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Influence of selected factors on the content and properties of starch in the grain of non-food wheat

O. Faměra[1], M. Mayerová[2], I. Burešová[3], L. Kouřimská[1], M. Prášilová[4]

[1]*Department of Quality of Agricultural Products, Czech University of Life Sciences Prague, Prague, Czech Republic*
[2]*Crop Research Institute, Prague, Czech Republic*
[3]*Department of Food Technology, Faculty of Technology, Tomas Bata University in Zlín, Zlín, Czech Republic*
[4]*Department of Statistics, Faculty of Economics and Management, Czech University of Life Sciences Prague, Prague, Czech Republic*

ABSTRACT

Three non-food cultivars of winter wheat (Biscay, Clarus, Rapsodia) were grown at four locations in the Czech Republic with different agro-ecological conditions in the years 2005–2008 and two levels of nitrogen fertilization – 90 and 120 kg N/ha were used. The influence of growing conditions (year, location, cultivation technology) on grain yield, starch content, protein, gluten, share of amylose in starch, falling number and amylographic characteristics. All qualitative indicators were significantly affected by the weather. Wheat grown in the areas 500 m a.s.l. had lower grain yield of 2.8 t/ha (8.4 t/ha), higher starch content of 2.1% (69.0%) and lower content of proteins by 1.8% (10.2%) in grain than wheat grown in the fertile conditions (below 300 m a.s.l.). The cv. Biscay had the highest starch content of 68%; the proportion of amylose in the starch was in the range 22–27%. The content of proteins was negatively correlated with starch content (r = –0.89). The level of damage of starch α-amylase was most affected by wet weather during ripening – falling number was 62 s, with a very low amylographic gelatinization maximum (30–155 Brabender units) and a low gelatinization temperature at the maximum (67–74°C). The respective values recorded in dry conditions were ca 300 s, AU 600 and 90°C. Most damaged starch was recorded in the cv. Clarus.

Keywords: *Triticum aestivum* L.; climatic conditions; flour; starch production; bioethanol

Wheat (*Triticum aestivum* L.) is one of the three most important crops worldwide and is one of the major cereal crops grown in the Czech Republic. Besides growing wheat for food purposes, there is also an interest in wheat cultivars for non-food and technical uses – starch and bioethanol production (Kůst and Potměšilová 2014).

Technological quality of grain is mainly genetically determined. Therefore, the selection of cultivars for a particular purpose is very important (Bergthaller et al. 1998). Cultivars with a high starch content and larger starch granules are suitable for starch production. Most native wheat starch is a mixture of amylose and amylopectin, in the ratio of 1:3 by weight. The content of amylose in wheat starch generally ranges from 20–26% (Burešová et al. 2010). However, there are cultivars with a high amylose content (60–70%, amylotypes) and cultivars with a very low amylose content (1–2%, waxy cultivars) (Ezekiel and Rani 2000, Velíšek 2014). Starch with a high content of amylopectin is preferred in paper and textile production. High amylose starch is suitable for biodegradable plastics production because the simple straight chains of amylose enable them to bind together to create synthetic polymers (Lindeboom et al. 2004). Some starch processing technologies require not only cultivars with a high starch content, but

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Table 1. Agro-ecological conditions at localities, their long-term average temperature and annual rainfall

Locality	GPS positions	Production region	Altitude (m a.s.l.)	Temperature (°C)	Annual rainfall (mm)	FAO classification
Hněvčeves	50°18'N, 15°43'E	A	265	8.2	573	Haplic Luvisol on loess
Kostelec nad Orlicí	50°12'N, 16°21'E	A	290	7.6	681	Haplic Luvisol on loess brown
Humpolec	49°32'N, 15°32'E	B	525	6.5	667	Haplic Cambisol on paragneiss
Pernolec	49°32'N, 15°32'E	B	530	7.1	559	Cambisol on orthogneiss

A – more fertile conditions; B – medium fertile conditions

also cultivars with high enzyme activity (ethanol production) or with a high gluten content and good gluten washability (the Martin process for washing out the starch from wheat dough with tap water while it passes through a tumbling cylindrical agitator) (Day et al. 2006).

