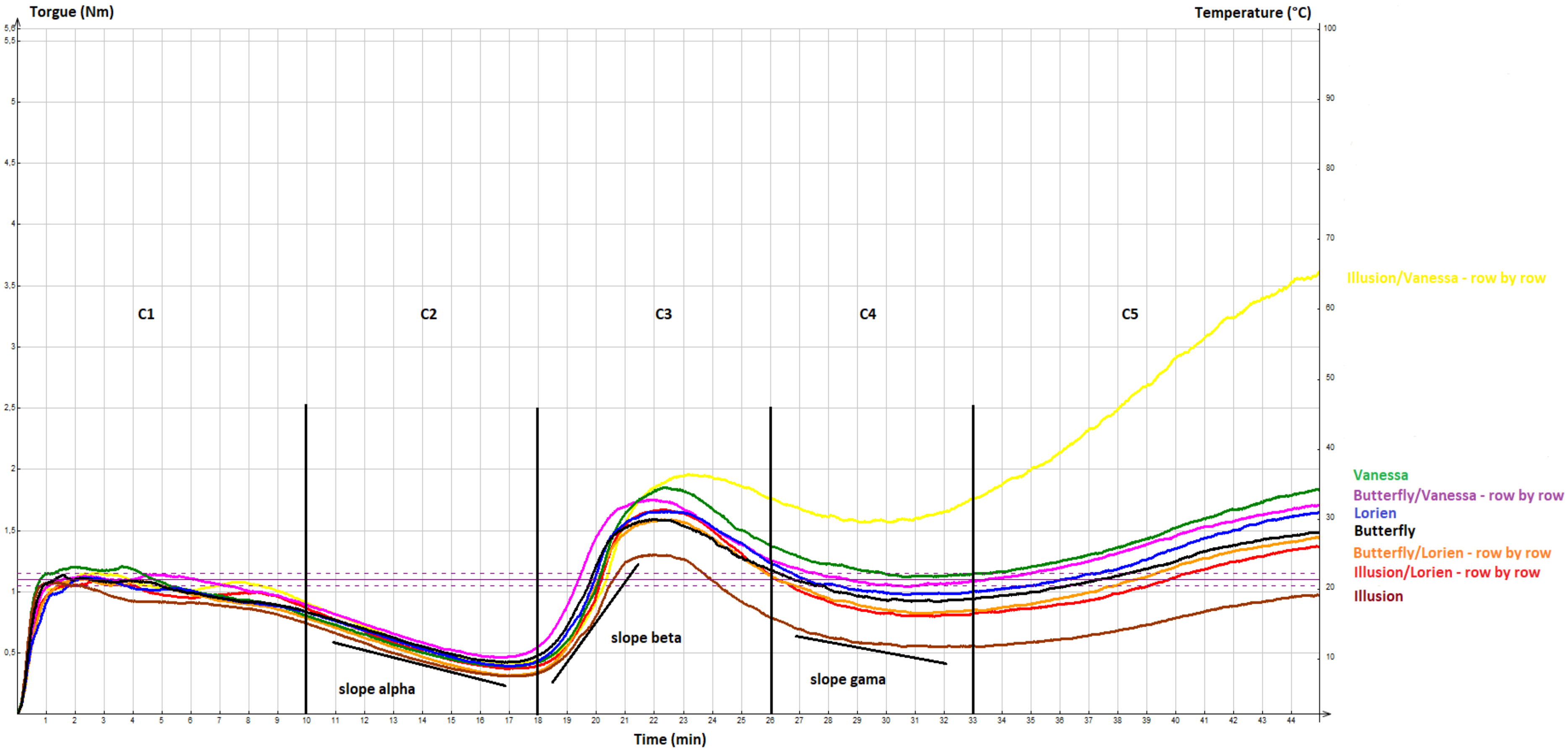

Figure 4: Mixolab curve – single varieties + varieties seeded row by row.

