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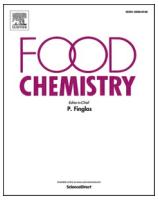

Maiorca wheat malt: A comprehensive analysis of physicochemical properties, volatile compounds, and sensory evaluation in brewing process and final product quality

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ABSTRACT

This study explores the potential of Maiorca wheat malt as an alternative ingredient in beer production, investigating its impact on the brewing process and beer quality at different recipe contents (50 %, 75 %, 100 %). The study encompasses a comprehensive analysis of key malt parameters, revealing Maiorca malt's positive influence on maltose, glucose, filterability, extract, free amino nitrogen, and fermentability. Notably, the malt exhibited heightened levels of α-amylase and β-amylase enzymes compared to conventional commercial malt. Furthermore, the analysis of aroma compounds and subsequent sensory evaluations unveiled a significant correlation between the proportion of Maiorca malt in the formulation and intensified estery, fruity, malty, honey, complemented by a reduction in attributes such as aromatic compounds, phenolic, yeasty, sulfury, oxidized, and solvent-like odors. This research underscores the favorable contribution of Maiorca wheat malt to enhancing both the brewing process and final beer quality, highlighting its potential as an innovative ingredient in brewing practices.

1. Introduction

Beer is one of the oldest and most consumed beverages worldwide. While wheat has historically been utilized for malt and beer production, it has received less research attention compared to barley, which is the predominant grain for brewing (Faltermaier et al., 2014). Wheat has been the most widely used cereal in bakery products, this has undoubtedly led research and breeding towards the selection of varieties that provide optimal characteristics for bread-making purposes and causes the proliferation of inappropriate genotypes to malt and beer production (Faltermaier et al., 2014). Recently, there has been a growing interest in using unconventional cereals for beer production, and some studies have been conducted on Italian wheat varieties to evaluate their malting and brewing performance (Alfeo et al., 2018a, 2021; Baiano, 2021; Blšáková et al., 2021; De Flaviis et al., 2021, De Flaviis et al., 2022a, 2022b, Mascia et al., 2014). As demonstrated by some researchers (Alfeo et al., 2021, De Flaviis et al., 2022, Mascia et al., 2014) many old landraces of wheat that have not been subjected to breeding programs assessing pivotal qualities for the malters and the brewers and could be use as innovative ingredients in brewing practices.

Moreover, the use of old landraces of wheat is strongly linked to the development of sustainable processes and productions, short supply chains, protection of local biodiversity, and reduction of transport and emission with the aim of increasing the well-being and improving the health of current and future generations.

Southern Italy, in particular Sicily, can be considered a remarkable part in terms of old landraces of wheat, barley and spelt (Gallo et al., 2010; Lo Bianco et al., 2017). Recent studies by Alfeo et al. (2018a, 2018b, 2021) on the screening of old Sicilian wheat landraces have shown their suitability in the malt industry and the potential use of some of them in the production of beer with 100 % malted wheat. Considering the most important parameters commonly used to define malt quality, some common and durum wheat varieties such as Romano, Maiorca, Bufala nera corta, and Bufala lunga corta showed excellent characteristics (Alfeo et al., 2021).

The assessment of malt quality hinges on factors such as proteins and

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non-starch polysaccharides, influenced by genetics, growth conditions, and malting processes. Excessive protein and non-starch polysaccharide content in cereals can lead to malt and brewing challenges, including poor modification, low solubility, and inefficient extraction (Jin et al., 2012; Alfeo et al., 2021). However, a balanced protein content contributes positively to foam stability, amino acid production, and Free Amino Nitrogen (FAN) generation, vital for yeast nutrition and fermentation (Hu et al., 2019; Hill & Stewart, 2019).

Some Sicilian varieties such as Maiorca, Romano and Martinello, known for their refined taste and widely used by bakers and pastry chefs, have also demonstrated great potential in malt production. In particular Maiorca (Triticum vulgare Host. Var. albidum Koern), one of the more diffuse in Sicily, has showed excellent characteristics (Alfeo et al, 2021; Benanti et al., 2023).

The malt obtained from Maiorca wheat, when subjected to various malting conditions, has showed a high content of amylolytic enzyme, low protein levels, and reduced non-starch polysaccharides content. These attributes highlight its suitability as base malt for beer production. In light of these observations, this study aims to explore the effects of substituting Maiorca malt at different levels (100 %, 75 %, and 50 %) in beer recipes, assessing changes in physicochemical and sensory attributes of wort and beer. The goal is to establish the viability of creating beer using Maiorca malt as the primary ingredient, a novel approach given the rarity of 100 % wheat malt-based beers.

Currently, Grodziskie beer, a Polish historical style, is the sole example of a 100 % wheat malt beer, featuring smoked wheat malt for a distinct profile. This research seeks to contribute to the craft beer sector by introducing a new 100 % wheat malted beer variety. By harnessing locally sourced wheat malt, this venture offers a unique sensory experience to the beer market and potential cost-saving opportunities for Sicilian breweries.

2. Materials and methods

2.1. Wheat grain

From November 2021 to June 2022, the old common wheat variety (Maiorca, *Triticum vulgare* Host. Var. albidum Koern), cultivated by adopting conventional agronomical management, has been supplied by a commercial cereal farm in Valledolmo (Palermo, Sicily) "MolinOro 100 % grano siciliano" (Lat.37°43' Long. 13°45', 450 m above sea level, sandy clay soil). Samples were harvested in June 2022 and stored at 4–6 °C until malting, which was carried out in October 2022. After malting the malt sample were stored under-vacuum at 12 °C for 3 weeks until the analysis.

2.2. Malting conditions

Malting tests on Maiorca wheat were performed in triplicate in an automatic malting system (Phoenix Biosystems, Adelaide, Australia) in Department of Agricultural, Food and Forest Sciences (SAAF) of the University of Palermo (Italy). Maiorca wheat samples were cleaned to remove the glumes and husks. Malting processes were carried out according to the conditions proposed by Alfeo et al. (2018b). For each basket, 800 g of grains steeped in water at 15 °C for 5 h, followed by 8 h of air-rest, and further 4 h in water, reaching steeping-out moisture of 41 %. The germination occurred after 120 h at 15 °C and 95 % of relative humidity, then the samples were dried and kilned for 34.5 h as follow: 3 h at 55 °C, 12 h at 60 °C, 10 h at 65 °C, 5 h at 70 °C, and 4,5h at 75 °C.

2.3. Malts, hops and yeast strain

Maiorca malt (MM) was used at different percentage (50, 75, 100 %) in the grist. Commercial wheat malt (CWM) (Wheat Best Maltz ®, Heidelberg, Germany) was used as wheat malt for the control beer sample. Commercial barley malt (CBM) (Pilsner Best Maltz ®, Heidelberg, Germany) was used as remaining percentage of grist in beer recipes with 50 and 75 % of wheat malt. Rice husk was also added to the mash as 5 % of wheat malt in the recipe. The German hop (Hallertau Hersbrucker, 4.5 % w/w alpha acids) and the *S. cerevisiae* top-fermented dry yeast of the US-05 strain (Fermentis Division of S.I.Lesaffre, Marcq-en-Baroeul, France) were used as hops and yeast strain, respectively.

2.4. Congress trial conditions

Wheat malts (MM and CWM, respectively) were blended at different percentages (50 % and 75 %) with commercial barley malt (CBM), except for samples with 100 % of wheat malt, respectively MM100 and CWM100, which were tested pure (without addition of barley malt). The six samples obtained, respectively MM100, MM75, MM50, CWM100, CWM75, CWM50, were mashed with I- CUBE MASH BATH - R8 in accordance with Analytica EBC method 4.4 (1997). A portion of 50.0 g of each sample was weight in the mash beakers and the mashing was performed. The filtration was performed using fluted filter paper (Whatman Schleicher & Schuell Qualitative Folded Filter Paper Grade 597 ½; 320 mm Diameter) in a 200 mm diameter funnel with a stem that reaches the bottom of the conical flask. After filtration, the wort was measured in terms of color, pH, extract, and specific gravity according to EBC method (respectively EBC method 8.3 (1997); EBC method 4.5.1 (2004); EBC method 4.4 (1997) and EBC method 8.2.2 (2004)).

The worts obtained (approximately 350 g) were boiled in round bottom flask as proposed in the work of Zdaniewicz et al., 2021 and at the begging of boiling, an amount of 1.5 g /L of the hop was added. After 60 min of boiling all wort samples were cooled at 20 °C and a portion of them was evaluated before inoculation of yeast. The aliquots of boiled and hopped wort (300 g for each recipe) were inoculated with *S. cerevisiae* top-fermented dry yeast of the US-05 strain by Fermentis. All experiments were performed in triplicate.

2.5. Micro-brewing conditions

In this study, the experimental beers were brewed using "all in one" microbrewery plant Klarstein mod. 10,031,629 (Chal-Tec GmbH Berlin, Germany) and fermented in stainless-steel fermenter with hermetic closure. Malts were grinded with a double roller mill (Mattmill Kompakt, Germany), by setting the distance of roller at 1.20 mm and then added to 24L of water. The mash was performed at 68 °C for 60 min, until the complete saccharification tested with iodine solution as reported by Mayer et al. (2016). To inactivate the enzymes, the mixture was heated to 78 °C for 10 min and then the lautering phase was performed with a fist recirculation of wort on the spent grain and rinsed it using 12 L of water heated to 78 °C. The wort obtained (30 L in volume) was boiled for 60 min and at the begin of boiling was added the hop at the quantity of 1.5 g/L, same concentration used in the laboratory scale tests. After boiling, the wort was cooled with stainless-steel chiller until 20 °C and transferred in stainless-steel fermenter to be inoculated with selected yeast strain. Standard quality parameters of beer wort were: 5.3 pH and 12 °P. During fermentation, a temperature of 18 °C was maintained for 16 days. The fermentation was considered complete when the specific gravity was constant for two consecutive days. The beer samples were stored at a temperature of 2 °C for 8 days in order to induce precipitation of suspended yeasts and trubs. The beers were bottled into brown glass bottles and sucrose (9 g/L) was added to perform fermentation in the bottle and to ensure the production of 3 vol of CO_2 per liter of beer.

2.6. Malt and wort analysis

The analyses were performed in triplicate according to the Analytica-EBC, (2007)Analytica European Brewery Convention (EBC) (2007). In details, the moisture (%) of malts was determined by EBC methods 4.2 (2000). Proteins and soluble proteins were calculated as total nitrogen

(TN, dry basis %, db %) and soluble nitrogen (SN, db %), respectively according to the EBC method 4.3.1 (2004) and 4.9.1 (1997) and then multiplied by 6.25. The Kolbach Index (%) was calculated in accordance with EBC methods 4.9.1(1997). The malt extract (db %), extract difference and pH were calculated respectively according to EBC method 4.4 (1997), EBC method 4.5 (1997) and EBC method 4.6 (1997). The saccharification rate, fermentability (%), free amino nitrogen (FAN, mg 100 g^{-1} db), and wort color were determined by EBC methods 4.4.1 (1997), 4.11.1 (1999), 4.10 (1997), and 8.3 (International Method, 1997), respectively. The speed of filtration was measured using EBC methods 4.4.3 (1997) with some modifications, in particular, the filtrate volume was determined at 10, 30, 60,75, and 85 min for each sample. The total volume of filtration was 350 mL for all samples. The Megazyme assay kit (Megazyme International, Ireland) was used to determine total starch content (db %) following the AOAC Method 996.11 (2005) supplied with the assay kit, and a malt amylase assay kit (Megazyme International) was employed to quantify α and β amylases in malt flours. The enzyme activities were measured by reading the assay absorbance using Beckmann DU650 spectrophotometer (Pasadena, California, US) and reported as units per gram of dry matter (U g^{-1}). One unit of activity is defined as the amount of enzyme required to release 1 μmol of reducing sugar equivalents per minute under the defined assay conditions. The β-glucan and β-glucanase content were determined using Megazyme assay kit (respectively K-BGLU and K-MBGL- Megazyme International) following EBC Methods 3.10.1 and 8.13.1 for malt and wort β-glucan content and Azo-barley glucan method for β-glucanase content.

