

## Brewing Trials with Spring and Winter Barley Varieties

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### Abstract

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The effects of a set of barley varieties on the brewing process and quality of beer production intermediaries were studied in trial brews (40 l) prepared using the two-mash decoction process. The varieties included in the trial were selected based on the starch granule size distribution determined previously. A significant effect of the varieties on the saccharification time of both mashes was determined. The highest saccharification rate in brews was achieved with the variety Jersey; the saccharification time of the 1<sup>st</sup> and 2<sup>nd</sup> mash with the variety Tiffany was markedly longer. The varieties with a greater fraction of large starch granules (Tiffany and Luxor) exhibited a higher haze with sweet wort as well as hopped wort compared to the varieties with a low fraction of large starch granules (Jersey and Tolar). The effect on the lautering time was not demonstrated. Pronounced varietal differences were determined in the extract balance of the brewing process. The varieties Tiffany and Luxor exhibited significantly lower extract yields. The malts from these varieties had lower laboratory extracts and higher extract losses in spent grains. The effect of the variety on the saccharide composition in hopped wort was confirmed. The proportion of fermentable saccharides in hopped wort extract rose from the variety Tiffany (66.9%) to the variety Jersey (83.6%). A significant difference in the final attenuation was also determined (76% in beers prepared from the varieties Tiffany and Luxor compared to 81.5% from the Jersey variety).

**Keywords:** *Hordeum vulgare* L.; saccharification; saccharide composition; beer

The main component of barley caryopses endosperm is starch in the form of large (type A) and small (type B) starch granules (MAY & BUTTROSE 1959). According to the literature, the small starch granules generally make about 90% of the total number of the granules present but only about 10% of the total starch weight (e.g. 1). The small starch granules tend to fast embedding in the protein matrix and inside the cell walls (PALMER 1972) and thus they can cause some problems in the production of the final product. TILLET and BRYCE (1993) assume that only about one half of the small starch granules are degraded in the course of malting and kilning, which means that approximately 5% of the total starch content is not degraded to

fermentable saccharides that are important for the consequent conversion to alcohol. The remaining small starch granules can form starch haze and can block filtration beds in lauter tuns.

Many different methods and techniques have been used for the measurement of the ratio of large and small starch granules, e.g. image analysis (BAUM & BAILY 1987), Coulter counter (SOUTH & MORISSON 1990), low angle laser light scattering (LALLS) (PSOTA *et al.* 2000), and field-flow fractionation (FFF) (CHMELÍK *et al.* 2001).

Starch granule size distribution is predominantly a varietal trait affecting technological parameters of barley varieties. The effects of locality and year on this trait are markedly lower but the statistical

dependence between the technological parameters of malt and starch granule size distribution was found to be relatively low (PSOTA *et al.* 2004).

The relationship between starch granule size distribution and technological quality of barley caryopses is affected by the binding of starch granules to the protein matrix, quality of the protein matrix and cell walls (BRENNAN *et al.* 1996, 1997) and caryopsis enzymatic apparatus.

Based on the previous results (PSOTA *et al.* 2004, 2008a, b; CHMELÍK *et al.* 2007), four barley varieties differing in starch granule size distribution were chosen for the brewing trial. In the present study, a comparison of the brewing qualitative parameters of these varieties is given. The aim of the study was to obtain information on the behaviour of spring and winter barley varieties selected on the basis of the above mentioned characteristics during the brewing process, and on the effects of these varieties on the final product quality.

## MATERIAL AND METHODS

Brewing quality of the selected barley varieties was evaluated in semi-pilot brews. Technological and analytical parameters were studied.

**Selection of the varieties.** Based on the research into starch granule size distribution carried out in the set of 12 barley varieties in 2001–2003 (PSOTA *et al.* 2008b), the spring barley varieties Jersey and Tolar and winter barley varieties Luxor and Tiffany were selected for this experiment. The variety Jersey had the smallest fraction of large starch granules; it was followed by the varieties Tolar and Luxor. The greatest fraction of large starch granules was determined in the variety Tiffany. The malts were prepared in the micromalting plant by the standard method (PSOTA *et al.* 2008a).

**Brewing trials.** For higher demonstrativeness of the differences studied, all-malt trial brews were prepared from the malts from the varieties mentioned above with the same quantity of grist and uniform hopping in all brews regardless of the malt extract content. Two brews were prepared with each malt; the reproducibility of the RIBM results was comparable with those of industrial breweries (ŠKACH & NIKOLAI 2008).

