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Hot Steep Malt Sensory Evaluation Method combined with Check-All-That-Apply – an approach for barley (*Hordeum vulgare* L.) malt characterisation and quality control?

The Hot Steep Malt Sensory Evaluation, a method approved by the American Society of Brewing Chemists, was combined with the Check-All-That-Apply (CATA) method to assess the flavour profile of brewing malt. Ancient spring barley varieties (*Hordeum vulgare* L.), including Mahndorfer Hanna, Kraffts Riedgerste, and Alpine Pfauengerste, alongside a modern spring barley variety from the 2020 harvest, were malted with a standardised, small-scale malting process. Kilning was performed to produce Pilsner pale malt. The results of this study demonstrate that the Hot Steep Malt Sensory Evaluation, in conjunction with the CATA method, is an effective tool for differentiating the flavour characteristics of brewing base malt due to its simplicity and rapid execution. Both methods also proved to be suitable for detecting off-flavours in base malt. However, successful implementation requires the identification of relevant attributes and sensory training of panellists. Consequently, these methods are highly applicable for quality control within malt houses and breweries to assess malt quality. Both methods combined might improve quality control even in small-scale breweries.

Descriptors: Hot steep, sensory, malt flavour, malt quality, spring barley

1 Introduction

For the malting and brewing industry as well as for distillers barley (*Hordeum vulgare* L.) is the most important raw material [1]. Normally the suitability of barley for brewing is based on specific malt quality parameters. Organisations around the world like the European Brewing Convention (EBC) set acceptable standards and approve new malting varieties by the use of guidelines and lengthy testing requirements to approve barley varieties for malting and brewing [1-3]. Malt quality is influenced by barley variety, environmental factors such as weather, type of soil and the application of fertilisers [4]. *Morrissy* et al. (2022) showed slight influence of barley genotype on beer flavour [5], while *Herb* et al. (2017) showed, in small-scale trials, clear influence of the genotypes tested [6]. Malt is a main contributor to aroma and flavour of the final beer [7]. To date only a few methods are available to assess the sensory properties of malt. Normally malt quality control involves the visual inspection of the product's colour as well as checking for defects, by smelling on the malt to determine off-notes. Further, the malt itself is chewed to determine its acceptability [8]. Assessing taste and texture of the malt itself, does not give insights into how the wort will taste, which might be the most relevant aspect for brewers. Malt evaluation methods according to the Central European Commission for Brewing Analysis (MEBAK) involve mechanical and physiological tests. The congress mashing procedure provides insights into analytical parameters such as extract, saccharification or filtration rate. At the end, appearance, aroma and taste of the congress wort are assessed, too. Analysis of these parameters is quite time consuming and will not provide a lot of information about the brewing malts flavour compounds. To overcome this, the so-called "Hot Steep Malt Sensory Evaluation Method" (HSME), was developed for Briess Malt & Ingredients in 2015 and was later validated and approved by the American Society of Brewing Chemists Sensory Technical Subcommittee (ASBC) [9]. It only takes about 30 minutes and reduces conversion time and temperature. This increases the chance to perceive malt flavour compounds without the intense sweetness of a congress wort. It is a quick and easy preparation method, which enables the assessment of the brewing malts' sensory profile in a quick and reproducible manner [10]. It is also favourable, if only a small quantity of brewing malt is available [11].

Craine et al. (2021) used HSME in combination with Check-All-That-Apply (CATA) using consumers to study, among other methods, the relationships between genotype and consumer acceptance of hot steep and beer. Consumers were not sensitive enough to differentiate between hot steep samples, but were able to differentiate between beer samples. In case of using consumers for HSME, elaborate training as well as the use of reference stand-

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ards or even the development of a lexicon was recommended for improved results [12]. *Windes* et al. (2021) used, among other methods, HSME to differentiate between various barley genotypes. HSME profiles, gathered by Projective Mapping combined with Ultra Flash Profiling, were found to be more distinct than the sensory profile of the resulting beers. According to the authors, further research is needed to investigate if HSME is suitable for predicting the aroma and flavour profile of resulting beers. Nevertheless, HSME might be useful to establish a common language for maltsters and brewers [11].

Morrissy et al. (2021) conducted HSME in combination with Difference from Control Test and CATA to study, if the incorporation of Maris Otter barley into breeding of experimental genotypes resulted in a distinctive hot steep and beer profile, characteristic for Maris Otter. The authors assume "that hot step sensory may identify malt aromas and flavors that do not readily carry on to finished beer" [13].

CATA Method is a rapid sensory profiling method. It is a frequency-based method and can be used for sensory description of food with trained assessors or with consumers. Sensory assessors are provided with a list of attributes relevant to describe the products tested and are supposed to tick the attributes that are most relevant to describe the individual tested products. The results show how often an attribute is used by the assessors. The more often an attribute is selected, the more relevant it is to describe the product [14]. CATA is considered as a method that is easy to understand and provides reproducible results, when the list of attributes is properly selected [15]. When products are more complex or when products are quite similar to each other, CATA provides comparable results with consumers, but trained assessors were able to identify more differences between the products tested [16]. CATA is widely applied in sensory science and for a wide range of products. *An* and *Lee* (2024) considered CATA as a suitable method to describe orange juice and yoghurt [17]. *Redon* et al. (2023) applied CATA for categorising large numbers of Bordeaux wine in a fast and reproducible way [18]. *Nguyen* et al. (2023) were able to characterise plant-based sausages and meat sausages using CATA in combination with rating of the products overall liking [19]. *Iwamura* et al. (2022) successfully applied CATA for sensory description of gluten-free bread [20], and *Rahardjo* et al. (2022) to characterise dark chocolate from cocoa beans fermented in different ways [21].

Ancient cereals are defined by *Giambanelli* et al. (2013), as "populations not subject to any modern breeding or selection, and sometimes retaining characters of wild ancestors, such as individual variability, height, brittle rachis, low harvest index and, in some taxa, hulled kernels" [22]. Along with the term ancient barley [23], the term heritage barley [24] is also used in scientific literature. According to *Perrino* (2022), there is no clear scientific way to differentiate modern cereal varieties from ancient ones, but often ancient cereal varieties are defined as those planted and processed before the start of the Green Revolution in the 1960s [25]. Despite the fact that ancient barley varieties often do not have the agronomic performance of modern bred varieties, they might be interesting for brewers, especially craft beer brewers, as they can bring in characteristic flavour properties specific to the respective variety [13]. Using ancient barley varieties might become more popular, as nowadays consumers are searching for new flavours and high-quality products [26]. This leads to a still growing market share of craft beer [27].