2.7. Congress wort sugar profile

The simple sugars were determined in the congress wort samples. The analysis of fructose, glucose, maltose, and sucrose were performed using the analyzer iCubio iMagic M9 (Shenzhen iCubio Biomedical Technology Co. Ltd. Shenzhen, China) as reported by Matraxia et al. (2021). All reagents and standards were purchased from R-Biopharm AG (Darmstadt, Germany).

2.8. Beer analysis

The measurement of pH of the experimental beer before and after fermentation was conducted with a pH meter Mod.70 XS/50010162 (Cheimika, Pellezzano, Italy). BeerFoss™ FT Go (FOSS Italia srl, Padova, Italy) was used to determine the following parameters of the experimental beer: alcohol (% vol), Original Gravity (SG), Final gravity (SG), Apparent attenuation (%), Real attenuation (%), Original extract (°P), Apparent extract (°P), Real extract (°P), and Alcohol (% Vol).

2.9. Volatile aroma compounds analysis

The aroma volatiles of worts and beers were extracted and analyzed by Headspace-Solid-Phase Microextraction technique (HS-SPME) coupled with Gaschromatography-Mass Spectrometry (GC–MS) following a previously optimized method (Cincotta et al., 2022; Verzera et al., 2021). In detail, a 40 mL vial equipped with a "mininert" valve (Supelco, Bellefonte, PA, USA) was filled with 15 mL of each wort and beer sample by adding 5 g of sodium chloride and stirred at 40 °C for 30 min. Afterwards a DVB/CAR/PDMS (Divinylbenzene/Carboxen/Polydimethylsiloxane) fiber with film thickness of 50/30 μm (Supelco, Bellefonte, PA, USA) was used to perform the extraction of the volatiles in the headspace of the vial for 30 min. After sampling, the SPME fiber was kept for 10 min in the splitless injector at 260 °C of a Shimadzu GC 2010 Plus gas chromatograph directly interfaced with a TQMS 8040 triple quadrupole mass spectrometer (Shimadzu, Milan, Italy) by using the following conditions: polar capillary column, VF-WAXms, 60 m, 0.25 mm i.d., 0.25 μm film thickness (Agilent Technologies Italia S.p.A., Milan, Italy); oven temperature, 45 °C held for 5 min, increased to 80 °C at 10 °C/min and up to 240 °C at 2 °C/min; carrier gas, helium at a constant flow of 1 mL/min; transfer line temperature, 250 °C; acquisition range, 30–400 *m/z*; scan speed, 1428 amu/s. Each compound was successively identified using mass spectral data, the NIST '18 (NIST/EPA/NIH Mass Spectra Library, version 2.0, USA) and FFNSC 3.0 databases, linear retention indices (LRIs) calculated according to the equation of Van Den Dool, & Kratz equation (Van Den Dool and Kratz, 1963), literature data, and the injection of standards. 2-Octanol was used as an internal standard and for quantitative purposes as reported by Medina et al. (2023). The aroma analysis was performed in triplicate.

2.10. Sensory analysis

Quantitative descriptive analysis has been carried out in order to understand the differences between samples and to define the color, odor and taste sensory descriptors, and the overall quality of beer. The selection and training of panelists were performed according to 13.4 EBC methods (1997). For descriptive tests, 10 trained panelists (5 males, 5 females, 23–30 years old) were selected. According to the ISO 8589:2007 standard, tests were carried out under controlled sensory laboratory circumstances. The assessors gave their written consent after receiving full information concerning sensory test. The subjects experienced no danger as a result of the samples odor and taste.

The test was carried out in individual booths under white light at room temperature in Sensory Analysis Laboratory of the University of Palermo (Italy). Approximately 80 mL of beer samples with a temperature of 8 °C were presented to assessors. The presentation order and the identification codes were randomized using Smart Sensory Box software (Smart Sensory Solutions S.r.L., Sassari, Italy). Water was provided to cleanse the palate between the samples.

2.10.1. Descriptive test

Assessors performed descriptive tests to provide a complete sensory description of the product, taking into account all perceived sensations: visual, olfactory, and gustatory features. Thirty-three attributes derived from Beer Flavour Whell developed by Meilgaard et al. (1979) of the America Society of Brewing Chemistry reported in EBC methods 10.12 (1979) were included in the evaluation process. Four attributes were chosen to evaluate visual characteristics including color, turbidity, foam persistency, and foam structure; fifteen were selected related to odor including odor intensity, estery, fruity, floral, hoppy, grainy, honey, malty, caramel, phenolic, solvent-like, diacetyl, sulfury, yeasty, and oxidized; and thirteen gustatory traits included taste intensity, acid, sweet, salty, bitter, estery, fruity, spicy, oxidised, astringent, carbonation, alcoholic, and body. The panelists were asked to rate their overall acceptance. The panelists were said to gradually sip the sample and describe it using the tablet connected to Smart Sensory Box. The sensory attributes were assessed using an unstructured nine-point scale anchored at the left end with "absent" and at the right end with "high".

2.11. Statistical analysis

All the data were evaluated by Matlab software (MathWorks Inc., Nutick, Massachusetts, United States). Sensory data were analyzed using Smart Sensory Box. One-way Analysis of Variance (ANOVA) with Tukey's post hoc test was used for multiple comparisons and a p-value $<$ 0.05 was considered significant. In order to individuate samples with similar characteristics, hierarchical cluster analysis was performed by Matlab software. All experiments were conducted in triplicate and results were reported as mean ± standard deviation.

3. Result and discussion

3.1. Quality parameter of malts

Malt samples were analysed, and the main parameters studied are summarized in Table 1.

Table 1
Malts quality parameters.

Parameter	Maiorca Malt	Commercial Wheat Malt	Commercial Barley Malt
Moisture (% ww-1)	5.29 ± 0.33[a]	5.53 ± 0.09[a]	6.01 ± 0.15[b]
Proteins (db %)	12.34 ± 0.44[b]	11.93 ± 0.06[b]	10.21 ± 0.07[a]
Sol.Proteins (db %)	4.71 ± 0.14[c]	4.17 ± 0.13[b]	3.49 ± 0.20[a]
Starch (db %)	62.60 ± 0.88[b]	58.46 ± 2.61[ab]	55.27 ± 1.12[a]
β-glucan (g 100 g^{-1} db)	0.33 ± 0.07[a]	0.36 ± 0.04[a]	0.74 ± 0.04[b]
Kolbach Index (%)	38.29 ± 2.54[a]	34.95 ± 0.94[a]	34.23 ± 1.80[a]
FAN (mg L^{-1})	104.27 ± 7.36[b]	90.66 ± 2.27[a]	118.60 ± 2.75[c]
β-amylase (BU g^{-1} db)	39.03 ± 0.08[c]	36.86 ± 0.64[b]	13.32 ± 0.99[a]
α-amylase (CU g^{-1} db)	200.95 ± 1.22[b]	71.20 ± 1.59[a]	203.08 ± 2.06[b]
endo- β-glucanases (U kg^{-1} db)	15.97 ± 2.49[a]	17.19 ± 2.35[a]	406.15 ± 3.13[b]
Endo-1,4-β D-xylanase (U g^{-1} db)	1.18 ± 0.02[c]	0.94 ± 0.03[b]	0.71 ± 0.14[a]
Diastatic power (WK)	375.59 ± 2.53[b]	374.51 ± 2.21[b]	357.80 ± 1.95[a]

db = dry basis; FAN = free amino nitrogen; BU = Betamyl Units; CU = Ceralpha Units; U = Units of enzyme; WK = Windish-Kolbach units;
Values in the same line followed by different letter are statistically different ($p < 0.05$).

The moisture content of all samples used in the experiment was in the optimal range to prevent fungal development and any other kind of damage during storage. Normally, the optimal range has a security value of 10–13 % of moisture (Kibar, 2015). Grain moisture is one of the important qualitative parameters; many qualitative attributes depend on this aspect and can be strongly modified during storage when the moisture content is not in the optimal range. Among the losses observed during the storage of cereals in inappropriate humidity conditions, may occur a weight loss due to the grain breathing, as well as a qualitative decrease in terms of taste, smell, nutritional value, metabolites, alteration of the physiological function of cereals (germination and vigor), increased risk of mycotoxins and reduced market value.

The protein content is one of the most important parameters that affect the choice of malt in beer production. As reported in Table 1 the protein content of MM (12.34 ± 0.44 db %) is comparable with the CWM (11.93 ± 0.06 db %). MM as old landraces are often cultivated minimising cultivation input, in particular fertilization has a positive effect on the wheat protein content for the malting process. It is known that the proteins of malt influence several quality attributes of wort and beer. The proteins content, type, and dimensions directly influence the filtration during wort production, the wort fermentability, the foam stability and haze in beer and wort (Faltermaier et al., 2014).

Proteolytic attributes as Kolbach Index (KI), soluble and total nitrogen was also used to characterize malt quality and can be consider a great indicator of malt modification. From the values of soluble and total nitrogen of wort sample, respectively 4.71 ± 0.14 and 4.17 ± 0.13 (db %) for MM and CWM malt, the Kolbach Index (KI) was calculated, which is considered by brewers to be the most important index to evaluate the degree of modification of malt. The samples MM100 and CWM100 showed a comparable value of KI. The KI value of MM was comparable than CWM, respectively 38.29 ± 2.54 % and 34.95 ± 0.94 %, the values obtained showed no statistically significant difference. In both cases, the value of KI indicates the good protein modification achieved by malting. KI is intended as a measure of the degree of protein modification of malt and can be controlled by steeping period; long steeping period results in high KI (Nischwitz et al, 1999). It is important that the KI value would not be excessively high; an optimal KI value for wheat malt is around 39,5 % (Jin et al., 2012). Several authors have found a negative correlation between quality attributes of beer such as foam stability and KI (Evans & Sheehan, 2002). There is also increasing of malting loss with increasing of KI (Alfeo et al., 2021).

Soluble proteins contribute to providing a base for yeast nutrition; generally, are influenced by the proteins type and content in malt, and its degree of modification as well. Within the soluble fraction of the protein constituents, the Free Amino Nitrogen (FAN) comprises individual amino acids and small peptides (consisting of one to three units) that are available for yeast to utilize for cell growth and proliferation (Hill, A. E., & Stewart, G. G., 2019). FAN, together with ammonia, constitutes the amount of yeast absorbable nitrogen that can be measured before fermentation begins. The type of malt proteins affects the FAN content, which especially shows a linear correlation with the wheat gliadin contents, probably due to the more efficient degradation of these types of proteins by aminopeptidase during the germination process FAN content in Maiorca malt showed significantly the highest value compared to CWM and lower value compared CBM.

The starch content is an important parameter because it is the main source of beer extract. The analyzed samples were in the range between 55.27 and 62.60 db % as reported in the Table 1, 62.60 ± 0.88 db % for MM, 58.46 ± 2.61 db % and 55.27 ± 1.12 db % for CWM and CBM, respectively. In our samples, the starch content was highest in the MM and CWM samples (62.60 ± 0.88 g db % and 58.46 ± 2.61 db %, respectively); the lowest value was obtained in the CBM sample (55.27 ± 1.12 db %). The total starch present in the malt is affected by the malting process since a certain quantity of starch is hydrolyzed during germination and is involved in the process of kernel development, as it is the principal source of nutrition and energy for the embryo. An important fraction of polysaccharides in malt is β-glucan; among the unmalted cereals, a higher percentage of this compound is found, between 2.5 and 11.5 %, typically in barley and oat, while in other cereals such as wheat, rye or rice the content of β-glucan is much lower and it is between 0.04 and 2.9 %. (Lazaridou et al., 2007). The β-glucans are water-soluble compounds present in the cell wall of endosperm; they are degraded during the germination process by the action of the β-glucanase enzyme, this is why their content in malt is very low. Low degradation of β-glucan during malting affects the viscosity of the wort, making it extremely viscous and difficult to filter during the filtration phase. A low content of β-glucans is desiderable. In this study, the content of β-glucans in CBM (0.74 ± 0.04 g 100 g^{-1} db) was average with values found in the literature for barley malt (Wang et al., 2004). The content of β-glucans in MM (0.33 ± 0.07 g 100 g^{-1} db) and CWM (0.36 ± 0.04 g 100 g^{-1} db) was lower than in CBM but was in line with values found in the literature for wheat malt (Li et al., 2020). Lower values do not adversely affect final beer qualities, and it has also been shown that the decrease in viscosity due to lower β-glucans content does not adversely affect foam stability (Evans & Sheehan, 2002).

