



Effects of brewing procedures and oenological yeasts on chemical composition, antioxidant activity, and sensory properties of emmer-based craft beers

Maria Tufariello^{a,1}, Francesco Grieco^{a,1}, Anna Fiore^b, Carmela Gerardi^a, Vittorio Capozzi^c, Antonietta Baiano^{b,*}

^a Institute of Sciences of Food Production, National Research Council, Via Prov.le, Lecce-Monteroni, 73100, Lecce, Italy

^b Dipartimento di Scienze Agrarie, Alimenti, Risorse Naturali e Ingegneria (DAFNE), University of Foggia, Via Napoli, 25, 71122, Foggia, Italy

^c Institute of Sciences of Food Production, National Research Council (CNR), C/o CS-DAT, Via Michele Protano, 71122, Foggia, Italy

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ABSTRACT

Emmer is among the most ancient domesticated grains. In craft brewing, emmer is used in an adjunct, due to its tannic astringency and typical nutty aroma. The aim of this study was to evaluate the effects on the quality of emmer-based craft beers exerted by the employment of two novel brewing procedures (BP1 and BP2) and four oenological *Saccharomyces cerevisiae* starter strains, namely 17,290, 14,061, 9502, and 9518. The two technological approaches differed for water conductivity (570 and 440 $\mu\text{S}/\text{cm}$), protein rest (30 and 10 min), boiling step (90 and 55 min), and Irish moss addition (only in BP1). The highest total phenolic concentrations were detected in the beers fermented by 17,290 and 14,061 strains. The beers fermented by 14,061 showed the highest contents of volatile esters, alcohols, and terpenes (the latter if produced according to BP1). The beers produced according to BP2 had the highest concentrations of volatile acids, norisoprenoids, hydrocarbons, and phenols with significant effects of the utilized starter strain. The highest overall sensory score (~ 4.5) was assigned to BP2-9502 beers and it was positively correlated with color, pH, foam amount and persistence, olfactory finesse, body/fullness, and negatively correlated with CO_2 , titratable and volatile acidity, saltiness, and sourness.

1. Introduction

Beers inspired by the belgian Witbier style can be produced from mixtures of malted barley with various unmalted cereals since they are a cheaper source of compounds that can impart new/better sensory and nutritional characteristics to the product (Cadenas et al., 2021; Yorke et al., 2021). In this regard, new brewing trends include the use of ancient unmalted wheat species (Marconi et al., 2013) capable of enriching these beers with higher amounts of antioxidants than modern cereal species. The reason is that, according to literature (Oliveira de Araújo Melo et al., 2020), the ethanol toxic effects are mediated by several mechanisms of oxidative stress (induction of oxidative damage, lipid peroxidation) and the adjuncts of alternative grains contribute to the beer antioxidant activity counteracting the adverse health effects of ethanol (Yang & Gao, 2021). Emmer (*Triticum turgidum* L. spp. *dicoccum*

Schrunk) is among the most common ancient wheat species, being a domesticated form of the wild emmer wheat (*T. turgidum* spp. *dicoccoides*). Emmer has a protein content of 11–12% and an onset gelatinization temperature of 58.8 °C, which make it suitable for brewing practices also in the unmalted form (Baillière et al., 2022). Moreover, it generally shows higher antioxidant contents than the other wheat species (Zrcková et al., 2019).

Brewing performed with the addition of unmalted cereals can be challenging and the main disadvantage is the low concentration of enzymes usually synthesized during the malting process such as amylase, protease, and cytase, which can have detrimental effects on beer quality parameters. Overcoming these problems requires changing the conditions of the brewing process, namely water quality, mashing times and temperatures, boiling duration, possible addition of adjuncts (Yorke et al., 2021). Moreover, the choice of the yeast starter stain is critical for

* Corresponding author.

E-mail addresses: maria.tufariello@ispa.cnr.it (M. Tufariello), francesco.grieco@ispa.cnr.it (F. Grieco), anna.fiore@unifg.it (A. Fiore), carmela.gerardi@ispa.cnr.it (C. Gerardi), vittorio.capozzi@ispa.cnr.it (V. Capozzi), antonietta.baiano@unifg.it (A. Baiano).

¹ The authors equally contributed to this work.

the beer sensory quality, because of the different ability of the various *Saccharomyces cerevisiae* strains to influence beer chemical composition (Cardoso Viana et al., 2021). In recent years, great efforts have been focused on the use of yeast starter strains of oenological origin as brewing starters (Iorizzo et al., 2021; Siesto et al., 2023; Vrñceanu et al., 2022). Nevertheless, as far as we know, the production of beer by the combination of oenological *S. cerevisiae* and unmalted emmer as adjuncts never been described.

This investigation aimed to test combinations of two novel brewing procedures and four oenological *S. cerevisiae* strains isolated from grape to overcome the critical issues arising from the use of an unmalted cereal, maximize antioxidant content, differentiate volatolomic profile, and improve sensory quality of beers produced with a mixture of malted barley and unmalted emmer.

2. Materials and methods

2.1. Brewing materials

Barley malt cv. Fortuna was supplied by Agroalimentare Sud (Melfi, Potenza, Italy). The unmalted dehulled emmer cv. Padre Pio (*Triticum dicoccum*) came from the experimental fields of CREA-CI Research Centre for Cereal and Industrial Crops (Foggia, Italy). The beers were manufactured using a mixture of 60% malted barley and 40% unmalted dehulled emmer, percentages chosen to emphasize the effects of the unmalted cereals but respecting the upper limit established by the Italian Law 1354 (1962). Birramia (Querceta, Lucca, Italy) supplied the following flavoring agents - dried hop cones of cv. Cascade (6.7% α -acid content), bitter orange peels, and coriander - and the Irish moss (marine algae, used to facilitate coagulation and sedimentation of proteins). The wort fermentation trials were carried out using the following four oenological *S. cerevisiae* strains of the ITEM Agro-Food Microbial Culture Collection (CNR-ISPA, 2023), which were already described by Tristezza et al. (2014) and Tufariello et al. (2014): 17,290 and 14,061, isolated from Negroamaro grape; 9502 and 9518, isolated from Susumaniello grape.

2.2. Formulation of recipe and brewing process

According to the optimized recipe of Baiano et al. (2024), the amounts of ingredients necessary to produce 100 L of finished beer were the following: water, 115 L for mashing, and 20 L for sparging; malted barley, 14.75 kg; unmalted emmer 9.75 kg; hop cones, 100 g; bitter orange peels, 100 g; coriander, 100 g. Before brewing, the malted and unmalted cereals were separately crushed with a 2-roller mill (Albrigi Luigi, Stallavena, Italy) under mill gaps of 0.5 ± 0.1 mm and then mixed together. The brewing trials were performed in a Braumeister system (Speidel Tank-und Behälterbau GmbH, Ofterdingen, Germany).

Two brewing procedures, referred to as BP1 and BP2, were tested. The BP2 brewing procedure - already successfully applied to the production of beers with unmalted durum and common wheat (Baiano et al., 2024) - was used as a control and compared with the B1 brewing procedure, which differed from BP2 for a higher water conductivity, longer protein rest and boiling step, and addition of Irish moss. The choice to vary precisely these parameters lies in the fact that: water conductivity is related to the ion content that in turn affects enzymatic and non-enzymatic reactions; protein rest length affects the hydrolysis rate of protein polymers; boiling affects the beer oxidation-reduction potential and, together with the Irish moss, precipitation of protein-polyphenol complexes.

Brewing Procedure 1 (BP1) – The cereal mixture was added to the mashing water (conductivity 570 ± 10 μ S/cm) previously heated at 47 °C. The mashing steps were the following: protein rest (54 °C; 30 min); β -amylase rest (63 °C; 50 min); α -amylase rest (70 °C; 50 min); mash-off (81 °C; 16 min). Temperature between rests increased at a rate of about 1.5 °C/min. The exhausted solid fraction was separated from

the wort, crossed by the sparge water at 81 °C, and left to drain. The final wort pH was close to 5.4 ± 0.1 . The resultant wort was boiled for 65 min, with the flavoring agents and the Irish moss (20 g/100 L) added 50 and 15 min before the end of boiling, respectively.

Brewing Procedure 2 (BP2) – The cereal mixture was added to the mashing water (conductivity of 440 ± 5 μ S/cm) previously heated at 47 °C. The mashing steps were the same as BP1 except for duration of protein rest (10 min instead of 30 min). The final wort pH was 5.3 ± 0.2 . The resultant wort was boiled for 55 min, with the flavoring agents added 50 min before the end of boiling. A final original gravity of 1.053 ± 0.001 was reached.

The worts resulting from BP1 or BP2 were cooled at room temperature, whirlpooled to remove solid residues and then divided into 4 aliquots, each of them separately inoculated with one of the four oenological yeast strains ($\sim 1 \times 10^7$ cells/mL). Fermentations were carried out at 20 ± 2 °C for 21 ± 1 days, until an original gravity value of 1.018 ± 0.002 was reached. After that, maturation was carried out at 4 ± 1 °C for 4 days. Finally, beers were racked, inoculated with the same yeast strain used for the first fermentation ($\sim 1 \times 10^5$ cells/mL), added with sucrose (6 g/L), and packaged into 750 mL glass brown bottles. The bottled beer was conditioned at 20 ± 1 °C for 1 month, and subsequently stored at 5 ± 1 °C until analyses. Eight types of craft beers were produced combining the two brewing procedures (BP1 or BP2) and the four *S. cerevisiae* strains (17,290, 14,061, 9502, or 9518). For each type of beer, three technological replicates were performed.

2.3. Analyses of starting ingredients and their mixture

Moisture, ash, and protein contents, expressed as %, were determined according to the AACC methods 44–15.02, 08–01.01, and 46–30.01 (Dumas combustion nitrogen method; the nitrogen was converted to protein using a factor of 5.7), respectively (AACC, 2012). The extraction of total phenolics was performed according to the optimized conditions found by Gandolpho et al. (2021) with some modifications. More in depth, 1 g of sample was added to 30 mL of a 58% ethanolic solution and an ultrasound-assisted extraction was applied (30 °C; 30 min, 34 KHz). The mixture was then centrifuged at 2000 rpm for 25 min at 20 °C, and the supernatant was filtered through 0.45 μ m nylon filters. Total phenolic content (TPC, mg of gallic acid equivalents/100 g of dry matter), phenolic profiles (mg/100 g dm), and antioxidant activity (AOA, mmol of Trolox/g dm) of the extracts were analyzed as described in section 2.5.

2.4. Technological and chemical analyses of the beers

The pH values, soluble solids (as Brix), carbon dioxide content (as mg CO₂/L), alcohol content (%), titratable acidity (g lactic acid/L), and volatile acidity (g acetic acid/L) were determined as described in Baiano et al. (2023). Beer color was determined at 430 nm according to the Method 9.6 (European Brewery Convention, 1975) on previously degassed and filtered (0.45 μ m) samples.

Organic acids, maltodextrin, maltotriose, maltose, glucose, fructose, and glycerol concentrations (mg/mL) were simultaneously determined according to Coelho et al. (2018) onto an Agilent Hi-Plex H (300 \times 7.7 mm) with internal particles of 8.0 μ m (Agilent Technologies, Santa Clara, CA, USA). Organic acids were detected through a Diode Array Detector at 210 nm, while the sugar detection was carried out through a Refractive Index Detector. Quantification of individual organic acids and sugars was directly performed through the ChemStation software (Agilent) using five-point regression curves ($r^2 \geq 0.99$) of the authentic standards.

2.5. Total phenolic content, phenolic profile, and antioxidant activity of the beers

The total phenolic content (TPC, mg gallic acid equivalents/L) was

determined through the Folin–Ciocalteu method (Singleton & Rossi, 1965) with some modifications. A mixture of 125 μL of the opportunely diluted sample, 0.5 mL of deionized water, and 125 μL of the Folin–Ciocalteu reagent was kept to react for 6 min. Successively, 1.25 mL of a 7% aqueous solution of Na_2CO_3 was added. And the final volume was adjusted to 3 mL with water. After 90 min, the absorption was read at 760 nm against water as a blank. TPC was quantified through a calibration curve of gallic acid (20–1000 mg/L range).

The phenolic profiles (mg/L) of the extracts were analyzed by a 1100 HPLC-DAD system (Agilent, Santa Clara, CA, USA) equipped with a 100 mm \times 4.6 mm \times 3 μm RP-C18 Gemini column (Phenomenex, Aschaffenburg, Germany) as described by Aliakbarian et al. (2011). The following conditions were applied: Solvent A (water solution of acetic acid, 1.0% v/v); Solvent B (50% methanol, 50% acetonitrile, v/v); injection volume 100 μL ; temperature 30 $^\circ\text{C}$; flow rate 1 mL/min; wavelengths 280 and 320 nm. The following linear gradient of Solvent B was applied: from 5 to 25% in min; from 25 to 30% in 5 min; from 30 to 40% in 10 min; from 40 to 48% in 5 min; from 48 to 60% B in 10 min; return to the initial conditions in 5 min and equilibration of column for 5 min. The identification of phenolic compounds was performed comparing their retention times and spectra with those of 18 pure standards while quantification was obtained on the basis of calibration lines built by injection of known amounts of pure standards.