The aims of this study are (1) to see if HSME in combination with CATA is a suitable method to differentiate barley varieties. Ancient spring barley varieties were used, where a distinctive sensory profile is expected, when compared to modern spring barley varieties. (2) To determine if HSME combined with CATA is suitable for quality control during incoming goods inspection and (3) if off-notes can be detected by applying HSME. (4) Finally, preservation trials with hot steep extracts were conducted, to see, if the samples sensory characteristics might be preserved longer than the four hours recommended between extract preparation and sensory analysis [28].

2 Material and Methods

2.1 Malting

Malting was carried out as a standardised, modified MEBAK small malting process using a rotating germination box. Steeping times were observed according to the regulations, but pre-sorting of the barley was omitted. A kilning programme for Pilsner malt was used, as the formation of melanoidins and other products of non-enzymatic browning reactions was not favoured. The aim was to keep the spring barley varieties' characteristic flavour by using lower kilning temperatures. The same, standardised malting process was applied for all varieties tested. Malting was carried out using three ancient spring barley (*Hordeum vulgare* L.) varieties: Mahndorfer Hanna, a two-row nutans barley variety; Kraffts Riedgerste, a two-row nutans form, and Alpine Pfauengerste, a two-row barley variety with characteristic pyramidal shaped ears. Further, the modern bred spring barley variety Avalon was integrated. All samples were grown in the Lower Rhine region in North Rhine-Westphalia and harvested in 2020. An Avalon industry standard (Durst Malz - Heinrich Durst Malzfabriken GmbH & Co. KG, Bruchsal, Germany, vintage 2020) was also incorporated.

Table 1 Malt analysis MEBAK reference values for an ideal pale malt

	Barley raw protein content (%)	Sorting 1st grade (%)	Moisture content (%)	TKW (g)	HW (kg)	Floating test (%)	Glassiness (%)	Friability value (%)
reference values for pale malt (MEBAK)	10 – 11	85	3.0 – 5.8	25 – 35	48 – 62	35	5	78

TKW = Thousand kernel weight, HW = Hectolitre weight

Table 2 **Congress Mashing Procedure – MEBAK reference values for an ideal pale malt**

	Extract malt dry matter (g/100g)	pH-value	Sacchari-fication time (min.)	Filtration time (min.)
reference values for pale malt (MEBAK)	79 – 82	5.6 – 5.9	< 10	< 60

2.2 Malt analysis

Malt analyses were carried out in triplicate according to MEBAK and involved visual inspection of the malt, taking aspects like odour, colour, shape and lustre into consideration. Further moisture content of malt was determined with an infrared dryer, mechanical and physiological tests involved sieving test for malt, thousand kernel weight (TKW) and hectolitre weight (HW), floating test, endosperm character - glassiness in malt, as well as friability of malt. Raw protein content of barley kernels was determined according to commission regulation (EC) 152/2009, annex III, C [29]. Ideal references values for pale malt according to MEBAK [30] are shown in table 1.

At the end, congress mashing procedure was applied, according to MEBAK [30]. The relevant MEBAK reference values for an ideal pale malt are summarised in table 2.

2.3 Hot steep malt extract preparation

Hot steep malt extract samples were prepared following the instructions of ASBC method Sensory Analysis 14 Hot Steep Malt Sensory Evaluation Method [28] at the Department of Beverage Technology, University of Geisenheim. After filtration, extracts were filled in 0.75 L glass bottles and closed with screw caps. Samples were stored at room temperature prior to sensory analysis, which took place within the recommended time of four hours after hot steep malt extract preparation.

2.4 Sensory analysis – Check-All-That-Apply

Sensory analysis was conducted in the sensory lab at the Department of Oenology, University of Geisenheim, using FIZZ Acquisition, Package 3.7, Couternon, France. The lab was equipped according to DIN EN ISO standard 8589 [31].

Samples were tasted by 16 trained panellists. Panellists underwent basic sensory training according to DIN EN ISO 8586 [32], beer specific training and training on attributes relevant to describe hot steep malt extracts from base malt. Panellist were trained on the execution of CATA method, too. The panellists tasted all samples in three replicates.

For neutralisation, water and unsalted crackers (matzo) were provided to cleanse the panellists' palate.

Hot steep malt extract samples were served in transparent glasses with an amount of 40 mL at room temperature. Samples were presented to panellists in randomised order, coded with three-digit codes and covered with petri dishes for aroma preservation. Panellists were provided with a modified attribute list from Craine et al. (2021), because these parameters -among others- were deemed relevant to describe hot steep malt extracts from ancient spring barley varieties [12]. An internal pre-tasting of hot steep malt extracts showed only minor adjustments were needed. For instance, attributes like "tea", "cucumber", "corn" and "berries" were added. Attributes shown in table I (supplementary material) were deemed appropriate to describe the Pilsner malts of this study. Attributes were presented to panellists in randomised order, to avoid predominant selection of the first few attributes on the list. It is recommended to vary the attribute list between evaluations to avoid bias in the data [33].

For statistical analysis of CATA data, Factorial Correspondence Analysis (FCA) was conducted, using FIZZ Calculation, Package 3.7, Couternon, France. Further, Cochran's Q-Test was applied. For CATA attributes with significant *p*-values, multiple pairwise comparisons using the critical difference (Sheskin) procedure was used, following the approach of Craine et al. (2021) [12] with XLSTAT, 2022.

2.5 Hot steep malt extract preservation

For preservation trials with potassium sorbate, hot steep malt extracts were prepared following the instructions of ASBC method Sensory Analysis 14 [28], using Vienna Malt (Bestmalz, Heidelberg, Germany), to which 200 mg / L potassium sorbate (Vinosorb, Erbslöh, Geisenheim, Germany) was added. Treated samples were filled in brown 0.75 L glass bottles, closed with screw caps and stored for three days at 4 °C.

For pasteurisation trials, hot steep malt extracts were produced using Vienna Malt (Bestmalz, Heidelberg, Germany) according to ASBC Method Sensory Analysis 14 [28]. The hot steep malt extract was filled in 0.75 L bottles, closed with screw caps and placed in a water bath for pasteurisation (20 minutes, 80 °C). Directly afterwards, bottles were cooled by rinsing with cold water and then stored for three days at 4 °C.

To determine if the preserved sample can be differentiated from a freshly produced hot steep malt extract, triangle tests according to DIN EN ISO 4120 [34] were conducted. 30 mL of hot steep malt extract was presented at room temperature in glasses covered with petri dishes for aroma conservation. Samples were coded with three-digit codes and served in randomised order to 16 trained panellists. Panellists underwent basic sensory and beer specific training as well as training on executing triangle tests. Data collection was done computer based using FIZZ Acquisition, package 3.7, Couternon, France. For statistical analysis, FIZZ Calculations, Package 3.7, Couternon, France was used.