3.2. Congress wort characteristics

Malt extract (% db) is one of the most important qualitative parameters to understand the suitability of malt in beer production.

In the conditions of Congress mash, the extract values of samples were in the range between 79 and 85 %. As shown in Table 2, the highest values in worts were obtained from MM samples. The extract content in the MM samples was higher than in the CWM samples, except for sample CWM50 in which the extract value (83.8 ± 0.37 db %) was comparable with samples MM50, MM75 and MM100. Fine-coarse difference (db %) can be used to determine the modification of malts, in our samples the values showed differences between samples but all within an optimal range for malts (Briggs, 1998). In particular, the lowest values were recorded for MM100, MM75 and MM50, which were very close to the value of the CBM sample. The highest value of was recorded for CWM100 followed by CWM75 and CWM50, which had higher values

Table 2
Quality parameters of congress worts.

Parameter	MM100	MM75	MM50	CWM100	CWM75	CWM50	CBM
Extract (db %)	84.7 ± 1.32^{b}	84.3 ± 0.49^{b}	83.8 ± 0.15^{b}	79.8 ± 1.13^{a}	80.5 ± 0.66^{a}	83.8 ± 0.37^{b}	80.3 ± 0.73^{a}
Fine-coarse difference (db %)	1.46 ± 0.03^{a}	1.43 ± 0.02^{a}	1.40 ± 0.04^{a}	1.60 ± 0.02^{c}	1.58 ± 0.06^{bc}	1.50 ± 0.05^{abc}	1.47 ± 0.04^{ab}
pH	6.14 ± 0.04^{ab}	6.24 ± 0.04^{c}	6.34 ± 0.02^{d}	6.09 ± 0.03^{a}	6.15 ± 0.04^{ab}	6.2 ± 0.02^{bc}	6.11 ± 0.01^{a}
Colour (EBC unit)	4.87 ± 0.11^{c}	4.62 ± 0.13^{bc}	4.45 ± 0.19^{bc}	4.42 ± 0.31^{bc}	4.21 ± 0.22^{ab}	4.37 ± 0.13^{abc}	3.86 ± 0.17^{a}
Fermentability (%)	81.9 ± 0.15^{b}	81.7 ± 0.35^{b}	81.1 ± 0.42^{ab}	80.4 ± 0.56^{a}	80.3 ± 0.7^{a}	81.5 ± 0.32^{ab}	81.7 ± 0.38^{b}
FAN (mg L^{-1})	104 ± 7.36^{bc}	107.9 ± 5.01^{bcd}	111.4 ± 2.72 ^{cd}	90.7 ± 2.31^{a}	97.6 ± 1.56^{ab}	104.6 ± 1.37^{bc}	118.6 ± 2.78^{d}
Saccarification time (min)	<10	<10	<10	10 < s < 15	10 < s < 15	<10	<10
Fructose (g L^{-1})	0.49 ± 0.16^{a}	0.51 ± 0.11^{a}	0.42 ± 0.02^{a}	0.49 ± 0.07^{a}	0.47 ± 0.09^{a}	0.38 ± 0.05^{a}	0.08 ± 0.00^{b}
Glucose (g L^{-1})	6.22 ± 0.53^{bc}	6.23 ± 0.30^{bc}	6.74 ± 0.14^{c}	5.08 ± 0.74^{a}	5.52 ± 0.15^{ab}	5.83 ± 0.13^{abc}	9.41 ± 0.31^{d}
Saccarose (g L^{-1})	5.36 ± 0.92^{ab}	5.55 ± 0.57^{a}	4.98 ± 0.08^{ab}	5.89 ± 1.47^{b}	5.88 ± 1.39^{b}	6.35 ± 0.36^{b}	2.93 ± 0.10^{a}
Maltose (g L^{-1})	36.11 ± 0.20^{a}	35.07 ± 0.92^{a}	35.10 ± 0.1^{a}	35.48 ± 0.52^{a}	35.46 ± 0.41^{a}	35.66 ± 0.55^{a}	40.32 ± 1.37^{b}
Tot. simple sugar (g L^{-1})	48.2 ± 0.61^{a}	47.4 ± 1.73^{a}	47.2 ± 0.25^{a}	46.9 ± 2.09^{a}	47.3 ± 1.62^{a}	48.3 ± 0.83^{a}	52.75 ± 1.79^{b}

s = saccarification; FAN = free amino nitrogen; Congress wort with different content (50, 75, 100 %) of Maiorca malt (MM) and Commercial wheat malt (CWM). CBM = commercial barley malt.
Values in the same line followed by different letter are statistically different ($p < 0.05$).

than the MM samples but still statistically similar to CBM (Table 2). The pH showed a similar value reported in other studies (Alfeo et al., 2021), which was between 6.09 and 6.34.

The fermentability of wort shows the amount of fermentable sugars produced during mashing by the combined effect of alpha and beta-amylase on the starch component.

In this research, fermentability was between 80 and 82 %. The highest level of fermentability was observed in the following samples, respectively: MM100, MM75, MM50, CWM50 and CBM.

All samples under congress mash conditions showed rapid conversion of starch in <10 min, except for the CWM100 and CWM75 in which saccharification took place between 10 and 15 min. As the saccharification is determined by the content of amylolytic enzymes and their catalytic actions, it is a parameter that is strongly influenced by the content of alpha-amylase. This parameter showed a correlation with the content of alpha-amylase in the CWM, which was lower compared to the other malts analyzed (Table 1).

The color of analyzed samples were in the range between 4.2 and 4.9 EBC unit.

In our samples the filtration of total wort volume (350 mL) was obtained between 60 and 82 min. The MM samples showed a good filterability of the congress wort in laboratory conditions. In addition, MM100 and MM75 showed the better filterability compared to others sample. The lowest filtration rate was observed in the CWM100 and CWM50, in which the filtration occurred respectively in 79 and 82 min (Supplementary file – Figure 3). The filterability of wort is often influenced by β-glucan content, as demonstrated in another study, the β-glucan content affects the filtration of beer with different effects in relation to the molecular weight classes. It seems that the low molecular weight β-glucan does not affect the viscosity and then filterability of wort and beer (Sadosky et al., 2002; Kupetz et al., 2015).

3.3. Enzyme activities

During germination, the activation of enzymes allows the hydrolysis of macromolecules that are easier to digest and help to produce compounds necessary for plant growth (Guzmán-Ortiz et al., 2019). The enzyme activity and final enzyme content of malt can be influenced by the kilning phase, especially the temperature used during this step, similar to the effect observed during germination (Briggs, 1998).

Based on the analysis conducted and presented in Table 1, there was a significant variation in the levels of α-amylase observed in the samples. The MM had a similar α-amylase content to that of BM, while the CWM had a low value. The low α-amylase content in CWM was associated with a longer saccharification time for this malt during the Congress mash test. The β-amylase content of malt samples showed similar values to those reported in previous studies on wheat and barley (Alfeo et al., 2018b, 2021; Charmier et al., 2021).

The diastatic power was studied to understand the catalytic activity of amilolytic enzyme. A correlation was found between the β-amylase content and diastatic power, as demonstrated in a previous study by Gibson et al. (1995). This suggests that the total β-amylase content can be utilized to investigate the diastatic power of malt, as they are positively correlated.

The content of β-glucans and β-glucanase is an important indicator of malting performance. In particular, a low level of β-glucans and a high content of β-glucanase are desirable in order to achieve malt with a good yield in brewing (Habschied et al., 2020).

The content of β glucans is influenced by genetics, environmental conditions and the malting process (Izydorczyk and Dexter, 2008). In the malt samples, the β glucans content ranged from 0.32 db % to 0.75 db %, while the content of β-glucanase showed a significant difference between the wheat and barley samples.

No significant difference was found in the β-glucanase level between MM and CWM, which were 16.02 U/kg and 16.82 U/kg, respectively. However, in CBM, the content was significantly higher compared to the wheat samples, precisely 406.85 U/kg. The result was in line with those found in the literature for wheat and barley malt (Alfeo et al., 2018b, 2021; Wang et al., 2004).

3.4. Congress wort sugar profile

The wort sugar profiles of the studied wheat malts did not show statistically significant differences, except for the glucose content. The levels of simple sugars in the analyzed wort ranged from 46.9 g/L to 48.3 g/L for CWM100 and CWM50, respectively, as shown in the Table 2. These values were obtained by summing the amounts of fructose, glucose, sucrose, and maltose. No significant differences were found in the fructose, saccarose and maltose content among the studied worts. The content of glucose was in the range of 5.08–6,74 g/L, respectively for CWM100 and MM50. In general, the higher content of glucose was found in the worts brewed with Maiorca. The beer wort usually contains 60 % maltose and only 10 % glucose (Younis and Stewart, 1998).

The glucose content can be increased to higher levels through specific mashing conditions that ensure maltase activity, which breaks down maltose into two molecules of glucose (Gresser, 2010). The higher content of glucose in the wort's sugar profile is important for modifying the secondary products of fermentation; allowing for an increase in the content of ethyl- and isoamyl-acetate (banana flavor), which are characteristic flavors of German wheat beer. The observed amount of maltose and in general the simple sugar composition in the wort were consistent with those derived from wheat malt in a previous study by Alfeo et al. (2021).

3.5. Physico-chemical characteristics of experimental brewing test

The physico-chemical characteristics of the experimental beer were reported in Table 3. All experimental worts and beers had similar pH values before and after fermentation, with the initial pH of the wort ranging between 5.32 and 5.37. After fermentation, the pH decreased to values between 4.29 and 4.39. No significant differences were observed among the different wheat beers produced. However, statistical significance differences ($p < 0.05$) were reported for original gravity. The higher values were found in MM50 and MM100, with values of 1.049 ± 0.01 and 1.050 ± 0.01, respectively, while the lower value was observed in CWM100, with a value of 1.047 ± 0.01. Similar significant differences were also observed in the beer samples for original extract (°P) and alcohol content (%Vol), as these parameters are positively correlated with original gravity. The original extract (°P) in the beer samples ranged from 11.75 ± 0.28 to 12.31 ± 0.14, with higher values observed in MM100 (12.31 ± 0.14 °P) and MM50 (12.23 ± 0.14 °P), and a lower in CWM100 (11.75 ± 0.28 °P). The alcohol content showed values between 4.8 ± 0.17 and 5.27 ± 0,15 (%Vol), with higher values in MM100 and MM50, and a lower level in CWM100. The results of apparent attenuation (Table 3) obtained in the experimental beer were in line with the fermentability values (Table 2) of the congress wort, demonstrating the same behavior of yeast in both congress and a micro-scale conditions. No significant differences were found among beers for density FG, real attenuation, apparent extract and real extract.

3.6. Volatile compound composition of congress wort

The results of the analysis of volatile compounds in the congress wort are presented in Table 4. Most of these compounds were confirmed using reference compounds from previous studies on the analysis of volatile compounds in beer wort (De Schutter et al., 2008a; De Schutter et al., 2008b). The identified compounds in the beer samples were categorized into eight classes: organic acids, alcohols, aldehydes, ketones, esters, terpenes, aromatic compounds and furans.

Among all the compounds, aldehydes were the most abundant, with contents ranging from 201.06 to 417.63 mg/L. The samples MM50, and CWM100 showed the highest values, with 417.63 and 349.20 mg/L, respectively, while the lowest values were found in MM100, MM75 with an amount of 201.06 and 235.73 mg/L, respectively. Among the aldehydes identified in the samples, the most abundant were 2-methyl-butanal, hexanal, and decanal; the latter was highest in MM100.

Alcohols were detected in the congress worts with a content ranging from 92.67 to 150.53 mg/L. The percentage content of total alcohols was the most abundant in the beers with Maiorca malt. Among the most abundant alcohols identified were 1-octanol, 1-octen-3-ol, 1-dodecanol and 2-ethyl-1-hexanol.