The antioxidant activity (AOA) was determined by 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical-scavenging activity (Brand-Williams et al., 1995). More specifically, 0.1 mL of sample was added to 3.9 mL of a methanolic DPPH solution (40 mg/L) and kept in the dark. A blank was prepared by adding 0.1 mL of distilled water to 3.9 mL of the DPPH solution. The absorbance values of both sample and blank were measured at 515 nm after 90 min. AOA was quantified as mmol of Trolox per L using a calibration lines prepared with known amounts of 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (Trolox). AOA was expressed as mmol of Trolox/L.

2.6. Volatolomic analysis of the beers

A head-solid phase micro-extraction combined with gas chromatography-mass spectrometry (HS-SPME-GC/MS) was applied according to Palombi et al. (2023). Briefly, 100 μL of the internal standard solution (IS, 4-methylpentan-2-ol, 200 mg/L) was added to a 5 mL of each beer in a 20 mL headspace vial (Alltech Corp., Deerfield, IL, USA). After equilibration of the sample for 20 min at 40 $^\circ\text{C}$, a 50/30 DVB-CAR-PDMS solid phase fibre (Supelco, Bellefonte, PA) was inserted into the vial and let to adsorb volatiles for 30 min at 40 $^\circ\text{C}$. After that, the fiber was inserted into the injector port (250 $^\circ\text{C}$) in <2 min (splitless mode) of a GC 6890 (Agilent Technologies, Palo Alto, CA) equipped with a HP-INNOWAX capillary column (60 m \times 0.25 mm, 0.25 μm , J&W Scientific Inc., Folsom, CA, USA) and coupled to an Agilent MSD 5973 Network detector (Tufariello et al., 2019). The MS analysis employed electron ionization (EI) mode at 70 eV over a scan mass range of 35–350 amu. The ion source temperature was 250 $^\circ\text{C}$ and MS source temperature at 280 $^\circ\text{C}$. Mass spectra of the volatile compounds were compared with: those of the data system library (NIST 98, $p > 90\%$); the retention data of commercially available standards MS data reported in the literature. Concentration of each volatile compound was assessed by the internal standard method.

2.7. Odor activity value and aromatic series

Odor activity value (OAV) was calculated by dividing the concentration of a specific aroma compound in a sample, by its odor threshold. The odor threshold was the minimum concentration at which a compound can be perceived by the human nose. If the OAV was greater than 1, it suggested that the aroma compound was present in a concentration higher than its odor threshold and was likely to contribute significantly to the overall aroma of the sample. By following this approach, it was

possible to quantitatively link the volatile composition of the beers to their aroma descriptors. The aromatic series values reflected the combined contribution of specific groups of compounds to the overall aroma profile.

2.8. Sensory descriptive analysis

The Quantitative Descriptive Analysis (QDA) of beers was performed by a panel of six trained judges between 25 and 65 years of age, experienced in alcoholic beverage sensory evaluation and in possession of a sommelier or technical wine taster certificate. Panelists performed a Quantitative Descriptive Analysis (QDA) as described by Baiano et al. (2023). They were asked to evaluate 5 visual (for foam: color, amount, and persistence; for liquid portion: color, and turbidity), 9 olfactory (overall flavor intensity, olfactory finesse, malty, hoppy, floral, fruity, spicy, yeast, aromatic herbs), 4 gustatory (sweetness, bitterness, saltiness, sourness), and 3 tactile (alcoholic, effervescence, and body/fullness) parameters, which had been previously selected among those found in the literature and those generated by the same panel. The panelists also gave a comprehensive and objective score of the sensory quality of each sample evaluated after its swallowing (overall quality). All descriptors and the overall quality were evaluated on a 5-point scale except for those referred to foam color (1 = white, 2 = rose, 3 = cream, or 4 = capuchin) and liquid color (1 = pale straw yellow, 2 = straw yellow, 3 = golden yellow, or 4 = amber).

2.9. Statistical analysis

Each analysis was replicated at least three times for each of the three technological replicates and then the averages and the standard deviations were calculated. A two-way ANOVA followed by LSD test ($p < 0.05$) was applied to highlight the single and interactive effects of brewing procedures (BP1 or BP2) and yeast strains (*S. cerevisiae* 17,290, 14,061, 9502, and 9518) on physico-chemical and sensory characteristics of the beers. Principal Component Analysis (PCA) was applied to verify the possibility of homogeneously grouping the height types of beers according to their organic acid, sugar, phenolic, and volatile contents. The Pearson correlation coefficient (R) at p -value < 0.05 was applied to individuate significant correlations among beer characteristics and the results are reported in Table S1. The package Statistica for Windows V. 7.0. (Statsoft, Tulsa, OK, USA) was applied to perform all statistical analysis.

3. Results and discussion

3.1. Characteristics of the cereal mixtures

In order to understand the changes induced by the partial replacement of malted barley with de-hulled unmalted emmer, some compositional characteristics of the two cereals and of their mixture were evaluated. Moisture, ash, and protein contents of the mixture were $4.7 \pm 0.1\%$, $2.32 \pm 0.04\%$, and $9.4 \pm 0.4\%$, respectively, i.e. values similar to those detected in both the malted barley ($4.5 \pm 0.4\%$, $2.11 \pm 0.06\%$, and $9.7 \pm 0.4\%$) and the unmalted emmer ($4.4 \pm 0.2\%$, $2.07 \pm 0.08\%$, and $9.2 \pm 0.5\%$) individually analyzed. Moisture and protein contents were within the ranges of EBC standard (3.5–8% and 9–12%, respectively) considered suitable for brewing (Deme et al., 2019). The ash content of the unmalted emmer and barley malt were very similar. According to literature (Fogarasi et al., 2015), the adjunct of unmalted emmer decreased the TPC of the mixture (408.0 ± 8.1 mg/100 g, barley malt; 314.6 ± 38.9 mg/100 g, emmer; 353.5 ± 12.9 mg/100 g, the mixture) and its antioxidant activity (1.98 ± 0.13 mmol/100 g, barley malt; 0.35 ± 0.01 mmol/100 g, emmer; 1.85 ± 0.00 mmol/100 g, the mixture), although to a lesser extent than the addition of other wheat species (Baiano et al., 2023; Zrcková et al., 2019). The phenolic profile of the cereal mixture included the following compounds: kaempferol

(14.3 ± 0.10 mg/100 g), *p*-coumaric acid (1.64 ± 0.06 mg/100 g) and sinapic acid (1.04 ± 0.02 mg/100 g), to which both emmer and malt contributed; gallic acid (3.28 ± 0.14 mg/100 g) mainly contributed by the barley malt; epicatechingallate (0.22 ± 0.01 mg/100 g) contributed only by emmer; 4-hydroxybenzoic acid (0.79 ± 0.05 mg/100 g), vanillic acid (0.89 ± 0.03 mg/100 g), and caffeic acid (0.80 ± 0.01 mg/100 g) supplied only by the barley malt. First of all, it can be stated that the phenolic profiles of cereals and corresponding mixtures depend on genotypic variations, environmental influences, and barley malting process. However, *p*-coumaric acid content is an index of a high quality barley malt, since it increases from unmalted to malted grains and it is also positively correlated with soluble nitrogen and Kolbach index of a malt due to concurrent biosynthesis of hydrolytic enzymes (proteases and esterases) that facilitate both proteolysis and the release of phenolic acids (Cai et al., 2015). Moreover, for the same reason, barley malt also provided a good contribution of several free phenolic acids (Šimić et al., 2017). Instead, emmer contributed a remarkable content of epicatechingallate to the mixtures, thus counterbalancing the reduction of catechins during malting (Leitao et al., 2012), thus highlighting the opportunity to add unmalted emmer although it caused a reduction in overall phenolic content and antioxidant activity.

3.2. Physico-chemical and composition of the beers

The data reported in Table 1 highlight that both brewing procedures and yeast strains affected color and concentrations of CO₂, sugars, and organic acids of the produced beers. The mean EBC color ranged from 5.4 to 6.8, with significant single effects of brewing procedure and yeasts. BP1-beers showed significantly lower EBC values even though the application of conditions (higher conductivity water, longer protein rest, longer boiling time) having darkening effect due to polyphenol oxidation, increased Maillard reaction, and sugar caramelization (Xu

et al., 2017). However, the addition of Irish moss made BP1 worts clearer and brighter. The highest and the lowest color intensity were detected in beers fermented with *S. cerevisiae* 9502 (6.73) and 17,290 (5.75), respectively, probably as a result of their different ability to adsorb colored compounds on their cell walls. However, all these beers showed darker color (higher EBC values) than those produced by Baiano et al. (2023) using the same proportion of unmalted emmer and malted barley in the mixtures because of the more intense heat treatments of both BP1 and BP2. The average alcohol content was comprised between 4.20% and 5.23%. A slight but significant higher alcohol content was detected in BP2 beers. BP1 beers showed higher concentrations of the residual sugars as a consequence of the higher starch degradation occurring during their brewing. These results were related to the more intense endo-β-glucanase and endo-1,4-β-D-xylanase activities occurring during the longer protein rest of BP1, since those enzymes had optimal temperatures similar to those of proteases. In fact, the best performance of amylases can be obtained only if the endosperm cell walls were previously degraded by β-glucanases and xylanases, thus making the starch more available (Alfeo et al., 2021). BP1 beers also showed the highest CO₂ (3.60 g/L) and glycerol (2.73 mg/L) contents. Regarding the effects of yeasts, the highest (4.81%) and the lowest (4.34%) alcohol contents were measured on beers fermented with *S. cerevisiae* 9518 and 14,061, respectively, and were related to their different sugar fermentation ability. The average soluble solids remained in the final products ranged from a maximum of 8.32 Brix (*S. cerevisiae* 9502) to a minimum of 8.10 Brix (*S. cerevisiae* 14,061). *S. cerevisiae* 14,061 also left the lowest residual concentrations of all sugars after fermentation. The *S. cerevisiae* 14,061 could produce a series of secondary metabolites such as fusel alcohols, esters, carbonyls, sulfur compounds, thiols, and terpenoids that can contribute to the organoleptic properties of the beer (Hirst & Richter, 2016). The fermentation with *S. cerevisiae* 17,290 produced beers with the highest CO₂ (3.86 g/L) and the lowest glycerol (2.49

Table 1

Influence of brewing procedures and yeast strains on some physico-chemical parameters and on the contents in sugars and glycerol of the beers.