The volumes of unboiled sweet wort and hot hopped wort were constant. The quantity of grist per a brew corresponded to the production of 12% hopped wort with the addition of standard malt.

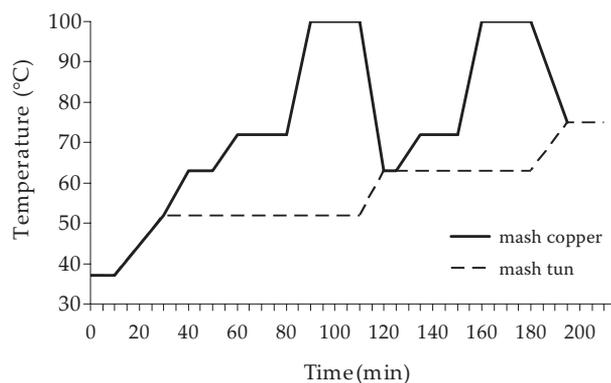


Figure 1. Mashing diagram of two-mash decoction procedure

The brews were prepared in a four-vessel brew-house facility, the hot hopped wort volume was ca 40 l. The sweet wort was produced using a double decoction procedure. The mash temperature was 37°C and the liquor temperature was adjusted to 52°C by hot water addition. The temperature and time course of mashing are given in the mashing diagram (Figure 1).

In addition, the volume of the spent grains in the lauter tank was measured when sparging was accomplished. The volume of sweet wort was constant (40 l).

The brews were hopped with Saaz hop pellets and hop CO<sub>2</sub> extract in a 1:1 ratio. The dose was 12 g of alpha acids per 1 hl of the hopped wort. Open stainless fermentation cylinders were used for primary fermentation. The hopped worts were pitched at 8°C with yeasts of the strain No. 95 of the RIBM yeast collection. The dose was 0.6 l/hl of the hopped wort. Maximal temperature did not exceed 12°C. The duration of the primary fermentation varied from 6 days to 8 days depending on the hopped wort extract. The green beers were transferred to lagering tanks at the difference of apparent and final attenuation of 10% to 15%.

The beers matured at a temperature of 1–2°C for 50 days. Final beers were filtered on a sheet filter and bottled.

**Analytical methods.** During mashing, the saccharification of mashes was determined with iodine solution from the fifth minute in one-minute intervals.

The haze of the first wort and wort of hot sweet was measured at an angle of 90° with a laboratory nephelometer in the range of up to 20 EBC units. Prior to the measurement, the samples were diluted with hot water. The hopped wort clarity was

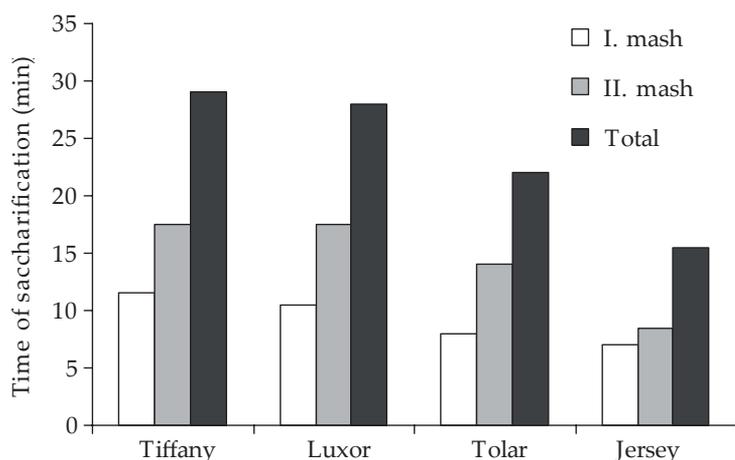


Figure 2. Saccharification of mashes in trial brews

assessed visually on a five-point scale from clear (1) to cloudy (5).

The beer analysis was carried out according to the Analytica EBC (ANONYMOUS 1998), spent grains analysis after the Brewing and Malting Analytica (ANONYMOUS 2001). Saccharides in hopped wort to the stage of 10 polymerisation units were determined using the HPLC method. The Rezex RSO-Oligosaccharide column in Ag cycle was used for the separation. The mobile phase was water at a flow rate of 0.3 ml/min, the column temperature was 80°C. Oligosaccharides were detected with a highly sensitive RI-101 detector (Shodex, Kanagawa, Japan). Maltose was used for calibration and oligosaccharide evaluation; the response factor corresponded to anhydrous maltose.