3 Results and Discussion

3.1 Malt analysis

3.1.1 Visual Inspection

The colour of Pilsner malt from the project's modern spring barley variety Avalon can be described as dark-yellow, glossy and mostly evenly coloured. The Avalon industry standard showed a straw-yellow colour, with some darker coloured kernels and a glossy surface.

The colour of malt from the ancient spring barley variety Alpine Pfauengerste can be described as dark-yellow with several darker, and some burnt kernels. The surface is less glossy and duller compared to the other varieties. The ancient spring barley variety Kraffts Riedgerste is slightly dark-yellow in colour with some darker kernels. Pilsner malt from the ancient spring barley variety Mahndorfer Hanna showed a quite even colour which can be described as yellow together with a glossy surface. Occasionally some darker kernels can be found.

The modern spring barley variety Avalon from the project showed long, round kernels. The kernels of the Avalon industry standard were larger and even rounder and very regularly shaped, which might be an indicator for a high extract content and might be easier to malt and mill. However, the size of the kernel also depends on barley variety [4].

The shape of the ancient spring barley varieties differed slightly from the modern variety Avalon. For instance, Alpine Pfauengerste showed small, elongated kernels. Some of them were slightly burnt. Normally, this is a result of high kilning temperatures, but here quite low kilning temperatures of 82 °C were used for the production of Pilsner malt [35]. Due to the size and shape of the kernels, the applied kilning temperature might have been sufficient to cause this undesired increase in colour and maybe the formation of unfavourable aroma active substances like Benzaldehyde, which is characterised by almond and burnt sugar notes [36].

The kernels of the ancient spring barley variety Kraffts Riedgerste are average in size but more flat than the modern variety Avalon.

Mahndorfer Hanna is characterised by smaller and slightly longer kernels. In general, the ancient spring barley varieties showed slimmer or smaller, less round kernels compared to the modern barely variety Avalon. Barley showing a larger uniform kernel size is desired by maltsters, because it enables homogenous water uptake as well as modification. Kernels with thin cell walls and loose packed endosperm, large mealy kernels, allow a rapid water up-take as well as a uniform distribution of water and enzymes synthesized during germination [37]. Therefore, it can be expected, that ancient barley varieties take up water in a more inhomogeneous way and show a lower degree of modification.

3.1.2 Aroma, taste and mouthfeel

The Pilsner malt of Avalon industry standard showed a very intense and typical malt aroma. The other malts all were less intense. Pilsner malt from the ancient spring barley varieties Mahndorfer Hanna and Kraffts Riedgerste had more vegetative notes. Especially the aroma of Kraffts Riedgerste was dominated by these vegetative notes. Vegetative notes are already formed during germination by lipoxygenases responsible for the production of the cucumber-like aroma compound (E,Z)-2,6-nonadienal, and other compounds such as hexanal (green-like aroma) and the cardboard-like aroma (E)-2-nonenal [36]. The aroma of the ancient barley variety Alpine Pfauengerste and the Avalon from the project was slightly dull.

All samples, with the exception of Alpine Pfauengerste, showed a relatively sweet taste and were mealy, while the kernels of Alpine Pfauengerste were much harder, less mealy and less sweet in taste.

3.1.3 Mechanical and chemical analysis

Raw protein content of the ancient barley varieties ranged between 12.9 % and 17.1 %. Mahndorfer Hanna showed with 12.9 % the lowest raw protein content of the ancient barley varieties, followed by Kraffts Riedgerste with 13.1 %. Alpine Pfauengerste clearly exceeds the ideal raw protein content or malting barley, with a protein content of 17.1 %. The protein content of the modern spring barley variety Avalon was with 13.0 % also above the ideal protein content for brewing malt suggested by MEBAK [30]. Protein content is an

Table 3 Results mechanical and chemical malt analysis – Pilsner malt, vintage 2020, mean and standard deviation of three replicates, expect for one repetition of barley raw protein content

Variety	Barley raw protein content (%)	Moisture content (%)	Sorting 1st grade (%)	TKW (g)	HW (kg)	Floating test (%)	Glassiness (%)	Friability value (%)
Avalon Industry Standard	–	8.2 ± 0.10	98.5 ± 0.24	43.1 ± 0.21	53.8 ± 0,13	12.3 ± 3.5	0.5 ± 0.5	97.5 ± 0.68
Avalon project	13.0	8.4 ± 0.53	91.7 ± 0.62	39.0 ± 0.36	52.7 ± 0.45	14.5 ± 4.8	2.5 ± 2.18	68.9 ± 0.29
Alpine Pfauengerste	17.1	7.9 ± 0.20	82.5 ± 0.78	32.5 ± 0,25	49.0 ± 0.39	23.2 ± 4.8	6.7 ± 0.58	40.2 ± 1.58
Kraffts Riedgerste	13.1	8.6 ± 0.84	86.7 ± 1,47	33.6 ± 0.15	55.1 ± 0.45	27.5 ± 10.1	5.8 ± 1.26	49.4 ± 0,50
Mahndorfer Hanna	12.9	8.3 0,10	90.6 ± 1,17	36.2 ± 0.34	57.7 ± 0.28	48.2 ± 6.0	8.3 ± 1.53	40.5 ± 1.16

TKW = Thousand kernel weight, HW = Hectolitre weight important parameter with regard to malt and beer production. Barley with high protein content is difficult to process and results in higher malting loss. Each percentage of protein content results in the same percentage of less extract. Protein influences, among other factors, beer foam stability [38], an important factor for German beer consumers [39]. Higher protein content in barley result in difficulties during filtration and might cause turbidity [37, 38]. For Alpine Pfauengerste, which exceeds the ideal protein content of 10 – 11 % by 6 %, it can be concluded that the extract content will also be lower by minimum 6 %. Based on the harvest reports of the last decades, protein content in barley malt decreases. Nevertheless, there are still some "high-protein years", like 2017 – 2020, where the protein content was on average very high [40, 41], this might also explain the high protein content of the modern barley variety Avalon grown within the project. Overall, ancient barley varieties tend to be higher in protein content. It can be assumed that brewing with ancient barley varieties leads to challenges in terms of filtration rate and lower extract of the final beer. This applies predominantly to Alpine Pfauengerste.

The moisture content of the malts studied ranged from 7.9 % – 8.6 % and is therefore higher than the MEBAK reference value [30]. Higher moisture content of malt might lead to lower extract yield per kg and lower milling performance. A higher moisture content will also reduce the malts storage capability [4].

