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Determination of Cytolytic Malt Modification – Part II: Impact on Wort Separation

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ABSTRACT

The cytolytic malt criteria viscosity and β-glucan are used as an integral part of routine laboratory control measures to classify the malt modification level and to ensure good processability. Part one of this two-part publication demonstrated that barley varieties with high cytolytic modification along with low β-glucan content tend to have a higher content of usually unconsidered water-soluble arabinoxylan. This study investigated the arabinoxylan molar mass distribution and its impact on processability. In order to observe the impact of arabinoxylans on separation processes in the brewhouse, lautering tests of different modified malt types were performed. The results suggest that in contrast to their lower β-glucan content, highly modified malt types had a higher average arabinoxylan content (767 mg/L) than medium modified samples (710 mg/L). Furthermore, differentiation of arabinoxylan molar mass resulted in a higher molar mass of 14–34% above 50 kDa in these samples. In terms of processability, flux through the spent grain cake was mainly influenced by wort viscosity ($r=-0.621$, $p < 0.01$), although arabinoxylan had little influence on the viscosity. However, 53% of highly modified malt samples had a flux of less than 4 L/h × m^2 (average value) despite low β-glucan levels. A principal component analysis identified arabinoxylans as the main driving force in connection with the permeability through the filter cake, whereby especially highly modified barley varieties (60%) were affected. Arabinoxylans may have a decisive effect, so it is not only β-glucan as a structural cell wall substance and present viscosity that should be considered as determining quality characteristics.

KEYWORDS

Arabinofuranosidase; β-glucanase; filtration; xylanase; lautering

Introduction

In malt quality control, friability, viscosity and β-glucan content are the main analyses for cytolytic malt evaluation. In this context, cytolysis describes the enzymatic breakdown of cereal cell wall polysaccharides, mainly β-glucan and arabinoxylan.[1] Cytolytic malt characteristics are important criteria in terms of processability in brewing processes such as lautering and beer filtration, but are also associated with positive product parameters such as palate fullness.[2] In beer production, filtration plays an important role in clarifying the product from yeast and haze.[3] Positioned upstream, another important separation step in the brewing process is lautering (wort separation), a special type of cake filtration. The purpose of lautering in beer brewing is to separate the wort, which contains soluble malt components, from the solids, the brewer's spent grains. Components of interest within the filter cake are starch polysaccharides (dextrins), non-starch polysaccharides such as β-glucan, and arabinoxylan, as well as proteins, lipids, polyphenols, and metal ions.[4,5] Polysaccharides are polymeric carbohydrates built from monosaccharides or monosaccharide derivatives linked by glycosidic bonds with a main source in malt. A differentiation between α-, β-glucans and arabinoxylans can be made in wort and beer. Besides the α-linked starch polysaccharides, β-linked glucose units originating in barley with β-1,3;1,4-glycosidic linear linkages (non-starch polysaccharides) are known as β-glucans. These polysaccharides contain up to 70% β-1,4-glycosidic bonds that are interrupted by at least 30% β-1,3-glycosidic bonds in barley.[3]

Based on their solubility, the non-extractable hemicelluloses and soluble gum can be differentiated in malt. While malting and mashing, non-water-soluble β-glucans are released from cereals such as barley by glucan-degrading enzymes, resulting in a reduction of molar mass.[6,7] Thus, molar masses between 2.0×10^3 and 40.0×10^6 g/mol have been detected in beer.[8] Furthermore, β-glucans are known to increase the turbidity and viscosity of beer due to their ability to form agglomerates known as β-glucan gels.[9,10] Another β-glycosidic bound polysaccharide of the cereal cell wall is arabinoxylan. This polymer consists of a backbone of xylopyranosyl residues linked by β-1,4-glycosidic bonds and β-d-xylopyranosyl residues substituted at O-2/O-3 or O-2 and O-3 with a varying amount of α-L-arabinose residue. These arabinose residues are linked with β-d-xylopyranosyl

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at O-3 and can be substituted with ferulic acid at O-5.[11] Arabinoxylans (210–500 mg/L) have been determined in lager beer.[12] A molar mass distribution in beer could not be found in the literature. About 25% of the barley endosperm cell wall is composed of arabinoxylans, and a similar amount is present in the aleurone and subaleurone layers (24%), whereas the lowest content of these compounds is found in the pericarp (only 6%).[3,13]

In the last few years, breeding progress has brought forth new malting barley varieties with high cytolytic modification identifiable by low β-glucan content and viscosity values. However, sometimes these varieties have not shown the intended optimized processability. This is due to the fact that the effect of other hemicellulose constituents such as arabinoxylan and their enzymatic hydrolysis are mostly disregarded in a standard malt analysis. This study was divided into two parts. The aim of the first part was to investigate the individual cytolytic composition of different malting barley varieties and to point out differences in cytolytic malt parameters depending on their genetically determined modification level. Appropriate analytical possibilities for arabinoxylan determination and significant extraction methods for cytolytic evaluation were investigated. In addition, the most important cytolytic enzymes were characterized. The second part refers to the resulting processability with a focus on wort separation. In many sources, soluble fiber is thought to increase wort viscosity and to result in prolonged lautering and filtration time.[6,14–17] Furthermore, different authors[18–20] have described an impact of water-soluble arabinoxylans on lautering and filtration. However, this approach only takes the total content of arabinoxylans (measured by different extraction methods) into account, independently of the molecular mass distribution.

Detailed data on molar mass distribution of arabinoxylans and information about corresponding cytolytic enzyme activity concerning their impact on wort separation could not be found in the current literature. Even though the cytolytic malt parameters depend on malt modification and variety characteristics, there appear to be no systematic investigations available about their influence on processability, in particular wort separation.