## RESULTS AND DISCUSSION

The malts made from the varieties tested differed in malting quality parameters describing cytolytic and proteolytic modifications (Table 1). The variety Jersey exhibited the highest modification degree, the variety Luxor the lowest one. The malt from the variety Tiffany had a higher diastatic power, that from the variety Luxor had the lowest value. The time of the sweet wort saccharification was the same with all the varieties (10 min) with the exception of the variety Luxor (10–15 min). The speed of saccharification is one of the basic process control parameters in the brewhouse. It is given by the activity of amylolytic enzymes and availability of the substrate, i.e. starch contained in the malt. The first mash saccharifies faster than the 2<sup>nd</sup> mash, all enzymes contained in the malt are active here. In the second mash, only ca 65% of the original

activity of amylolytic enzymes is maintained. Approximately 1/3 of the total volume of the liquor is boiled, hence part of the enzymes is inactivated. Boiling, however, leads to a better splitting of the substrate. In terms of intensity of the enzymatic effect on the substrate, two opposite impacts are regarded. The results given in Table 2 and Figure 2 show the saccharification rate in the Tiffany and Luxor varieties. The highest saccharification rate was achieved in the brews with the variety Jersey; in the variety Tiffany, saccharification times of the 1<sup>st</sup> and 2<sup>nd</sup> mash were markedly longer (Table 2).

The differences in the quality of malts from the tested barley varieties did not affect lautering rate significantly; our experiences show that with a thin layer of spent grains in a lauter tun, the time of lautering is significantly affected only when raw

Table 1. Results of malt analysis

	Tiffany	Luxor	Tolar	Jersey
Saccharification (min)	10–10	10–15	10–10	10–10
Viscosity (mPa.s)	1.543	1.736	1.497	1.471
Extract (% DM)	78.1	74.6	81.2	82
Relative extract 45°C	37.1	27.8	34.5	43.7
Diastatic power (WK)	438	394	429	416
Degree of attenuation (%)	78.2	77.2	80.6	82.1
Protein (%)	12.3	11.5	10.3	10.5
Soluble nitrogen (mg/100 ml)	86.2	62.6	71.2	83.7
Kolbach index (%)	40.4	31.5	40	46.1
Friability (%)	64	38.6	86.7	82.3
Homogeneity (%)	83.1	52	99	99
Glassy corns (%)	2.3	4.4	0.6	0.2
β-Glucan (mg/l)	465	1211	196	189

Table 2. Technological parameters of trial brews

Brew	Tiffany		Luxor		Tolar		Jersey	
	a	b	a	b	a	b	a	b
Saccharification 1 <sup>st</sup> mash (min)	12	11	10	11	8	8	7	7
2 <sup>nd</sup> mash (min)	18	17	17	18	14	14	9	8
Total (min)	30	28	27	29	22	22	16	15
Lauter time (min)	34	34	32	32	34	33	32	34
Haze First wort (EBC U.)	105.5	88.8	52.6	62.8	29.6	28.0	31.2	30.0
Sweet wort (EBC U.)	46.4	40.0	31.2	32.4	26.4	26.8	27.6	26.8
Wort*	4	4	4	4	3	3	2	2
Primary fermentation (days)	6	6	6	6	7	7	8	7

a – brew a; b – brew b; \*visual evaluation by 5-point scale (clear–cloudy)

material with a high concentration of substances impeding filtration is processed.

Significant differences were found out in the sweet wort clarity (Table 2). The clarity of the first wort and sweet wort was affected by the quality of malts from different barley varieties, the highest haze having been measured in the sweet worts from the variety Tiffany, followed by variety Luxor. The haze in the varieties Tolar and Jersey was low and comparable. The clarity of wort during lautering is one of the basic indicators for sweet wort quality technological control, haze-forming substances affecting the fermentation process, production of secondary fermentation metabolites, and beer filterability. In the varieties with a greater fraction of large starch granules, Tiffany

and Luxor, a significantly higher sweet wort haze was determined and also, as discussed further, a significantly higher content of oligosaccharides with the number of glucose units higher than ten, sweet wort haze corresponding to the saccharide composition. The wort haze particles consist of protein-polyphenol-polysaccharide complexes. The malts from Tiffany and Luxor varieties had higher values of soluble nitrogen and they were less cytolytically modified (Table 1).