4.4 Evaluation of varieties by mixolab profiler

Mixolab Profiler is a feature of the Mixolab System uses the standard ICC N°173 protocol for a complete characterization of flours (protein network, starch and enzyme activity) and produces a simplified graphic interpretation of the results.

The standard curve (figures 9,10,11,12) is converted into a set of six scores graduated from 0 to 9 to characterize a flour by six fundamental criteria.

Absorption potential or Water Absorption Index: This is a function of the composition of the flour (protein, starch, fiber…). It affects dough yield (profit).

From the figures 9,10,11,12, we can see the water absorption is low in case of all the tested varieties.

Behavior in mixing or Mixing Index: This represents the behavior of the dough during mixing at 30°C (stability, development time and weakening). α A high value corresponds to high dough stability in mixing. From our results we can see the best mixing index in Lorien variety (figure 7). Other varieties had low mixing index.

Gluten strength or Gluten Index: This represents the behavior of the gluten when heating the dough. α A high value corresponds to high gluten resistance to heating. Two evaluated varieties (Butterfly, Illusion) had strength of gluten in target we need (figure 5, figure 6). The gluten of varieties Lorien and Vanessa will be difficult to process, because it will be hard (figure 7, figure 8).

Maximum viscosity or Viscosity Index: This represents the increase in viscosity during heating. It depends on both amylase activity and starch quality. α A high value corresponds to high dough viscosity during heating. Viscosity index of Butterfly, Lorien and Illusion variety was low. Vanessa showed a high viscosity of dough during the heating.

Amylase activity or Amylolysis Index: This is a function of the starch's ability to withstand amylolysis. A high value corresponds to low amylase activity. Varieties Butterfly and Illusion had high amylase activity. Varieties Lorien and Vanessa were in optimal scale (figure 7, figure 8).

Retrogradation or Retrogradation Index: This is a function of the characteristics of the starch and its hydrolysis during the test. A high value corresponds to a low shelf life of the end product. The results showed the same as amylase activity – the best values had varieties Lorien and Vanessa.

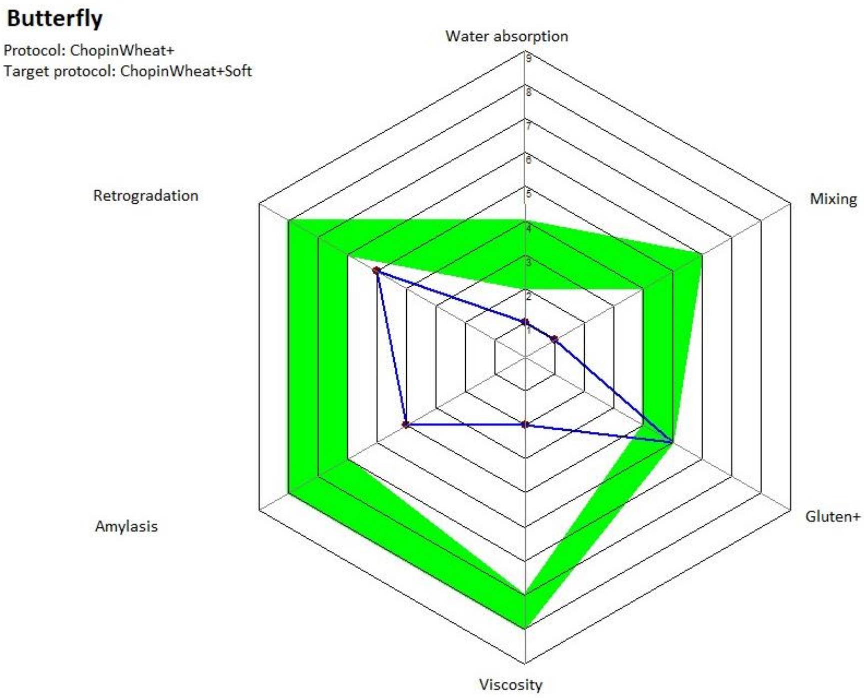

Figure 5: Complete characterization of flour by Mixolab Profiler – Butterfly.