Beer acronyms	Color (EBC)	Alcohol content (%)	CO ₂ (g/L)	Soluble solids (Brix)	Sugars (mg/mL)					Glycerol (mg/L)
					Maltodextrins	Maltotriose	Maltose	Glucose	Fructose	
Interactive effects (Brewing Procedure × Yeast Strain)										
BP1-17,290	5.41 ± 0.21 ^a	4.20 ± 0.02 ^a	4.14 ± 0.35 ^f	8.47 ± 0.21 ^e	67.70 ± 2.58 ^e	27.51 ± 0.04 ^e	5.50 ± 0.39 ^{cd}	2.26 ± 0.19 ^e	2.12 ± 0.03 ^c	2.68 ± 0.13 ^d
BP1-14,061	5.80 ± 0.10 ^b	4.39 ± 0.01 ^{cd}	2.90 ± 0.10 ^c	8.33 ± 0.15 ^{de}	61.03 ± 0.16 ^d	21.82 ± 0.91 ^c	5.33 ± 0.10 ^{cd}	1.39 ± 0.26 ^d	1.24 ± 0.08 ^{ab}	2.52 ± 0.0 ^{ac}
BP1-9502	6.69 ± 0.51 ^{de}	4.46 ± 0.10 ^d	3.63 ± 0.08 ^{de}	8.43 ± 0.06 ^e	60.64 ± 2.21 ^d	24.85 ± 0.73 ^d	5.75 ± 0.19 ^e	1.00 ± 0.07 ^{bc}	1.17 ± 0.08 ^a	2.66 ± 0.12 ^{cd}
BP1-9518	5.85 ± 0.20 ^b	4.40 ± 0.05 ^{cd}	3.75 ± 0.10 ^{de}	8.47 ± 0.06 ^e	78.63 ± 1.75 ^f	31.82 ± 0.21 ^f	6.61 ± 0.43 ^f	0.90 ± 0.01 ^{bc}	1.36 ± 0.18 ^b	3.07 ± 0.08 ^e
BP2-17,290	6.09 ± 0.01 ^{bc}	4.76 ± 0.06 ^e	3.58 ± 0.08 ^d	8.07 ± 0.06 ^{bc}	55.91 ± 1.10 ^c	21.59 ± 0.24 ^c	5.00 ± 0.65 ^c	1.05 ± 0.11 ^c	1.16 ± 0.05 ^a	2.30 ± 0.13 ^a
BP2-14,061	6.23 ± 0.02 ^{cd}	4.29 ± 0.04 ^{ab}	3.95 ± 0.20 ^{ef}	7.87 ± 0.12 ^a	41.77 ± 2.21 ^a	14.44 ± 0.97 ^a	3.03 ± 0.66 ^a	nd ^a	1.11 ± 0.02 ^a	2.63 ± 0.35 ^d
BP2-9502	6.77 ± 0.10 ^e	4.37 ± 0.0 ^{bc}	2.00 ± 0.20 ^a	8.20 ± 0.0 ^{cd}	50.98 ± 0.59 ^b	20.21 ± 0.34 ^b	8.96 ± 0.12 ^e	0.81 ± 0.08 ^b	1.30 ± 0.17 ^{ab}	2.94 ± 0.11 ^{de}
BP2-9518	6.48 ± 0.02 ^{de}	5.23 ± 0.06 ^f	2.45 ± 0.25 ^b	8.00 ± 0.0 ^{ab}	54.12 ± 0.11 ^c	19.35 ± 0.45 ^b	4.24 ± 0.20 ^b	1.11 ± 0.04 ^c	1.25 ± 0.18 ^{ab}	2.37 ± 0.17 ^{ab}
Significance	*	*	*	*	*	*	*	*	*	*
Single effect of Brewing Procedure										
BP1	5.94 ^a	4.36 ^a	3.60 ^b	8.42 ^b	67.00 ^b	26.50 ^b	5.80 ^b	1.39 ^b	1.47 ^b	2.73 ^b
BP2	6.40 ^b	4.66 ^b	2.99 ^a	8.03 ^a	50.69 ^a	18.90 ^a	5.31 ^a	0.74 ^a	1.20 ^a	2.56 ^a
Significance	*	*	*	*	*	*	*	*	*	*
Single effect of Yeast Strain										
17,290	5.75 ^a	4.47 ^c	3.86 ^d	8.27 ^b	61.80 ^c	24.55 ^c	5.25 ^b	1.65 ^c	1.64 ^b	2.49 ^a
14,061	6.02 ^b	4.34 ^a	3.42 ^c	8.10 ^a	51.40 ^a	18.13 ^a	4.18 ^a	0.69 ^a	1.23 ^a	2.58 ^{ab}
9502	6.73 ^c	4.41 ^b	2.81 ^a	8.32 ^b	55.81 ^b	22.53 ^b	7.35 ^c	0.91 ^b	1.17 ^a	2.80 ^c
9518	6.17 ^b	4.81 ^d	3.10 ^b	8.23 ^b	66.37 ^d	25.58 ^d	5.42 ^b	1.00 ^b	1.31 ^a	2.72 ^{bc}
Significance	*	*	*	*	*	*	*	*	*	*

In column, different letters indicate significant differences at $p < 0.05$ by LSD multiple range test; The asterisks indicate significant differences at $p < 0.05$ by LSD multiple range test.

ns: not significant, nd: not detected.

mg/L) contents. *S. cerevisiae* 14,061 produced a medium-to-high amount of CO₂ accompanied by a medium glycerol production (CO₂, 3.4 g/L; glycerol, 2.58 mg/L). *S. cerevisiae* 9502 exhibited an opposite behavior, producing beers with the lowest CO₂ (2.81 g/L) and the highest glycerol (2.80 mg/L) concentrations. This behavior is a phenotypic trait of each strain and is due to the High Osmolarity Glycerol pathway, i.e. a diversion of part of the carbon flux from CO₂ to glycerol synthesis and accumulation, a biochemical mechanism that prevent cellular dehydration (Aslankoohi et al., 2015). CO₂ and glycerol are two important components of a craft beer because the first influences bubbling process parameter (that in turn act on trigeminal and gustatory receptors) while the second enhances flavor intensity, reduces perceived roughness, and increases body/fullness. The optimum carbon dioxide content for a white beer is around 4.5 g/L (Belcar et al., 2022) while the typical content of glycerol ranged between 1 and 3 g/L (Zhao et al., 2015). Based on this information, BP1 brewing procedure together with fermentations carried out by the oenological *S. cerevisiae* strains 17,290 and 14,061 gave the best results in producing an emmer-based beer inspired by the belgian white style. In any case, due to the negative correlation between alcohol and CO₂ (R = -0.43) or glycerol (R = -0.53), highly carbonated beer styles such as white, lager, and pils, may be only moderately alcoholic and full.

Type and quantity of organic acids strongly affect quality and shelf life of a beer, contributing to its freshness (most organic acids) but also to its off flavors (mainly acetic acid) (Rodrigues et al., 2010). Indeed, both brewing procedures and yeast strains exerted significant single and interactive effects on pH, acidity, and organic acid profile (Table 2). The average pH (3.97–4.20) was in typical range of white beers, and it was lower in BP1 beers than in BP2 ones. BP1 beers also showed the highest concentration of acetic acid, whereas BP2 beers showed a higher amount of the other organic acids. Independently on the combination of brewing procedure and yeast strain and in agreement with Cootte and Kirsop (1974), succinic acid was the most representative acid in all beers (2.36–6.98 mg/mL), since it is the acid usually excreted to the highest extent by yeast cells. It was followed by acetic acid (1.16–2.91 mg/mL), while the concentration of malic acid in beer is determined by its concentration in wort (Li & Liu, 2015). Fumaric acid was never detected. Most beer organic acids are excreted by yeasts as a result of deamination of amino acids present in the starting wort. As a consequence, the

different organic acid profile between BP1 and BP2 beers can be due to two opposite phenomena: the prolonged protein rest of BP1 favored an intense proteolysis; the prolonged boiling of BP1 amino acid-phenol interactions (Peixoto et al., 2021). Beers fermented by 17,290 and 14061 yeast strains had lower pH than those fermented by Susumaniello strains. More in depth, the beers fermented by *S. cerevisiae* 17,290 showed the highest titratable and volatile acidity, and the highest concentrations of succinic, acetic, and citric acids. According to Selecký et al. (2008), this behavior is typical of strains deficient in the tricarboxylic acid cycle enzymes. The lowest volatile acidity (R = 0.61 for correlation between this acid and the beer sourness), together with the lowest concentration of all the organic acids (except malic) were detected in samples fermented by *S. cerevisiae* 9502 strain.

Phenolic compounds not only play an active role in haze formation and preservation of main technological properties of beer, but they also act as flavor precursors in beer. As can be inferred from Table 3, the average of TPC content was between 327.32 mg/L (BP2-9518) and 411.35 mg/L (BP2-17,290). However, since TCP represents an estimation of all compounds that act as reducers (not only phenolics), the study of the beer phenolic profiles is required to understand their influence of the brewing procedure. The beer produced through BP2 had the highest contents of 9 phenolic compounds, namely rosmarinic acid, catechin, gallic acid, epicatechin, quercetin, syringic acid, ferulic acid, caffeic acid, and *p*-coumaric acid. The concentrations of other 8 compounds (epicatechingallate, epigallocatechin, chlorogenic acid, sinapic acid, kaempferol, rutin, vanillic acid, and 4-hydroxybenzoic acid) were higher in BP1 beers. Resveratrol concentration was similar between the two brewing procedures. The differences in individual phenolic concentrations between the two brewing procedures are explained by to opposite phenomena: the prolonged resting time of BP1 favored the release of the bound phenolic acids (Schwarz et al., 2012) while the following boiling promotes the thermal decarboxylation of phenolic acids in volatile monophenols, the formation of polyphenol-protein complexes (hot trub) and of polyphenol-polysaccharide aggregations (Wannenmacher et al., 2018). Furthermore, the amount of hot trub formed depends on boiling time and a duration higher than 60 min (as in BP1) determines a decrease in polyphenol content (Muñoz-Insa et al., 2015). Data were further processed and, for each sample, the sum (SUM) of the concentrations of all phenolic compounds detected by HPLC-DAD was

Table 2

Influence of brewing procedures and yeast strains on pH, acidity, and the organic acid profiles of the beers.

Beer acronyms	pH	Titratable acidity (g/L)	Volatile acidity (g/L)	Organic acids (mg/mL)				
				Citric	Malic	Succinic	Lactic	Acetic
Interactive effects (Brewing Procedure × Yeast Strain)								
BP1-17,290	4.01 ± 0.05 ^{ab}	2.14 ± 0.02 ^d	1.35 ± 0.03 ^f	0.84 ± 0.03 ^f	0.81 ± 0.02 ^d	6.98 ± 0.08 ^b	0.71 ± 0.01 ^c	2.91 ± 0.03 ^g
BP1-14,061	4.05 ± 0.03 ^{bd}	1.92 ± 0.03 ^b	0.94 ± 0.03 ^c	0.69 ± 0.03 ^{de}	0.73 ± 0.00 ^c	6.80 ± 0.17 ^g	1.06 ± 0.01 ^b	1.98 ± 0.03 ^c
BP1-9502	4.03 ± 0.03 ^{ac}	2.60 ± 0.01 ^f	1.10 ± 0.02 ^d	0.46 ± 0.03 ^a	0.57 ± 0.01 ^a	2.98 ± 0.03 ^b	0.43 ± 0.01 ^b	2.11 ± 0.04 ^d
BP1-9518	4.09 ± 0.04 ^{cd}	2.40 ± 0.01 ^e	1.36 ± 0.02 ^f	0.48 ± 0.02 ^a	0.55 ± 0.03 ^a	2.36 ± 0.01 ^a	0.33 ± 0.01 ^a	2.23 ± 0.03 ^c
BP2-17,290	3.97 ± 0.01 ^a	2.95 ± 0.02 ^g	1.40 ± 0.02 ^g	0.55 ± 0.04 ^b	0.70 ± 0.00 ^b	6.71 ± 0.07 ^f	0.86 ± 0.02 ^e	2.50 ± 0.05 ^f
BP2-14,061	4.00 ± 0.00 ^{ab}	2.01 ± 0.02 ^c	1.30 ± 0.02 ^e	0.63 ± 0.01 ^c	0.57 ± 0.02 ^a	4.13 ± 0.02 ^c	0.83 ± 0.00 ^d	2.18 ± 0.04 ^c
BP2-9502	4.20 ± 0.04 ^e	1.34 ± 0.08 ^a	0.39 ± 0.03 ^a	0.68 ± 0.02 ^d	0.98 ± 0.01 ^e	4.28 ± 0.01 ^d	0.93 ± 0.00 ^f	1.16 ± 0.01 ^a
BP2-9518	4.09 ± 0.04 ^d	1.33 ± 0.02 ^a	0.55 ± 0.01 ^b	0.74 ± 0.03 ^e	0.74 ± 0.00 ^c	4.68 ± 0.01 ^e	1.01 ± 0.00 ^g	1.63 ± 0.03 ^b
Signif.	*	*	*	*	*	*	*	*
Single effect of Brewing Procedure								
BP1	4.03 ^a	2.27 ^b	1.18 ^b	0.62 ^a	0.64 ^a	4.78 ^a	0.63 ^a	2.31 ^b
BP2	4.07 ^b	1.91 ^a	0.91 ^a	0.65 ^b	0.75 ^b	4.95 ^b	0.91 ^b	1.87 ^a
Signif.	*	*	*	*	*	*	*	*
Single effect of Yeast Strain								
17,290	3.99 ^a	2.54 ^c	1.37 ^d	0.70 ^d	0.75 ^b	6.84 ^d	0.78 ^b	2.71 ^d
14,061	4.02 ^a	1.96 ^b	1.12 ^c	0.66 ^c	0.65 ^a	5.47 ^c	0.95 ^c	2.08 ^c
9502	4.11 ^b	1.97 ^b	0.74 ^a	0.57 ^a	0.77 ^b	3.63 ^b	0.68 ^a	1.63 ^a
9518	4.09 ^b	1.86 ^a	0.95 ^b	0.61 ^b	0.64 ^a	3.52 ^a	0.67 ^a	1.93 ^b
Signif.	*	*	*	*	*	*	*	*

In column, different letters indicate significant differences at $p < 0.05$ by LSD multiple range test.

The asterisks indicate significant differences at $p < 0.05$ by LSD multiple range test.

ns: not significant.

Table 3
Influence of brewing procedures and yeast strains on the total phenolic content, antioxidant activity, and phenolic profile of the beers.