The two-row spring barley varieties Jersey and Tolar with smaller fractions of large starch granules, as determined in the previous experiment (Psota *et al.* 2008a), had a more active enzymatic apparatus. The greater fractions of large starch granules in the winter barley varieties Luxor and

Table 3. Extract balance of trial brews

Brew	Tiffany		Luxor		Tolar		Jersey	
	a	b	a	b	a	b	a	b
<b>Malt</b>								
Weight (kg)	6.5		6.5		6.5		6.5	
Extract (% DM)	78.1		74.6		81.2		82.0	
Moisture (%)	7.8		7.9		7.5		7.6	
Extract yield (kg)	4.68		4.47		4.88		4.93	
<b>Wort</b>								
Volume (l)	37	37	37	37	37	37	37	37
Extract (%)	11.22	11.35	10.03	9.95	12.30	12.23	12.43	12.37
Extract yield (kg)	4.34	4.39	3.86	3.83	4.78	4.75	4.83	4.81
Loss laboratory/brewhouse (kg)	0.34	0.29	0.61	0.64	0.10	0.13	0.10	0.12
(%)	7.3	6.2	13.6	14.3	2.0	2.7	2.0	2.4

a – brew a; b – brew b

Table 4. Results of spent grain analyses

		Tiffany	Luxor	Tolar	Jersey
Moisture	(%)	78.4	79.2	74.6	73.9
Soluble extract	(% as in)	3.1	2.9	2.2	2.9
	(% DM)	14.3	13.9	8.7	11.4
Unaccharified extract	(% as in)	0.8	2.1	0.9	0
	(% DM)	4	10.3	4.1	0
Total extract	(% as in)	3.9	5	3.1	2.9
	(% DM)	18.3	24.2	12.8	11.4
Volume of spent grains	(l)	16.2	18.8	14.0	14.3

% as in – % in original matrix

Tiffany were not utilised for the extract formation, probably due to lower activities of cytolytic and proteolytic enzymes.

Visual evaluation of the hopped wort clarity corresponded to the results of the clarity measurements of the first wort and of the sweet wort. The hopped worts from the variety Jersey were classified with grade 2, the clarity of the varieties Tiffany and Luxor with grade 4 on a five-point scale.

The expected differences between the extract yields in the malts from the tested varieties were reflected in the hopped wort extract concentra-

tions, the extract values corresponding to the differences between the malt extracts and extract losses in the brewhouse.

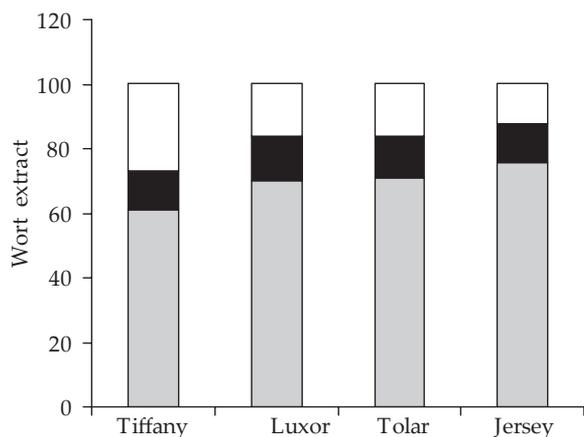
The extract balance shows that the brewhouse yield was high and comparable with the varieties Jersey and Tolar (Table 3). Laboratory-brewhouse loss was 2.3% on average. A significantly lower extract yield was determined with the variety Tiffany (average loss 6.7%), the lowest brewhouse yield was determined with the variety Luxor (loss on average 13.9%). The malt from the variety Luxor showed a very low cytolytic modification.

The extract losses in the brewhouse are those in nonsparged and nonsaccharified extract retained in the spent grains. The spent grain analysis performed in series “b” brews only (Table 4) documents the differences in the release and saccharification of starch malt extract. The highest content of soluble extract and total extract in the spent grains were found with the varieties Tiffany and Luxor. The spent grains from malts from these varieties had higher volumes and water contents than the spent grains from the varieties Tolar and Jersey. This corresponded to a higher retention of the dissolved extract in the spent grains. In the brew made from the malt from the variety Jersey, saccharification proceeded faster, no nonsaccharified extract having been found in the spent grains. On the contrary,

Table 5. Saccharides in hopped wort (g/100 ml)