Climatic conditions and the course of the weather affect starch content in wheat grain as well. Higher starch content in wheat was recorded in crops grown in cooler regions at higher altitudes (Petr et al. 2001). In contrast, dry weather conditions caused a decrease in yield and a higher proportion of crude protein in the grain (Erekul and Köhn 2006). Cox et al. (1985) reported that low-input cultivation technology significantly affects grain yield, protein and starch content. High doses of nitrogen increased crude protein content, and reduced the starch content in the grain (Vnuk and Ložek 1995). Studies focused on the effect of nitrogen fertilization showed a high positive correlation between gluten and protein content in flour (Szentpétery et al. 1992). Increasing nitrogen fertilization also increased the proportion of water-insoluble proteins, which enhanced the gluten washability (Kelfkens and Hamer 1991).

As it is possible to grow non-food cultivars with a high starch content in areas unsuitable for food wheat, the aim of the investigation was to compare starch production in selected non-food wheat cultivars in relation to the intensity of cultivation technology and the agro-ecological conditions in the growing regions.

MATERIAL AND METHODS

Plant material and localities. Three winter wheat cultivars (Biscay, Clarus and Rapsodia) classified as category C (not suitable for bread-baking) were grown in localities Hněvčeves and Kostelec nad Orlicí in more fertile conditions (region A, Tables 1 and 2) and in localities Humpolec and Pernolec medium fertile conditions (region B, Tables 1 and 2).

The weather in individual years and different attempts to influence the formation of wheat yield, and grain quality were observed. Year 2006 was above normal rainfall. Rainy weather during wheat ripening caused damage to enzymatic starch. In 2007, warm weather created favourable conditions for harvesting grain whereas in 2008, wheat was harvested at temperatures above normal.

Field experimental methods. Wheat cultivars were grown in field plots (harvest area of 12.5 m^2) using two production technologies. The technology 1 (T1) used nitrogen fertilization at a total dose of 90 kg N/ha. The technology 2 (T2) used a total

Table 2. Average annual temperatures (°C) and annual rainfall (mm) at experimental localities during 2005–2008

Locality	2005		2006		2007		2008	
	temperature	rainfall	temperature	rainfall	temperature	rainfall	temperature	rainfall
Hněvčeves	8.5	502	8.8	460	9.9	431	9.5	406
Kostelec nad Orlicí	8.5	731	8.9	750	9.7	771	9.6	618
Humpolec	8.0	864	8.4	792	9.2	709	9.1	626
Pernolec	7.6	403	8.8	607	10.6	724	8.2	546

Data are from local weather stations located in the experimental fields

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dose of 120 kg N/ha. In both technologies 60 kg N/ha was applied as ammonium nitrate with lime for regeneration in early spring and 30 kg N/ha in 29 BBCH (BBCH – scale of growth stages for cereals), together with weed control and one treatment against fungal diseases. In case of T1 30 kg N/ha of urea in 36 BBCH, another fungicide Tango Super 1 L/ha (*Blumeria graminis*), growth regulator Cycocel 750 SL (BASF, Germany, active substance chlormequat chloride 65.8%) in 29 BBCH and insecticide protection as necessary were applied – Decis Flow 2.5, 0.3 L/ha (Oulema). Spring barley was grown as a pre-crop cereal.

Analytical methods. Three kilograms of grain from each locality were harvested according to the ISO 24333, 2009. The following parameters were determined: grain yield, starch content (ISO 10520, 1997), proportion of amylose in the starch, protein content (ICC 105/2, 1994), wet gluten content in the grain (ICC 155, 1994), and falling number (ISO 3093, 2009). Amylose proportion in starch was determined according to the modified method of Yun and Matheson (1990).

Statistical analysis. Statistical analyses, multiple-way ANOVA method and the Scheffe's *HSD* test with 5% level of significance (α), were performed using the software Statistica 9 (StatSoft, Inc., Tulsa, USA).

RESULTS AND DISCUSSION

Grain yield was significantly affected by weather conditions and by agro-ecological production region (interaction $P = 0.0021$, Table 3). More fertile conditions in lower-altitude regions (A regions) increased the wheat yield (the average value was 11.2 t/ha), while in B regions the average yield was lower by 2.6 t/ha. Technology 2 showed only an average yield increase of 0.4 t/ha over the technology 1.

The content of the main wheat grain components, starch and protein, depended on several factors – year, production area, cultivars and technology, but the interactions among them were not significant. The starch and protein contents negatively correlated ($r = -0.84$). According to Muchová (2003), weather affected all quality indicators of wheat. The least favourable conditions for the formation of starch grains were in 2006 when the starch contents were around 66%, but the protein content was the highest (12.3%).