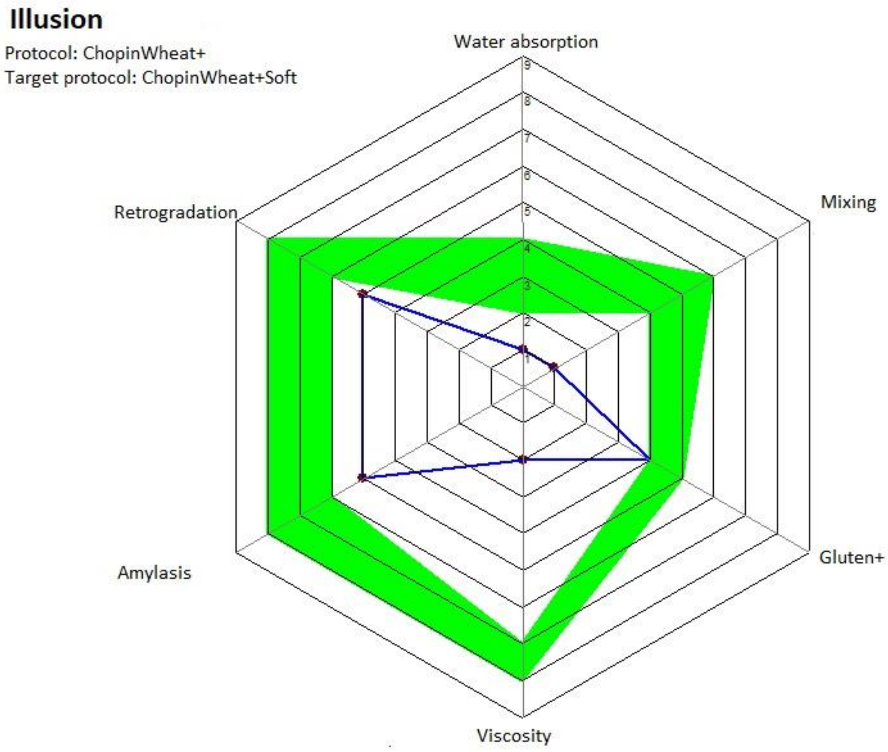

Figure 6: Complete characterisation of flour by Mixolab Profiler – Illusion.