The class of esters showed statistically significant differences, as indicated by the clustergram analysis (Fig. 2a). The content of these compounds ranged from 103.52 to 2011.71 mg/L. Specifically, the highest value was found in CWM100, followed by CWM75 and CWM50 while the lowest values were observed in MM samples. These compounds are initially present in small amounts in the wort and increase during fermentation as by-products of yeast metabolism (He et al., 2014; Hiralal et al., 2014). Among the different volatile compounds identified in beer worts, ketones were among the most significant constituents. Their content in congress wort samples ranged from 89.60 to 11.19 mg/L. Overall, these compounds were found to be more abundant in worts with Maiorca malt. Nerylacetone and 6-methyl-5-hepten-2-one were identified as the dominant ketones in this samples.

3.7. Volatiles organic compound composition of beer

The volatile composition of the experimental beers is reported in Table 4. The analysis revealed more than 90 volatile compounds belonging to eight classes, including organic acids, alcohols, aldehydes, esters, ketones, terpenes and aromatic compounds. Many of the compounds identified are known to contribute to the characteristic's flavors of wheat beer, as demonstrated by several articles in the literature (De Flaviis et al., 2021; De Flaviis et al., 2022; Li et al., 2012; Langos et al., 2013).

The beers analyzed showed distinct aroma profiles, as depicted in the clustergram (Fig. 2b). Among the various compound classes, esters were the most prominent in terms of number of compounds and abundance with ethyl acetate, isoamyl acetate, ethyl hexanoate, ethyl octanoate, isoamyl formate, ehtyl hexanoate, 2-phenethyl acetate, ethyl-9-decanoate, and ethyl dodecanoate the main compound. Ethyl octanotate was particularly prevalent in MM100 and MM75 reaching the amount of 3.6 g/L. Esters are the main contributors of sweet and fruity aroma and most of them exceeded in a remarkable way their Odor Threshold Value (OTV) has happened for ethyl acetate and isoamyl acetate, and ethyl hexanoate and ethyl octanoate (Supplementary file – Table 5).

The presence of organic acids was prominent, too. CWM100 displayed the lowest amount of organic acids among the experimental beers, reaching the value of 1.38 g/L. The octanoic, decanoic and nonanoic acid were identified as the most abundant. Notable variations in octanoic acid content were observed among the different experimental beers, ranging from 353 to 701 mg/L, with CWM100 having the lowest value and MM beers the highest. Additionally, statistically significant differences were noted in the content of hexanoic acid, which content was extremely lower in MM samples. 3 methyl butanoic acid was another important contributor in the aroma of beer with and OAV ranged between 788 and 996 (Supplementary file – Table 5) being responsible of sweet and cheese flavor.

Alcohols were mainly characterized by the presence of 2-methyl-1-propanol. This compound, along with esters, is well-documented as typical components of wheat beers (De Flaviis et al., 2022a, Mascia

Table 3
Beer and wort quality parameters.

Parameter	MM100	MM75	MM50	CWM100	CWM75	CWM50
Wort pH	5.32 ± 0.07^{a}	5.36 ± 0.07^{a}	5.34 ± 0.08^{a}	5.37 ± 0.06^{a}	5.35 ± 0.04^{a}	5.32 ± 0.05^{a}
Beer pH	4.31 ± 0.05^{a}	4.41 ± 0.04^{a}	4.39 ± 0.01 ^{a}	4.29 ± 0.06^{a}	4.37 ± 0.04^{a}	4.39 ± 0.10^{a}
Density OG	1.050 ± 0.01^{b}	1.049 ± 0.01^{ab}	1.049 ± 0.01^{b}	1.047 ± 0.01^{a}	1.048 ± 0.01^{ab}	1.049 ± 0.01^{ab}
Density FG	1.010 ± 0.01^{a}	1.010 ± 0.01^{a}	1.009 ± 0.01^{a}	1.011 ± 0.01^{a}	1.011 ± 0.01^{a}	1.010 ± 0.01^{a}
Apparent attenuation (%)	79.67 ± 1.15^{a}	79 ± 0.00^{a}	80.33 ± 1.15^{a}	77 ± 2.65^{a}	76.33 ± 4.16^{a}	79.67 ± 1.15^{a}
Real attenuation (%)	64.67 ± 1.15^{a}	64 ± 0.00^{a}	65.33 ± 1.15^{a}	62.33 ± 2.08^{a}	62 ± 3.61^{a}	64.67 ± 1.15^{a}
Original extract (°P)	12.31 ± 0.14^{b}	12.07 ± 0.14^{ab}	12.23 ± 0.14^{b}	11.75 ± 0.28^{a}	11.83 ± 0.37^{ab}	12.15 ± 0.24^{ab}
Apparent extract (°P)	2.48 ± 0.14^{a}	2.56 ± 0.01^{a}	2.39 ± 0.14^{a}	2.73 ± 0.29^{a}	2.82 ± 0.44^{a}	2.48 ± 0.14^{a}
Real extract (°P)	4.25 ± 0.11^{a}	4.28 ± 0.02^{a}	4.17 ± 0.11^{a}	4.36 ± 0.27^{a}	4.44 ± 0.36^{a}	4.23 ± 0.13^{a}
Alcohol (% Vol)	5.27 ± 0.15^{b}	5.07 ± 0.06^{ab}	5.27 ± 0.15^{b}	4.8 ± 0.17^{a}	4.83 ± 0.31^{ab}	5.2 ± 0.17^{b}

OG = original gravity; FG = final gravity; °P = Plato;
Worts and beers produced by micro-brewing trials with different content (50, 75, 100 %) of Maiorca malt (MM) and Commercial wheat malt (CWM).
Values in the same line followed by different letter are statistically different ($p < 0.05$).

Table 4
Volatile compounds, linear retention indices and odor descriptors identified in worts and beers.