As a consequence, the aim of the second part of the investigation was to examine the impact of cytolytic malt composition and enzyme activity on wort separation performance. The wort separation (lautering) of malting barley varieties with different cytolytic modification levels was compared in a laboratory lauter tun and process data, such as average filter cake permeability and flux, were calculated. These data should not only be compared with the commonly used cytolytic parameters of the malts; in particular, detailed analysis of the substance group arabinoxylans with respect to their molar mass distribution was an important new issue. In addition, it should be determined to what extent varietal characteristics can be influenced during malting using the steeping degree as the technological process parameter relevant in practice and analogous to common German breeding program evaluation procedures for variety differentiation.

Experimental

Apart from the methods described in the preceding article–namely the evaluation of cytolytic malt characteristics depending on different extraction methods, polysaccharide concentration (arabinoxylan and β-glucan content) and enzyme activity (β-glucanase, endo-xylanase and arabinofuranosidase) – the samples were analyzed using the following methods.

Sample material and preparation

In total, 28 barley malt samples from the 2017 harvest (spring barley) were analyzed with the following modification distribution classified based on the barley variety characteristics: 15 highly modified, 1 minimally modified and 12 medium modified samples (HM – highly modified, MM – medium modified, LM – minimally modified ≙ varieties with high/medium/low levels of modification). A classification according to cytolytic modification characteristics of the varieties was made on the basis of expert knowledge and with the help of the breeders' expertise by means of their cytolytic quality parameter results (friability, β-glucan and viscosity) of recent years. Twelve samples were malted identically according to MEBAK R-110.00.008 [03-2016].[21] Of the remaining 16 samples (two patterns each highly modified and medium modified samples) a standardized malting procedure with falling germination temperature (18 to 14.5 °C) and six days' germination time was used under variation of the degree of steeping (39, 41, 43, 45% moisture content – after 48 h by spraying the required amount of water – of the green malt at the start of the kilning process) analogous to the German breeding program evaluation procedure. The aim was to investigate malt samples with a wide range of cytolytic specifications. For the extraction of polysaccharides in barley malt an isothermal 65 °C mashing procedure according to MEBAK R-207.00.002 [2016-03] (fine grist 0.2 mm, grist:water ratio 1:6, 1 h at 65 °C, after 30 min add 50 ml of water, weight up to 450 g) was used,[21] because this method provided the best comparability (see part one).

Molar mass distribution of arabinoxylans

The molar mass distributions of the arabinoxylans were analyzed with a fractionation using gel permeation chromatography (GPC) and a subsequent determination of arabinoxylan content in the collected fractions.[22,23] Fractionation was performed with an Äkta Prime (GE Health Care, Germany) using a Hiload 26/600 Superdex 200PG column. With a constant flow of 2.2 mL/min 500 mL phosphate buffer pH 7.0 (Honeywell Chemicals) was used as eluent to separate the sample into 30 fractions of 5.5 mL each. The sample was concentrated tenfold before separation using a vacuum evaporation at 60 °C. Then 3 mL of concentrated wort sample was injected into the system.

The calibration of the GPC system was performed with a medium-viscosity arabinoxylan standard (wheat arabinoxylan, Megazyme, Ireland). This standard was fractionated with the program described above and the molar mass in

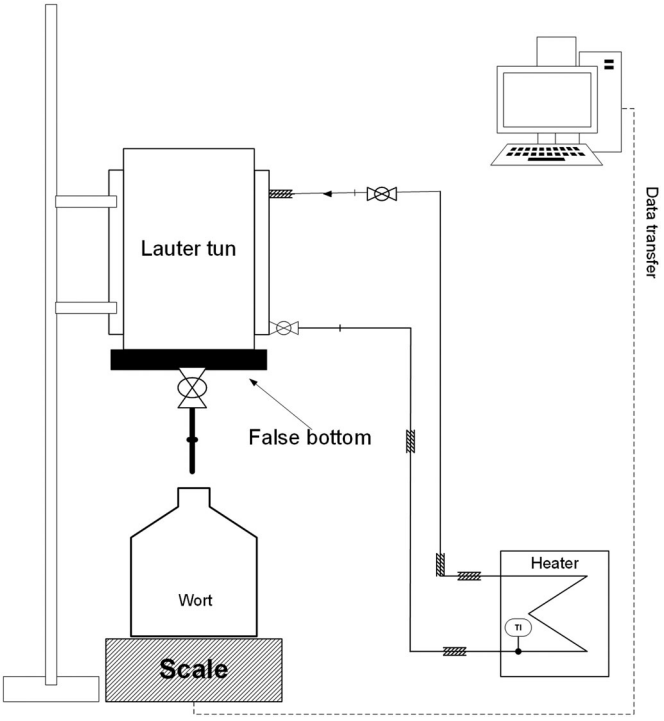

Figure 1. Experimental setup for the determination of the lautering performance on a laboratory scale.

the fractions was determined in duplicate with an asymmetric field-flow fractionation (AsFiFFF; Wyatt Technology, Germany) coupled with multiangle laser light scattering (Dawn, Heleos II, Wyatt Technology, Germany) and refractive index (RI) as quantitative detector (Agilent Series 1200 G1362A, Agilent Technologies, Germany) according to Kupetz et al..[24] This resulted in an 11-point linear calibration of molar mass Mn [kDa] in dependence on the fraction number of the GPC with r= −0.985 ($P < 0.05$). Using this calibration curve, classification of the molar mass into the three ranges <10 kDa, 10–50 kDa and >50 kDa could be performed. The arabinoxylan content in the fractions was determined as described in part one.

