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POTENTIAL OF GRAIN HARDNESS OF BARLEY (*HORDEUM VULGARE* L.) AS A SELECTION TRAIT IN BREEDING PROGRAMME

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Abstract

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The study presents the results of research on the use of barley caryopses hardness for prediction of the malting quality. Grain hardness was determined by three methods: Particle size index (PSI), hardness according to Brabender (BRA) and grain milling energy (GME). The correlations among methods for determining the kernel hardness were estimated as well among selected technological traits of barley and malt and kernel hardness. Significant correlations were found among PSI values and Final attenuation of laboratory wort from malt (0.73*) a Glycidic extract (0.70*). Values of kernel hardness measured by BRA significantly correlated with Final attenuation of laboratory wort from malt (–0.80*) and Soluble nitrogen of malt (0.64*). Grain milling energy values were statistically significant correlated with Nitrogen content in malt (0.64*). The preliminary results confirmed the possibility to use kernel hardness for prediction of some malting quality traits. Further studies will be done on larger number of samples.

hardness, milling energy, quality, particle size index, do-corder, barley

Physical and mechanical properties of a barley caryopsis and consequently of malt are a reflection of their chemical composition and internal structure. Hard grain can be defined as a grain resistant to penetration of a foreign matter or resistant to destruction and breakdown to particles. On the contrary, soft grain can be defined as a grain easily breaking down under pressure. Texture, i.e. organization of individual grain components, first of all endosperm, decides whether the grain will be hard or soft (ALLISON et al., 1976; BRENNAN et al., 1996; CHANDRA et al., 1999; MACGREGOR, 1991; PALMER, 1999). There were developed many methods and devices for hardness analysis (MOLL, 1996). Three of ones were used in this work.

For the production of quality malts it is necessary to modify cell walls, starch and surrounding protein matrix during malting (PALMER, 1993). This enzymatic destruction is significantly affected both by the activity of the enzymatic apparatus and quality of the substrate affected by the enzymes. Physical and mechanical characters of a hulled barley caryopsis and malt are most significantly affected by the characters of endosperm and hulls. The terms mealiness and glassiness are used for the description of endosperm characteristic. The glassy endosperm is degraded more slowly than the mealy one (CHANDRA et al., 1999). Varieties suitable for the production of good quality malts have a similar endosperm structure. Based on the current results, this type of the structure is denoted as mealy. Subjective evaluation of mealiness and glassiness is replaced by objective methods (KOLIATSOU and PALMER, 2003; OSBORNE and ANDERSSEN, 2003).

β-glucans and arabinoxylans are the main components of cell walls of barley endosperm. An increased amount of non-starch polysaccharides in the endosperm creates an obstacle for the enzymatic destruction of the cell content. The increased content of β-glucans and arabinoxylans is therefore connected with the reduced degree of modification of malt (MARTIN and BAMFORTH, 1980). High β-glucan content in wort increases its viscosity, which leads to filtration problems (BAMFORTH and BARCLAY, 1993). β-glucans and arabinoxylans also affect physical properties of grain, first of all its hardness (SWANSTON, 1997; TOHNO-OKA et al., 2004).

We tried to detect serviceability of three methods characterized caryopses hardness for selection in breeding programme. There were calculated the correlations between caryopses hardness and selected technological traits of barley and malt.

MATERIALS AND METHODS

Cultivars

Particle size index (PSI), hardness according to Brabender (BRA) and grain milling energy (GME) were determined in ten varieties of spring barley with different malting quality (Tab. I). Seed samples were obtained from testing station of the Central and Testing Institute in agriculture of the Slovak Republic. Monitoring was held for one year (2003). The samples were graded and sieving fractions above 2.5 mm were used for the assessment.