Beer acronyms	TPC (mg/L)	Phenolics (mg/L)																		AOA (mmol Trolox/ L)
		Gallic- acid	4- HBA	Catechin	Vanillic- acid	Caffeic- acid	Syringic- acid	Epicatechin	Chlorogenic- acid	EGC	Ferulic- acid	p- Coumaric- acid	Sinapic- acid	EG	Rutin	Resveratrol	Rosmarinic- acid	Quercetin	Kaempferol	
Interactive effects (Brewing Procedure × Yeast Strain)																				
BP1- 17,290	390.01 ± 8.90 ^c	21.88 ± 0.15 ^c	2.16 ± 0.16 ^b	5.19 ± 0.15 ^b	8.36 ± 0.45 ^f	1.86 ± 0.06 ^c	0.13 ± 0.04 ^a	15.21 ± 0.37 ^d	21.99 ± 0.82 ^{cd}	12.94 ± 0.95 ^a	1.99 ± 0.02 ^a	2.46 ± 0.04 ^{bc}	14.71 ± 0.95 ^d	37.95 ± 0.45 ^f	9.09 ± 0.38 ^d	1.43 ± 0.01 ^{bc}	7.15 ± 0.79 ^a	2.73 ± 0.22 ^b	9.69 ± 0.05 ^b	0.94 ± 0.01 ^{bc}
BP1- 14,061	385.71 ± 14.30 ^c	29.52 ± 0.13 ^f	4.44 ± 0.06 ^e	32.35 ± 0.05 ^e	4.72 ± 0.14 ^e	1.77 ± 0.04 ^b	11.47 ± 0.19 ^e	16.54 ± 0.87 ^e	25.47 ± 5.04 ^f	27.06 ± 1.39 ^d	2.48 ± 0.02 ^d	1.61 ± 0.20 ^a	12.82 ± 0.22 ^c	45.01 ± 1.74 ^g	9.76 ± 0.65 ^d	1.45 ± 0.01 ^{cd}	37.34 ± 2.48 ^b	2.92 ± 0.11 ^{bc}	22.13 ± 1.40 ^f	1.03 ± 0.10 ^e
BP1-9502	370.76 ± 6.74 ^{bc}	8.81 ± 0.45 ^a	3.68 ± 0.08 ^d	4.31 ± 0.10 ^{ab}	2.59 ± 0.06 ^c	0.87 ± 0.00 ^a	1.21 ± 0.31 ^b	9.56 ± 0.54 ^a	10.62 ± 0.76 ^{a b}	32.92 ± 1.91 ^e	2.07 ± 0.01 ^b	2.27 ± 0.15 ^b	13.92 ± 1.38 ^{cd}	26.35 ± 1.21 ^c	7.59 ± 0.30 ^c	1.30 ± 0.01 ^a	8.11 ± 0.78 ^a	2.97 ± 0.07 ^{bc}	5.53 ± 0.33 ^a	0.88 ± 0.03 ^{ab}
BP1-9518	354.03 ± 19.70 ^b	9.20 ± 0.27 ^a	4.45 ± 0.30 ^e	3.07 ± 0.12 ^a	1.09 ± 0.01 ^a	0.90 ± 0.07 ^a	1.59 ± 0.22 ^b	10.33 ± 0.38 ^a	17.73 ± 4.70 de	12.42 ± 2.15 ^a	2.39 ± 0.02 ^c	1.52 ± 0.07 ^a	17.85 ± 0.65 ^e	32.42 ± 0.41 ^d	0.29 ± 0.00 ^a	1.63 ± 0.01 ^e	8.67 ± 0.24 ^a	3.14 ± 0.17 ^c	20.09 ± 0.83 ^{ce}	0.84 ± 0.00 ^a
BP2- 17,290	411.35 ± 6.73 ^d	22.95 ± 0.09 ^d	3.11 ± 0.11 ^c	37.43 ± 1.90 ^f	1.41 ± 0.02 ^b	3.08 ± 0.07 ^e	1.39 ± 0.21 ^b	15.67 ± 0.60 ^d	12.63 ± 0.83 bc	23.17 ± 1.43 ^c	3.01 ± 0.00 ^g	3.12 ± 0.06 ^d	14.95 ± 0.71 ^d	36.11 ± 7.20	3.20 ± 1.09 ^c	1.48 ± 0.06 ^d	13.73 ± 0.40 ^a	5.74 ± 0.35 ^d	12.70 ± 0.18 ^c	1.13 ± 0.07 ^d
BP2- 14,061	364.00 ± 12.50 ^b	12.85 ± 0.38 ^b	0.86 ± 0.14 ^a	11.78 ± 1.05 ^c	1.43 ± 0.01 ^b	2.30 ± 0.05 ^d	1.48 ± 0.03 ^b	14.29 ± 0.16 ^c	13.99 ± 1.21 bd	11.90 ± 0.06 ^a	2.61 ± 0.00 ^f	1.74 ± 0.23 ^a	11.27 ± 0.53 ^b	33.00 ± 1.90 ^d	5.33 ± 0.41 ^b	1.40 ± 0.00 ^b	60.65 ± 9.93 ^c	1.72 ± 0.02 ^a	14.53 ± 1.03 ^d	1.17 ± 0.06 ^d
BP2-9502	357.70 ± 10.50 ^b	24.23 ± 0.56 ^c	4.17 ± 0.28 ^c	42.78 ± 2.17 ^g	4.44 ± 0.07 ^e	2.26 ± 0.06 ^d	8.25 ± 0.48 ^c	12.97 ± 0.27 ^b	16.22 ± 0.81 ^{cd}	19.37 ± 1.33 ^b	2.52 ± 0.04 ^e	2.56 ± 0.12 ^c	9.03 ± 0.28 ^a	17.99 ± 0.00 ^b	6.08 ± 0.08 ^b	1.70 ± 0.00 ^f	42.19 ± 3.41 ^b	7.94 ± 0.20 ^c	9.71 ± 0.06 ^b	0.83 ± 0.01 ^a
BP2-9518	327.32 ± 7.44 ^a	44.56 ± 0.20 ^g	4.35 ± 0.22 ^e	24.22 ± 0.62 ^d	3.01 ± 0.05 ^d	2.25 ± 0.04 ^d	9.68 ± 0.60 ^d	12.22 ± 0.28 ^b	6.98 ± 0.36 ^a	22.08 ± 0.39 ^c	2.49 ± 0.02 ^{de}	2.47 ± 0.02 ^{bc}	10.74 ± 0.12 ^b	14.98 ± 1.31 ^a	6.15 ± 0.57 ^b	1.30 ± 0.02 ^a	35.84 ± 3.32 ^b	7.79 ± 0.07 ^e	11.82 ± 0.27 ^c	0.99 ± 0.04 ^c
Signif.	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*
BP1	375.08 ^a	17.35 ^a	3.68 ^b	11.23 ^a	4.19 ^b	1.35 ^a	3.60 ^a	12.91 ^a	18.95 ^b	21.34 ^b	2.23 ^a	1.96 ^a	14.83 ^b	35.43 ^b	6.68 ^b	1.45 ^a	15.32 ^a	2.94 ^a	14.36 ^b	0.92 ^a
BP2	365.08 ^a	26.14 ^b	3.17 ^a	29.05 ^b	2.57 ^a	2.47 ^b	5.20 ^b	13.79 ^b	12.45 ^a	19.13 ^a	2.66 ^b	2.47 ^b	11.50 ^a	25.52 ^a	6.19 ^a	1.47 ^a	38.10 ^b	5.80 ^b	12.19 ^a	1.03 ^b
Signif.	ns	*	*	*	*	*	*	*	*	*	*	*	*	*	*	ns	*	*	*	*
17,290	400.67 ^c	22.42 ^c	2.64 ^a	21.31 ^b	4.89 ^d	2.47 ^c	0.76 ^a	15.44 ^b	17.31 ^b	18.06 ^{ab}	2.50 ^c	2.79 ^d	14.83 ^b	37.03 ^c	8.15 ^c	1.45 ^b	10.44 ^a	4.23 ^b	11.19 ^b	1.04 ^c
14,061	374.83 ^b	21.18 ^b	2.65 ^a	22.06 ^b	3.07 ^b	2.03 ^b	6.47 ^d	15.41 ^b	19.73 ^c	19.48 ^b	2.54 ^d	1.62 ^a	12.05 ^a	39.01 ^d	7.54 ^c	1.42 ^a	48.99 ^c	2.32 ^a	18.33 ^d	1.10 ^d
9502	364.17 ^b	16.53 ^a	3.92 ^b	23.55 ^c	3.51 ^c	1.57 ^a	4.73 ^b	11.26 ^c	13.42 ^a	26.14 ^c	2.29 ^a	2.42 ^c	11.47 ^a	22.17 ^a	6.84 ^b	1.50 ^c	25.15 ^b	5.46 ^c	7.62 ^a	0.85 ^b
9518	340.67 ^a	26.88 ^d	4.41 ^c	13.65 ^a	2.05 ^a	1.57 ^a	5.64 ^c	11.27 ^c	12.35 ^a	17.25 ^a	2.44 ^b	2.00 ^b	14.30 ^b	23.70 ^b	3.22 ^a	1.47 ^b	22.25 ^b	5.47 ^c	15.96 ^c	0.91 ^b
Signif.	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*

In column, different letters indicate significant differences at $p < 0.05$ by LSD multiple range test. The asterisks indicate significant differences at $p < 0.05$ by LSD multiple range test. ns: not significant. TPC: Total Phenolic Content. AOA: Antioxidant activity. **IUPAC names:** gallic acid, 3,4,5-trihydroxybenzoic acid; 4-HBA, 4-hydroxybenzoic acid; catechin, (2R,3S)-2-(3,4-dihydroxyphenyl)-3,4-dihydro-2H-chromene-3,5,7-triol; vanillic acid, 4-hydroxy-3-methoxybenzoic acid; caffeic acid, (E)-3-(3,4-dihydroxyphenyl)prop-2-enoic acid; syringic acid, 4-hydroxy-3,5-dimethoxybenzoic acid; epicatechin, (2R,3R)-2-(3,4-dihydroxyphenyl)-3,4-dihydro-2H-chromene-3,5,7-triol; chlorogenic acid, (1S,3R,4R,5R)-3-[(E)-3-(3,4-dihydroxyphenyl)prop-2-enoyl]oxy-1,4,5-trihydroxycyclohexane-1-carboxylic acid; ECG (epigallocatechin), (2R,3R)-2-(3,4,5-trihydroxyphenyl)-3,4-dihydro-2H-chromene-3,5,7-triol; ferulic acid, (E)-3-(4-hydroxy-3-methoxyphenyl)prop-2-enoic acid; p-Coumaric acid, (E)-3-(4-hydroxyphenyl)prop-2-enoic acid; sinapic acid, (E)-3-(4-hydroxy-3,5-dimethoxyphenyl)prop-2-enoic acid; EG (epicatechingallate), [(2R,3R)-2-(3,4-dihydroxyphenyl)-5,7-dihydroxy-3,4-dihydro-2H-chromen-3-yl] 3,4,5-trihydroxybenzoate; rutin, 2-(3,4-dihydroxyphenyl)-5,7-dihydroxy-3-[(2S,3R,4S,5S,6R)-3,4,5-trihydroxy-6-[[[(2R,3R,4R,5R,6S)-3,4,5-trihydroxy-6-methyloxan-2-yl]oxymethyl]oxan-2-yl]oxy]chromen-4-one; resveratrol, 5-[(E)-2-(4-hydroxyphenyl)ethenyl]benzene-1,3-diol; rosmarinic acid, (2R)-3-(3,4-dihydroxyphenyl)-2-[(E)-3-(3,4-dihydroxyphenyl)prop-2-enoyl]oxypropanoic acid; quercetin, 2-(3,4-dihydroxyphenyl)-3,5,7-trihydroxychromen-4-one; kaempferol, 3,5,7-trihydroxy-2-(4-hydroxyphenyl)chromen-4-one.

calculated, obtaining values ranging from 144.70 mg/L of BP1-9502 to 288.85 mg/L of BP1-14,061. Data concerning the effect of the brewing procedures on SUM (189.82 mg/L for BP1 beers and 219.83 mg/L for the BP2) and on the various classes of polyphenols (hydroxycinnamic acids, hydroxybenzoic acids, flavanols, and flavonols) confirm this trend. Regarding the effects of yeast strains, the highest and the lowest TPC were detected on beers fermented by *S. cerevisiae* 17,290 (400.67 mg/L) and 9518 (340.67 mg/L), respectively. Data concerning the single effect of the yeasts on the individual phenolic compounds do not show a constant behavior for each strain. Instead, the yeast effects depended of on the nature of the phenolic classes. Concerning the sum of hydroxycinnamic acid contents (HCA SUM), the highest (87.02 mg/L) and the lowest (50.33 mg/L) concentrations were detected in beers fermented by *S. cerevisiae* 14,061 and 17,290, respectively. The beers fermented by *S. cerevisiae* 9518 had the highest concentrations of total hydroxybenzoic acids (HBA SUM, 38.96 mg/L) and total flavonols (FLAVON