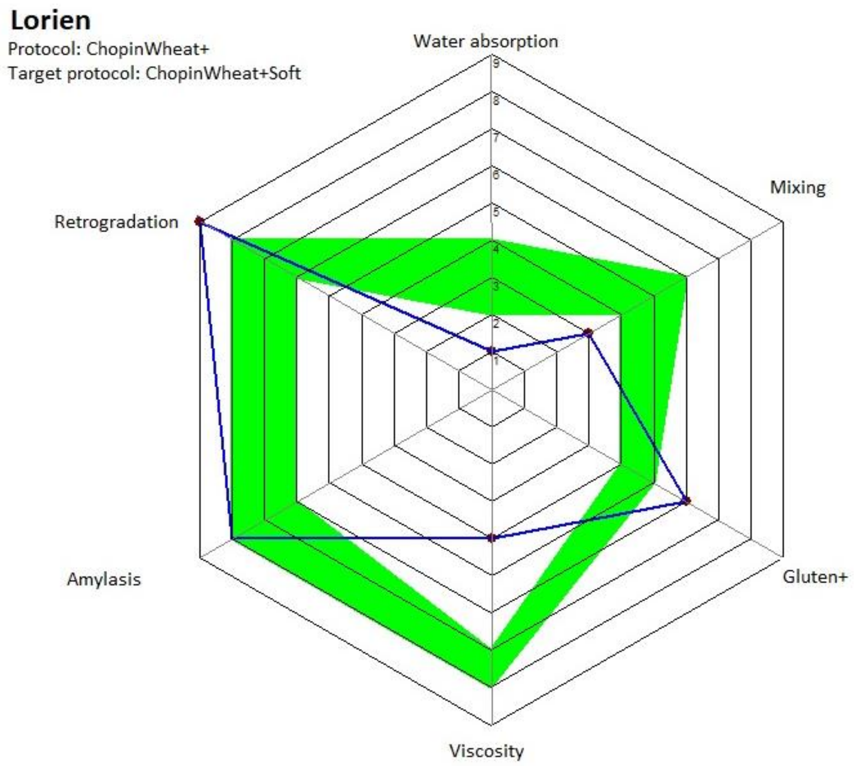

Figure 7: Complete characterisation of flour by Mixolab Profiler – Lorien.