As shown in table 3, results of the sorting test indicate that the modern variety Avalon showed the highest amount of malt of 1st grade. According to Kunze (2016), it can be considered as malt of the highest quality level [38]. The ancient spring barley varieties Mahndorfer Hanna and Kraffts Riedgerste showed 90.6 % and 86.5 % of 1st grade malt, and therefore exceeded the MEBAK minimum requirements [30]. The ancient spring barley variety Alpine Pfauengerste showed only 82.4 % of 1st grade malt and was therefore below the MEBAK minimum requirements. The ancient spring barley varieties showed higher amounts of tailings, formed by small kernels, steam parts and awns, as pre-sorting of the raw material was omitted, while the industry standard was pre-sorted. Possible consequences of malt with lower percentages of 1st grade might be lower extract yields, difficulties during filtration, as well as higher extraction of polyphenols due to other plant materials or broken husks, which might cause bitter flavour and turbidity in the final beer [38]. It can be stated that there is a clear difference in malt quality between the ancient spring barley varieties and the modern variety Avalon, where the ancient spring barley varieties showed a lower malt quality.

None of the samples were below the MEBAK reference value for thousand kernel weight (TKW) [30]. Alpine Pfauengerste (32.5 g) as well as Kraffts Riedgerste (33.6 g) were within the TKW reference value. The two samples of the modern variety Avalon as well as the ancient variety were above the reference value. Kunze (2016) considers TKWs of 30 – 45 g as suitable values for brewing malt [38]. See also table 3. Within these limits, it can be assumed that an increasing TKW correlates with an increasing extract. As all malts tested were within this range, they can be considered as suitable brewing malts, when assessed solely on their TKW.

In terms of hectolitre weight (HW), all samples were within the MEBAK reference value [30]. While Mahndorfer Hanna was at the upper end of the value together with the modern variety Avalon, the ancient variety Alpine Pfauengerste was at the lower end of the reference value (Table 3). HW is not a sufficient indicator for high quality malt [30]. A high HW can be the result of well-modified heavy barley malt, or under-modified light barley malt. Based on the results of the TKW, it can be assumed that the high hectolitre weight of the ancient spring barley varieties Kraffts Riedgerste and Mahndorfer Hanna are based on poor modification and a low kernel weight. However, aspects like the growing environment, as well as other factors like crop management practices and plant diseases will negatively influence yield and grain size and will further lead to increased protein content [37]. In Germany, 2020 was the second warmest year since recording the temperature in 1881 and North Rhine-Westphalia was considered as one of the warmest regions in Germany [42], influencing HW and malt quality, as all samples were from 2020. All samples might have been negatively influenced by above average temperature and dryness.

The amount of kernels floating is an indicator how far the acrospire is developed and how well the malt is modified. Therefore, a high amount of floating kernels is desired. As shown in table 3, the ancient barley variety Mahndorfer Hanna showed the highest percentage (48.2 %) of malt that sinks in water and is therefore above the tolerable level for pale malt, according to MEBAK standards [30]. Based on the results, it looks like malt from Mahndorfer Hanna is not well-modified. The two samples of the modern variety Avalon showed the lowest percentage of malt kernels that sink in water, which indicates a high degree of solubility and a high extract yield. A high degree of solubility and the resulting degradation of β-glucan reduces the risk of filtration problems caused by β-glucan residues or by an increased viscosity and gel formation [38]. In contrast, the ancient barley variety Mahndorfer Hanna has a low degree of solubility resulting in low extract yields.

The malt from all ancient spring barley varieties showed more than 5 % glassy kernels, while the two samples from the modern variety Avalon were below 5 % (Table 3). Ancient barley varieties showed higher levels of glassy kernels than recommended by MEBAK standards [30].

The so-called friability value indicates the degree of modification and measures the homogeneity or evenness of modification, and is an important measurement to predict brewing performance and extract yield [4]. All malt samples from the ancient barley varieties showed lower friability values than the MEBAK reference value, while the industry standard of the modern variety Avalon showed a friability value of 97.5 % (Table 3) [30].

The amount of well-modified kernels shows again a clear difference between the modern spring barley variety Avalon and the ancient varieties. The results of the friability test confirm the previously stated assumption that Mahndorfer Hanna has a high hectolitre weight due to a low degree of solubility caused by a high level of glossy kernels. According to *Narziss* and *Back* (2012), friability values are an important indicator for filtration problems, especially a glassiness above 5 % is an important indicator for filtration problems [35].

Malt with low friability values and a high percentage of glassiness can lead to problems during the lautering process, wort clarification as well as fermentation, beer clarification and filtration. Year of harvest and barley variety might also influence friability values [30]. Based on the results given, it is quite likely that brewing with Pilsner malt from ancient spring barley varieties might cause challenges within the brewing process and lead to lower extract yields due to the uneven breakdown of the cell walls [4]. Therefore, parameters have to be adjusted, to ensure a more even breakdown of the cell walls. It might also be useful to adjust the brewing process, for instance the mashing process. When using ancient spring barley varieties, a traditional standard three-step infusion mashing should be applied, which includes a β-glucanase and protein rest at around 45 °C, an β-amylase rest at approximately 62 °C, and an α-amylase rest at 70 °C [43]. Nowadays often only a two-step infusion mashing is applied, because modern bred barley varieties and highly-modified malt allow mashing in directly at temperatures < 60 °C, because proteins are already broken down [44]. The so-called decoction mashing method, which is suitable for under-modified or enzymatically weak malt and mainly used to produce lager beer [45] might also be an option when brewing with ancient spring barley varieties. An advantage of the decoction method is the support of an enzymatic breakdown by physical-thermal decomposition of starch with the help of the boiling parts of the mash [46]. Of course, the three-step infusion mashing, as well as the decoction mashing are less efficient in terms of time, labour and energy consumption [47]. It might also be recommended to use ancient spring barley varieties for the production of unfiltered beer, as filtration rates during congress mashing procedure were relatively low.

3.1.4 Congress mashing procedure

The saccharification time was below ten minutes for all samples tested, because the relatively low kilning temperatures for Pilsner malt will result in malt with strong enzymatic activity [36]. Based on the implicated enzyme activity, all malts tested can be considered as base malts which will not result in low saccharification times within the brewing process.

In terms of the filtration rate, which predicts lautering performance, clear differences between the samples tested could be found. The filtration rate of the two modern variety Avalon samples can be considered as normal as it was finished after 60 min. The ancient spring barley varieties Mahndorfer Hanna and Kraffts Riedgerste showed a low filtration rate as it took more than 60 minutes to finish. Filtration of Alpine Pfauengerste was stopped after two hours and was therefore considered as too slow. Narziss and Back (2009) reported filtration problems due to a high β-glucan content in inhomogeneous and poorly modified malt [48]. Filtration times of more than 60 minutes will delay the brewing process significantly [37]. Longer filtration and lauter times can be expected when brewing with base malt from ancient spring barley varieties. Therefore, it is recommended to use ancient barley varieties for unfiltered beer and in breweries with a focus on beer specialties and less pressure on production efficiency.