Lautering trials on the laboratory scale

To investigate the lautering performance of the different barley malt samples, a laboratory filter test (membrane filter test module) was rebuilt and equipped with a perforated plate as a false bottom with a 1 mm pore size. The lauter test consisted of a heatable stainless-steel vessel and a scale with a connection to a computer (see Figure 1). The double-walled housing of the filter vessel was tempered to 78 °C. For a double test, 50 g malt grist was milled with a DLFU mill of Bühler (milling gap 0.2 mm, Bühler GmbH, Beilngries, Germany). Per trial, 22.2 g malt grist was mixed with 177 mL hot water (65 °C) and isothermal 65 °C mashes were performed according to MEBAK.[21] After 1 h of mashing time, the sample was transferred into the filter. The filtration was started after a 10 min lauter rest. The filtrate weight was recorded in dependence on time and flux, and filter cake permeability was calculated using MATLAB2016a. The pressure difference (Δp) on the filter cake was calculated using the formula $\Delta p = \rho g h$.[25] Cake height of the spent grain cake was determined after each trial.

Statistics

Statistical analyses, to determine Pearson correlation coefficients, averages, and standard deviations as well as principal component analysis, were carried out using OriginPro 2018G (OriginLab Cooperation, Northampton, U.S.A.).

Results

To characterize the malt samples with different levels of modification, cytolytic malt parameters and enzyme activities were analyzed according to the methods described in part one. In addition, based on these results the classification into modification level of the malting barley varieties could be reviewed by two harvest years, 2016 and 2017. Based on the data, it was to be determined whether there are differences in varieties with different cytolytic modification levels in both the enzyme activity and the composition of the polysaccharides. Besides the characterization of the cytolytic malt composition, their impact on wort separation (lautering) was the main focus of this work. In addition, it was to be determined to what extent these varietal characteristics could be influenced during the malting procedure using variations of steeping degree for differentiation. As in the first part, the activities of the cytolytic enzymes were measured and their impact on variety differences were examined.

The β-glucanase activity (see Figure 2a) ranged between 88 and 385 U/kg with an average of 211 U/kg. The highly modified samples had an average of 169 U/kg (88–354/U/kg), whereas medium modified malt samples had an average of 262 U/kg (93–385 U/kg). The minimally modified sample had a β-glucanase activity of 183 U/kg. This confirms the previous study and shows that highly modified barley malts have lower β-glucanase activity than medium modified ones. Endo-xylanase (see Figure 2b) activity had an average of 36 U/kg (24–61 U/kg). Differences in the mean endo-xylanase activities between highly and medium modified samples could be examined. The highly modified barley malt had a lower average activity (32 U/kg, range: 24–43 U/kg) than medium modified samples (average: 41 U/kg, range: 27–61 U/kg). Arabinofuranosidase activity, shown in Figure 2c, achieved an average of 187 U/kg (range: 74–379 U/kg). Highly modified samples (161 U/kg, range: 74–249 U/kg) again had lower activity than the medium modified samples (218 U/kg, range: 123–379 U/kg). These results show clear differences in the activity of the cytolytic enzymes as a function of the modification level of the malt. When considering the full set of samples, β-glucanase and xylanase activity ($r = 0.583$, $P < 0.05$) as well as β-glucanase and arabinofuranosidase activity ($r = 0.587$, $P < 0.05$) correlated significantly. In addition, an evaluation of the samples with different steeping degrees (modification level) can be performed. In the case of β-glucanase, a lower steeping degree resulted in higher activity. Xylanase and arabinofuranosidase activity did not show this trend. Since the enzyme activity has a great impact on the