I: *Characterization of tested cultivars*

Varieties	Malting quality index	Quality in Slovak Republic (SR)	Country of origin
Atribut	3	A	CZ
Danuta	3	B	DE
Ludan	2	B	SK
Annabell	3	A	DE
Expres	3	A	SK
Jubilant	2	A	SK
Pedant	2	A	CZ
Celinka	3	A	FR
Nitran	5	A	SK
Ebson	4	A	CZ

Note: Quality SR: A = excellent; B = standard
MQI: 9 = the best quality; 1 = without malting quality

Micromalting and Analyses of malt

A micromalting test procedure follows the traditional method used in the Malting Institute of the RIBM in Brno and the trial malting method accepted on the 102nd session of the EBC Barley and Malt Committee in Perugia on May 31, 2000. Temperature of steeping and germination was 14.5 °C, time of steeping and germination was 144 hours. Temperature at the beginning of kilning was 50 °C and at the end 80 °C, time of kilning was 22 h. The malt samples produced were analyzed according to the EUROPEAN BREWERY CONVENTION ANALYSIS (1998) and ANALYSENKOMMISSION (1997) methods. The malting quality index (MQI) (PSOTA and KOSAŘ, 2002) was used for the assessment of the total malting quality. For the MQI calculation the following parameters were used: content of nitrogenous substances (protein) in barley grain, extract in malt dry matter, relative extract at 45 °C, Kolbach index, diastatic power, apparent final attenuation, malt friability and β-glucan content in wort.

Hardness measurement

There were used three technically different methods for determining the caryopses hardness.

Particle Size Index Measurements

Particle size index method is based on different ability to disintegrate of caryopses during the milling (AACC, 2000). Relative hardness of barley was detected through determination of the particle size index (PSI – Particle size index for hardness) by milling the sample and sieving on the sieve 75 μm (0.075 mm) (WILLIAMS and SOBERING, 1986). Twenty-three grams of grain was milled on the laboratory mill LM 3303 (Perten Instruments) with head no. 2. Ten grams of the milled sample was taken and placed into the sifter Swing 200 (Mezos, CZ) and sieved at 180 [$1*min^{-1}$] for 10 min. The particles that fell through the sieve (through) were weighed. Each sample was measured six times. The particle size index (PSI) was calculated using the following equation:

PSI [%] = (throughs [g] / sample mass [g]) × 100

Lower PSI values mean harder endosperm (AACC, 2000) (in the text and tables, the results obtained by this method are marked with the abbreviation "PSI").

Hardness according to Brabender

The method is based on measure of resistance, which are originated in conic mill during the milling. From each sample four 50 g of barley grain were taken for measurement. Hardness was assessed on the apparatus Do–Corder of the firm Brabender. Each sample was measured four times. Grain grinding was recorded graphically and hardness was expressed by the peak area in cm^2 (in the text and tables, the results obtained by this method are marked with the abbreviation "BRA"). Higher BRA values mean harder endosperm.

Determination of grain milling energy

The method is based on measure of required energy to milling the sample. Output of alternating current was measured with the device HR 2672/mini (1-Cube Ltd., CZ), connected in electrical network before the mill SJ 500 (Swantech Int., F). Periodicity of measurement was 200 ms. Energy (in joules) necessary for milling the sample was calculated from the achieved values. Each sample was measured three times. Aperture 7.75 was used for milling 10 g of samples.

Moisture of the samples was measured using a non-destructive moisture meter Supermatic 13610 (Foss, DK) and it ranged from 10.5 to 11.8 %. (In the text and tables, the results obtained by this method are marked with the abbreviation "GME"). Higher GME values mean harder endosperm.

The achieved data were evaluated using one factorial analysis of variance and Tukey HSD test with the statistical program STATISTICA, Ver. 7.0. ($P \leq 0.05$). Relationships among PSI, BRA, GME and selected technological parameters of barley and malt were evaluated using the correlation analysis of mean values.

RESULTS AND DISCUSSION

Effect of the variety on the barley caryopsis hardness

The methods used for characterization of caryopses hardness are based on technically different principles. This is the reason, why we found only low correlations among these methods (Tab. IV) and why the order of varieties is not similar at each of the method.

Hardness of a barley grain is affected by a number of factors, first of all by the variety (ALLISON, 1986). In the studied set, statistically significant difference between the varieties was found in all methods used for the measurement of hardness (Tab. II).