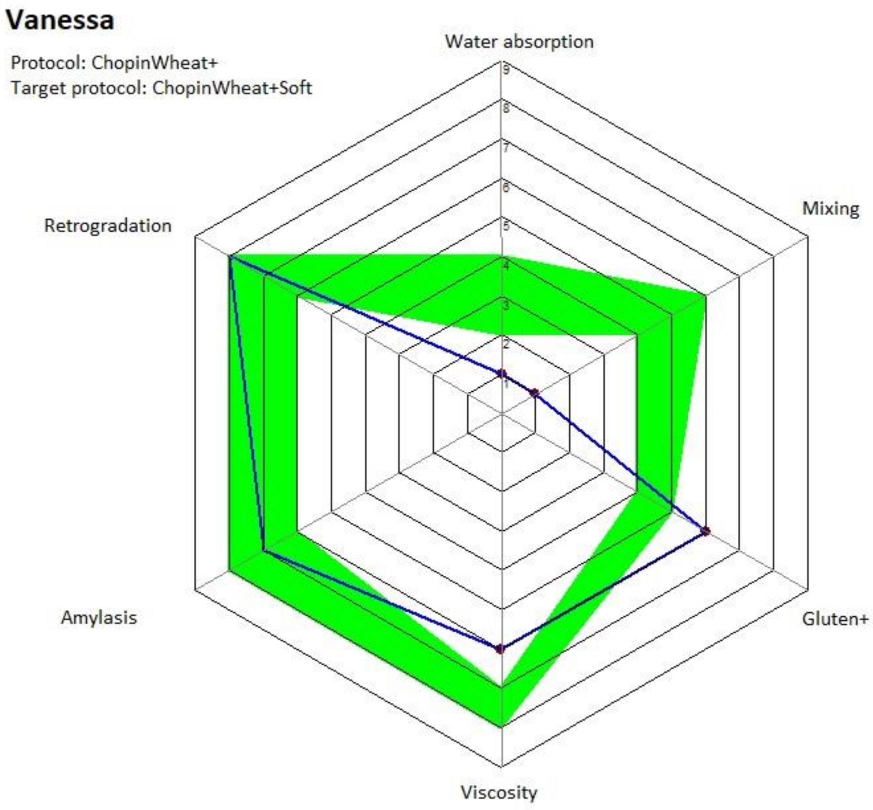

Figure 8: Complete characterisation of flour by Mixolab Profiler – Vanessa

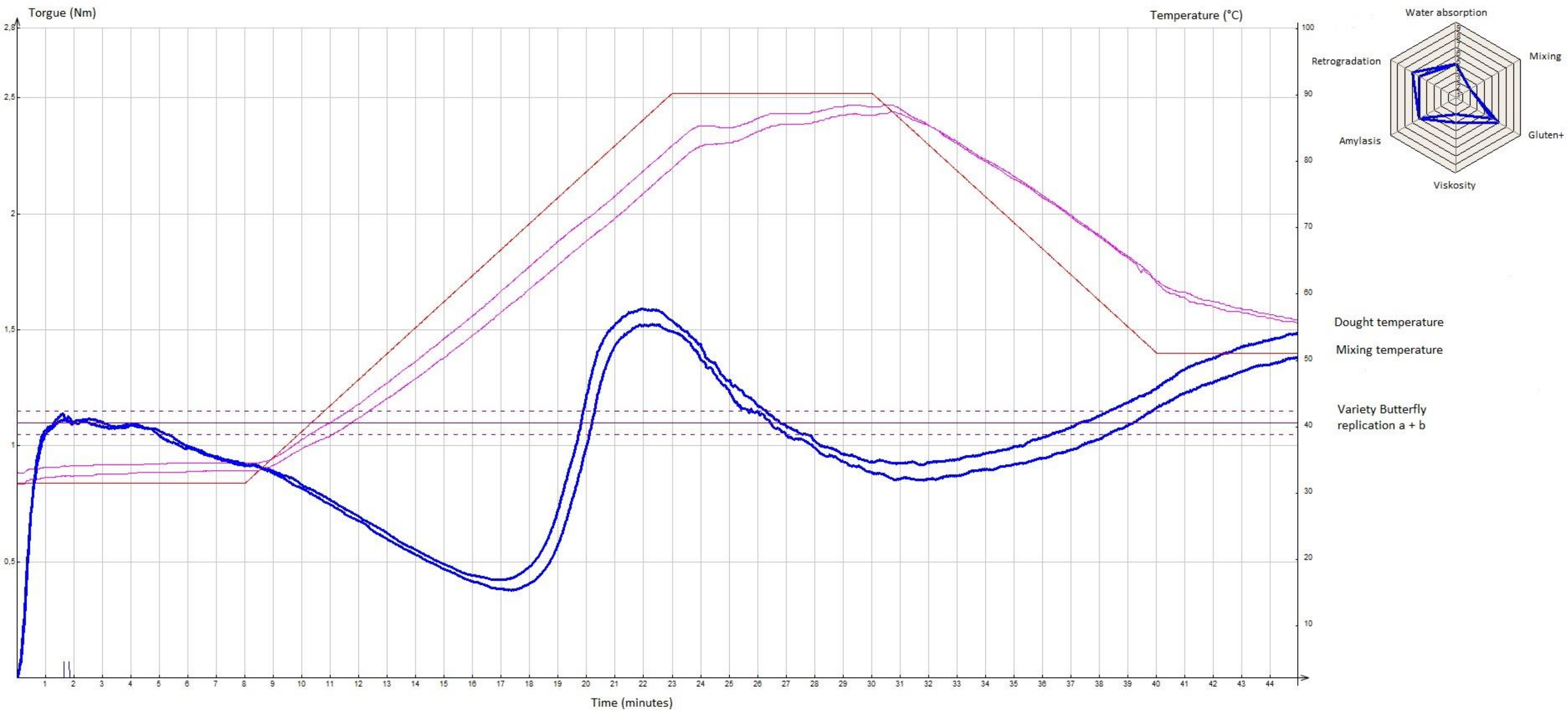

Figure 9: Standard curve of Butterfly variety.