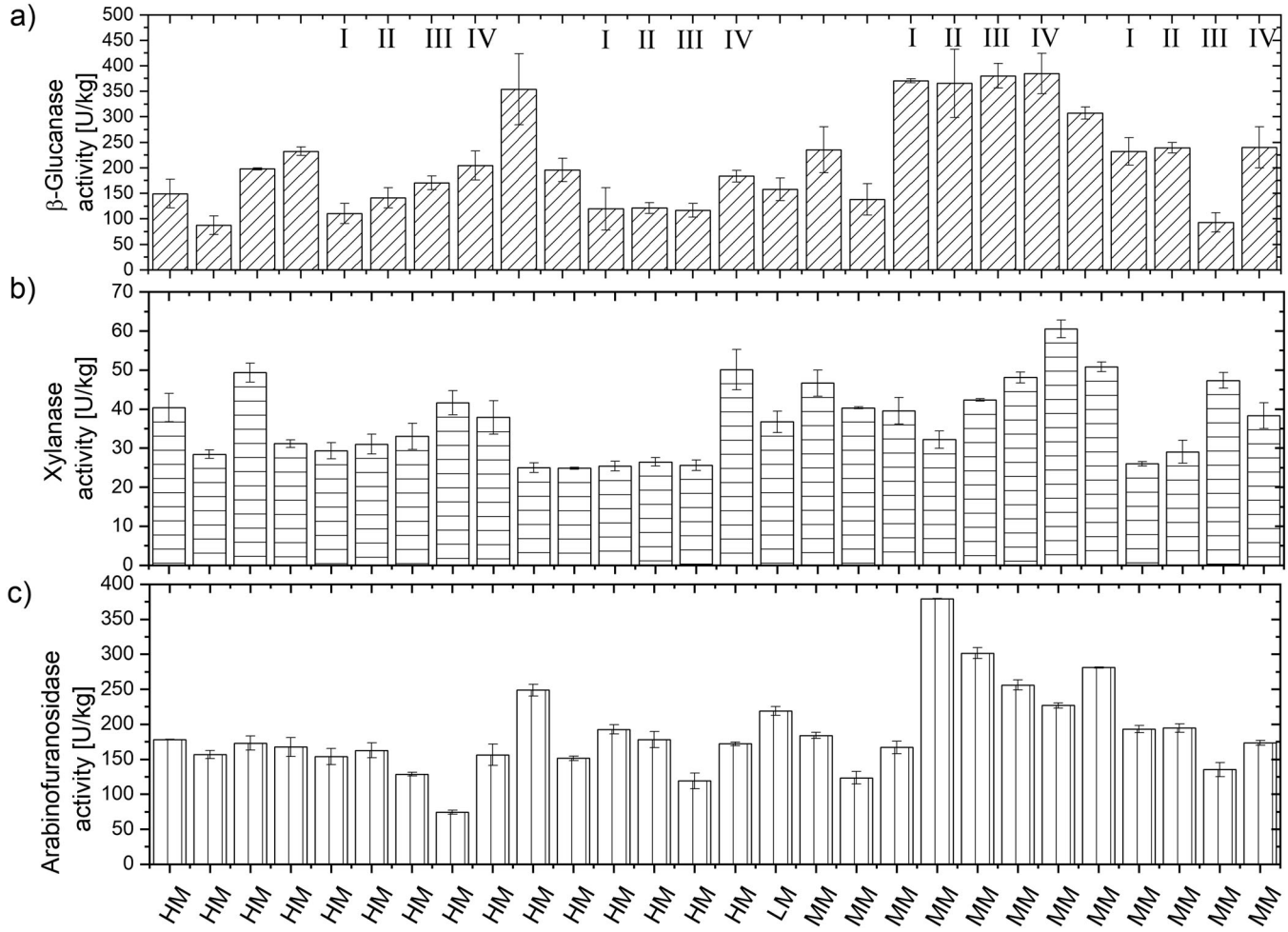

Figure 2. β-Glucanase, xylanase, and arabinofuranosidase activity (n = 3) of the investigated malt samples – classification of modification level by cytolytic malt parameters; variations in degree of steeping: I: 45%, II: 43%, III: 41%, IV: 39%; legend: HM – highly modified, MM – medium modified, LM – minimally modified (≙ varieties with high/medium/low levels of modification (HM/MM/LM)).

concentration of the water-soluble polysaccharides and the viscosity, these characteristics of the malt samples in laboratory wort were also investigated. The results are shown in Figure 3.

The β-glucan content (see Figure 3a) ranged between 30 and 673 mg/L with an average of 285 mg/L. In comparison to medium modified barley malt samples (402 mg/L, range: 68–673 mg/L), highly modified malt had a lower average content of 197 mg/L (range: 30–475 mg/L), which confirms the correct classification according to their modification level. Arabinoxylan content ranged between 367 and 979 mg/L (average: 742 mg/L). Again, highly modified malt had a higher average arabinoxylan content (767 mg/L, range: 367–979 mg/L) than medium modified samples (710 mg/L, range: 554–859 mg/L). Viscosity had an average of 1.497 mPa × s in all samples. With 1.499 mPa × s, medium modified samples had a slightly higher average viscosity than highly modified malt (1.497 mPa × s, see Figure 3c). The only significant correlation between the non-starch polysaccharides and the viscosity could be determined with the β-glucan content ($r = 0.387$, $p < 0.05$). Similar to the β-glucanase activity, an influence of the steeping degree could also be determined for the β-glucan content. A lower steeping degree resulted in higher β-glucan content and higher viscosity. This is consistent with the results of the enzyme activity, since higher enzyme activities can cause a higher concentration of water-soluble β-glucan. In contrast, total water-soluble arabinoxylan content was not affected by the steeping degree.

Besides the total soluble arabinoxylan content, investigation of molar mass distribution in the different cytolytic modified varieties was a main focus of this study. This was necessary since the applied arabinoxylan measurement method determines total content like an enzymatic assay (non-starch mono-, oligo- and polysaccharides). However, in the context of β-glucans, it is not the total content but high molar mass β-glucan (>10 kDa) that has a significant impact in the context of beer and wort filtration.[26,27] Thus, differentiation of arabinoxylan molar mass was performed using gel permeation chromatography. To compare the malt samples, molar mass was clustered into three ranges: <10 kDa: low molar mass, 10–50 kDa: medium molar mass and >50 kDa: high molar mass fractions. The molar mass distribution of arabinoxylan is shown in Figure 4. The highest proportion of arabinoxylan was detected in the low molar mass range. About 50–76% (226–617 mg/L) of soluble arabinoxylan had a molar mass of less than 10 kDa. High molar mass arabinoxylan ranged between 9 and 34%