Brew	Tiffany		Luxor		Tolar		Jersey	
	a	b	a	b	a	b	a	b
Fructose	0.26	0.26	0.21	0.22	0.29	0.23	0.26	0.35
Glucose	1.08	0.97	0.88	1.04	1.30	1.13	1.29	1.21
Maltose	5.03	4.29	5.14	4.82	6.43	5.91	6.60	7.04
Maltotriose	1.32	1.22	1.15	1.07	1.56	1.37	1.58	1.42
DP4	0.42	0.41	0.51	0.31	0.46	0.46	0.45	0.37
DP5	0.14	0.16	0.24	0.12	0.21	0.21	0.16	0.24
DP6	0.19	0.23	0.21	0.15	0.23	0.26	0.19	0.18
DP7	0.20	0.16	0.17	0.12	0.19	0.19	0.18	0.16
DP8	0.14	0.10	0.15	0.17	0.18	0.14	0.20	0.17
DP9	0.19	0.14	0.20	0.17	0.21	0.22	0.17	0.23
DP10	0.19	0.18	0.18	0.21	0.20	0.25	0.21	0.18
Sum (DP1–DP10)	9.16	8.12	9.04	8.40	11.26	10.37	11.29	11.55
Fermentable saccharides (DP1–DP3)	7.69	6.74	7.38	7.15	9.58	8.64	9.73	10.02
Oligosaccharides (DP4–DP10)	1.47	1.38	1.66	1.25	1.68	1.73	1.56	1.53

a – brew a; b – brew b; DP – degree of polymerisation (number of glucose units)



□ – rest of wort extract; ■ – oligosaccharides (DP4–DP10);  
 ■ – fermentable saccharides (DP1–DP3)

Figure 3. Hopped wort saccharides composition

a high amount (10.3% in DM) of nonsaccharified extract in the spent grains was detected in the brew from the malt from the variety Luxor.

In the trial brews, the intensive double decoction procedure was used for the sweet wort production. The infusion technology of processing malts from the varieties Jersey and Tolar would provide even lower brewhouse yields.

Starch granule size distribution also affected the composition of low molecular sugars in the hopped wort. The results of the determination are given in Table 5. Table 6 shows the relative distribution of the individual saccharides in the hopped wort extracts.

Despite a certain variability in repeated brews, the results clearly show the effect of variety on the hopped wort saccharide composition as well as the tendency to a higher proportion of fermentable sugars in the varieties, on average 75.9% in the variety Jersey and 61.2% in the variety Tiffany (Figure 3). The average proportion of the most important fermentable sugar, maltose, in total hopped wort extract declined from the variety Jersey (52.4%) to the variety Tiffany (39.5%). The differences between the contents of oligosaccharides with four to ten glucose units (DP4–DP10) were not significant. These oligosaccharides formed approximately 12–15% of the hopped wort extract. The rate of other compounds, polysaccharides and non-saccharide material, rose from the hopped worts from the variety Jersey (12.4%) to the hopped wort from the variety Tiffany (26.8%). Small size starch granules are less accessible to amylolytic enzymes.

Table 6. Percentage share of saccharides in hopped wort extract

Series	Tiffany		Luxor		Tolar		Jersey	
	a	b	a	b	a	b	a	b
Fructose	2.2	2.2	2.0	2.1	2.2	1.8	2.0	2.7
Glucose	9.2	8.2	8.4	10.0	10.1	8.8	9.9	9.3
Maltose	42.9	36.1	49.3	46.6	49.8	46.1	50.6	54.2
Maltotriose	11.3	10.3	11.0	10.3	12.1	10.7	12.1	10.9
DP4	3.6	3.5	4.9	3.0	3.6	3.6	3.4	2.8
DP5	1.2	1.3	2.3	1.2	1.6	1.6	1.2	1.8
DP6	1.6	1.9	2.0	1.4	1.8	2.0	1.5	1.4
DP7	1.7	1.3	1.6	1.2	1.5	1.5	1.4	1.2
DP8	1.2	0.8	1.4	1.6	1.4	1.1	1.5	1.3
DP9	1.6	1.2	1.9	1.6	1.6	1.7	1.3	1.8
DP10	1.6	1.5	1.7	2.0	1.5	1.9	1.6	1.4
Sum (DP1–DP10)	78.1	68.3	86.5	81.0	87.2	80.8	86.5	88.8
Fermentable saccharides (DP1–DP3)	65.6	56.8	70.7	69.0	74.2	67.4	74.6	77.1
Oligosaccharides (DP4–DP10)	12.5	11.5	15.8	12.0	13.0	13.4	11.9	11.7
Rest of wort extract*	21.9	31.7	13.5	19.0	12.8	19.2	13.5	11.2