Based on testing with the Tukey HSD method, the set can be split into three different groups (Tab. III). Classification of the varieties in the groups was very similar in methods. These methods assigned Nitran to the softer varieties. On the contrary, the variety Atribut was assigned to the harder varieties. ALLISON et al. (1979, 1976) as well classified the varieties with different malting quality using milling energy. Other varieties created an intermediate group. The disagreement among methods was registered at Danuta variety. The PSI method and BRA method assigned the variety Danuta to the harder varieties while the GME method assigned it to the varieties with a softer grain.

Effect of the grain composition on its hardness

Quality and quantity of protein in a caryopsis significantly affect its physical characters (ALLISON et al., 1979; LEACH et al., 2002; PALMER and SHIRAKASHI, 1994). The results of the research (BROADBENT and PALMER, 2001) suggest that although the glassiness is connected with a high nitrogen content and mealiness, on the contrary, with low nitrogen content, glassy and mealy endosperm can have grains with similar nitrogen content (HOLOPAINEN et al., 2005). Hordein proteins of mealy endosperms modify faster than hordein proteins of glassy endosperms (HOWARD et al., 1996).

Range of the protein content of the individual samples of the followed set was narrow (10.8–11.9 %). The effect of protein content on the caryopsis hardness, expressed by PSI, BRA and GME was not statistically significant (Tab. IV).

Effect of the caryopsis hardness on the selected malting parameters

The glycidic extract statistically significant increased (0.70*) with the increasing of PSI values (declined caryopses hardness), which also corresponds with the correlations found by HOME and ELAMO (1993) and TAYLOR and SWANSTON (1987). There were not found relationship between extract ($-0.02^{n.s.}$) as well glycidic extract ($-0.09^{n.s.}$) and grain hardness by GME method, which also corresponds with the correlations found by ELLIS et al. (1999) and SWANSTON et al. (1992).

Protein modification expressed by amount of Soluble nitrogen in wort (0.64^{*}) was also significantly affected by the caryopsis hardness determined by BRA (Tab. IV).

Lower level of modification of starch, protein and cell walls conditioned by the enhanced hardness of barley caryopses affects negatively the quality of wort composition and therefore also the fermenting process. With the increased caryopses hardness by PSI and BRA, the wort composition deteriorates and thus the values of Final attenuation of laboratory wort decline (PSI 0.73^{*}, BRA -0.80^{*}).

In the samples of the varieties observed, a number of parameters with direct or indirect impact on the variety utilization in the malt house were determined. The Malting Quality Index can be used for the measurement of the differences in quality between varieties. With the increasing level of hardness of caryopses, the MQI value declined, however the correlations were statistically not significant (Tab. IV).

II: *Analysis of variance for three parameters of grain hardness*

	PSI		BRA		GME	
Source of variation	d.f.	Mean square	d.f.	Mean square	d.f.	Mean square
Variety	9	9.12***	9	20.91***	9	8192***
Residual	50	0.12	30	1.28	20	180

III: *Multiple range analysis for three parameters of grain hardness*

PSI [%]	n = 6		BRA [cm^2]	n = 4		GME [J]	n = 3	
Varieties	average		Varieties	average		Varieties	average	
Atribut	12.45	a	Nitran	35.31	a	Nitran	574.4	a
Danuta	13.25	b	Jubilant	37.37	a b	Danuta	624.9	b
Ludan	13.95	c	Annabell	37.65	a b	Ebson	632.6	b
Annabell	14.22	c d	Ebson	38.24	b c	Expres	645.2	b c
Expres	14.32	c d e	Ludan	38.87	b c	Pedant	662.7	b c d
Jubilant	14.47	c d e	Celinka	39.37	b c	Annabell	681.3	c d
Pedant	14.82	d e f	Expres	39.60	b c	Celinka	683.4	c d
Celinka	14.93	e f	Pedant	40.75	c	Jubilant	697.4	d e
Nitran	15.18	f	Danuta	40.81	c	Ludan	735.9	e f
Ebson	17.10	g	Atribut	43.67	d	Atribut	747.2	f

Note: Average values indicated by various letters are statistically different (P = 0.05)