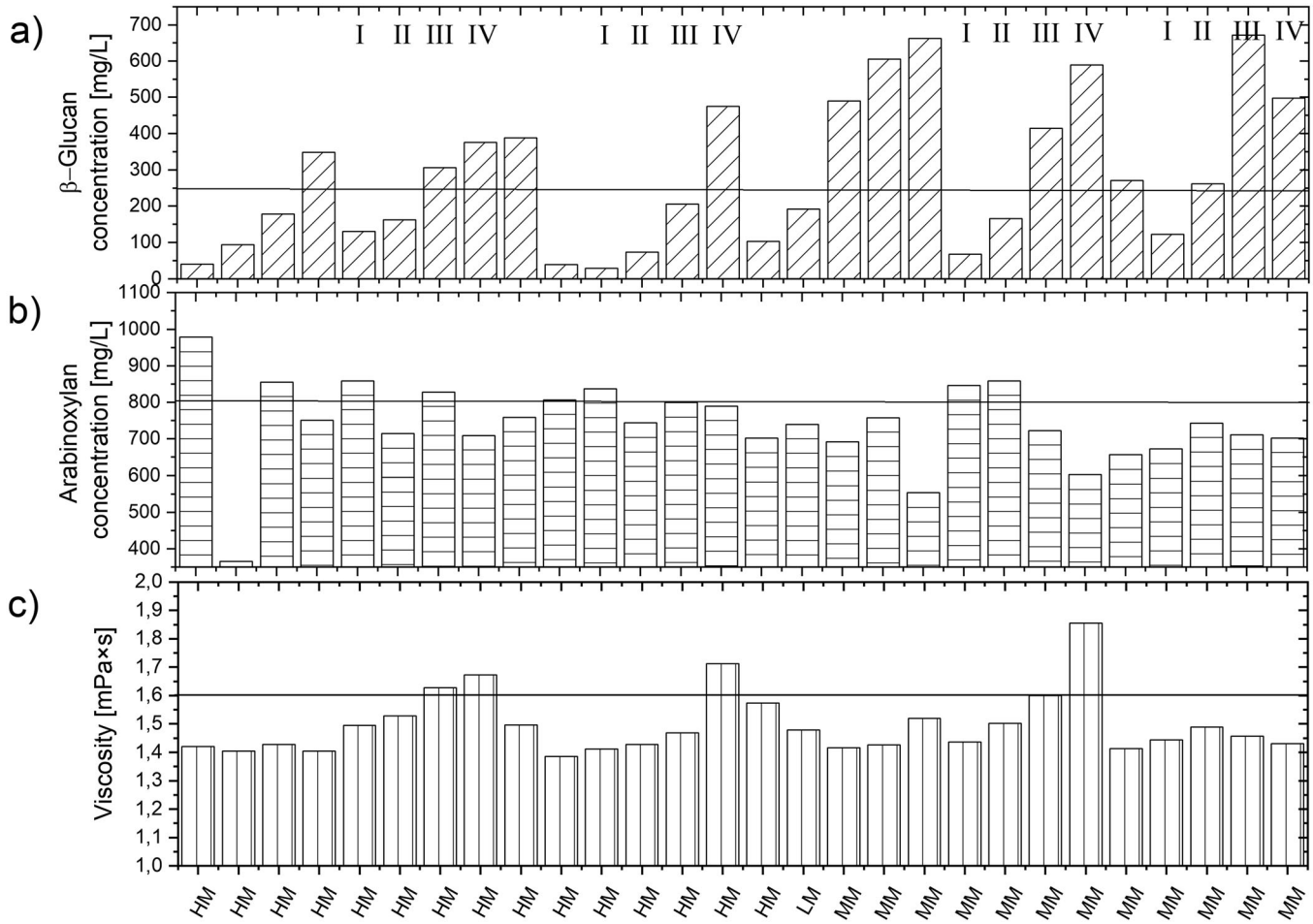

Figure 3. β-Glucan concentration (a, n = 4), arabinoxylan concentration (b, n = 4) and viscosity (c, n = 3) of the investigated malt samples – classification of modification level by cytolytic malt parameters; variations in degree of steeping: I: 45%, II: 43%, III: 41%, IV: 39%; legend: HM – highly modified, MM – medium modified, LM – minimally modified (≙ varieties with high/medium/low levels of modification (HM/MM/LM)).

(163 mg/L, range: 62–265 mg/L). Examining only the arabinoxylan greater than 10 kDa (289 mg/L, range: 141–432 mg/L), comparable concentrations to β-glucan content could be determined. Similar to the total arabinoxylan, differences were found depending on the modification level of the malt samples. Highly modified samples had a slightly lower share of low molar mass arabinoxylans (<10 kDa: 50–72%, 226–617 mg/L), but a bit higher high molar mass content of 14–34% (>50 kDa: 86–251 mg/L). In comparison, medium modified samples had a low molar mass fraction between 50 and 76% (320–570 mg/L) and a high molar mass fraction between 9–31% (62–265 mg/L).

Only two samples had a proportion of 50% of medium plus high molar mass arabinoxylan (one highly and one medium modified malt). These two samples also had the highest percentage share of high molar mass fraction. In total 39% of arabinoxylans were above 10 kDa, with highly modified samples having a share of 41% and medium modified samples 37%. This corresponds to a concentration of 289 mg/L arabinoxylan in all samples.

In contrast to the total arabinoxylan concentration, the molar mass distribution of arabinoxylan was influenced by the steeping degree. However, the different varieties had no uniform behavior. Highly modified malts had an increasing proportion of high molar mass arabinoxylan with decreasing steeping degree (corresponding to lower modification in the malting procedure), whereas medium modified malts did not allow a clear assignment. Due to these large differences between the individual samples, a possible connection between the total arabinoxylan content and the lautering performance was investigated. The lautering tests were performed on a laboratory scale on a temperature-controlled stainless-steel filter unit. Depending on filtration time and weight, permeability of the filter cake [mDarcy] as well as flux in $[L/(h \times m^2)]$ were calculated. The permeability ranged between 4.07 and 6.77 mDarcy (average: 4.03 mDarcy). Highly modified malt samples had an average permeability of 5.11 mDarcy (4.07–6.39 mDarcy), whereas medium modified samples had an average of 5.71 mDarcy (4.52–6.77 mDarcy). The minimally modified sample had a permeability of 4.91 mDarcy. There was no linear correlation between the analytical cytolytic malt parameters and the permeability of the filter cake. Malting procedure influenced the permeability only to a small extent. The highly modified malt samples had an average of 0.12 mDarcy between highest and lowest permeability, with no discernible dependence on steeping degree. Only in the medium modified sample types could a decrease of the permeability