With the exception of Alpine Pfauengerste, the appearance of all congress worts was clear. According to Narziss and Back (2009) a high β-glucan content might be the reason for the turbidity of the congress wort [48] of the ancient spring barley variety Alpine Pfauengerste. In addition, the previously discussed low filtration rate indicates that the ancient spring barley variety Alpine Pfauengerste has a high β-glucan content. All samples showed typical wort aroma notes without any off-notes.

The malt extract value of the industry standard Avalon (82.9 %) ranks above the reference value, while all other samples are below the MEBAK reference value [30]. The modern spring barley variety Avalon showed the second highest extract value. The lowest extract value was determined for the malt from Alpine Pfauengerste (67.0 %), which might also be due to the high protein content, which results in lower extract values [38]. The other two ancient spring barley varieties Kraffts Riedgerste and Mahndorfer Hanna showed relatively similar extract values of 72.2 % and 72.3 % (see Table 4).

The measured pH-value of the congress wort for the two Avalon samples as well as Alpine Pfauengerste (5.8) were within the MEBAK range [30], while Kraffts Riedgerste as well as Mahndorfer Hanna showed an average pH-value of 6.0 and were therefore slightly higher than the ideal pH-range of 5.6 - 5.9. *Kreisz* (2009) considers pH-values between 5.8 and 5.95 as suitable for pale barley malt, as too low values might indicate a strong modification of the malt [49]. Low pH-values will result in a low mashing pH, but other factors like hardness of the brewing water will also have an influence on mashing pH [38]. Results of the congress mashing procedure are summarised in table 4.

Based on the results, it can be stated that the produced Pilsner malt from ancient spring barley varieties showed minor malt quality in comparison to the modern spring barley variety, for instance in terms of lower friability values and lower extract yields and slower filtration rates. Nevertheless, malt quality is, apart from barley variety, also influenced by environmental factors such as weather, type of soil as well as the application of fertilisers [4]. So, for further studies, it is recommended to test malt quality of the same barley

Table 4 Results Congress Mashing Procedure – Pilsner malt, vintage 2020, mean and standard deviation of three replicates

Variety	Extract malt dry matter (g/100g)	pH-value	Saccharification time (min.)	Filtration time (min.)
Avalon Industry Standard	82.9 ± 0.20	5.8 ± 0.01	< 10	< 60 “normal”
Avalon Project	77.0 ± 0.20	5.8 ± 0.04	< 10	< 60 “normal”
Alpine Pfauengerste	67.0 ± 0.93	5.8 ± 0.04	< 10	Stopped after 120
Kraffts Riedgerste	72.2 ± 0.78	6.0 ± 0.02	< 10	> 60 – 120 “slow”
Mahndorfer Hanna	72.3 ± 0.64	6.0 ± 0.01	< 10	> 60 – 120 “slow”

Fig. 1 Changes in colour by pasteurisation: fresh hot steep malt extract (left) and pasteurised hot steep malt extract (right)

variety grown at different locations, treated with different types and concentrations of fertilisers, to see how these parameters influence malt quality of ancient barley varieties.

It can be expected that brewing with Pilsner malt from ancient spring barley varieties might cause difficulties and needs an adjusted brewing process. During mashing, it is recommended to ensure sufficient cytolysis to break down glucans and other filtration inhibitory cell substances in the cell walls, as well as an extended proteolysis. Both are uncommon today due to the use of highly modified malts from modern barley varieties, as with modern brewing barley varieties sufficient cytolysis and proteolysis takes place during the malting process. Due to that, mashing for light beers only needs adjustment for amylolysis [49]. Because of the lower filtration rates seen during the congress mashing procedure, it might be feasible to use ancient spring barley varieties for production of unfiltered beer. The use of ancient spring barley varieties might therefore be interesting for craft brewers as craft beer consumers evaluate the use of ancient barley varieties as beneficial, especially when grown locally [50]. Craft beer consumers are also willing to pay higher prices [51, 52], which makes the use of ancient barley varieties despite higher production costs profitable. Brewing with ancient spring barely varieties might not be relevant for large industry scaled breweries. Craft brewers might use these varieties to bring in the characteristic flavour properties of the respective variety [13] to gain a competitive advantage over other breweries. This might be feasible as nowadays consumers are searching for new flavours and high-quality products [26] leading to a still growing market share of craft beer [27].

3.2 Hot steep malt extract sensory evaluation

The HSME, approved by the ASBC, can be considered as a method easy to execute. It enables a standardised hot steep malt extract preparation for further sensory analysis. However, HSME extract preparation with malt from ancient spring barley varieties might take slightly longer compared to modern varieties, due to lower filtration rates. Sensory analysis of hot steep malt extracts at room temperature is easy to execute and the time of four hours between hot steep malt extract preparation and sensory analysis is feasible. It is helpful to cover the glasses with petri dishes for aroma preservation.

3.2.1 Hot steep malt extract preservation

Results show that hot steep malt extract preservation by pasteurisation is not recommended as this causes changes in colour and taste. A triangle test with 16 trained panellists showed significant differences (*p*-value 0.0159) between a fresh hot steep malt extract and a preserved version. Panellists described differences in sweetness and colour. The change in colour is due to the pasteurisation process (Fig. 1) and might be explained by the formation of Maillard products like melanoidins. *Zufall* (1997) observed an increase in colour by 0.25 European Brewery Convention (EBC) units with 500 pasteurisation units (PU). Within this trial, higher PU were applied, so it can be assumed that the increase in colour will be even higher as stated by Zufall (1997) [53].

Statistical analysis of a triangle test with 16 panellists showed that conserving hot steep malt extracts for three days with potassium sorbate resulted in no significant difference (*p*-value 0.1265) between samples tested. So, the application of potassium sorbate might be a useful method to conserve hot malt steep extracts, when sensory analysis cannot be conducted within the four hours recommended by the ASBC method. Nevertheless, it is still advocated to verify the results with other malt types as well as other types and concentrations of potassium sorbate. Before adding preservatives, it is advisable to check the legal requirements in the respective country, as in some countries the addition of preservatives for experimental sensory analysis may not be acceptable or typical.