IV: *Correlation coefficient between three parameters of grain hardness and parameters of grain and malt*

Parameter	**PSI**	**BRA**	**GME**
BRA	–0.60 n. s.		
GME	–0.51 n. s.	0.54 n. s.	
Malting quality index	0.49 n. s.	–0.35 n. s.	–0.56 n. s.
Grain moisture content (%)	0.13 n. s.	0.15 n. s.	0.38 n. s.
Protein content (N × 6.25) (%)	–0.32 n. s.	0.16 n. s.	0.57 n. s.
Extract of malt. congress mash (%)	0.56 n. s.	–0.39 n. s.	–0.02 n. s.
Mash method according to Hartong and Kretschmer VZ 45 °C (%)	–0.37 n. s.	0.51 n. s.	–0.19 n. s.
Kolbach index (%)	–0.39 n. s.	0.49 n. s.	–0.16 n. s.
Diastatic power (jWK)	0.33 n. s.	–0.15 n. s.	–0.11 n. s.
Final attenuation of laboratory wort from malt (%)	0.73 *	–0.80 *	–0.62 n. s.
Friability (%)	0.31 n. s.	–0.47 n. s.	–0.32 n. s.
High molecular weight b-glucan content of malt. FIA (mg/l)	0.15 n. s.	–0.01 n. s.	–0.06 n. s.
Nitrogen content in malt (%)	–0.30 n. s.	0.17 n. s.	0.64 *
Soluble nitrogen of malt. Kjeldahl method (mg/l)	–0.62 n. s.	0.64 *	0.27 n. s.
Glycidic extract (%)	0.70 *	–0.53 n. s.	–0.09 n. s.

Note: *... P=0.05; n. s. ... not significant

CONCLUSIONS

Statistically significant differences among varieties were found in hardness values determined by three technically different principles. The correlations among methods for determining the kernel hardness were counted as well among selected technological traits of barley and malt and kernel hardness. Significant correlations were found among PSI values of hardness and Final attenuation of laboratory wort from malt (0.73*) a Glycidic extract (0.70*). Values of kernel hardness measured by BRA statistically correlated with Final attenuation of laboratory wort from malt (–0.80*) and Soluble nitrogen of malt (0.64*). Grain milling energy values had statistically significant correlations with Nitrogen content in malt (0.64*). Other relationships among kernel hardness expressed by three different method and selected technological traits of barley and malt were not significant. These results are preliminary and further studies are necessary to confirm the results.

SOUHRN

Potenciál tvrdosti zrna ječmene (*Hordeum vulgare* L.) jako selekčního znaku v procesu šlechtění

Přesné stanovení kvality ječmene mikrosladováním je pro šlechtitelskou selekci nákladné, a proto jsou hledány vhodné levné metody odhadu kvality. Cílem bylo zjistit, zdali je možné podle úrovně tvrdosti obilek ječmene selektovat na sladovnickou kvalitu. Byly analyzovány vzorky obilek souboru deseti odrůd ječmene. Tvrdost obilek byla stanovena metodami PSI (Particle size index), tvrdostí dle Brabendera (BRA) a metodou stanovení energie potřebné k mletí vzorku (GME). Vzorky byly následně mikrosladovány a u sladů byly stanoveny vybrané technologické parametry.
Byly zjištěny statisticky průkazné rozdíly mezi odrůdami v úrovni tvrdosti. Byly stanoveny korelace mezi úrovní tvrdosti a vybranými technologickými parametry ječmene a sladu. Hodnoty PSI byly v průkazné korelaci s dosažitelným stupněm prokvašení (0.73*) a glycidovým extraktem (0.70*). Hodnoty BRA byly v průkazné korelaci s dosažitelným stupněm prokvašení (–0.80*) a obsahem rozpustného dusíku ve sladu (0.64*). Hodnoty GME byly ve významné korelaci s obsahem dusíkatých látek ve sladu (0.64*). Po ověření na rozsáhlejším souboru vzorků může být tvrdost zrna použita jako selekční znak v časných fázích selekce.

tvrdost, energie potřebná k mletí, kvalita, ječmen, particle size index, do-corder

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