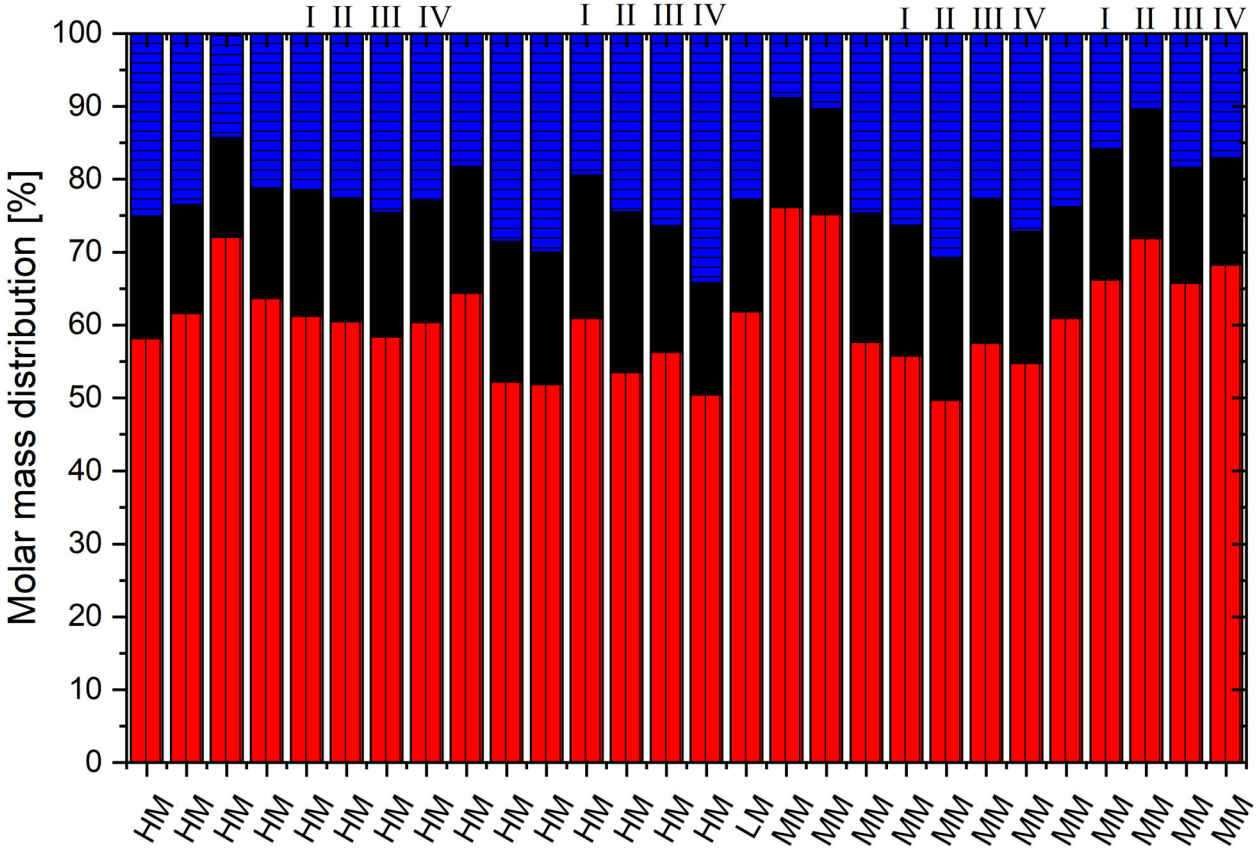

Figure 4. Percentage molar mass distribution of arabinoxylans (n = 4) in different modified malt samples using a standard malting procedure and with variations in degree of steeping: I: 45%, II: 43%, III: 41%, IV: 39%; legend: <10 kDa (▮), 10–50 kDa (▮) and >50 kDa (▮), legend: HM – highly modified, MM – medium modified, LM – minimally modified (△ varieties with high/medium/low levels of modification (HM/MM/LM)).

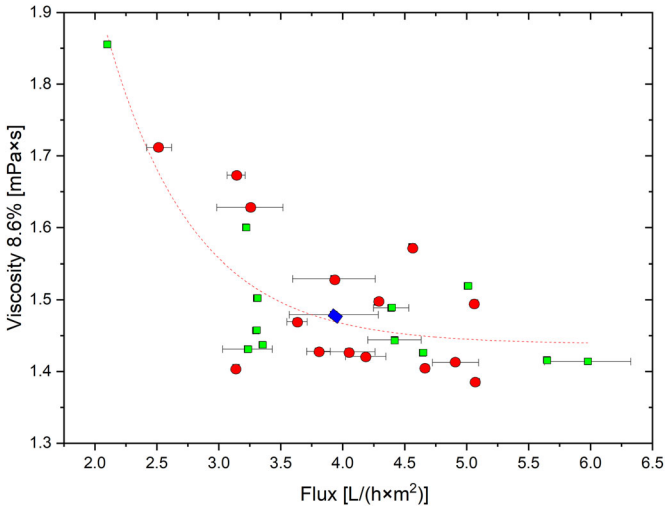

Figure 5. Flux through the spent grain cake (n = 2) in dependence on wort viscosity (calculated to 8.6%, n = 2) of the investigated malt samples; legend: ● highly modified malt samples, ■ medium modified malt samples, ◆ minimally modified malt samples (△ varieties with high/medium/low levels of modification (HM/MM/LM)).

with a lower steeping degree be determined (difference 1.32 mDarcy).