SUM, 21.42 mg/L) while the corresponding lowest concentrations were detected in beers inoculated with *S. cerevisiae* 9502 (28.68 and 13.08 mg/L). The highest total concentration of flavanols (FLAVAN SUM) and rutin were detected in beers fermented with *S. cerevisiae* 14,061 (95.97 mg/L) and *S. cerevisiae* 17,290 (8.15 mg/L), respectively. Instead, the beers fermented with *S. cerevisiae* 9518 had the lowest FLAVAN SUM (65.87 mg/L) and rutin (3.22 mg/L). Finally, the average sum (SUM) of the concentrations of all phenolic compounds detected by HPLC-DAD was in the following decreasing order: 14,061 (245.98 mg/L), 17,290 (197.91 mg/L), 9502 (189.55 mg/L), and 9518 (185.86 mg/L). Consistently with data related to the phenolic component, the antioxidant activity was significantly higher in BP2 than in BP1 beers, in agreement with Fantozzi et al. (1998). Concerning the effects of yeasts, AOA followed the same decreasing order of SUM and showed a high correlation coefficient ($R = 0.837$) to which caffeic acid, ferulic acid, and epicatechin have significantly contributed (Table S1). These high

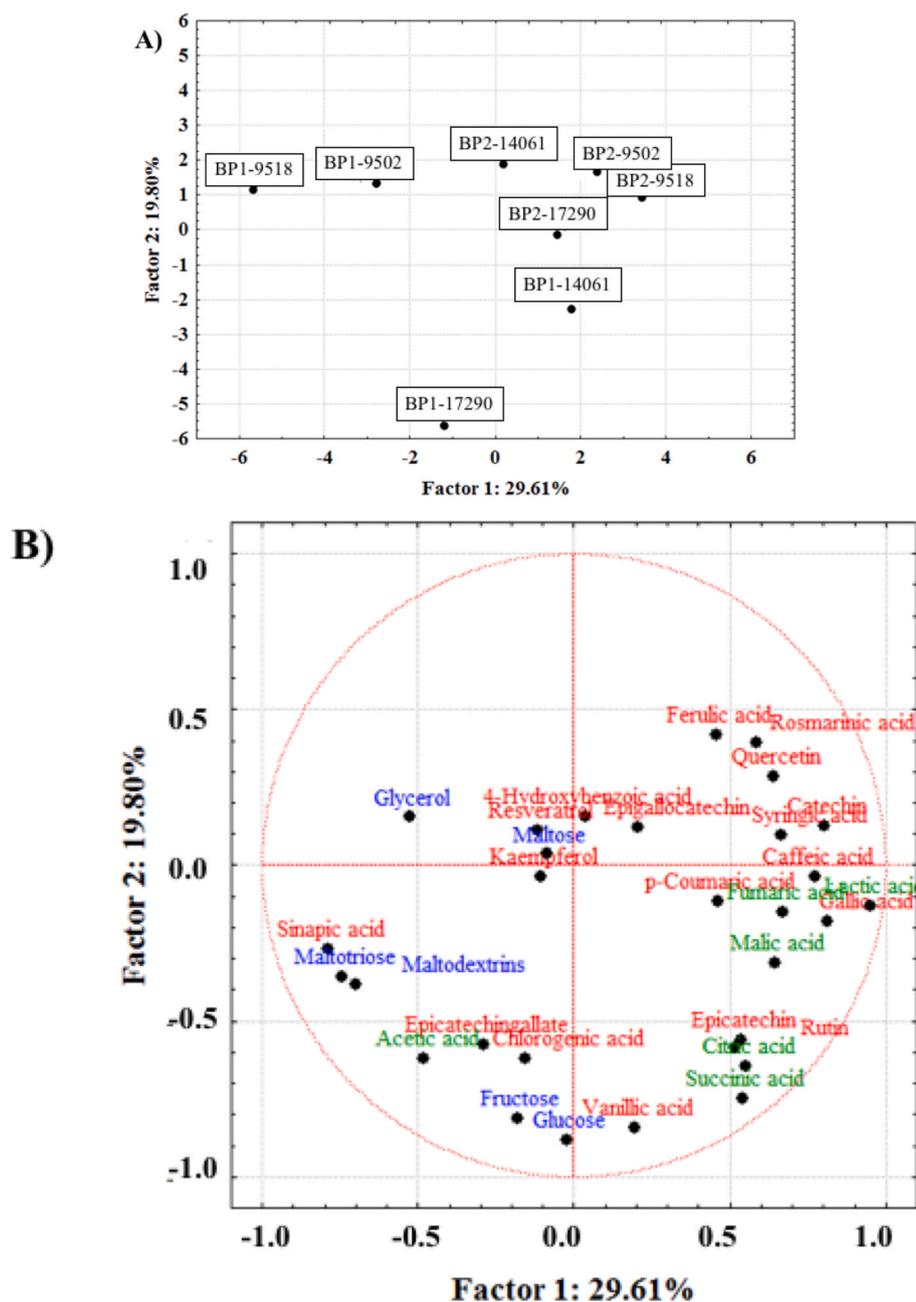


Fig. 1. PCA analysis of non-volatile compounds: scores (A) and correlation circles (B). Sugars and glycerol are in blue, organic acids in green, and phenolic compounds are in red font.

correlations depended on both the relative antioxidant efficiency of the individual phenolics (Anitha et al., 2020; Martins et al., 2016)) and their concentrations.

PCA was applied to the data set including all the non-volatile compounds (Fig. 1), with the first two principal components explaining 49.41% of the total variance. The scores on the factorial plane (Fig. 1A) highlighted that the beers homogeneously grouped according to both the brewing procedure and the fermenting strain. Coming from negative to positive scores, Factor 1 allowed to separate BP1-9518 (around -6), BP1-9502 (around -3), BP1-17,290 (around -1), BP2-14,061 (around 0), the couple BP2-17,290/BP1-14,061 (range 1–2), and the couple BP2-9502/BP2-9518 (range 2.5–3.5). However, most of them are very close from each other due to the similar scores of Factor 2, which were comprised in a narrow range (1–2.2). BP2-17,290 and BP1-14061 were clearly separated from each other along Factor 2, thanks to: the highest concentrations of lactic acid, syringic acid, epicatechin, chlorogenic acid, epicatechigallate, and kaempferol detected in BP1-14,061; the highest concentrations of succinic acid, acetic acid, caffeic acid, ferulic acid, and *p*-coumaric acid and the lowest glycerol content detected in BP2-17,290. BP1-17,290, which had Factor 2 scores around -1, were characterized by the lowest contents of syringic and ferulic acids and by the highest concentrations of glucose, fructose, citric acid, malic acid, succinic acid, acetic acid, and vanillic acid.

3.3. Volatile molecules composition of beers

Thirty seven volatile molecules belonging to different chemical classes were identified (Table 4). The ester group was characterized by a large number of molecules distributed between the two subgroups of ethyl esters and acetate esters which contribute notes of fruitiness. This result was in agreement with previous studies on the aromatic characterization of wheat beers (De Flaviis et al., 2022; Gugino et al., 2024; Palombi et al., 2023). Esters can penetrate yeast cell membranes and permeate beer, which explain their increasing content during fermentation (Paszko et al., 2023). Among esters, isoamyl acetate (1.12–1.62 mg/L), phenyl acetate (1.05–2.30 mg/L), ethyl hexanoate (1.40–1.81 mg/L), ethyl octanoate (0.40–3.56 mg/L) and ethyl decanoate (1.17–3.14 mg/L) were detected in the highest concentrations. Based on the information provided by authors (Hiralal et al., 2014; Pires et al., 2014), it seems that ethyl hexanoate is a common compound found in top fermentation beers, also known as ales. Additionally, other studies have reported the presence of compounds such as ethyl decanoate, phenylethyl alcohol, and ethyl octanoate in these types of beers. These compounds are likely derived from the fermentation process, where yeast metabolizes sugars and produces various by-products that contribute to the aroma and flavor characteristics of the beer.

Fatty acid esters (ethyl hexanoate-octanoate-decanoate) were quantified at concentrations above their respective perception thresholds (0.005 mg/L, 0.5 mg/L, 1.5 mg/L, respectively), suggesting a significant and positive impact on the overall odor profile (Mastrangelo et al., 2023; Pietrafesa et al., 2023). Significant differences ($p < 0.05$) were found both as a function of the technological process (except for ethyl hexanoate) and as a function of the yeast strain used for all esters. *S. cerevisiae* 14,061 and 9518 showed significantly higher concentrations (11.26 and 10.95 mg/L, respectively) of the total esters identified.

Among the fermentation by-products, alcohols are known to play a key role in the overall volatolomic profile. According to Pires et al. (2014), alcohols were the predominant group of volatile compounds found in the evaluated Blond Ale and IPA beers, which is a common characteristic in beers. This suggests that alcohols contribute significantly to the aroma and flavor profiles of these beer styles.

In emmer beers, this class of molecules, originate from amino acid metabolism through a sequence of decarboxylation and reduction reactions, is quantitatively predominant (De Flaviis et al., 2022; Gugino et al., 2024). In particular, the higher alcohols (mainly 1-propanol, 2-methyl-1-propanol, 3-methyl-1-butanol, and phenylethanol) affect

both directly and indirectly the sensory profile. Concentrations of higher alcohols lower than 400 mg/L confer fruity characters (Swiegers et al., 2005). In the beer samples under investigation, significant differences between the total concentrations of higher alcohols were found, depending on the yeast strain used, and ranging from 28.76 mg/L (*S. cerevisiae* 17,290) to 38.52 mg/L (*S. cerevisiae* 14,061). Higher alcohols were also involved in esterification reactions, thus contributing to the production of flower/fruity odors. Phenylethanol, a molecule characterized by a typical rose odor, was detected in all the samples tested, except BP2-9518, at concentrations above the threshold of perception (10 mg/L), proving to exert a strategic role in the beer flavor profile.

The contribution of hops to the aromatic complexity of beers is expressed through the production of terpenes and terpenoids, the third most important class of compounds in our samples. Differences in terpene and terpenoid content may be related to the role of yeast strains in the biotransformation of monoterpenes. In beer, terpenes contribute to the aroma and flavor profile, adding complexity and character to the finished product. Terpenes are responsible for the distinctive floral, citrus, pine, herbal, and spicy notes often associated with different beer styles. Ten terpenes were identified and the interaction between the brewing procedure and the strain used significantly influenced the total terpene content and the individual molecules. BP1-170,461 and BP1-9502 showed the highest (17.59 mg/L) and the lowest (3.39 mg/L) terpene concentration, respectively. Linalool, considered an indicator compound in the analysis of hop aroma, was detected in quantities ranging from 0.27 mg/L (BP2-17,061) to 2.95 mg/L (BP1-9518).

As suggested by Fritsch and Schieberle (2005), linalool, which is claimed to be a key molecule contributing to hoppy flavor. This abundance could be attributed to its polar nature and higher solubility compared to the less polar and more volatile monoterpene and sesquiterpene hydrocarbons. These hydrocarbons are typically lost during boiling due to evaporation with wort steam and adsorption on the trub (Dresel et al., 2015). It is important to note that the specific terpene profile of a craft beer can vary widely depending on factors such as hop selection, brewing techniques, and recipe formulation. Brewers often experiment with different hop combinations and processes to create unique flavor profiles in their beers, which may emphasize certain terpenes over others.

Among the volatile acids, hexanoic, octanoic, nonanoic and decanoic acids were identified in the beer headspace. Significant differences were observed among beers with total values ranging from 0.98 mg/L (BP1-17,290) to 4.26 mg/L (BP2-9518). A significant portion of the total organic acids in beer, approximately 50%, originates from the wort itself. The remaining portion is either produced or transformed through yeast metabolism during the fermentation process (Yamauchi et al., 1995). Concerning hydrocarbons, styrene was detected in concentration varying from 0.14 mg/L (BP1-9518) to 0.62 mg/L (BP2-17,290) as a result of the decarboxylation of free cinnamic acid (Rossi et al., 2014) while it was under the detection limit in BP1-17,290 and BP1-9502. Regarding volatile phenols, the enzymatic decarboxylation of ferulic acid during the fermentation process resulted in the formation of 4-vinylguaiacol, which was identified in all beers, with values significantly higher in BP2 samples, especially if fermented by *S. cerevisiae* 14, 061. (E)- β -Damascenone was the only norisoprenoid identified, with the highest concentrations detected in BP2-17,290 beers (0.006 mg/L).

A PCA was computed to explore the correlations between the volatile molecules identified and the experimental beers. The first two factors explained 46.37% of the total variance (Fig. 2). Factor 1 explains a larger percentage of the total variance (30.57%) and allowed the separation of BP2-17,290 beers, which are placed in correspondence with extremely negative scores, from all the other samples, which are located in the space delimited by scores comprised between 0 and 4. The distinctive elements of BP2-17,290 beers were the following: lower concentrations of esters and alcohols; higher concentrations of hydrocarbons and norisoprenoids; concentrations under the detection limits of the esters

Table 4
Influence of brewing procedures yeast strain on the concentrations (mg/L) of volatile compounds in beers.