3.2.2 Check-All-That-Apply with hot steep malt extract

The results of the FCA for the modality aroma showed that the first axis explains 61.6 % and the second axis further 23.3 % of the data. Based on the FCA plot, the aroma of the modern spring barley variety Avalon (industry standard) is mainly described by a fruity and honey like aroma. One possible aroma active substance which might cause these impressions might be β-damascenone, which is formed in malt from carotenoids [36]. Phenylacetaldehyde and Nonanal are responsible for honey like notes in malt [8]. Benzenealdehyde can be detected in barely itself, as well as 3-Methylbutanal. While Benzenealdehyde is described by honey-like notes, 2-Methylbutanal is responsible for a peach-like aroma [36]. Another substance responsible for fruity notes in malt is 2-Ethylhexanol. Cocal, another substance causing a fruity impression, was only identified in specialty brewing malts and might not be relevant to describe the fruity notes of the Pilsner malt used within this study [8].

The modern spring barley variety Avalon cultivated in the project was described by a bread-like aroma and notes of berries. 2,3,5-Trimethylpyrazin or 2-Ethyl-3,5-Dimethylpyrazin are prevalent substances causing bread-like notes in malt. However, these substances are mainly formed at temperatures at 100 °C and to a higher extent at 180 °C. Therefore, these substances are mainly found in special malts. As the Pilsner malts of this project are kilned at 82 °C, it is more likely that furfural is responsible for the bread-like notes [36] found in malt from the modern barley variety Avalon. Furfural is

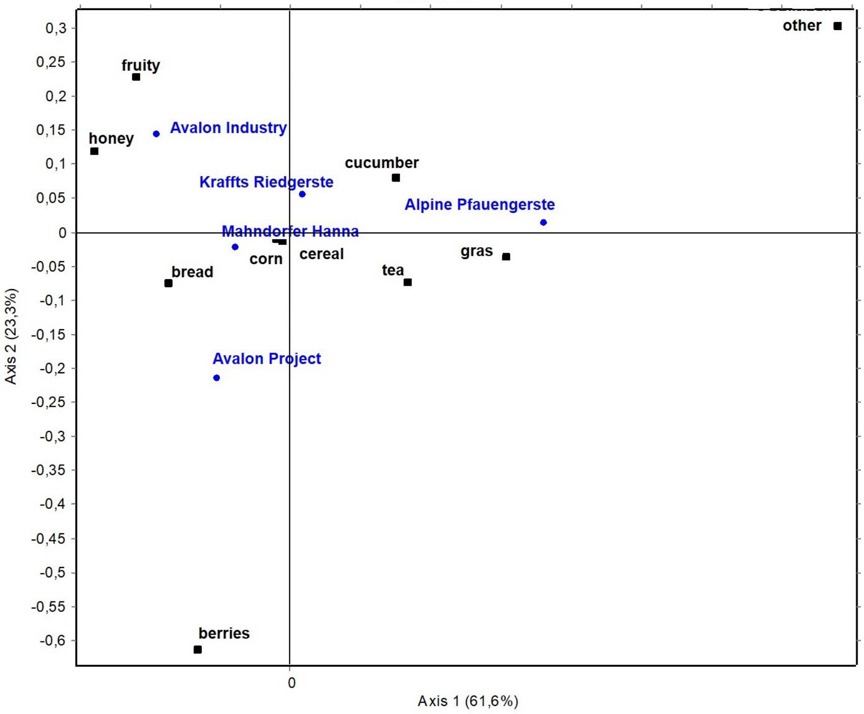

Fig. 2 Results Factorial Correspondence Analysis of Check-All-That-Apply data – Aroma (n = 16 trained panellists, three replicates)

formed within the Maillard reaction or through sugar caramelisation already at lower temperatures of 65 °C during the stewing phase [54]. Substances that might contribute to an aroma associated with berries are 2-Furaldehyde and 5-Methyl-3-Hexen-2-on [11].

The ancient spring barley variety Mahndorfer Hanna is also described with bread-like notes, along with notes of corn and cereal. However, the last two attributes were not relevant to differentiate the samples tested. A corn-like aroma might be caused by dimethyl sulphide, a characteristic compound for off-notes in beer. Dimethyl sulphide can be formed from two precursors: S-methyl methionine, which is mainly formed during malt kilning by thermal decomposition of the malt non-protein amino acid S-methyl methionine [55]. Another relevant precursor is dimethyl sulfoxide, which is reduced to dimethyl sulphide by bacteria in the wort [56]. All samples tasted were described by the attribute cereal. Therefore, this attribute was not relevant to differentiate the samples. One aroma active compound causing cereal-like aroma notes might be 4-Methyl-2-phenyl-2-pentenal [8].

The ancient spring barley variety Alpine Pfauengerste showed a very different sensory profile, with dominant vegetative notes (grass, tea and cucumber). These notes were also found in the ancient spring barley variety Kraffts Riedgerste, but Alpine Pfauengerste is also described by the attribute "others", which includes atypical notes, or even off-notes like dull or musty notes (Fig. 2). One aroma active compound which causes green, grassy notes is Hexanal, which can already be found in the raw barley and its concentration increases during the malting process and will be reduced during thermal treatment within the kilning process [36]. Due to the low kilning temperature of 82 °C, it can be assumed that still perceivable amounts of Hexanal can be found in the hot steep malt extracts. However, *Bettenhausen* et al. (2021) identified 23 compounds which were strongly associated with grassy aroma of hot steep malt extracts [57]. These included alkane/alkenes, benzenoids, organic acids, lipids, and fatty acid esters. Other substances which might be responsible for vegetative notes in wort are 1-Octen-3-ol, Trans-2-Nonenal and Geranylaceton [8]. Tea-like aroma notes in wort might be caused by E-Methylgeranat [11]. To determine which volatile compounds are responsible for the aroma of ancient spring barley varieties, it is recommended to incorporate headspace solid-phase microextraction gas chromatography-mass spectrometry (HS/SPME-GC-MS) as done by Morrissy et al. (2021) [13]. The results of this study indicate that the ancient spring barley variety Alpine Pfauengerste was, in addition to vegetative notes, predominantly described by the attribute "others", which included aromas that were not typical for base malt as well as off-notes. Due to that, HSME in combination with CTA can be considered as suitable approach to test for off-notes in base malt like Pilsner malt. One potential application of both methods could be in quality control. This quick and easy approach to the test for off-notes in base malt, might amend basic malt analysis in the lab. When testing for off-notes, the attribute list must be extended by integrating relevant off-notes like "mouldy", or "lactic". It is also important to train panellists on relevant off-notes by preparing hot steep malt extracts with faulty, i.e. sour our mouldy base malt in different concentrations. Panellists must be able to perceive off-notes even in low concentrations. Benefits of using HSME in combination with CATA for quality control are in the quick and easy procedure that gives maltsters and brewers a good indication on the sensory quality of the malt tested. As it does not need expensive equipment, it might also be feasible for small-scale breweries and malt houses. This approach might also be suitable for breweries during incoming goods inspection to determine the quality of the incoming raw material in a quick and reproducible manner as no time-consuming congress mashing procedure or brewing trials are required to test for off-notes. Off-notes can be detected quickly, even before the start of the actual brewing process, which saves time, money and resources. HSME might also be combined with other sensory evaluation methods applied in quality control, like difference from control test or (descriptive) In Out Test. When working with those tests, an intensive training on malt aroma and off-notes as well as the correct use of the scale has to be done prior to tasting [58]. As this study was solely conducted on base malt, it might also be relevant look into the application of HSME and CATA with speciality malts. The characteristic intense roasted and/or smoked aromas, might over-power some off-notes. This has to be verified in additional research projects.