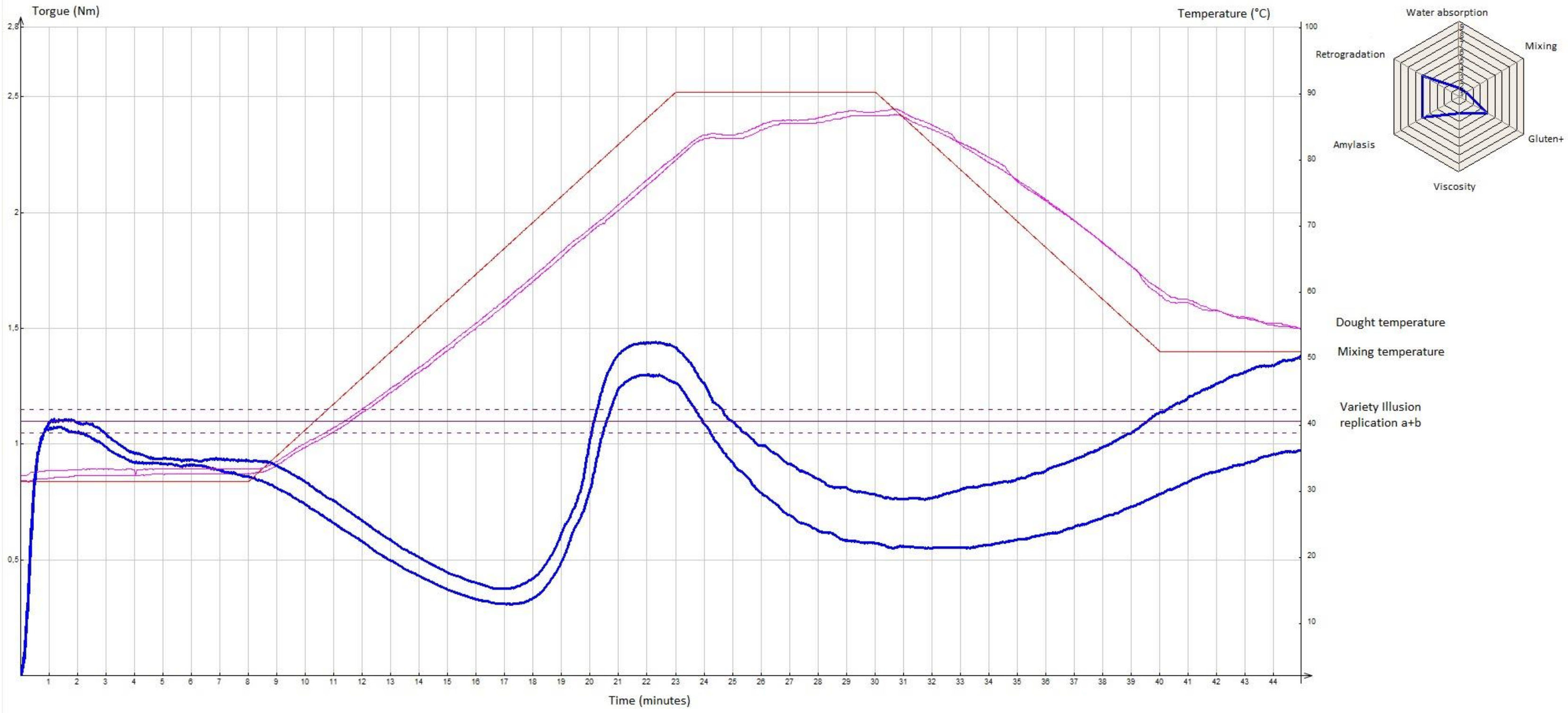

Figure *10*: Standard curve of Illusion variety.