The high starch content in 2007 (67.9%) and 2008 (68.5%) resulted in the lowest protein content (10.6% and 10.3%, respectively). Higher starch content in the grain (69.0%) occurred in B regions than in A regions (66.9%). These findings are consistent with those of Petr et al. (2001). The lowest starch

Table 3. Average results of selected quality indicators of wheat in relation to agro-ecological factors

Factor	Factor specification	Grain yield (t/ha)	Starch (%)	Amylose (%)	Protein (%)	Gluten (%)	Falling number (s)	Amgr. max. (AU)	Amgr. temp. (°C)
Growing region	A	11.2^{a}	66.9^{a}	24.8^{a}	12.0^{b}	31.0^{a}	219^{a}	323^{a}	78^{a}
	B	8.4^{b}	69.0^{b}	25.3^{a}	10.2^{a}	22.4^{b}	221^{a}	253^{a}	78^{a}
Year	2006	8.9^{a}	65.9^{a}	24.3^{a}	12.3^{b}	27.7^{a}	97^{a}	106^{a}	73^{a}
	2007	10.2^{ab}	67.9^{b}	25.6^{b}	10.6^{a}	25.0^{a}	263^{b}	324^{b}	81^{b}
	2008	10.3^{b}	68.5^{b}	25.2^{ab}	10.3^{a}	27.4^{a}	299^{c}	434^{b}	80^{b}
Cultivar	Biscay	10.0^{a}	68.0^{b}	24.9^{a}	10.8^{a}	28.1^{a}	277^{c}	431^{c}	84^{c}
	Clarus	9.3^{a}	67.3^{a}	25.3^{a}	11.5^{b}	27.0^{a}	163^{a}	120^{a}	71^{a}
	Rapsodia	10.1^{a}	66.9^{a}	24.9^{a}	11.0^{ab}	25.0^{a}	220^{b}	312^{b}	79^{b}
Technology	1	9.6^{a}	67.7^{a}	24.9^{a}	10.9^{a}	25.8^{a}	215^{a}	283^{a}	78^{a}
	2	10.0^{a}	67.2^{b}	25.2^{a}	11.3^{a}	27.6^{a}	225^{a}	293^{a}	79^{a}
	SD	2.2	1.5	1.43	1.5	5.99	112*	247*	9*

Values with different letters (a, b, c) are significantly different (α = 0.05). Technology: 1 – 90 kg N/ha, 2 – 120 kg N/ha; SD – standard deviation; Amgr. max. – gelatinization maximum measured by amylograph; Amgr. temp. – gelatinization temperature at maximum measured by amylograph; AU – Brabender units; * results were affected by high α-amylase activity in 2006

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content (66.8%) was in grain from the locality with the highest grain yields (Hněvčeves). In more fertile areas the increased intake of nitrogen promotes the formation of protein at the expense of starch (Erekul and Köhn 2006). Lower nitrogen fertilization with low input technologies resulted in higher starch content (by 0.5% on average) compared to intensive nitrogen fertilization. Similar results were obtained by Crista et al. (2012). Capouchová et al. (2002) stated that starch is formed mainly at the milk stage of grain ripening. The highest starch content (68.0%) was found in cv. Biscay grain. Other cultivars showed values which were lower by 0.6–1.0%. The average protein content in cv. Biscay was 10.8%. Cvs. Rapsodia and Clarus had higher protein contents (11.0% and 11.5%, respectively) and simultaneously lower starch contents.

Gluten content affects the suitability of wheat grain for industrial production of wheat starch using the Martin process (Van Der Borght et al. 2005). Gluten content was closely related to the protein content (r = 0.71), in agreement with the observations of Szentpétery et al. (1992). Significantly higher levels of gluten (31.0%) were found in A regions than in B regions (23.3%). Study of Konvalina et al. (2014) shows protein genetic sources of protein for low intensity of cultivation (organic farming).

Significant interaction (P = 0.0133) was found between location, year of cultivation and amylose content. The proportion of amylose ranged from 22–27%, similarly to Velíšek (2014). The average amylose content during the three-year evaluation was almost identical in Kostelec nad Orlicí (25.4%), Humpolec (25.2%) and Pernolec (25.2%). The cultivation technology, locality and wheat cultivar did not affect the proportion of amylose in the starch. Blazek et al. (2009) reported that the amylose content affected quality of noodles and bread and in the soft wheat it strongly correlated with the diameter of cookies (r = 0.969; P < 0.001).