FCA results of CATA data for the modality taste show that the first axis explains 90.5 % of the data. The second axis adds only 9.4 %. It can be stated that there is a clear distinction between the samples tested. The ancient spring barley variety Mahndorfer Hanna and the modern variety Avalon were described by an umami and sweet taste, the industry standard Avalon as well as the ancient barley variety Kraffts Riedgerste were mainly described by its sweetness. In contrast, the ancient spring barley variety Alpine Pfauengerste showed again a very different taste profile. Here the hot steep malt extract is characterised by a bitter and sour taste (Fig. 3). The dominant sweet taste indicates that the hot steep process modifies a lot of starch out of the malt, which is then degraded by the enzymes α-amylase and β-amylase to sugar. A precondition for this is a well-modified malt with advanced cytolytic and glucan degrading processes, as well as a high enzyme activity. An indicator for a high enzyme activity is a low steeliness as it is found within the Avalon industry standard.

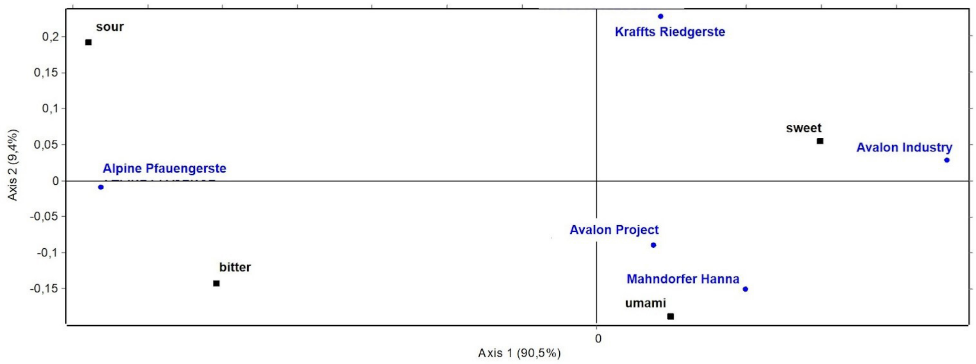

Fig. 3 Results Factorial Correspondence Analysis of Check-All-That-Apply data – Taste (n = 16 trained panellists, three replicates)

Based on figure 4 there are also clear differences in colour of the hot steep malt extract samples. The hot steep malt extract from the ancient spring barley varieties Kraffts Riedgerste and Mahndorfer Hanna are paler than the hot steep malt extract from the modern barley variety Avalon. The darkest and most turbid hot steep malt extract is from the ancient barley variety Alpine Pfauengerste. Therefore, it might also be feasible to include the modality appearance into further CATA studies with hot steep malt extracts. It might also be interesting to see if the colour of the hot steep malt extracts also resulted in beer with comparable colour nuances. So far, studies have focused predominantly on aroma and taste of hot steep malt extracts [12, 13].

Results of the Cochran's Q-Test on CATA data (Tab. II supplementary material) show significant differences in several CATA attributes tested, indicating that HSME in combination with CATA is a suitable procedure to describe hot steep malt extract of Pilsner malt from ancient spring barley as well as modern spring barley varieties. As both modern Avalon samples do not show a very similar sensory profile, especially in aroma, it might also be a suitable method to reflect the influence of growing location as well as type of malting on the sensory profile of hot seep malt extracts. Morrissy et al. (2022) determined a nuanced influence of barley variety as well as growing location on beer flavour [5]. Also, *Stewart* et al. (2022) showed that variety and/or growing environment, or malthouse can lead to differences in malt flavour in wort [59]. Further research might focus on testing the same barley variety grown at different locations, to study, if growing location and environmental factors have an influence on the sensory profile of hot steep malt extracts. It might also be relevant to study, how the malting process influences HSME.

Based on the results of this study, it can be stated, that HSME in combination with CATA is suitable to differentiate Pilsner pale malt. It became obvious that some Pilsner malt from ancient origin showed a distinctive profile to the modern spring barley variety Avalon. The ancient spring barley variety Mahndorfer Hanna was characterised by a relatively similar sensory profile as the modern spring barley variety Avalon, while Kraffts Riedgerste as well as Alpine Pfauengerste had a relatively different sensory profile with more vegetative notes, which were not found in the modern spring barley variety Avalon. Using malt from ancient barley varieties might be of interest for brewers who would like to transfer specific flavours that are characteristic to this variety into the finished beer. One example is the use of Maris Otter, a variety that is no longer recommended due to its poor yield and agronomic performance. The integration of barley varieties specific sensory characteristics into the final beer is often the aim of traditional ale brewers [4]. When applying HSME in combination with CATA to describe malt, the attributes presented to panellists must be selected carefully prior to the actual tasting. The list of attributes may vary based on the sensory profile of the type of malt tested. Speciality malts like roasted, smoked or sour malts show a very distinctive sensory profile in comparison to base malts. Pyrazines, pyridines, and furanes become more abundant in roasted products under dry conditions, moving their flavour towards intense roasted notes. In smoked malt, phenols like guaiacol and eugenol are responsible for the product's characteristic smoky notes [36]. So, when specialty malts have to be evaluated with HSME and CATA, the attribute list has to be adapted and should include aroma attributes like "roasted", "chocolate", "caramel", "burnt", "smoke" or even "lactic" when sour malt has to be assessed. To select appropriate attributes in a pre-tasting, the use of a sensory wheel for brewing malt as developed by *Su* et al. (2022) might be helpful [8].

Fig. 4 Hot steep malt extract colour differences – Pilsner malt

The use of HSME in combination with CATA might be suitable to test for off-notes or mouldy notes in base malt. This might be relevant for instance during incoming goods inspection. This approach might also be helpful for small-scale breweries. Based on the results of this study, and others, HSME can be considered as a method, which enables the assessment of the brewing malts' sensory profile in a quick and reproducible manner [10]. For brewers, it is an opportunity to align the sensory profile of the previously sent malt sample with the sensory quality of the actual received product. This is possible as only a small quantity of malt is required for HSME [11]. Using HSME extracts instead of congress wort makes sensory assessment easier, as the extracts do not show the predominant sweetness of a congress wort and takes less time for preparation [10].