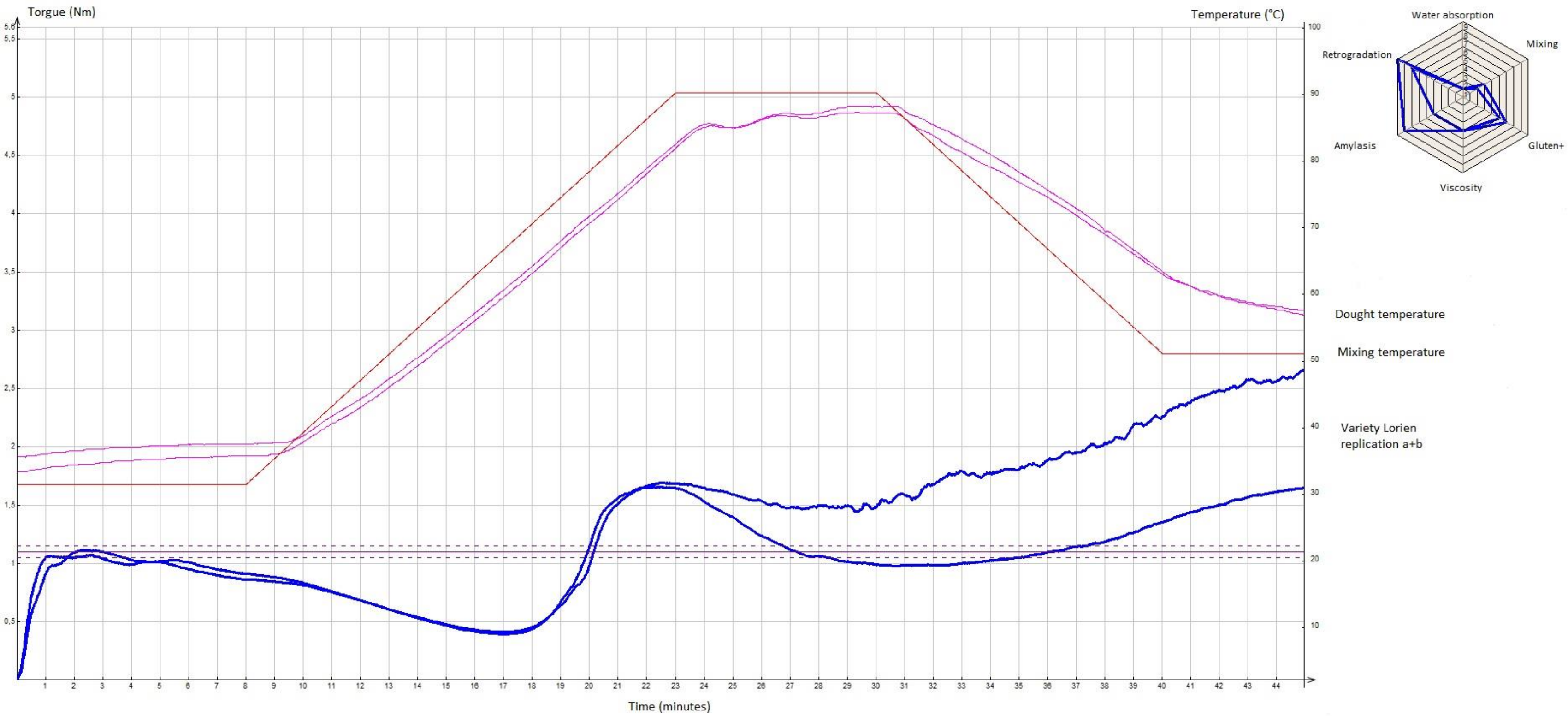

Figure 11: Standard curve of Lorien variety.