Damage to the structure of starch granules by amylases plays a significant role in the use of wheat starch. Grains with a high activity of amylases (low falling number) are technologically less suitable for starch production (Every et al. 2002). Long periods of rainy weather and harvest delay in 2006 were responsible for high α-amylase activity and starch damage in the caryopses, as well as for low falling number value (62 s) at three localities (Hněvčeves, Kostelec nad Orlicí and Humpolec). This was associated with a very low amylographic gelatinization maximum (30–155 Brabender units) as well as a low gelatinization temperature at the maximum (67–74°C). If grain maturation takes place in dry conditions the activity of α-amylase is low: falling number, usually above 350 s and amylographic gelatinization maximum over 600 Brabender units at a temperature above 90°C (Faměra et al. 2011).

Falling number value closely correlates with amylographic results (r > 0.80) (Table 4). Cv. Clarus was the most susceptible to the enzymic starch damage. In 2007 and 2008 the falling number values ranged from 280–340 s except for cv. Clarus (207 s). In relation to these factors, significant interactions between cultivar and year of cultivation (P = 0.0025) and between location and year (P = 0.0003) were found (Table 5).

Table 4. Pair correlation coefficients (r) of wheat quality parameters

Parameter	Grain yield	Starch	Amylose	Protein	Gluten	Falling number	Amgr. max.	Amgr. temp.
Grain yield	–							
Starch	–0.003	–						
Amylose	–0.001	0.258	–					
N-comp.	0.309	**–0.839**	–0.242	–				
Gluten	0.543	–0.422	–0.146	**0.712**	–			
Falling number	0.341	0.600	0.330	–0.461	0.050	–		
Amgr. max.	0.386	0.357	0.219	–0.279	0.187	**0.848**	–	
Amgr. temp.	0.246	0.282	0.195	–0.253	0.095	**0.817**	**0.883**	–

Strong relationships (r > 0.7) are in bold. Amgr. max. – gelatinization maximum measured by amylograph; Amgr. temp. – gelatinization temperature at maximum measured by amylograph

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Table 5. Multi-way analysis of variance with interactions (α = 0.05) of the relationships of individual quality indicators and characteristics of wheat and their interactions

Factor	*P*							
	grain yield	starch	amylose	protein	gluten	falling number	Amgr. max.	Amgr. temp.
Growing region	**< 0.001**	**< 0.001**	0.114	**< 0.001**	**< 0.001**	0.837	0.054	1.000
Cultivar	0.315	**0.001**	0.462	**0.024**	0.092	**< 0.001**	**< 0.001**	**< 0.001**
Year	**0.023**	**< 0.001**	**0.007**	**< 0.001**	0.111	**< 0.001**	**< 0.001**	**< 0.001**
Technology	0.295	**0.032**	0.333	0.051	0.101	0.388	0.794	0.705
Growing region × cultivar	0.871	0.839	0.368	0.795	0.257	0.800	0.232	0.183
Growing region × year	**0.002**	0.407	**0.013**	0.160	0.799	**< 0.001**	**< 0.001**	**0.001**
Cultivar × year	0.940	0.634	0.392	0.888	0.996	**0.005**	**0.014**	0.052
Growing region × technology	0.785	0.083	1.000	0.086	0.237	0.490	0.705	0.737

Statistically significant values ($P < \alpha$) are in bold. Amgr. max. – gelatinization maximum measured by amylograph; Amgr. temp. – gelatinization temperature at maximum measured by amylograph

In conclusion, the weather conditions (year) and location were the most important factors of the measured parameters of winter wheat. These factors affected both the grain yield and its quality. Poor agro-ecological conditions and a lower level of nitrogen nutrition resulted in an increase in starch content in the caryopses and in a reduction of protein and gluten content (significant negative correlation), making the grain suitable for the production of bioethanol. Fertile conditions are more suitable for the production of starch because of the increased gluten content and its better washability.

Cv. Biscay had a higher starch content than the other tested cultivars. Cv. Clarus had the lowest grain yield and was the most susceptible to starch damage by α-amylase. The average content of amylose in the starch was 25%. Not only the starch content, but also other quality indicators (protein and gluten content, gluten washability) should be taken into account when selecting wheat cultivars suitable for starch or bioethanol production. The total dose of nitrogen should be up to 100 kg N/ha in more fertile conditions and up to 120 kg N/ha in medium fertile marginal localities. Last nitrogen fertilization should be done in 32 BBCH.

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Corresponding author:

Ing. Oldřich Faměra, CSc., Česká zemědělská univerzita v Praze, Fakulta agrobiologie, potravinových a přírodních zdrojů, Katedra kvality zemědělských produktů, Kamýcká 129, 165 21 Praha 6 – Suchdol, Česká republika
e-mail: famera@af.czu.cz