Besides the permeability of the spent grain cake, the flux is of particular interest for the evaluation of wort separation. In total, the flux ranged between 2.1 and 6.0 L/(h × m^2) with an average of 4.0 L/(h × m^2) (see Figure 5). Both highly modified (range: 2.5–5.1 L/(h × m^2)) and medium modified (range: 2.1–6.0 L/(h × m^2)) samples had an average flux of 4.0 L/(h × m^2). Malting procedure (modification during the malting procedure) had a clear impact on flux, whereas a higher steeping degree resulted in a higher flux (data not shown) in all investigated barley varieties.

Using the average flux as the evaluation criteria for a good wort separation, 53% of the highly modified and 50% of the medium modified samples had a smaller flux. In addition, it was noted that the highly modified samples had no flux above 5 L/(h × m^2). The flux of the samples did not correlate with any malt characteristics, apart from the viscosity of the wort (r=-0.621, $p < 0.01$). The coefficient of determination of the exponential function could be determined with R^2=0.65.

Since average flux had no differences within the barley varieties, chemometric investigation of the influencing factors on permeability was performed. Figure 6 shows the PCA biplot. Two principal components with a cumulative variance of 53.3% are plotted on the x- and y-axes. The first component is mainly defined by the arabinoxylan and its molar mass distribution as well as permeability through the spent grain cake (almost 31%). Mostly highly modified malt samples are influenced by arabinoxylan, which also had an impact on the permeability of the wort samples. About 60% of highly modified samples are influenced by arabinoxylan and permeability. Due to the distribution of the vectors, it becomes apparent that arabinoxylans, especially those

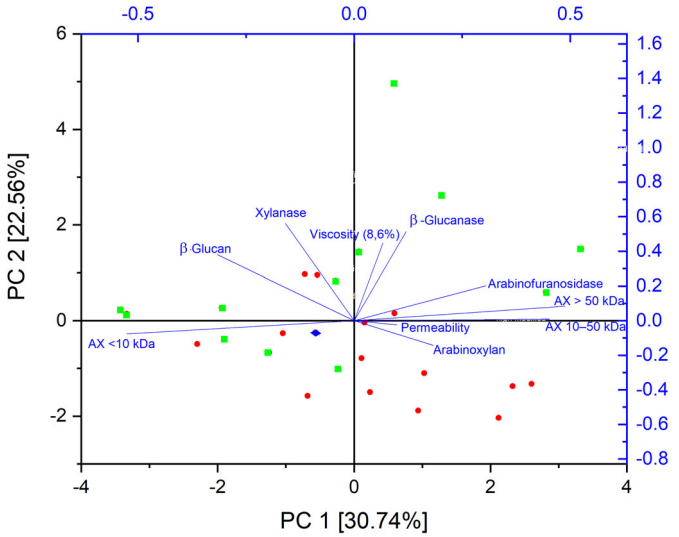

Figure 6. Principal component analysis of cytolytic malt characteristics and lautering performance; legend: ● highly modified malt samples, ■ medium modified malt samples, ◆ minimally modified malt samples (△ varieties with high/medium/low levels of modification (HM/MM/LM)).

between 10 and 50 kDa or greater than 50 kDa, have an influence on permeability. This impact of arabinoxylan concentration was evident. The lowest permeability malt sample had the highest content of high molecular weight arabinoxylan (>50 kDa) in combination with a β-glucan content of only 100 mg/L. Arabinofuranosidase, the enzyme which cleaves off the side chains of the β-1,4-xylan backbone so that no cross-linking of the ferulic acids takes place via oxidation, is likewise influenced by component one. The second component is mainly defined by the viscosity and malt enzyme activity of β-glucanase and xylanase (almost 23%). This statistical evaluation is consistent, since the enzymes determined can impact the viscosity of the wort, and the degradation of the β-glucans had a greater impact on the viscosity than arabinoxylans.

The principal component analysis shows that the permeability of the filter cake depends on the concentration of arabinoxylan, in particular high molar mass arabinoxylan. In addition, initial information can be found that the side chains of arabinoxylan, which contain arabinose and ferulic acid, have an impact on permeability. The literature shows that ferulic acid tends to cross-link under the action of oxygen.[28,29] These gels are temperature independent, but have a comparable impact on the filter performance like β-glucan gel.

Discussion

The results document that significant breeding progress has been achieved for malting barley varieties in recent years. Since the 1970s, different studies have investigated the impact of different mashing procedures on wort composition to estimate the processability of different barley varieties.[30–34] Over the years, however, the variety characteristics have changed due to the breeding process, requiring adapted extraction procedures. In consequence, there exist several extraction procedures, which are difficult to compare due to different processes and features despite a large amount of data available. It was found that the breeding of highly modified varieties had an impact on the cytolytic parameters as well as cytolytic enzyme activities. Not only the β-glucanase had significantly lower activities, but also xylanase and arabinofuranosidase. This low β-glucanase activity could be one reason for the demonstrably lower soluble β-glucan concentrations in wort produced by highly modified malts. The results could be confirmed in both crop years, although 2017 had more pronounced results. Comparable data has not been presented in the literature to date.

In addition to the enzyme activities, significant differences in the composition of the polysaccharides (β-glucan and arabinoxylan) between the varieties were found. Variety characteristics were verified by the commonly used analytical parameters viscosity and β-glucan content, confirming that highly modified varieties show lower values. However, the result suggests that the concentration of water-soluble arabinoxylan in the malting barley varieties had marked differences. It was demonstrated for the first time that cytolytically highly modified varieties have a higher concentration of water-soluble arabinoxylan. In addition, differences in the molar mass distribution of arabinoxylan were proven. Highly modified varieties had the largest proportion of arabinoxylan greater than 50 kDa (41%), whereas medium modified varieties only had a share of 37%. This shows that not only the enzyme activities in the barley varieties vary significantly, but also the proportion of water-soluble polysaccharides and the resulting molar mass distribution after mashing. These differences in malting barley varieties had not yet been shown in the literature.