Compound	Odor	LRI	MM100	MM75	MM50	CWM100	CWM75	CWM50	Reference
Wort volatile compounds									
Acids									
3-methyl-butanoic acid	sweaty	1674	5.96 ± 0.14^{c}	5.39 ± 0.11^{b}	6.15 ± 0.13^{c}	3.65 ± 0.07^{a}	3.51 ± 0.07^{a}	3.23 ± 0.06^{e}	8
Hexanoic acid	sweaty	1844	10.27 ± 0.21^{b}	11.08 ± 0.19^{b}	11.83 ± 0.2^{b}	13.02 ± 0.27^{c}	13.85 ± 0.29^{c}	7.93 ± 0.16^{a}	2,5,6,8,9
Nonanoic acid	waxy	2167	23.68 ± 0.50^{b}	19.13 ± 0.29^{a}	17.15 ± 0.36^{a}	38.86 ± 0.82^{e}	32.61 ± 0.26^{d}	27.62 ± 0.58^{c}	2,5,9
Decanoic acid	fatty	2275	4.51 ± 0.09^{a}	4.73 ± 0.10^{a}	4.33 ± 0.09^{a}	20.38 ± 0.43^{c}	6.63 ± 0.14^{b}	6.39 ± 0.13^{b}	2,5,8,9
Pentadecanoic acid	waxy	2782	3.54 ± 0.07^{b}	2.66 ± 0.05^{a}	2.63 ± 0.05^{a}	17.77 ± 0.37^{e}	12.75 ± 0.05^{d}	5.35 ± 0.11^{c}	8,9
Total			***47.98 ± 1.03^{b}***	***43.01 ± 0.76^{a}***	***42.11 ± 0.89^{a}***	***93.71 ± 1.98^{d}***	***69.37 ± 0.83^{c}***	***50.54 ± 1.07^{b}***	
Alcohols									
1-Penten-3-ol	ethereal	1158	3.52 ± 0.07^{c}	5.46 ± 0.11^{d}	5.86 ± 0.12^{e}	2.03 ± 0.04^{a}	2.70 ± 0.05^{a}	2.19 ± 0.04^{b}	8,9
1-Pentanol	yeasty	1224	3.27 ± 0.06^{a}	11.58 ± 0.24^{c}	17.35 ± 0.36^{d}	11.25 ± 0.23^{c}	5.16 ± 0.10^{b}	2.60 ± 0.05^{a}	8,9
(Z)-2-Penten-1-ol	fruity	1319	2.92 ± 0.06^{c}	4.89 ± 0.10^{d}	6.38 ± 0.13^{e}	1.50 ± 0.03^{a}	2.75 ± 0.05^{c}	2.20 ± 0.04^{b}	9, 10
1-Heptanol	green	1448	2.87 ± 0.0^{a}	2.86 ± 0.03^{a}	2.61 ± 0.05^{a}	4.5 ± 0.09^{b}	4.16 ± 0.08^{b}	2.99 ± 0.06^{a}	3,4,9
1-Octanol	green	1453	9.02 ± 0.19^{bc}	6.90 ± 0.14^{a}	8.41 ± 0.17^{b}	10.19 ± 0.21^{d}	9.79 ± 0.20 ^{cd}	11.17 ± 0.23^{e}	9, 10
1-Octen-3-ol	mushroom	1556	70.61 ± 1.49^{e}	60.40 ± 1.28^{d}	61.88 ± 1.31^{d}	37.64 ± 0.79^{c}	27.03 ± 0.99^{b}	24.06 ± 0.50^{a}	9, 10
1-Nonanol	aldehydic	1659	6.57 ± 0.13^{c}	3.98 ± 0.08^{a}	4.11 ± 0.08^{a}	5.25 ± 0.11^{b}	6.83 ± 0.14^{d}	6.36 ± 0.13^{c}	9, 10
Phenethyl alcohol	floral	1912	5.69 ± 0.12^{a}	7.43 ± 0.15^{b}	7.62 ± 0.16^{b}	8.45 ± 0.17^{c}	8.44 ± 0.17^{c}	9.22 ± 0.19^{d}	9, 10
1-Dodecanol	honey	1965	30.79 ± 0.56^{d}	29.85 ± 0.63^{d}	22.93 ± 0.48^{c}	16.13 ± 0.34^{ab}	17.75 ± 0.37^{b}	15.39 ± 0.32^{a}	3,4,9
2-Ethyl-1-hexanol	citrus	1488	16.21 ± 0.34^{d}	14.48 ± 0.30^{c}	11.95 ± 0.25^{b}	8.04 ± 0.17a	15.93 ± 0.33^{d}	16.43 ± 0.34^{d}	1,9
Total			***150.53 ± 3.12^{e}***	***147.89 ± 3.10^{d}***	***149.14 ± 3.15^{d}***	***105.04 ± 2.22^{c}***	***100.59 ± 2.55^{b}***	***92.67 ± 1.95^{a}***	
Aldehydes									
2-methyl-propanal	aldehydic	815	2.27 ± 0.04^{b}	2.32 ± 0.04^{b}	2.19 ± 0.04^{b}	11.01 ± 0.23^{d}	2.93 ± 0.06^{c}	1.04 ± 0.02^{a}	3,9
2-methyl-butanal	aldehydic	917	13.83 ± 0.29^{d}	12.13 ± 0.25^{c}	11.61 ± 0.24^{c}	15.43 ± 0.32^{e}	10.59 ± 0.22^{6b}	5.75 ± 0.12^{a}	3,9
3-methyl-butanal	chocolate	921	22.99 ± 0.48^{b}	25.18 ± 0.53^{b}	29.77 ± 0.63^{c}	46.29 ± 0.98^{d}	31.13 ± 0.65^{c}	17.12 ± 0.36^{a}	3,4,9
Pentanal	fermented	984	4.99 ± 0.10^{a}	6.44 ± 0.13^{b}	8.68 ± 0.18^{c}	13.60 ± 0.28^{e}	12.60 ± 0.26^{d}	5.96 ± 0.12^{b}	3,4,9
Hexanal	green	1084	28.19 ± 0.59^{a}	94.42 ± 2.00^{b}	246.96 ± 5.23^{e}	116.54 ± 2.47^{c}	114.55 ± 4.55^{c}	137.03 ± 2.90^{d}	3,4,9
Heptanal	green	1188	2.81 ± 0.05^{a}	2.78 ± 0.05^{a}	9.12 ± 0.19^{b}	17.11 ± 0.36^{d}	15.69 ± 0.28^{c}	14.48 ± 0.30^{c}	3,4,9
Octanal	aldehydic	1293	6.97 ± 0.14^{a}	6.98 ± 0.14^{a}	6.68 ± 0.14^{a}	9.09 ± 0.19^{b}	9.04 ± 0.19^{b}	13.05 ± 0.27^{c}	3,4,9
(Z)-2-Heptenal	–	1329	2.70 ± 0.05^{a}	5.57 ± 0.11^{b}	12.43 ± 0.26^{d}	6.20 ± 0.13^{c}	6.25 ± 0.13^{c}	5.23 ± 0.10^{b}	3,4,9
Nonanal	aldehydic	1398	19.51 ± 0.41^{a}	24.54 ± 0.51^{b}	24.41 ± 0.51^{b}	40.66 ± 0.86^{c}	46.97 ± 0.78^{d}	56.71 ± 1.20^{e}	3,4,8,9
Decanal	aldehydic	1501	88.08 ± 1.86^{c}	42.44 ± 0.68^{a}	42.63 ± 0.90^{a}	44.49 ± 0.94^{a}	35.04 ± 0.74^{b}	32.44 ± 0.68^{b}	3,4,8,9
Benzaldehyde	almond	1530	1.95 ± 0.04^{a}	5.71 ± 0.11^{b}	11.95 ± 0.25^{c}	14.13 ± 0.29^{d}	13.18 ± 0.34^{d}	11.93 ± 0.25^{c}	3,4,9
(E)-2-Nonenal	fatty	1539	6.67 ± 0.14^{a}	7.17 ± 0.15^{a}	11.15 ± 0.23^{b}	14.60 ± 0.30^{c}	14.98 ± 0.31^{c}	18.52 ± 0.39^{d}	3,4
Total			***201.06 ± 4.24^{a}***	***235.73 ± 4.77^{b}***	***417.63 ± 8.84^{e}***	***349.20 ± 7.39^{d}***	***313.02 ± 8.55^{c}***	***319.31 ± 6.75^{c}***	
Ketones									
Heptan-2-one	cheesy	1185	6.58 ± 0.13^{a}	7.57 ± 0.15^{b}	9.40 ± 0.19^{c}	11.06 ± 0.23^{d}	12.35 ± 0.26^{e}	17.18 ± 0.15^{f}	5,8,9,10
2-Octanone	earthy	1288	6.60 ± 0.13^{b}	6.51 ± 0.13^{b}	5.69 ± 0.01^{a}	8.57 ± 0.18^{d}	8.90 ± 0.23^{d}	7.51 ± 0.15^{c}	9
2,5-Octanedione	sweet	1325	4.79 ± 0.10^{a}	6.38 ± 0.13^{c}	7.48 ± 0.15^{d}	5.46 ± 0.11^{b}	6.23 ± 0.19^{c}	6.20 ± 0.13^{c}	5,9,10
6-Methyl-5-Hepten-2-one	citrus	1339	32.00 ± 0.67^{d}	25.83 ± 0.33^{c}	21.04 ± 0.44^{b}	23.92 ± 0.50^{bc}	18.01 ± 0.31^{a}	17.60 ± 0.37^{a}	5,9,10
Oct-3-en-2-one	earthy	1412	2.64 ± 0.05^{a}	3.54 ± 0.07^{b}	7.01 ± 0.14^{d}	3.70 ± 0.07^{b}	4.93 ± 0.10^{c}	2.99 ± 0.06^{a}	5,9
Nerylacetone	fatty	1855	52.85 ± 1.12^{c}	52.82 ± 1.11^{c}	60.54 ± 1.28^{d}	47.46 ± 1.00^{b}	45.78 ± 0.97^{b}	38.09 ± 0.80^{a}	na
Total			***105.50 ± 2.23^{d}***	***102.67 ± 1.95^{c}***	***111.19 ± 2.24^{e}***	***100.19 ± 2.11^{c}***	***94.22 ± 2.07^{b}***	***89.60 ± 1.68^{a}***	
Esters									
Ethyl acetate	fruity	891	2.79 ± 0.05^{c}	1.55 ± 0.03^{b}	1.26 ± 0.02^{a}	1.38 ± 0.02^{ab}	1.27 ± 0.02^{a}	4.64 ± 0.09^{d}	8,9
Isoamyl acetate	sweaty	1122	11.09 ± 0.23^{a}	11.34 ± 0.15^{a}	11.29 ± 0.23^{a}	15.11 ± 0.31^{b}	17.96 ± 0.16^{c}	23.73 ± 0.50^{d}	5,6,8,10
Ethyl hexanoate	green	1233	9.13 ± 0.19^{b}	9.74 ± 0.12^{b}	8.15 ± 0.17^{a}	13.33 ± 0.28^{c}	17.15 ± 0.15^{d}	23.42 ± 0.49^{e}	8, 9
Hexyl formate	fruity	1350	11.08 ± 0.23^{a}	11.52 ± 0.24^{a}	14.02 ± 0.29^{b}	42.36 ± 0.89^{d}	40.97 ± 0.86^{d}	16.76 ± 0.35^{c}	8,9
Ethyl octanoate	waxy	1436	37.74 ± 0.79^{a}	41.21 ± 0.44^{a}	55.61 ± 1.17^{b}	92.35 ± 1.95^{d}	87.26 ± 0.57^{c}	86.58 ± 1.83^{c}	4,5,8
Ethyl decanoate	waxy	1639	21.77 ± 0.46^{b}	19.18 ± 0.12^{a}	19.34 ± 0.40^{a}	24.63 ± 0.52^{c}	23.64 ± 0.50^{c}	29.04 ± 0.61^{d}	8
Ethyl 9-decenoate	fruity	1691	2.06 ± 0.04^{b}	1.18 ± 0.02^{a}	1.18 ± 0.02^{a}	7.5 ± 0.15^{d}	6.79 ± 0.03^{c}	6.22 ± 0.13^{c}	8,9
Ethy dodecanoate	waxy	1843	5.64 ± 0.11^{c}	3.19 ± 0.06^{b}	2.87 ± 0.05^{a}	7.65 ± 0.16^{d}	7.32 ± 0.04^{d}	11.68 ± 0.24^{e}	na
Ethyl palmitate	–	2253	2.95 ± 0.06^{a}	3.55 ± 0.03^{b}	3.64 ± 0.07^{b}	7.34 ± 0.15^{d}	5.76 ± 0.12^{c}	5.43 ± 0.11^{c}	na
Total			***104.30 ± 2.20^{a}***	***103.52 ± 1.25^{a}***	***117.41 ± 2.47^{b}***	***211.71 ± 4.47^{d}***	***208.16 ± 2.49^{c}***	***207.55 ± 4.39^{c}***	
Terpenes									
Eucalyptol	camphoreous	1211	0.0027 ± 0.0001^{e}	0.0019 ± 0.0000^{c}	0.0017 ± 0.0000^{b}	0.0015 ± 0.0000^{a}	0.0017 ± 0.0000^{bc}	0.0021 ± 0.0000^{d}	na
Dihydromyrcenol	citrus	1466	0.0068 ± 0.0001^{e}	0.0061 ± 0.0001^{d}	0.0064 ± 0.0001^{de}	0.0009 ± 0.0000^{a}	0.0046 ± 0.0001^{b}	0.0053 ± 0.0001^{c}	na
Linalool	floreal	1546	0.0034 ± 0.0001^{b}	0.0026 ± 0.0001^{a}	0.0026 ± 0.0001^{a}	0.0032 ± 0.0001^{b}	0.0026 ± 0.0001^{a}	0.0029 ± 0.0001^{a}	7,9
Total			***0.0129 ± 0.0003^{d}***	***0.0106 ± 0.0002^{c}***	***0.0108 ± 0.0002^{c}***	***0.0056 ± 0.0001^{a}***	***0.0089 ± 0.0002^{b}***	***0.0103 ± 0.0002^{c}***	
Aromatic compounds									
Styrene	balsamic	1262	2.68 ± 0.05^{c}	1.84 ± 0.037^{a}	2.03 ± 0.04^{a}	2.38 ± 0.04^{b}	2.24 ± 0.04^{b}	2.89 ± 0.06^{d}	3,4

(*continued on next page*)

Table 4 (*continued*)