	Interactive effects (Brewing Procedure × Yeast Strain)									Single effect of Brewing Procedure			Single effect of Yeast Strain				
	BP1-17,290	BP1-174,061	BP1-9502	BP1-9518	BP2-17,290	BP2-174,061	BP2-9502	BP2-9518	Sign.	BP1	BP2	Sign.	17,290	14,061	9502	9518	Sign.
ESTERS																	
Ethyl Acetate	0.64 ± 0.17 ^{bc}	0.39 ± 0.11 ^a	0.68 ± 0.15 ^{bc}	0.54 ± 0.11 ^{ac}	0.52 ± 0.17 ^{ac}	0.72 ± 0.11 ^c	0.63 ± 0.11 ^{bc}	0.48 ± 0.11 ^{ab}	*	0.56 ^a	0.59 ^a	ns	0.58 ^a	0.56 ^a	0.65 ^a	0.51 ^a	ns
3-Methylbutyl Acetate (Isoamyl Acetate)	1.62 ± 0.33 ^d	1.32 ± 0.14 ^{bc}	1.12 ± 0.14 ^b	1.61 ± 0.14 ^{cd}	nd ^a	1.55 ± 0.14 ^{cd}	1.43 ± 0.14 ^{cd}	1.50 ± 0.14 ^{cd}	*	1.41 ^b	1.12 ^a	*	0.81 ^a	1.43 ^{bc}	1.27 ^b	1.56 ^c	*
Ethyl Hexanoate	1.40 ± 0.17 ^{bc}	1.40 ± 0.25 ^{bc}	1.35 ± 0.31 ^{bc}	1.22 ± 0.22 ^b	nd ^a	1.81 ± 0.25 ^d	1.66 ± 0.25 ^{cd}	1.50 ± 0.22 ^{bd}	*	1.34 ^a	1.24 ^a	ns	0.70 ^a	1.60 ^b	1.50 ^b	1.36 ^b	*
Ethyl Heptanoate	nd ^a	0.02 ± 0.01 ^a	nd ^a	0.02 ± 0.01 ^a	1.54 ± 0.21 ^b	0.03 ± 0.01 ^a	0.02 ± 0.01 ^a	0.02 ± 0.01 ^a	*	0.01 ^a	0.40 ^b	*	0.77 ^b	0.03 ^a	0.01 ^a	0.02 ^a	*
Methyl Octanoate	0.10 ± 0.02 ^a	0.11 ± 0.02 ^a	0.12 ± 0.03 ^a	0.13 ± 0.04 ^a	0.14 ± 0.02 ^a	0.15 ± 0.02 ^{ab}	0.12 ± 0.02 ^a	0.19 ± 0.04 ^b	*	0.12 ^a	0.15 ^b	*	0.12 ^a	0.1 ^{3ab}	0.12 ^a	0.16 ^b	*
Ethyl Octanoate	1.44 ± 0.27 ^b	1.90 ± 0.21 ^{bc}	1.21 ± 0.17 ^b	1.68 ± 0.84 ^b	0.40 ± 0.07 ^a	3.56 ± 0.21 ^d	1.82 ± 0.21 ^{bc}	2.48 ± 0.84 ^c	*	1.56 ^a	2.06 ^b	*	0.92 ^a	2.73 ^d	1.51 ^b	2.08 ^b	*
Methyl Decanoate	0.14 ± 0.03 ^{bc}	0.01 ± 0.00 ^a	0.21 ± 0.04 ^c	0.04 ± 0.02 ^a	0.86 ± 0.13 ^d	0.08 ± 0.00 ^{ab}	0.03 ± 0.00 ^a	0.16 ± 0.02 ^{bc}	*	0.10 ^a	0.28 ^b	*	0.50 ^c	0.04 ^a	0.12 ^b	0.10 ^{ab}	*
Ethyl Decanoate	2.95 ± 0.51 ^d	3.14 ± 0.34 ^d	2.62 ± 0.34 ^{cd}	2.96 ± 0.51 ^d	1.17 ± 0.51 ^a	2.55 ± 0.34 ^{cd}	1.94 ± 0.34 ^{bc}	1.73 ± 0.51 ^{ab}	*	2.92 ^b	1.85 ^a	*	2.06 ^a	2.84 ^b	2.28 ^a	1.34 ^{ab}	*
Diethyl Butanedioate (Diethyl Succinate)	0.16 ± 0.03 ^{bd}	0.15 ± 0.04 ^{bd}	0.26 ± 0.04 ^e	0.13 ± 0.03 ^{bc}	nd ^a	0.11 ± 0.04 ^b	0.18 ± 0.04 ^{cd}	0.20 ± 0.03 ^d	*	0.17 ^b	0.12 ^a	*	0.08 ^a	0.13 ^b	0.22 ^c	0.16 ^b	*
Ethyl dec-9-enoate (Ethyl-9-Decenoate)	0.25 ± 0.04 ^{cd}	0.13 ± 0.02 ^b	0.17 ± 0.03 ^b	0.30 ± 0.04 ^d	0.04 ± 0.02 ^a	0.30 ± 0.02 ^d	0.12 ± 0.02 ^b	0.23 ± 0.04 ^c	*	0.21 ^b	0.17 ^a	*	0.14 ^a	0.21 ^b	0.15 ^a	0.26 ^c	*
Benzyl Acetate (Benzene Acetate)	0.02 ± 0.01 ^a	0.01 ± 0.00 ^a	0.11 ± 0.05 ^b	0.11 ± 0.00 ^a	0.21 ± 0.07 ^c	0.05 ± 0.01 ^a	0.01 ± 0.01 ^a	0.03 ± 0.01 ^a	*	0.04 ^a	0.07 ^b	*	0.11 ^c	0.03 ^{ab}	0.06 ^b	0.02 ^a	*
Methyl Dodecanoate	0.05 ± 0.02 ^b	0.01 ± 0.00 ^a	nd ^a	0.04 ± 0.02 ^b	nd ^a	0.04 ± 0.00 ^b	0.01 ± 0.00 ^a	0.05 ± 0.02 ^b	*	0.03 ^a	0.03 ^a	ns	0.03 ^b	0.03 ^b	0.01 ^a	0.05 ^c	*
Phenyl Acetate	1.14 ± 0.11 ^b	1.05 ± 0.24 ^b	nd ^a	2.30 ± 0.16 ^d	1.13 ± 0.11 ^b	1.71 ± 0.24 ^c	2.10 ± 0.24 ^d	2.14 ± 0.16 ^d	*	1.12 ^a	1.77 ^b	*	1.13 ^a	1.38 ^b	0.05 ^a	2.22 ^c	*
Ethyl Dodecanoate	0.10 ± 0.03 ^{bc}	0.06 ± 0.02 ^{ab}	0.55 ± 0.07 ^c	0.09 ± 0.03 ^a	0.04 ± 0.02 ^a	0.16 ± 0.02 ^d	0.07 ± 0.02 ^{ac}	0.12 ± 0.03 ^{cd}	*	0.20 ^b	0.16 ^a	*	0.07 ^a	0.11 ^a	0.31 ^b	0.10 ^a	*
Total Esters	10.00 ± 1.26^c	9.70 ± 0.53^{bc}	8.40 ± 0.19^b	11.06 ± 1.32^c	6.05 ± 0.05^a	12.82 ± 0.11^d	10.14 ± 1.03^c	10.84 ± 1.08^c	*	9.79^a	9.96^a	ns	8.02^a	11.26^c	9.27^b	10.95^c	*
ALCOHOLS																	
Propan-1-ol	0.41 ± 0.06 ^f	0.51 ± 0.12 ^f	0.26 ± 0.04 ^{cd}	0.22 ± 0.07 ^{bc}	0.17 ± 0.03 ^{ac}	0.33 ± 0.04 ^{de}	0.10 ± 0.02 ^a	0.12 ± 0.07 ^{ab}	*	0.35 ^b	0.18 ^a	*	0.29 ^b	0.42 ^c	0.18 ^a	0.17 ^a	*
2-Methylpropan-1-ol	0.55 ± 0.12 ^e	0.13 ± 0.02 ^a	0.24 ± 0.07 ^{bd}	0.11 ± 0.03 ^a	0.32 ± 0.07 ^d	0.14 ± 0.02 ^{ab}	0.28 ± 0.07 ^{cd}	0.18 ± 0.03 ^{ac}	*	0.26 ^a	0.23 ^a	ns	0.43 ^c	0.13 ^a	0.26 ^b	0.14 ^a	*
3-Methylbutan-1-ol	21.74 ± 5.24 ^b	21.80 ± 4.21 ^b	21.44 ± 4.21 ^b	21.02 ± 4.17 ^b	12.0 ± 3.00 ^a	22.40 ± 6.07 ^b	22.96 ± 5.14 ^b	22.56 ± 4.17 ^b	*	21.50 ^a	19.98 ^a	ns	16.87 ^a	22.10 ^a	22.20 ^a	21.79 ^a	ns
2-Phenylethanol	10.80 ± 2.17 ^a	16.70 ± 0.14 ^c	10.35 ± 2.17 ^a	10.83 ± 2.14 ^a	11.54 ± 2.51 ^{ab}	15.04 ± 3.04 ^{bc}	10.61 ± 2.07 ^a	9.57 ± 2.14 ^a	*	12.17 ^a	11.69 ^a	ns	11.77 ^a	15.87 ^b	10.48 ^a	10.20 ^a	*
Total Alcohols	33.50 ± 7.35^{ab}	39.14 ± 3.97^b	32.29 ± 2.01^{ab}	32.18 ± 1.99^{ab}	24.03 ± 5.55^a	37.91 ± 9.09^b	33.95 ± 3.16^{ab}	32.43 ± 6.41^{ab}	*	34.28^a	32.08^a	ns	28.76^a	38.52^b	33.12^{ab}	32.31^{ab}	*
TERPENES																	
7-Methyl-3-methylideneocta-1,6-diene (β-Myrcene)	0.50 ± 0.14 ^a	11.70 ± 5.14 ^b	nd ^a	nd ^a	0.11 ± 0.04 ^a	0.11 ± 0.02 ^a	0.17 ± 0.04 ^a	nd ^a	*	3.05 ^b	0.10 ^a	*	0.30 ^a	5.90 ^b	0.08 ^a	nd ^a	*
(4 R)-1-Methyl-4-prop-1-en-2-ylcyclohexene (d-Limonene)	nd ^a	0.14 ± 0.03 ^b	0.14 ± 0.05 ^b	nd ^a	0.30 ± 0.07 ^c	nd ^a	nd ^a	0.16 ± 0.03 ^b	*	0.07 ^a	0.11 ^b	*	0.15 ^b	0.07 ^a	0.07 ^a	0.08 ^a	*

(continued on next page)

Table 4 (continued)