An impact of malting procedure (varying steeping degree to modify the modification intensity) on cytolytic malt composition – especially β-glucan as well as arabinoxylan and its molar mass distribution – could be observed. In total, little is known about the effects of malting procedure on arabinoxylans.[18,20,35,36] According to the literature, an increase in steeping degree results in a decrease in high molar mass β-glucan content in malt.[37] A similar phenomenon could not be determined for total-water-soluble arabinoxylan. However, a small impact on arabinoxylan molar mass could be examined by modification intensity, although not all tested samples reacted the same way. β-Glucanase and xylanase activity increased with lower steeping degree. Li et al.[36] showed that during the germination period, the content of total arabinoxylan in barley grain sometimes decreased, while water-soluble arabinoxylan content increased. The steeping degree had no impact during their experiments. An impact on molar mass fractions could not be shown. In difference to the xylanase activities shown in this article, the authors examined much higher values with no impact of steeping degree. In addition, Lee et al.[20] detected an influenced arabinoxylan level in dependence on barley genotype characteristics, while growing conditions influenced both β-glucan and arabinoxylan levels. This difference in genotype and its effect on arabinoxylan content was also confirmed in this article. Variations of the malting parameters were not examined by the authors. The results

show that in comparison to β-glucan, arabinoxylan (soluble content and molar mass distribution) cannot be influenced to such a great extent during the malting process. Furthermore, a correlation between β-glucan content and β-glucanase activity was examined. The proportion of arabinoxylan degraded correlated only to arabinofuranosidase, but not to xylanase activity. Comparable correlations could not be determined in this study. However, the arabinoxylan content measured by Lee et al.[20] was very low in comparison to the results of this article. One reason for the low contents could have been the applied precipitation or extraction from the malt, which was not needed in this study. In addition, the authors investigated the wort filtration rate and the impact of polysaccharide contents in the mash. They were able to determine a significant correlation to the β-glucan ($r = 0.84$), arabinoxylan ($r = 0.54$) and viscosity ($r = 0.89$) of an isothermal wort (65 °C – comparable mashing procedure to the one applied in this article), whereas the polysaccharides measured in a Congress mash had no correlation. In comparison, Holtz et al.[38] showed that solely malt β-glucan content does not provide reliable information for the lautering process. Their pilot trials showed that good lautering was possible, even though the β-glucan content of the malt (370 mg/L) was increased. However, other samples of significantly lower concentration (<140 mg/L) had lower lautering performance.

The filtration data (permeability and flux) presented in this article correlated neither with the β-glucan nor with the arabinoxylan content of the wort. Only the viscosity of the wort had a significant correlation to the wort flux. However, the highly modified malt samples had a lower permeability with minor impact on flux. PCA analysis showed that the substance group arabinoxylan in particular influences the permeability of the filter cake. In addition, an impact of the debranching enzyme arabinofuranosidase could be determined. Since arabinoxylan can form linkages via the ferulic acid side chains with proteins and other ferulic acid side chains, the influence of this debranching enzyme is quite sensitive.[39,40] The resulting oxidative linkages between arabinoxylan molecules can result in a longer lautering process due to a lower permeability of the spent grain filter cake.[29] Thus, arabinoxylan can have a great influence on wort separation, since more than 80% of arabinoxylan remains in the spent grains.[4] The results suggest that not only the water-soluble content of arabinoxylans, but the total concentration in the grain should be examined, because the total water-soluble arabinoxylan had no correlation to the flux of the wort. In comparison approximately 70% of the β-glucan is extracted, while 30% remains in the spent grains.[41] In this context, Krahl et al.[17] demonstrated that water-soluble arabinoxylan is washed out during lautering, with the highest content in the first wort and a drop with each sparging step. This phenomenon was not taken into account in this study, since the focus was on the variety characteristics and the molecular weight distribution of arabinoxylan.

The results show that significant differences in varieties with regard to the cytolytic composition could be identified in current malting barley varieties and that the breeding progress did not yield procedural advantages for all varieties. Generally, breeding progress to improve processability aims to reduce specific commonly known risk factors. In the case of the cytolytic parameter, as the proportion of soluble β-glucan was reduced, arabinoxylan, representing a mostly disregarded substance group, increased as a result. In addition, it was possible to identify varieties that had a significantly lower lautering performance in the form of permeability or flux. Initial influencing factors were identified in this study that can be used for further breeding approaches.

Conclusion and outlook

The presented results confirm a significant varietal impact of malting barley malt with different modification levels on their cytolytic composition as well as the resulting processability. In this case, the focus was on lautering performance. These results are based on a large sample size across two crop years, although the 2017 harvest showed more pronounced differences in enzyme activity and concentration of water-soluble β-glucans and arabinoxylans. It could be shown that cytolytically highly modified malting barley varieties have a lower enzyme activity, lower concentration of water-soluble β-glucan, but a higher concentration of total arabinoxylan. However, the better processability during lautering targeted by the lower β-glucan levels could not be confirmed for all samples. On the contrary, clear varietal differences could be shown, with some newly bred cytolytically highly modified varieties exhibiting low permeability during lautering. This confirms that more process markers have to be considered to guarantee good processability beyond the commonly used cytolytic analytical parameters. This article identified some potential analytical indicators for the lautering process that can be used for new breeding approaches. A deeper insight into the structure, especially the side chains, of the arabinoxylans still needs to be gained, in order to more precisely investigate possible interactions that affect the permeability of the filter cake.

Disclosure statement

No potential conflict of interest was reported by the authors.

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