Compound	Odor	LRI	MM100	MM75	MM50	CWM100	CWM75	CWM50	Reference
Total			***2.68 ± 0.05^{c}***	***1.84 ± 0.037^{a}***	***2.03 ± 0.04^{a}***	***2.38 ± 0.04^{b}***	***2.24 ± 0.04^{b}***	***2.89 ± 0.06^{d}***	
Furans									
2-ethyl-furan	–	957	0.0081 ± 0.0002^{d}	0.008 ± 0.0002^{d}	0.0071 ± 0.0001^{c}	0.0067 ± 0.0001^{bc}	0.0062 ± 0.0001^{b}	0.0053 ± 0.0001^{a}	na
2-pentyl-Furan	–	1231	0.0139 ± 0.0003^{c}	0.0172 ± 0.0004^{e}	0.0164 ± 0.0003^{de}	0.0153 ± 0.0003^{d}	0.0123 ± 0.0003^{b}	0.0073 ± 0.0002^{a}	na
			0.0220 ± 0.0005^{c}	***0.0252 ± 0.0005^{d}***	***0.0235 ± 0.0005 ^{cd}***	***0.0220 ± 0.0005^{c}***	***0.0185 ± 0.0004^{b}***	***0.0126 ± 0.0003^{a}***	
Beer volatiles compounds									
Acids									
Butanoic acid	sweaty	1038	45.47 ± 2.34^{b}	51.11 ± 0.72^{bc}	53.12 ± 1.97^{c}	37.77 ± 0.35^{a}	47.64 ± 3.01^{bc}	48.22 ± 0.02^{bc}	2,5,6,8
2-Methylbutanoic acid	sweaty	1053	0.60 ± 0.69^{a}	0.76 ± 0.01^{a}	0.71 ± 0.05^{a}	2.17 ± 0.05^{b}	0.98 ± 0.03^{a}	1.11 ± 0.18^{ab}	8
2-Methylpropanoic acid	sweaty	1194	1.02 ± 0.24^{a}	1.45 ± 0.03^{ab}	1.71 ± 0.16^{ab}	1.69 ± 0.04^{ab}	2.09 ± 1.68^{c}	2.24 ± 0.03^{c}	8
Acetic acid	acidic	1457	7.97 ± 0.02^{a}	8.76 ± 7.37^{a}	11.85 ± 1.71^{b}	6.41 ± 0.31^{a}	8.58 ± 3.30^{a}	8.39 ± 3.17^{a}	2,5
Isobutyric acid	waxy	1572	14.18 ± 1.67^{a}	13.76 ± 6.58^{a}	8.38 ± 2.89^{a}	29.79 ± 2.11^{b}	5.96 ± 1.34^{a}	8.73 ± 0.45^{a}	2,5
Butanoic acid	sweaty	1632	2.53 ± 0.15^{a}	2.99 ± 0.24^{a}	3.30 ± 0.31^{a}	3.02 ± 0.47^{a}	3.05 ± 0.19^{a}	2.55 ± 0.07^{a}	2,5
3-Methylbutanoic acid	sweaty	1674	26.01 ± 0.57^{b}	18.75 ± 2.50^{a}	16.76 ± 1.99^{a}	32.87 ± 0.93^{d}	30.75 ± 0.48^{c}	30.95 ± 0.63^{c}	2,5,6,8
Hexanoic acid	sweaty	1847	0.16 ± 0.07^{a}	0.02 ± 0.001^{a}	0.016 ± 0.01^{a}	64.74 ± 1.4^{b}	131.43 ± 60.42^{c}	135.97 ± 2.14^{c}	2,5,8,
2-Ethyl-hexanoic acid	cheese	1950	5.68 ± 0.98^{a}	10.00 ± 0.34^{b}	9.98 ± 1.07^{b}	3.31 ± 0.12^{a}	4.98 ± 0.28^{a}	6.33 ± 1.08^{a}	2,5,6,8
Heptanoic acid	–	1954	6.09 ± 0.25^{a}	7.00 ± 0.36^{a}	7.04 ± 1.54^{a}	6.01 ± 0.28^{a}	9.58 ± 2.95^{a}	10.29 ± 1.44^{a}	6,8
Octanoic acid	cheesy	2011	632.55 ± 9.77^{bc}	701.28 ± 18.51^{c}	689.99 ± 54.42^{c}	353.03 ± 20.79^{a}	533.02 ± 21.34^{b}	599.43 ± 23.87^{bc}	2,5,8
Nonanoic acid	cheesy	2166	660.63 ± 44.47^{a}	911.99 ± 12.98^{b}	1317.77 ± 7.05^{c}	701.91 ± 55.88^{a}	700.59 ± 10.24^{a}	707.72 ± 104.88^{ab}	2,5,8
Decanoic acid	waxy	2272	137.45 ± 1.02^{a}	209.95 ± 20.25^{b}	278.19 ± 29.96^{c}	74.09 ± 11.33^{a}	110.02 ± 5.80^{a}	115.79 ± 9.60^{a}	2,5,8
Dec-9-enoic acid	fatty	2332	10.67 ± 2.14^{a}	10.99 ± 7.71^{a}	10.93 ± 5.47^{a}	34.64 ± 0.1652^{b}	34.88 ± 0.30^{b}	32.96 ± 2.31^{b}	2,5,8
Dodecanoic acid	waxy	2484	26.67 ± 7.70^{a}	63.83 ± 18.84^{a}	67.34 ± 30.51^{a}	25.73 ± 6.27^{a}	17.63 ± 4.84^{a}	13.86 ± 1.59^{a}	5,8
Tetradecanoic acid	fatty	2695	27.19 ± 8.80^{c}	30.93 ± 27.62^{c}	32.74 ± 0.90^{c}	10.60 ± 5.33^{a}	12.38 ± 3.26^{b}	12.10 ± 0.21^{b}	2,5,8
Pentadecanoic acid	waxy	2787	23.46 ± 12.95^{c}	8.24 ± 6.91^{b}	5.54 ± 0.38^{ab}	7.87 ± 0.16^{b}	3.86 ± 1.83^{a}	4.70 ± 0.09^{a}	2,5,8
Total			***1628.40 ± 17.07^{b}***	***2051.88 ± 40.73^{c}***	***2515.45 ± 134.78^{d}***	***1395.72 ± 83.63^{a}***	***1658.51 ± 70.61^{b}***	***1741.43 ± 45.40^{b}***	
Alcohols									
2-Methyl-1-propanol	Ethereal	1096	1129.23 ± 10.45^{b}	951.40 ± 21.51^{a}	973.23 ± 17.81^{a}	2768.81 ± 11.60^{c}	1206.30 ± 12.12^{b}	1163.19 ± 11.63^{b}	1,3,6,8
1-Octen-3-ol	Balsamic	1449	63.95 ± 3.66^{bcd}	92.73 ± 5.01^{d}	91.99 ± 16.97 ^{cd}	22.99 ± 4.37^{a}	54.11 ± 2.53^{ab}	58.75 ± 8.64^{bc}	2,8
1-Heptanol	Earthy	1454	75.87 ± 3.82^{b}	81.93 ± 3.95^{b}	86.07 ± 7.12^{b}	65.14 ± 19.82^{a}	257.05 ± 20.60^{c}	598.31 ± 27.36^{d}	3,4
2-Nonanol	Green	1518	67.51 ± 8.05^{bc}	48.59 ± 1.39^{ab}	44.57 ± 6.49^{a}	48.91 ± 2.24^{ab}	84.37 ± 7.17^{c}	77.48 ± 2.41^{c}	3,4,8
1-Octanol	Waxy	1557	343.44 ± 10.21^{b}	221.46 ± 2.61^{ab}	221.87 ± 37.60^{ab}	161.64 ± 4.61^{a}	253.15 ± 54.52^{ab}	294.56 ± 59.36^{ab}	5,8
2-Decanol	waxy	1618	59.30 ± 0.89^{b}	28.54 ± 1.22^{a}	28.97 ± 2.66^{a}	64.69 ± 2.25^{bc}	62.49 ± 5.97^{bc}	68.12 ± 2.65^{c}	1,8
1-Decanol	–	1761	209.20 ± 29.49^{c}	173.63 ± 14.26^{b}	169.26 ± 10.60^{a}	157.95 ± 10.79^{a}	163.83 ± 3.25^{a}	177.39 ± 3.03^{b}	8
Phenethyl alcohol	Floral, honey	1914	7.32 ± 0.26^{ab}	7.42 ± 0.01^{b}	7.43 ± 0.26^{b}	6.72 ± 0.06^{a}	6.97 ± 0.03^{ab}	7.37 ± 0.11^{b}	1,3,4,8
Dodecanol	honey	1966	1.39 ± 0.01^{a}	1.37 ± 0.14^{a}	1.36 ± 0.09^{a}	61.09 ± 6.45^{b}	69.93 ± 10.60^{c}	76.26 ± 14.75^{c}	3,4,8
Total			***1957.23 ± 59.22^{b}***	***1607.11 ± 29.09^{a}***	***1624.78 ± 99.44^{a}***	***3357.97 ± 26.77^{d}***	***2158.25 ± 110.30^{c}***	***2521.47 ± 112.69^{c}***	
Aldehydes									
Octanal	Aldehydic	1294	3.80 ± 0.61a	7.29 ± 2.39^{a}	7.86 ± 0.71^{a}	6.89 ± 2.35^{a}	6.60 ± 2.39^{a}	27.64 ± 2.47^{b}	3,4,8
Nonanal	Aldehydic	1398	69.86 ± 44.99^{a}	68.10 ± 0.53^{a}	239.04 ± 21.87^{b}	53.96 ± 22.33^{a}	71.14 ± 36.71^{a}	184.90 ± 18.23^{b}	1,8
Decanal	Aldehydic	1503	60.21 ± 9.87^{a}	102.05 ± 61.29^{b}	122.66 ± 0.09^{c}	99.15 ± 3.64^{b}	123.81 ± 0.08^{c}	218.83 ± 19.11^{d}	1,3,4,8
Total			***133.88 ± 54.24^{a}***	***177.46 ± 63.15^{b}***	***369.57 ± 22.68^{d}***	***160.01 ± 21.04^{b}***	***201.56 ± 39.19^{c}***	***431.37 ± 3.35^{e}***	
Esters									
Ethyl Acetate	ethereal	890	337.32 ± 25.01^{b}	366.94 ± 10.12^{b}	383.46 ± 15.49^{b}	250.97 ± 7.72^{a}	344.23 ± 9.56^{b}	377.35 ± 25.06^{b}	1,6
Ethyl propanoate	fruity	961	38.27 ± 7.14^{a}	54.50 ± 2.96^{b}	55.13 ± 8.73^{b}	45.70 ± 11.66^{a}	48.37 ± 6.22^{a}	47.04 ± 22.34^{a}	1,6,8
Ethyl 2-methylpropanoate	fruity	969	32.15 ± 6.29^{a}	38.07 ± 3.86^{a}	34.84 ± 0.72^{a}	60.15 ± 7.20^{b}	35.38 ± 6.85^{a}	40.09 ± 1.09^{ab}	8
Propyl acetate	fruity	978	26.81 ± 0.36^{b}	28.97 ± 2.45^{b}	25.55 ± 0.36^{b}	17.48 ± 0.03^{a}	25.37 ± 1.27^{b}	20.06 ± 0.12^{b}	8,9

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Table 4 (*continued*)