a – brew a; b – brew b; \*polysaccharides and non-saccharide compounds

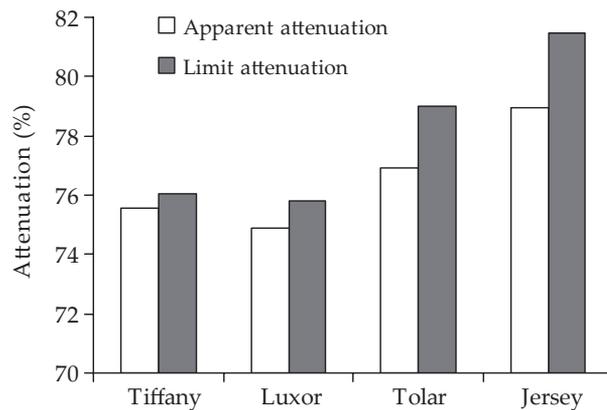


Figure 4. Attenuation degree of trial beers

The final attenuation agreed with the saccharide composition of the hopped wort, the highest value having been determined in the beers produced from the variety Jersey (81.5%). The final attenuation of the beers produced from the varieties Tiffany and Luxor was on the level of 76% (Table 6, Figure 4). The differences determined in the apparent attenuation of beers were significantly lower, the beers from the malts from the varieties Jersey and Tolar had a higher difference (2.0–2.7%) between the apparent and final attenuation than the beers from the malts from the varieties Tiffany and Luxor (0.3–0.9%). Thus we can state that the hopped worts of the varieties Tiffany and Luxor exhibited a lower final attenuation and the apparent attenuation approached the limits of the final attenuation. The value of the final attenuation and

difference between apparent and final attenuation of beers affect the sensory properties of beer.

The varieties included in the trial were selected based on the starch granule size distribution determined in the previous experiments. Starch granule size distribution in caryopses is significantly affected by the variety, the level of relationships between the technological parameters and starch granule size distribution is relatively low (PŠOTA *et al.* 2008a). For this reason, the results achieved with the varieties under study cannot be considered to be only the result of the starch granule size distribution in caryopses of the varieties studied. The results achieved were definitely affected by other characters of the varieties studied, first of all the activities of hydrolytic enzymes and compactness of the embedding of the starch granules in the protein matrix.

## CONCLUSION

The mashes from the malts from the varieties Jersey and Tolar saccharified considerably faster than those from the varieties Tiffany and Luxor.

The malts produced from the varieties Tiffany and Luxor exhibited a higher haze in the intermediate products (sweet wort, hopped wort) in comparison with the malts from the varieties Jersey and Tolar.

Significant differences were found between the malts produced from the tested varieties in the extract balance of the brewing process. The varieties Tiffany and Luxor exhibited substantially

Table 7. Results of beer analyses

Brew	Tiffany		Luxor		Tolar		Jersey	
	a	b	a	b	a	b	a	b
Apparent extract (% w/w)	2.68	2.92	2.56	2.59	2.77	2.84	2.54	2.63
Real extract (% w/w)	4.35	4.58	4.01	4.13	4.58	4.62	4.41	4.48
Final extract (% w/w)	2.59	2.84	2.48	2.50	2.53	2.59	2.29	2.31
Alcohol (% w/w)	3.59	3.58	3.08	3.28	3.92	3.86	4.06	4.01
(% v/v)	4.59	4.58	3.93	4.19	5.01	4.94	5.18	5.13
Apparent attenuation (%)	76.4	74.7	74.4	75.4	77.2	76.6	79.3	78.6
Real attenuation (%)	63.0	61.8	61.3	62.2	63.9	63.3	65.5	64.9
Final attenuation (%)	77.1	75	75.3	76.3	79.4	78.6	81.6	81.3
Attenuation difference (%)	0.7	0.3	0.9	0.9	2.2	2.0	2.3	2.7
Original extract (Plato) (% w/w)	11.3	11.5	10.0	10.5	12.2	12.1	12.3	12.3

a – brew a; b – brew b

lower amounts of the extract obtained. The malts from these varieties had lower extract contents determined in a laboratory and higher extract losses in a brewery.

For the brews from the varieties Jersey and Tolar, higher proportions of fermented saccharides in the sweet wort and higher final apparent attenuation were determined.

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