However, if the sensory characteristics of the malts described in the present study can be transferred into the final beer will be evaluated in a follow up project, as this might be a very relevant and interesting aspect for maltsters and brewers, too. Predicting the flavour of the final beer based on the sensory profile of HSME extracts would be very beneficial for maltsters and brewers, as no time and resource consuming brewing trials are required.

4 Conclusion

HSME can be considered as an efficient and easily executable method for assessing the sensory profile of brewing malt. HSME in combination with CATA can be considered as a suitable method to determine the sensory profile of Pilsner malt. As hypothesised, some ancient spring barley varieties showed a distinctive sensory profile compared to the modern barley variety Avalon. Both methods can be applied in quality control for maltsters and brewers. Maltsters might use both methods to determine the quality of their final product in terms of aroma and flavour. The application of HSME together with CATA might also be relevant for brewers: As only a small amount of malt is required, and both methods are suitable for brewers to test the sensory quality of their incoming malt. It is an opportunity to align the sensory profile of the previously sent malt sample with the sensory quality of the actual received product. The use of both methods in quality control for base malt might be beneficial, as they are helpful to test for off-notes in base malt, too. If the sole objective is to test for off-notes with CATA, the relevant off-notes must be incorporated into the attribute list. It is crucial that the attributes are carefully selected and tailored to the specific type of malt presented. For instance, if specialty malt has to be described, the list should include attributes like “chocolate” or “caramel”. Another very important aspect is to train panellists on the attributes provided, using reference standards or malt showing specific aroma notes or even off-notes. It is not recommended to conduct CATA with untrained panellist, when HSME combined with CATA is considered as a quality control test. It is more reasonable to describe the sensory profile of the malts with trained panellists, as they should be able to differentiate between small variations in the malts sensory profile and should be able to detect off-notes even in lower concentrations.

The addition of potassium sorbate might be a useful method to conserve hot steep malt extracts when sensory analysis cannot be conducted within the four hours recommended by the ASBC method. However, the results should be verified with other malt types as well as other types and concentrations of potassium sorbate. Nevertheless, care must be taken, as in some countries the addition of preservatives for experimental sensory analysis may not be acceptable or typical. Therefore, it is advisable to check on regulatory requirements prior to the addition of preservatives.

The two methods in combination were also able to show slight influences of the malting process. For further studies, it might be useful to incorporate analytical tools like headspace solid-phase microextraction gas chromatography-mass spectrometry (HS/SPME-GC-MS) to determine which volatile compounds are responsible for the aroma of ancient spring barley varieties.

Pilsner malt from the three ancient spring barley varieties tested within this study showed minor malt quality in comparison to the modern spring barley variety Avalon, for instance in terms of lower friability values and lower extract yields as well as slower filtration rates. It can be expected that brewing with ancient spring barley varieties might be challenging and needs an adjusted brewing process.

If the sensory profile of malts tested within this study can be transferred to the final beer will be investigated in a subsequent research project, which will also examine consumer acceptance of beer brewed with Pilsner malt from ancient spring barley varieties.

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Declaration of interest

The authors report no conflict of interest. The authors alone are responsible for the content and writing of the paper.

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Supplementary material

Table I Attribute list for Check-All-That-Apply, modified from Craine et al. (2021)

Modality	Attribute	Definition
Aroma	Cereal	Reminiscent of rice, millet, whole grain pasta, slightly earthy
	Cucumber	Reminiscent of fresh cut cucumber
	Bread	Reminiscent of bread crust aroma
	Gras	Reminiscent of freshly cut grass or plants
	Fruity	Reminiscent of fruits like apple, plum, banana or pineapple
	Berries	Reminiscent of raspberries, black berry or red fruit infusions
	Corn	Reminiscent of cooked corn, canned corn
	Tea	Reminiscent of green or black tea
	Honey	Reminiscent of honey
	Others	Missing attributes, and non-characteristic notes like "mouldy", "stale" etc.
Taste	Sweet	Basic taste, reminiscent of sucrose
	Bitter	Basic taste, reminiscent of caffeine
	Sour	Basic taste reminiscent of citric acid
	Umami	Basic taste, reminiscent of monosodium glutamate

Table II Independent comparison of barley varieties hot steep extract (Pilsner malt) for each attribute using Cochran's Q-test to analyse Check-All-That-Apply data, (n = 16 trained panellists, three replicates)

Modality	Attribute	Avalon project	Avalon industry standard	Alpine Pfauengerste	Kraffts Riedgerste	Mahndorfer Hanna	*p*-value
Aroma	Cucumber	0.245 (a)	0.306 (ab)	0.429 (bc)	0.469 (c)	0.327 (ab)	< 0.0001
	Cereal	0.837 (ab)	0.939 (b)	0.857 (ab)	0.755 (a)	0.837 (ab)	< 0.0001
	Fruits	0.082 (a)	0.204 (b)	0.082 (a)	0.112 (ab)	0.143 (ab)	0.000
	Berries	0.122 (b)	0.020 (a)	0.041 (ab)	0.041 (ab)	0.082 (ab)	0.005
	Tea	0.327 (b)	0.265 (ab)	0.388 (b)	0.286 (ab)	0.184 (a)	< 0.0001
	Corn	0.429 (a)	0.469 (ab)	0.429 (a)	0.449 (ab)	0.510 (b)	0.024
	Bread	0.510 (b)	0.510 (b)	0.306 (a)	0.388 (ab)	0.510 (b)	< 0.0001
	Honey	0.347 (ab)	0.571 (c)	0.184 (a)	0.408 (bc)	0.367 (b)	< 0.0001
	Gras	0.265 (a)	0.204 (a)	0.490 (b)	0.347 (ab)	0.286 (a)	< 0.0001
	Others	0.000 (a)	0.041 (ab)	0.122 (b)	0.020 (a)	0.041 (ab)	0.002
Taste	Sweet	0.796 (b)	0.980 (b)	0.449 (a)	0.857 (b)	0.796 (b)	< 0.0001
	Sour	0.143 (ab)	0.041 (a)	0.490 (c)	0.224 (b)	0.082 (ab)	< 0.0001
	Bitter	0.245 (a)	0.082 (a)	0.531 (b)	0.143 (a)	0.204 (a)	< 0.0001
	Umami	0.286 (b)	0.245 (ab)	0.245 (ab)	0.163 (a)	0.306 (b)	0.000

Note: For CATA attributes with significant *p*-values, multiple pairwise comparisons using the critical difference (Sheskin) procedure allow separation of hot steep extract from barley varieties by attribute. Values within a row, which do not share the same letter, are significantly different.