	Interactive effects (Brewing Procedure × Yeast Strain)									Single effect of Brewing Procedure			Single effect of Yeast Strain				
	BP1-17,290	BP1-174,061	BP1-9502	BP1-9518	BP2-17,290	BP2-174,061	BP2-9502	BP2-9518	Sign.	BP1	BP2	Sign.	17,290	14,061	9502	9518	Sign.
(3Z)-3,7-Dimethylocta-1,3,6-triene (cis-β-Ocimene)	nd ^a	nd ^a	0.10 ± 0.02 ^b	0.20 ± 0.05 ^c	nd ^a	nd ^a	nd ^a	0.11 ± 0.05 ^b	*	0.07 ^b	0.03 ^a	*	nd ^a	nd ^a	0.05 ^b	0.15 ^c	*
2-[(2S,5S)-5-Ethenyl-5-methylxolan-2-yl]propan-2-ol (trans-Linalool Oxide)	0.46 ± 0.17 ^b	nd ^a	nd ^a	nd ^a	0.94 ± 0.17 ^c	0.10 ± 0.02 ^a	0.84 ± 0.17 ^c	nd ^a	*	0.11 ^a	0.47 ^b	*	0.70 ^c	0.05 ^a	0.42 ^b	nd ^a	*
(1R,4R)-1,7,7-Trimethylbicyclo[2.2.1]heptan-2-one (Camphor)	1.51 ± 0.35 ^{cd}	0.40 ± 0.07 ^a	0.29 ± 0.05 ^a	nd ^a	1.71 ± 0.35 ^{cd}	1.45 ± 0.34 ^c	1.94 ± 0.35 ^d	0.94 ± 0.21 ^b	*	0.55 ^a	1.51 ^b	*	1.61 ^c	0.93 ^b	1.11 ^b	0.47 ^a	*
3,7-Dimethylocta-1,6-dien-3-ol (Linalool)	nd ^a	1.63 ± 0.24 ^d	1.41 ± 0.17 ^d	2.95 ± 0.14 ^e	nd ^a	0.27 ± 0.05 ^b	0.80 ± 0.11 ^c	1.51 ± 0.14 ^d	*	1.50 ^b	0.64 ^a	*	nd ^a	0.95 ^b	1.10 ^b	2.23 ^c	*
4-Methyl-1-propan-2-ylcyclohex-3-en-1-ol (Terpinen-4-ol)	1.90 ± 0.51 ^c	0.17 ± 0.06 ^a	nd ^a	nd ^a	1.51 ± 0.51 ^{bc}	1.32 ± 0.07 ^b	1.64 ± 0.41 ^{bc}	nd ^a	*	0.52 ^a	1.12 ^b	*	1.70 ^c	0.74 ^b	0.82 ^b	nd ^a	*
2-(4-Methylcyclohex-3-en-1-yl)propan-2-ol (α-Terpineol)	1.43 ± 0.28 ^a	1.90 ± 0.35 ^d	1.45 ± 0.22 ^{ac}	1.75 ± 0.17 ^{bd}	1.26 ± 0.24 ^{ab}	1.13 ± 0.04 ^a	1.63 ± 0.37 ^{cd}	1.20 ± 0.17 ^{ab}	*	1.63 ^b	1.30 ^a	*	1.34 ^a	1.51 ^a	1.54 ^a	1.47 ^a	ns
3,7-Dimethyloct-6-en-1-ol (Citronellol)	0.20 ± 0.05 ^{ab}	1.48 ± 0.41 ^d	nd ^a	1.40 ± 0.17 ^c	0.24 ± 0.08 ^{ab}	0.40 ± 0.11 ^b	0.15 ± 0.04 ^{ab}	1.11 ± 0.17 ^c	*	0.77 ^b	0.47 ^a	*	0.22 ^a	0.94 ^b	0.07 ^a	1.25 ^c	*
(2E)-3,7-Dimethylocta-2,6-dien-1-ol (Geraniol)	nd ^a	0.17 ± 0.04 ^b	nd ^a	0.01 ± 0.00 ^a	nd ^a	nd ^a	nd ^a	nd ^a	*	0.04 ^b	nd ^a	*	nd ^a	0.08 ^b	nd ^a	nd ^a	*
Total Terpenes	6.00 ± 0.22^{ab}	17.59 ± 4.84^c	3.39 ± 0.03^a	6.31 ± 0.43^{ab}	5.96 ± 0.60^{ab}	4.78 ± 0.15^{ab}	7.17 ± 1.19^b	5.03 ± 0.49^{ab}	*	8.32^b	5.74^a	*	5.98^a	11.19^b	5.28^a	5.67^a	*
VOLATILE ACIDS																	
Hexanoic Acid	0.12 ± 0.08 ^a	0.10 ± 0.02 ^a	0.45 ± 0.11 ^{cd}	0.67 ± 0.17 ^e	0.16 ± 0.07 ^a	0.38 ± 0.07 ^b	0.62 ± 0.24 ^{de}	0.18 ± 0.06 ^{ab}	*	0.34 ^a	0.33 ^a	ns	0.14 ^a	0.24 ^a	0.53 ^b	0.43 ^b	*
Octanoic Acid	0.56 ± 0.21 ^d	0.45 ± 0.08 ^{cd}	0.12 ± 0.08 ^{ab}	0.36 ± 0.07 ^{cd}	0.03 ± 0.00 ^a	0.28 ± 0.07 ^{bc}	2.71 ± 0.21 ^e	0.28 ± 0.21 ^e	*	0.37 ^a	0.75 ^b	*	0.29 ^a	0.22 ^a	0.20 ^a	1.54 ^b	*
Nonanoic Acid	0.15 ± 0.05 ^a	1.70 ± 0.37 ^d	1.22 ± 0.17 ^{bc}	0.96 ± 0.25 ^b	1.49 ± 0.34 ^{cd}	0.32 ± 0.05 ^a	1.47 ± 0.34 ^{cd}	1.15 ± 0.14 ^{bc}	*	1.01 ^a	1.11 ^a	ns	0.82 ^a	1.01 ^a	1.34 ^b	1.05 ^{ab}	*
Decanoic Acid	0.15 ± 0.07 ^a	0.10 ± 0.04 ^a	0.36 ± 0.11 ^{ab}	1.14 ± 0.34 ^d	0.17 ± 0.07 ^a	0.60 ± 0.14 ^{bc}	0.84 ± 0.18 ^c	0.22 ± 0.06 ^a	*	0.44 ^a	0.46 ^a	ns	0.16 ^a	0.35 ^a	0.60 ^b	0.68 ^b	*
Total Volatile Acids	0.98 ± 0.41^a	2.35 ± 0.51^{bc}	2.15 ± 0.47^{bc}	3.13 ± 0.83^c	1.85 ± 0.48^b	1.30 ± 0.26^{ab}	3.21 ± 0.83^{cd}	4.26 ± 0.47^d	*	2.16^a	2.65^b	*	1.41^a	1.82^b	2.68^c	3.70^d	*
HYDROCARBONS																	
Ethenylbenzene (Styrene)	nd ^a	0.21 ± 0.06 ^{bc}	nd ^a	0.14 ± 0.03 ^{ab}	0.62 ± 0.20 ^f	0.55 ± 0.08 ^{ef}	0.30 ± 0.07 ^d	0.44 ± 0.10 ^e	*	0.09 ^a	0.48 ^b	*	0.31 ^b	0.38 ^b	0.15 ^a	0.29 ^b	*
Total Hydrocarbons	nd^a	0.21 ± 0.06^{bc}	nd^a	0.14 ± 0.03^{ab}	0.62 ± 0.20^f	0.55 ± 0.08^{ef}	0.30 ± 0.07^d	0.44 ± 0.10^e	*	0.09^a	0.48^b	*	0.31^b	0.38^c	0.15^a	0.29^b	*
VOLATILE PHENOLS																	
4-Ethenyl-2-methoxyphenol (4-Vinylguaiacol)	0.17 ± 0.06 ^a	0.11 ± 0.04 ^a	0.22 ± 0.08 ^a	0.16 ± 0.04 ^a	0.40 ± 0.20 ^b	0.77 ± 0.12 ^c	0.15 ± 0.04 ^a	0.43 ± 0.08 ^b	*	0.16 ^a	0.44 ^b	*	0.28 ^b	0.44 ^c	0.18 ^a	0.29 ^b	*
Total Volatile Phenols	0.17 ± 0.06^a	0.11 ± 0.04^a	0.22 ± 0.08^a	0.16 ± 0.04^a	0.40 ± 0.20^b	0.77 ± 0.12^c	0.15 ± 0.04^a	0.43 ± 0.08^b	*	0.16^a	0.44^b	*	0.28^b	0.44^c	0.18^a	0.29^b	*
NORISOPRENOIDS																	
(E)-1-(2,6,6-Trimethyl-1-cyclohexa-1,3-dienyl)but-2-en-1-one ((E)-β-Damascenone)	0.01 ± 0.00 ^a	0.01 ± 0.00 ^a	0.01 ± 0.00 ^a	0.02 ± 0.00 ^{ab}	0.09 ± 0.02 ^d	0.05 ± 0.02 ^c	0.01 ± 0.00 ^a	0.04 ± 0.01 ^{bc}	*	0.01 ^a	0.05 ^b	*	0.05 ^c	0.03 ^b	0.01 ^a	0.03 ^b	*
Total Norisoprenoids	0.01 ± 0.00^a	0.01 ± 0.00^a	0.01 ± 0.00^a	0.02 ± 0.00^{ab}	0.09 ± 0.02^d	0.05 ± 0.02^c	0.01 ± 0.00^a	0.04 ± 0.01^{bc}	*	0.01^a	0.05^b	*	0.05^c	0.03^b	0.01^a	0.03^b	*

isoamyl acetate, ethyl hexanoate, diethyl succinate, methyl dodecanoate, and of the terpenes linalool and geraniol. The Factor 2 allowed a better separation of BP1-14,061 type from other beers, thanks to their most positive loadings deriving from the lowest ethyl acetate content and the highest concentrations of phenylethanol and terpenes (mainly β -myrcene, citronellol, and geraniol). The beers subjected to BP1 treatment and fermented by *S. cerevisiae* 17,290, 9518, 9502 as well as those brewed according to BP2 and fermented by *S. cerevisiae* 9502, 9518, 14,061 clusterize in the region in which more esters, and 3-methyl-1-butanol prevail. Integrated considerations on the first two factors reveal the weight of the interactive effect between brewing procedure and yeast strain on the release of volatile molecules.

3.4. Odor activity value and aromatic series

The Odor Activity Value (OAV) is a metric used to assess the contribution level of a specific compound to the overall aroma of a substance, being the ratio between concentration and Odor Threshold (OT) of a specified volatile compound. Therefore, OAVs can get an idea of the most active odorants (Francis & Newton, 2005, Gómez García-Carpintero et al., 2011; Gómez-Miguez et al., 2007). Table 5 reports odor thresholds, sensory descriptors and OAVs of the volatile compounds identified in all samples. Twenty of 37 compounds identified had OAVs ≥ 1 , thus playing a crucial role in shaping the perceived odor characteristics. This information is valuable for understanding the specific compounds that contribute the most to the overall aroma profile of sample. In the present work, the major contributors to the odor perception were compounds generated during the alcoholic fermentation. These compounds included esters associated with fruity and floral notes, phenyl ethanol, short-chain acids, and 4-vinyl guaiacol, known to be important contributors to the aroma of young red wines, particularly those with limited varietal aromatic potential (Gomez-Miguez et al., 2007). Esters are associated with fruity and floral notes (Capone et al., 2013). In all beers except BP2-17,290, esters such as isoamyl acetate, ethyl hexanoate, ethyl octanoate, ethyl-9-decanoate, and phenylacetate were the major contributors, having OAV ranging from 1.2 to 178. As demonstrated in wines (Capone et al., 2013), esters are volatile molecules with low perception thresholds, making them highly sensorially impactful compounds that contribute to fruity notes.

In beer as well, the fruity aromatic series proves to be the series characterized by the greatest intensity (Sánchez-Palomo et al., 2010).

β -Damascenone is the volatile compound with the highest OAV in all samples, ranging from 200 to 1800. This molecule has been identified as a powerful odorant in Belgian commercial beers (Chevance et al., 2002) and wheat beers (Langos et al., 2013). The authors proposed an alternative origin for the norisoprenoid (E)- β -damascenone, suggesting it could stem from the degradation of the carotenoid neoxanthin. Additionally, according to Kollmannsberger et al., 2006 (E)- β -damascenone may also derive from the β -D-glucoside of 3-hydroxy- β -damascenone found in hops.

Among terpenes, linalool was a potent odorant in all samples (except beers fermented by *S. cerevisiae* 17,290), with OAV varying from 10.80 to 118.0. Our data confirm the idea that linalool is a key molecule in beer odor, contributing in particular to floral notes. Linalool is a terpene alcohol found in various plants, including hops and is known for its distinctive floral and citrus aroma (Chen et al., 2023). Citronellol is identified as the second most important sensorially terpene in all samples except in BP1-9502 (where it was not detected) with values ranging from 2.0 to 14.80. This compound, being derived from geraniol, contributes to the overall sensory characteristics, potentially adding floral and citrusy notes (Chen et al., 2023). The total intensities for every aromatic series were calculated as the sum of the OAVs of each compound assigned to this series (Capone et al., 2013; Sánchez-Palomo et al., 2010) to provide a visual representation of the cumulative impact of compounds within each aromatic series (Fig. 3). The obtained results suggested that the intensity patterns observed across beer aroma series

In line, different letters indicate significant differences at $p < 0.05$ by LSD multiple range test.

The asterisks indicate significant differences at $p < 0.05$ by LSD multiple range test.

ns: not significant.

nd: not detected.

(The IUPAC names are reported. The common names are shown in brackets).