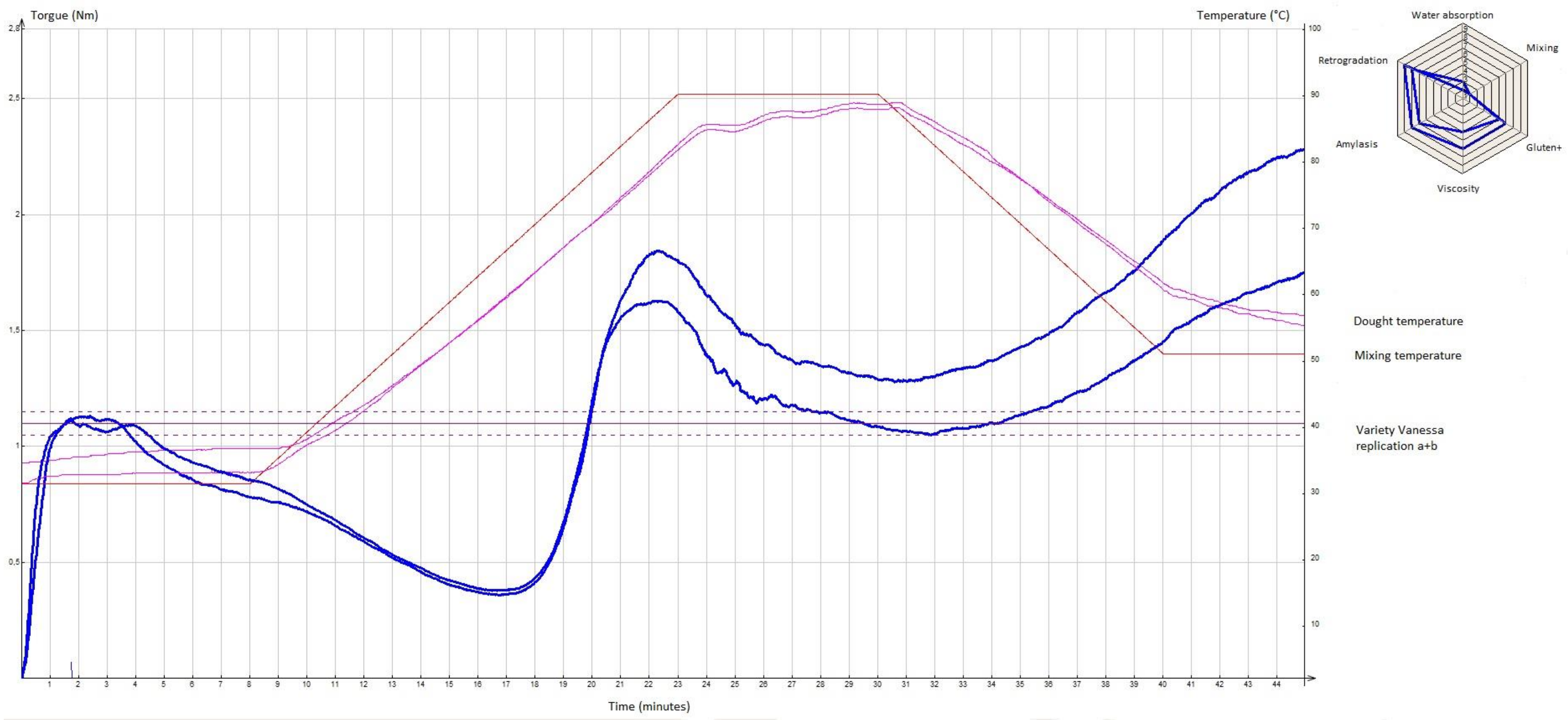

Figure 12: Standard curve of Vanessa variety.

5 Conclusions

The thesis presented a data of one-year experiment with four winter wheat varieties that were sown in a different ways of seeding/mixture. The varieties were seeded as single, two mixed varieties and two seeded varieties in row by row. The agronomically important basic characteristics were evaluated, The basic parameters of baking quality and rheological properties of flour (dough) by Mixolab II.

The length of stalk was influenced by the single variety and by a mixture of two varieties. The number of spikes before the harvest was influenced by the spacing, but the influence was low, because of the high autoregulation ability of wheat. The yield was more influenced by the variety or its combinations. The good yields were recorded in case of single grown varieties “Lorien, Butterfly and Vanessa”, and the lowest yields were in combinations of the variety “Lorien”.

The basic baking quality parameters were influenced mainly by the variety and less by the interactions between mixtures. The positive effect of seeding of row by row we saw in the volume weight parameter and protein content in the grain. The highest baking quality reached by the single variety “Butterfly” and mixtures of this variety.

The influence on the rheological properties of dough was low. There is a possibility to see some trends, but usually they are not significant statistically. The results of rheological properties were influenced more by the features of high quality of variety and less by the different seeding or mixtures.

The results were influenced by the climatic conditions. For the deep understanding of mechanism how the single/mixture/row by row seeding of varieties does influence its important agronomical characteristics, the basic baking quality parameters and rheological properties of flour need to replicate for the experiments of next two years.

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List of abbreviations

ANOVA: analysis of variance.

CP: crude protein.

FAOSTAT: food and agriculture organization corporate statistical database.

FN: falling number.

HFN: hagberg falling number.

HSD: honest significant difference.

N: nitrogen.