Compound	Odor	LRI	MM100	MM75	MM50	CWM100	CWM75	CWM50	Reference
2-Methylpropyl acetate	fruity	1015	29.57 ± 2.19^{a}	32.60 ± 0.21^{ab}	36.14 ± 1.99^{ab}	81.93 ± 1.44^{c}	37.36 ± 4.70^{ab}	40.81 ± 2.42^{b}	1,6,8
Ethyl isovalerate	fruity	1067	5.57 ± 0.66^{b}	1.58 ± 0.14^{a}	1.56 ± 0.31^{a}	5.06 ± 0.01^{b}	1.34 ± 0.03^{a}	3.43 ± 1.36^{ab}	8
Isoamyl acetate	fruity	1124	644.78 ± 25.99^{b}	713.00 ± 18.18^{bc}	759.92 ± 24.31^{c}	522.0 ± 6.31^{a}	697.59 ± 3.48^{bc}	705.90 ± 7.29^{bc}	8
Ethyl pentanoate	fruity	1134	4.82 ± 0.05^{a}	10.54 ± 4.35^{a}	11.44 ± 5.30^{a}	10.51 ± 1.45^{a}	11.73 ± 2.83^{a}	12.09 ± 2.48^{a}	8
Isoamyl butanoate	plum	1197	8.24 ± 1.29^{a}	27.57 ± 0.76^{b}	32.23 ± 1.17^{bc}	6.65 ± 0.13^{a}	42.69 ± 6.23^{cd}	46.65 ± 0.51^{d}	8
Isoamyl formate	sweet	1209	946.70 ± 9.16^{a}	959.7 ± 16.93^{a}	980.10 ± 20.97^{ab}	1194.24 ± 3.94^{d}	1026.50 ± 3.47^{bc}	1076.78 ± 13.48^{c}	2,5,6,8,
Ethyl hexanoate	sweet	1236	601.04 ± 22.69^{a}	775.52 ± 22.00^{b}	797.42 ± 19.86^{b}	624.57 ± 4.52^{a}	809.55 ± 58.47^{b}	753.21 ± 32.70^{b}	8
Hexyl acetate	fruity	1274	16.83 ± 0.13^{a}	33.05 ± 0.97^{b}	33.11 ± 0.36^{b}	20.50 ± 0.25^{a}	33.67 ± 3.61^{b}	37.76 ± 1.27^{b}	6,8
Ethyl 3-pentenoate	fruity	1335	13.50 ± 0.88^{d}	4.46 ± 1.73^{a}	4.33 ± 1.11^{a}	11.46 ± 0.11 ^{cd}	5.92 ± 0.97^{ab}	8.98 ± 0.72^{bc}	4,8
Ethyl heptanoate	fruity	1338	25.59 ± 1.54^{a}	45.16 ± 1.4004^{ab}	49.32 ± 0.70^{ab}	46.1604 ± 0.6853^{ab}	77.8433 ± 5.3007^{bc}	93.952 ± 21.3286^{c}	8
Methyl hexanoate	green	1350	9.70 ± 0.05^{b}	17.85 ± 2.9003^{c}	16.77 ± 0.30^{c}	3.25 ± 0.01^{a}	21.48 ± 2.11^{cd}	26.43 ± 0.72^{d}	8
Hexyl formate	sweet	1375	5.68 ± 0.20^{a}	9.57 ± 0.72^{ab}	8.75 ± 1.29^{ab}	14.45 ± 1.33^{c}	14.15 ± 1.66^{c}	10.41 ± 0.72^{bc}	2,5,6,8
Heptyl acetate	fruity	1444	9.34 ± 0.31^{a}	21.38 ± 4.37^{bc}	25.95 ± 1.32^{bc}	15.53 ± 2.14^{ab}	31.06 ± 3.29^{cd}	39.75 ± 3.72^{d}	8
Ethyl octanoate	coconut	1461	3655.05 ± 118.60^{bc}	3558.08 ± 142.35^{bc}	3877.4551 ± 30.69^{c}	2485.41 ± 75.49^{a}	3370.78 ± 25.48^{b}	3528.21 ± 66.30^{b}	8
Isoamyl hexanoate	fruity	1478	11.51 ± 0.51^{ab}	10.58 ± 1.30^{a}	10.89 ± 3.54^{ab}	12.05 ± 0.33^{ab}	16.00 ± 1.15^{ab}	17.48 ± 0.86^{b}	8
Octyl acetate	coconut	1529	19.86 ± 1.12^{bc}	14.71 ± 0.45^{abc}	15.15 ± 4.71^{abc}	18.09 ± 0.56^{bc}	9.45 ± 0.74^{a}	11.36 ± 0.2^{ab}	8
Ethyl nonanoate	fruity	1521	83.91 ± 7.29^{b}	202.67 ± 7.29^{c}	181.27 ± 29.16^{c}	1.41 ± 0.01^{a}	1.58 ± 0.02^{a}	0.82 ± 0.03^{a}	8
Propyl octanoate	fruity	1553	5.93 ± 0.75^{a}	6.55 ± 1.18^{a}	7.08 ± 0.48^{a}	5.22 ± 0.66^{a}	5.94 ± 0.73^{a}	7.07 ± 0.84^{a}	2,5,8
Isobutyl octanoate	fruity	1659	17.00 ± 2.75^{a}	10.54 ± 0.14^{a}	12.45 ± 0.25^{a}	35.10 ± 4.88^{b}	17.54 ± 0.61^{a}	19.66 ± 2.27^{a}	8
Isoamyl octanoate	fruity	1694	49.14 ± 9.20^{a}	49.64 ± 3.39^{a}	56.27 ± 4.26^{a}	55.29 ± 9.73^{a}	63.81 ± 6.86^{a}	68.79 ± 8.61^{a}	1,6,8
Ethyl 9-decenoate	fruity	1697	235.44 ± 34.81^{a}	249.10 ± 70.42^{a}	269.57 ± 8.04^{a}	898.96 ± 79.71^{d}	604.15 ± 75.39^{b}	720.67 ± 84.67^{c}	8
trans-Geranic acid methyl ester	green, fruity	1688	43.90 ± 4.14^{b}	45.36 ± 1.72^{b}	51.08 ± 2.88^{7b}	14.99 ± 0.67^{a}	66.15 ± 1.65^{c}	82.07 ± 2.27^{d}	8
Ethyl *trans*-4-decenoate	soapy	1741	2.86 ± 0.07^{a}	2.78 ± 0.0401^{a}	4.59 ± 0.36^{a}	12.50 ± 0.42^{b}	49.39 ± 3.26^{c}	50.26 ± 2.40^{c}	8
Ethyl-undecanoate	floral	1818	12.16 ± 1.09^{8a}	7.57 ± 1.44^{a}	6.79 ± 0.89^{a}	17.20 ± 7.60^{a}	9.15 ± 0.36^{a}	7.21 ± 0.32^{a}	8
2-Phenethyl acetate	honey	1846	391.03 ± 17.7^{93b}	565.22 ± 18.07^{c}	549.78 ± 20.12^{c}	277.05 ± 11.59^{a}	379.20 ± 8.96^{b}	414.76 ± 5.23^{b}	8
Ethyl-Dodecanoate	fruity	1861	211.61 ± 41.91^{b}	853.99 ± 2.99^{c}	971.74 ± 26.41^{d}	706.84 ± 163.01^{c}	227.26 ± 4.33^{b}	180.03 ± 41.17^{a}	8
Isoamyl decanoate	coconut	2029	4.79 ± 0.95^{a}	11.03 ± 0.58^{bc}	12.87 ± 1.36^{c}	14.81 ± 1.45^{c}	7.38 ± 0.94^{ab}	6.05 ± 0.33^{a}	2,5,6,8
Ethyl-tetradecanoate	sweet	2051	23.94 ± 4.98^{b}	31.92 ± 4.93^{c}	35.83 ± 3.48^{c}	38.01 ± 8.06^{d}	19.64 ± 0.34^{ab}	16.79 ± 0.08^{a}	8
Ethyl-hexadecanoate	waxy	2253	14.58 ± 0.42^{c}	16.60 ± 8.32^{d}	19.06 ± 1.20^{e}	14.65 ± 0.86^{c}	9.9164 ± 2.36^{b}	3.11 ± 1.09^{a}	8
Propyl acetate	fruity	1089	26.81 ± 0.36^{c}	28.97 ± 2.4535^{c}	25.55 ± 0.36^{c}	17.48 ± 0.03^{a}	20.37 ± 1.27^{b}	20.06 ± 0.12^{b}	8
Isobutyl isobutyrate	sweet	982	26.28 ± 1.44^{e}	22.64 ± 3.59^{d}	11.46 ± 0.44^{c}	1.79 ± 0.22^{a}	4.73 ± 2.43^{b}	8.62 ± 1.00^{c}	8
Total			***7591.91*** ± ***164.73^{a}***	***8828.61*** ± ***270.45^{b}***	***9375.07*** ± ***85.94^{c}***	***7557.62*** ± ***333.094^{a}***	***8131.84*** ± ***0.074^{b}***	***8503.88*** ± ***116.32^{b}***	
Ketones									
2-Pentanone	herbal	1010	18.28 ± 0.03^{a}	18.65 ± 0.03^{a}	18.11 ± 1.18^{a}	18.52 ± 0.13^{a}	21.56 ± 0.01^{b}	21.49 ± 0.14^{b}	8
4-Methyl-2-pentanone	cheesy	1185	9.22 ± 0.32^{c}	1.08 ± 0.03^{a}	1.05 ± 0.03^{a}	3.63 ± 0.04^{b}	1.02 ± 0.03^{a}	1.54 ± 0.03^{a}	2,5,8
2-Heptanone	citrus	1340	1.71 ± 0.10^{b}	1.34 ± 0.0729^{ab}	1.03 ± 0.0729^{a}	1.92 ± 0.22^{b}	1.59 ± 0.23^{ab}	1.37 ± 0.17^{ab}	2,5,8
6-methyl-5-Hepten-2-one	citrus	1810	8.52 ± 0.01^{c}	1.74 ± 0.07^{a}	1.5883 ± 0.07^{a}	10.15 ± 0.76^{d}	4.48 ± 0.40^{b}	3.23 ± 0.07^{b}	2,5,8
β-Damascenone	honey	1821	0.0028 ± 0.0004^{b}	0.0021 ± 0.0001^{ab}	0.002 ± 0.0002^{a}	0.0018 ± 0.0001^{a}	0.0013 ± 0.0000^{a}	0.0015 ± 0.0001^{a}	7,8
Total			***37.75*** ± ***0.17^{e}***	***22.82*** ± ***0.06^{b}***	***21.79*** ± ***1.36^{a}***	***34.23*** ± ***0.71^{d}***	***28.66*** ± ***0.59^{c}***	***27.64*** ± ***0.13^{c}***	
Terpenes									
trans-Farnesol	spicy	2352	0.0124 ± 0.0036^{a}	0.0123 ± 0.0022^{a}	0.0149 ± 0.0053^{a}	0.0117 ± 0.0016^{a}	0.0061 ± 0.0011^{a}	0.0036 ± 0.0003^{a}	8
Linalool	floral	1546	0.1226 ± 0.0131^{b}	0.1687 ± 0.0008^{c}	0.1728 ± 0.0156^{c}	0.0505 ± 0.0081^{a}	0.2028 ± 0.0138^{cd}	0.2264 ± 0.0062^{d}	8
Citronellol	floral	1760	0.0058 ± 0.0011^{a}	0.0837 ± 0.0049^{cd}	0.075 ± 0.0089^{c}	0.0365 ± 0.0000^{b}	0.0982 ± 0.0011^{de}	0.1185 ± 0.0092^{e}	7,8
Geranyl acetate	floral	1757	0.0029 ± 0.0002^{a}	0.0046 ± 0.0003^{b}	0.0050 ± 0.0001^{b}	0.0052 ± 0.0003^{b}	0.0046 ± 0.0004^{b}	0.0051 ± 0.0001^{b}	8

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Table 4 (*continued*)

Compound	Odor	LRI	MM100	MM75	MM50	CWM100	CWM75	CWM50	Reference
Geraniol	floral	1799	0.0206 ± 0.0018^{b}	0.0229 ± 0.0004^{b}	0.0216 ± 0.0035^{b}	0.012 ± 0.0013^{a}	0.0202 ± 0.0003^{b}	0.0258 ± 0.0023^{b}	8
Humulene epoxide II	floral	2037	0.2025 ± 0.0266^{c}	0.0066 ± 0.0003^{a}	0.0076 ± 0.0001^{a}	0.1206 ± 0.0057^{b}	0.1092 ± 0.0006^{b}	0.0076 ± 0.0021^{a}	8
Citronellyl acetate	floral	1662	0.0178 ± 0.0017^{a}	0.0198 ± 0.0001^{ab}	0.0243 ± 0.0001^{c}	0.0226 ± 0.0017^{bc}	0.0227 ± 0.0007^{bc}	0.0182 ± 0.0006^{a}	8
Total			***0.3847 ± 0.0374^{bc}***	***0.3185 ± 0.003^{ab}***	***0.3213 ± 0.0332^{ab}***	***0.2591 ± 0.0121^{a}***	***0.4639 ± 0.0133^{c}***	***0.4053 ± 0.02^{bc}***	
Aromatic compounds									
4-Viny guaiacol	spicy	2196	12.42 ± 0.06^{a}	12.58 ± 0.56^{a}	12.25 ± 3.61^{a}	29.28 ± 1.48^{b}	7.02 ± 1.06^{a}	7.12 ± 0.06^{a}	8
Styrene	balsamic	1262	0.82 ± 0.04^{b}	0.56 ± 0.01^{a}	0.56 ± 0.01^{a}	1.21 ± 0.03^{c}	0.66 ± 0.07^{a}	0.53 ± 0.02^{a}	3,4
Total			***13.24 ± 0.10^{a}***	***13.14 ± 0.57^{a}***	***12.81 ± 3.62^{a}***	***30.49 ± 1.51^{c}***	***7.68 ± 1.12^{b}***	***7.65 ± 0.08^{b}***	

Beers and worts produced by micro-brewing trials with different content (50, 75, 100 %%) of Maiorca malt (MM) and Commercial wheat malt (CWM). LRI = Linear retention index; na = not aviable. Values in the same line followed by different letter are statistically different ($p < 0.05$).
Reference: (1) De Flaviis et al., 2022b; (2) Medina et al., 2023*; (3)* Langos et al.2013*; (4)* Mascia et al., 2014*; (5)* Li et al., 2012*; (6)* De Flaviis et al., 2021*; (8)* Zunkel et al., 2011*; (9)* De Schutter et al., 2008a*, (10)* De Schutter et al., 2008b*; (11)* Alves et al.,2020.

et al.,2014). Significant statistical variations were observed for 2-methyl-1-propanol, with content ranging from 951 to 2768 mg/L. In general, this compound was more prevalent in commercial wheat malted beers especially in CWM100. 2-Methyl-1-propanol and phenethyl alcohol were among the two main contributor of floral notes as they exceeded their OTV.

Styrene is an aromatic compound, and several studies in the literature indicate that it is a typical compound found in wheat beers. It is formed through the decarboxylation of cimmanic acid during boiling or through enzymatic process through the time of fermentation. Styrene can be found in small amounts in grains, as well as in coffee or dried fruits, according to several conducted studies. The toxicity of styrene is primarily associated with its potential carcinogenic effects on humans, although research findings on this matter have sometimes been contradictory (Roe, 1994, Schwarz et al., 2012). On the organoleptic level, styrene brings balsamic and almond flavours that enhance the phenolic flavour of wheat beers. In the analyzed wheat beer samples, the styrene content exceeded its OTV from 28 to 61 times ranging from 0.53 to 1.21 mg/L and the highest value was found in CWM100 and the lowest in MM50. It is worth noting that there is a positive correlation between the styrene content and the percentage of wheat used in the beer recipe. In the case of Maiorca beers, the styrene content was consistently lower compared to beers made with commercial wheat malt, except in MM100 (0.82 mg/L), where the styrene content was higher but still lower than in CWM100.

Among the main identified compounds in the experimental beers, aldehydes such as nonanal and decanal prevailed. These compounds are commonly found in wheat beers and are known to contribute to citrus and fruity aromas. In beer samples, nonanal showed statistically significant differences, being more abundant in MM50 and CWM50 (239 and 184 mg/L, respectively).

The 4-vinyl guaiacol (4-VG) is an aromatic compound that contributes to their spicy and phenolic odor often desired in specific beer styles, as top-fermented wheat beer (Xu et al., 2020). It is formed during fermentation through the decarboxylation of ferulic acid, which is produced during the mashing process (McMurrough et al., 1996). In beer samples, the 4VG content showed statistically significant differences, ranging from 7.02 to 1.21 mg/L. The lowest values were observed in CWM75 and CWM50, while the highest were found in CWM100, whereas no variations were observed in MM samples. Similar to styrene, the 4VG content also displayed a positive correlation with the percentage of wheat malt used in the brewing process.

Ketones are another important class of carbonyl compounds found in beer. The overall content of compounds belonging to this class ranged from 21 to 37 mg/L, with the lowest value observed in MM50 and CWM50 and the highest in MM100 and CWM100. Among the identified ketones, β-damascenone and 2-pentanone were the most abundant. β-damascenone exceeded its OTV and it is responsible of honey notes in beer.

3.8. Sensory evaluation of experimental beer

The results of the sensory analysis for the experimental beers were presented in radar charts (Fig. 1a, 1b, 1c) and organized by visual, odor and taste attributes. Each chart also included an overall sample acceptance. Regarding visual attributes, the beers displayed statistically significant differences ($p < 0.01$) in terms of color and foam persistence. MM100 and CWM50 beers exhibited a more intense yellow color compared to the other beer samples. Foam persistence was positively influenced by the percentage of wheat malt used in the recipe, with the highest values observed in MM100, CWM100, and MM75. Considering the odor attributes, the beer samples showed significant differences ($p < 0.001$) in terms of the honey and phenolic attributes. MM100 and MM75 beers displayed the highest values for the honey attribute probably due to the highest content in phenyl ethyl alcohol and phenyl ethyl acetate and β-Damascenone in these samples, while CWM75 and CWM100 had the highest for the phenolic attribute as also found in earlier work by Mascia, I. et al., 2014 on the preliminary characterization of an Italian durum wheat craft beer. This could be associated to the higher content in aromatic compounds such as 4VG and styrene in those samples. The beer samples showed variations in odor intensity, estery ($p < 0.05$) and yeasty ($p < 0.01$). MM100 exhibited the most intense values for estery and fruity odor intensity followed by MM75 and CWM75. CWM75 beer scored the highest value of yeasty attribute. The evaluated experimental beers demonstrated differences in terms of taste features, particularly in relation to the fruity and bitter characteristics ($p < 0.001$), as well as taste intensity ($p < 0.01$). MM100 beer had the highest taste intensity, while CWM100 had the lowest. Additionally, MM100 beer showed higher values for bitter and fruity compared to the other samples.

Based on the overall acceptance assessment, it appears that consumers generally preferred Maiorca malt beers.

3.9. Effect of different content of wheat malt on wort quality parameter

To assess the impact of varying wheat malt content on selected quality parameters a clustergram was illustrated in Fig. 2a. Samples grouped in the same cluster demonstrate similar characteristics for the analyzed variables. The clustergram analysis allows us to observe how different malt content and types influence the studied technological parameters in the congress worts. It is noteworthy that the use of Maiorca malt had a positive effect on several technical-quality parameters of the wort.

The analysis of aromatic compounds in worts revealed that worts produced with Maiorca malt (MM) were more abundant in furans,

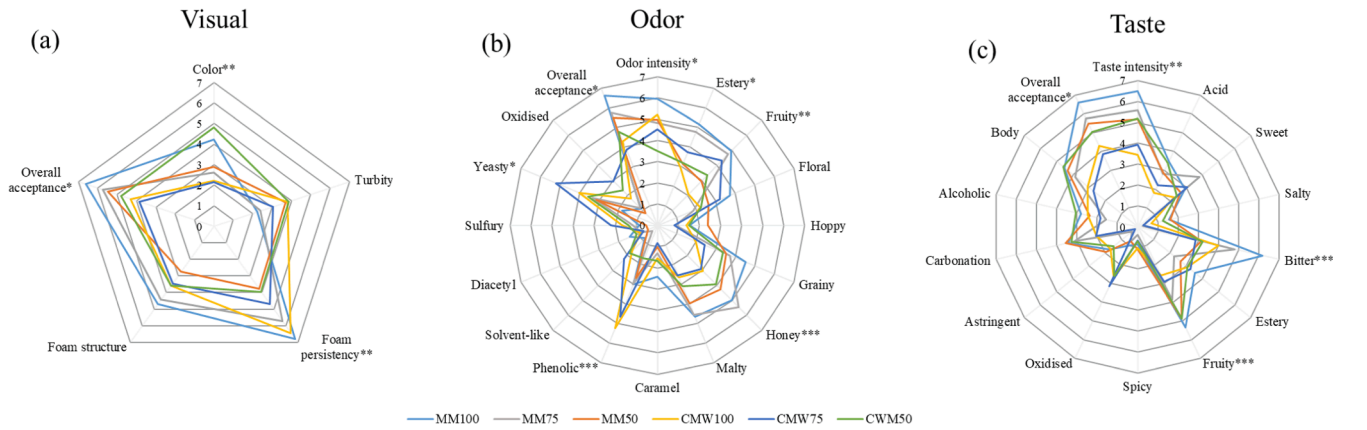

Fig. 1. Sensory analysis performed on visual (a), odor (b) and taste (c) of beers: spider plot of average scores for aroma determined by judges during tasting sessions. Beers produced by micro-brewing trials with different content (50, 75, 100 %) of Maiorca malt (MM) and Commercial wheat malt (CWM). Symbols: ***, $P < 0.001$; **, $P < 0.01$; *, $P < 0.05$.

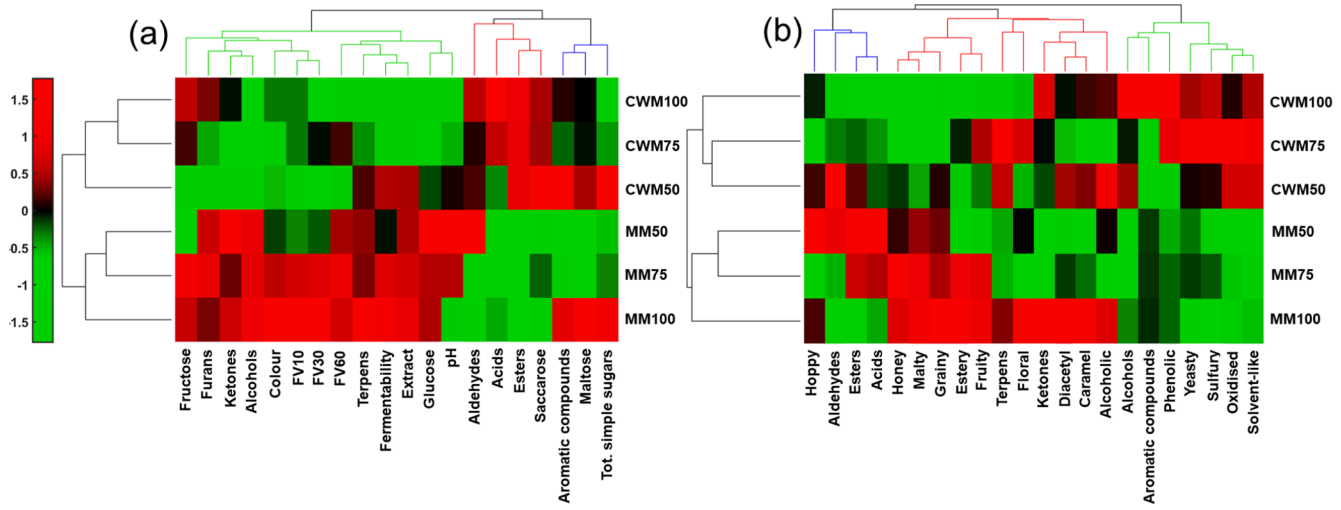

Fig. 2. Clustergram of worts (a) and beers (b) quality parameters. FV = filtration value at 10, 30 and 60 min; (a) Data source: quality parameters of congress worts and GC–MS data of wort samples as average total amount of each class of substances; (b) Data source: GC–MS data as average total amount of each class of substances and sensory analysis data of experimental beers.

ketones, and alcohols compared to worts made with commercial wheat malt (CWM). Worts from CWM displayed elevated levels of aldehydes, acids, esters, and sucrose. The clustergram also indicates that CWM50 and MM100 share similarities in terms of aromatic compounds, maltose, and total simple sugars, both of which were found to be higher compared to the other samples.

3.10. *Effect of different content of wheat malt on beer sensory parameter*

To evaluate effect of wheat malt on beer sensory parameter the olfactory attributes obtained from sensory evaluation were analyze together with the results of VOCs analysis in the form of a heat map (Fig. 2b). The heat maps provide insights into how varying percentages of wheat malt had a significant impact on the odor component of the studied beers.

Attributes such as honey, malty, grainy, estery, fruity, floral, caramel odor, as well as terpenes and ketones content tend to increase with the higher inclusion of Maiorca malt in the recipe. Conversely, in beers brewed with commercial wheat malt (CWM), attributes such as aromatic compounds, phenolic, yeasty, sulfury, oxidized, and solvent-like odors are more prominent in comparison to MM beers. The clustergram also illustrates that MM50 and CWM50 stand out from the other samples due to their elevated levels of aldehydes, esters, acids, and hoppy notes. The increase in estery and fruity odors can be attributed to the higher glucose content detected in the wort of Maiorca malt beers. Previous studies have shown that, under the same fermentation conditions, yeast produces more esters in wort that is richer in glucose (Lei et al., 2016; Gresser, 2010).

The clustergram reveals that both sample MM100 and CWM50 exhibited high values for the diacetyl attribute. However, the presence of vicinal diketones was not detected in our samples. Diacetyl aroma was also assessed through sensory analysis. As depicted in Fig. 1, the assessors assigned low scores to the diacetyl aroma, and no significant differences emerged between the samples for the diacetyl parameter.

4. Conclusion

The findings of this study indicate that MM possesses outstanding qualities concerning the technological parameters required for brewing cereals. Analyses conducted on the malts and congress wort revealed that MM showed not significance difference on Kolbach Index values compared to CWM and CBM, a higher FAN content when compared to CWM, the highest β-amylase content and α-amylase content that closely resembled the values recorded for CBM. Based on the analyses

conducted on diastatic power, both wheat malts demonstrated slightly higher values compared to barley malt. This can be attributed the β-amylase content, which is positively correlated with this parameter. The superior enzymatic activity of MM compared to commercial wheat malt is evident from the higher extract and fermentability values observed in the respective musts obtained with varying percentages of MM. The analysis of the sugar profile revealed no significant differences among the various samples except for the glucose content, which was higher in MM50, MM75 and MM100, respectively. The analysis of volatile compounds in the worts revealed a predominant presence of aldehydes, which were higher in CWM samples and MM50. Most of the compounds identified in the experimental beers were characteristic of wheat beers, as supported by various studies in the literature. The studied beers showed distinct aroma profiles, with esters being the most abundant class of compounds of which most exceed their odor threshold contributing to the fruity aroma of beer. When comparing the results of the volatile analysis with the descriptive sensory analysis of the experimental beers, it was observed that beers brewed with MM displayed different attributes compared to those brewed with CWM. The features of estery, fruity, grainy, malty, and honey tend to increase with a higher proportion of MM in the recipe, while attributes as aromatic compounds, phenolic, yeasty, sulfury, oxidized, and solvent-like odors were notably lower in comparison to CWM beers. The increase in estery and fruity odors could be due to the higher glucose content detected in the wort of MM beers. In terms of overall acceptance, MM beers, in particular MM100, were favored by the panelists. The results indicated that MM is a suitable malt for brewing 100 % malted wheat beer. Technologically, there are no apparent limitations in using MM for this purpose, as certain characteristics such as extract, filterability, enzymatic power are improved compared to CWM and very similar to CBM.

CRediT authorship contribution statement

Ignazio Maria Gugino: Conceptualization, Data curation, Investigation, Methodology, Writing – original draft, Writing – review & editing. **Vincenzo Alfeo:** Data curation, Investigation, Methodology, Software. **Mansour Rabie Ashkezary:** Data curation, Investigation, Writing – review & editing. **Ombretta Marconi:** Supervision, Validation. **Antonino Pirrone:** Methodology, Formal analysis. **Nicola Francesca:** Validation, Supervision. **Fabrizio Cincotta:** Data curation, Formal analysis, Investigation, Methodology, Writing – review & editing. **Antonella Verzera:** Supervision, Validation. **Aldo Todaro:** Conceptualization, Investigation, Methodology, Supervision, Resources, Validation.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

Appendix A. Supplementary material

Supplementary data to this article can be found online at https://doi.org/10.1016/j.foodchem.2023.137517.

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