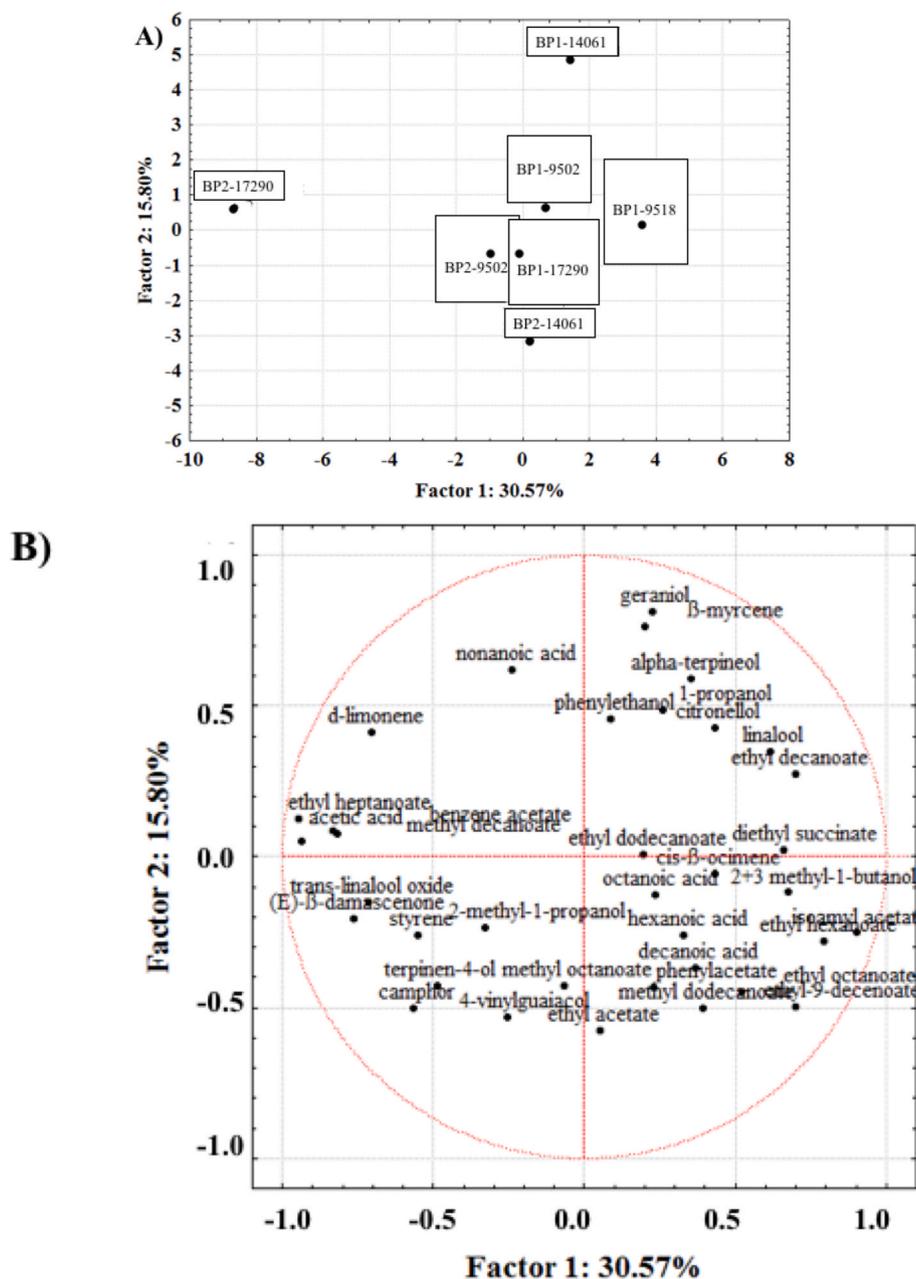


Fig. 2. PCA applied to the volatolomic profile: scores (A) and correlation circles (B).

indicate that the main flavor characteristics of the experimental beers consist of fruity, floral, fatty, woody, and herbaceous notes. Important differences were detected between the samples, particularly in terms of odor intensity of the dominant series. The dominant fruity and floral series, associated with chemical classes such as esters, terpenes, and phenylethanol, were clearly represented in beers produced according both brewing procedures, particularly if fermented with *S. cerevisiae* 174,061, 9518, and 9502. The freshness linked to acidic notes is more accentuated in BP2-9518 suggesting that interactions between strain and brewing processes contribute to a perception of freshness and acidity in the resulting beer. Clove and woody notes associated with 4-vinylguaiaicol have a higher intensity in BP2 beers fermented by *S. cerevisiae* 174,061, 17,290, and 9518. The herbaceous series shows great intensity in BP1 beers inoculated with *S. cerevisiae* 9518 and 9502, as well as in BP2-9518. Independently on brewing procedure, the beers fermented with *S. cerevisiae* 9518 showed more complex profiles due to the presence of different odor families with discrete olfactory intensities.

The above evidences indicated that the specific combinations of process and yeast strain produced a peculiar aroma profile. Understanding the mechanisms of aroma formation is crucial for brewers aimed to tailor novel beer able to meet consumer preferences. Many studies have underlined the role of Amadori products in enhancing the flavor (Beksan, et al., 2003; Yamamoto et al., 2012). Flavor is an important factor in attracting consumers and optimizing food quality, and the Maillard Reaction (MR) plays a crucial role in flavor development. However, MR products have a significant disadvantage due to their limited stability during heat treatment and storage. Amadori Rearrangement Products (ARPs), intermediates of the Maillard reaction that offer greater stability and a fresh flavor profile, are a promising alternative as flavor enhancers. Thanks to advances in analytical technologies, accurate characterization of ARPs is now possible, while improved preparation methods, including synthesis, separation and drying techniques, have increased the yield of ARPs by up to 95%. In reality, the stability of ARPs depends on various factors, such as chemical composition, pH levels,

Table 5
Odor Thresholds, sensory descriptors and Odor Activity Values (OAVs) of volatiles compounds identified in beers.

	Odor Threshold (mg/L)	Odor Descriptors	BP1-17,290	BP1-174,061	BP1-9502	BP1-9518	BP2-17,290	BP2-174,061	BP2-9502	BP2-9518
ESTERS										
Ethyl Acetate	12.26	Pineapple	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
3-Methylbutyl Acetate (Isoamyl Acetate)	0.030	Fruity	54.05	44.06	37.30	53.71	<0.1	51.75	47.75	50.00
Ethyl Hexanoate	0.050	Apple, banana, wine-like	28.00	28.00	27.06	24.45		36.27	33.28	30.06
Ethyl Heptanoate	0.22	Apricot, cherry, raspberry		<0.1		<0.1	7.00	<0.1	<0.1	<0.1
Methyl Octanoate	not found									
Ethyl Octanoate	0.02	Banana, floral, pear, pineapple, wine-like	72.00	95.00	60.50	84.00	20.00	178.00	91.00	124.00
Methyl Decanoate	not found									
Ethyl Decanoate	0.20	Floral	14.75	15.70	13.10	14.80	5.85	12.75	9.70	8.65
Diethyl Butanedioate (Diethyl Succinate)	200.00	Apple; apricot; chocolate;	<0.1	<0.1	<0.1	<0.1		<0.1	<0.1	<0.1
Ethyl dec-9-enoate	0.10	Fruity	2.50	1.30	1.70	3.00	0.40	3.00	1.20	2.30
Benzyl Acetate	not found									
Methyl Dodecanoate	not found									
Phenylacetate	0.25	Floral	4.56	4.20		9.20	4.52	6.84	8.40	8.56
Ethyl Dodecanoate	0.50	Fruity/floral	0.2	0.12	1.10	0.18	<0.1	0.32	0.14	0.24
ALCOHOLS										
Propan-1-ol	306.00	Alcohol, ripe fruit	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
2-Methylpropan-1-ol	40.00	Alcohol, solvent	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
3-Methylbutan-1-ol	30.00									
2-Phenylethanol	10.00	Floral	1.08	1.67	1.04	1.08	1.15	1.50	1.06	0.96
TERPENES										
7-Methyl-3-methylideneocta-1,6-diene (β -Mircene)	0.10	green mango, fresh green grass-like	5.00	117.00			1.10	1.10	1.70	
(4R)-1-Methyl-4-prop-1-en-2-ylcyclohexene (d-Limonene)	0.20	Lemon, orange, citrus, sweet		0.70	0.70		1.00			0.53
(3Z)-3,7-Dimethylocta-1,3,6-triene (<i>cis</i> - β -Ocimene)	0.034	herbaceous			2.94	5.88				3.24
2-[(2S,5S)-5-Ethenyl-5-methylxolan-2-yl]propan-2-ol (trans-Linalool Oxide)	3.00	Floral, green	0.15				0.31	0.03	0.28	
(1R,4R)-1,7,7-Trimethylbicyclo[2.2.1]heptan-2-one (Camphor)	not found									
3,7-Dimethylocta-1,6-dien-3-ol (Linalool)	0.025	refreshing floral, lemon, woody		65.20	56.40	118.00		10.80	32.00	60.40
4-Methyl-1-propan-2-ylcyclohex-3-en-1-ol (Terpinen-4-ol)	1.20	Floral	1.58	0.14	nd	nd	1.26	1.10	1.37	nd
2-(4-Methylcyclohex-3-en-1-yl)propan-2-ol (α -Terpineol)	5.00	Floral	0.29	0.38	0.29	0.35	0.25	0.23	0.33	0.24
3,7-Dimethyloct-6-en-1-ol (Citronellol)	0.10	Floral	2.00	14.80	nd	14.00	2.40	4.00	1.50	11.10
(2E)-3,7-Dimethylocta-2,6-dien-1-ol (Geraniol)	0.03	Apple, apricot, berry, rose	nd	5.67	nd	0.33	nd	nd	nd	nd
VOLATILE ACIDS										
Hexanoic Acid	0.42	Cheese, fatty, sour	0.29	0.24	1.07	1.59	0.38	0.90	1.48	0.43
Octanoic Acid	0.50	Fatty acid, dry, dairy	1.12	0.90	0.24	0.72	0.06	0.00	0.56	5.42
Nonanoic Acid	not found									
Decanoic Acid	1.40	Fatty acid, dry, woody	0.11	0.07	0.26	0.81	0.12	0.43	0.60	0.16
HYDROCARBONS										
Ethylbenzene (Styrene)	not found									
VOLATILE PHENOLS										
4-Ethenyl-2-methoxyphenol (4-Vinylguaiaicol)	0.04	clove, curry	4.25	2.75	5.50	4.00	10.00	19.25	3.75	10.75
NORISOPRENOIDS										
(E)-1-(2,6,6-Trimethyl-1-cyclohexa-1,3-dienyl)but-2-en-1-one ((E)- β -Damascenone)	0.00005	Apple, herbaceous, woody	200.00	200.00	200.00	400.00	1800.00	1000.00	200.00	800.00

References for odor threshold and sensory descriptors: Baiano et al. (2017, 2023); Capone et al., 2013; Gómez García-Carpintero et al., 2012; Tamura et al., 2001; Katarfina et al., 2014. OAVs >1 are reported in bold character.

(The IUPAC names are reported. The common names are shown in brackets).

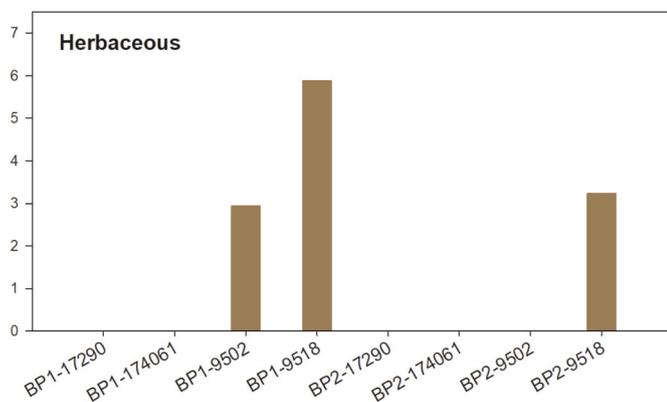


Fig. 3. Sum of the OAVs of the compounds belonging to each aromatic series.

temperature, water content and food matrix. Furthermore, there have not yet been in-depth studies on the toxicity and stability of ARPs, capable of using them (Luo et al., 2021). For this reason it is important to act on the technological and fermentation process and on the quality of the raw materials to produce beers of good acceptability.

3.5. Sensory characteristics of the finished beers

As can be inferred from Table 6, all the produced beers were characterized by: a white foam; a straw to golden yellow color; intermediate scores for the overall flavor intensity, the aromatic herb flavor, and the alcohol perception. Contrary to what was found through the instrumental measurement of color and alcohol content, their sensorial evaluation did not highlight significant single and interactive influences of brewing procedures and yeast strain, to indicate that the sensorial evaluation of these parameters was less sensitive.

The brewing procedure significantly affected amount and persistence of foam (higher in BP2 beers due to lower proteolysis induced by the brief protein rest), sweetness (higher in BP2 beers regardless of their lower residual sugar content, $R = -0.54$), saltiness and sourness (higher in BP1 beers, consistently with their lower pH and higher concentrations of CO_2 and acetic acid; $R = -0.75$, $R = 0.55$, and $R = 0.61$, respectively). However, our beers did not develop a substantial amount of foam because, according to Bravi et al. (2017), the ratio between unsaturated and saturated free fatty acids typical of the unmalted emmer has adverse effects on its ability to produce a stable foam.

Yeast strain strongly influenced most sensory attributes. The beers fermented by *S. cerevisiae* 17,290 obtained the lowest scores for: amount and persistence of foam; olfactory finesse; malty, hoppy, floral, and fruity intensity; sweetness; effervescence; and body/fullness. The same beers showed an excessive yeast flavor intensity and the highest score for sourness. *S. cerevisiae* 9502 imparted the best sensorial characteristics in terms of: greater amount and persistence of foam; higher malty, hoppy, floral, and fruity intensity together with a reduced yeast flavor intensity; greater and longer effervescence; greater body/fullness. Bitterness was increased by both 17,290 and 9502 strains. These effects of yeasts on the beer sensory properties are related to their different ability to synthesize volatile compounds as well as fatty acids during fermentation (Bravi et al., 2017). *S. cerevisiae* 17,290 synthesized less saturated fatty acids than the other strains (Table 5), thus explaining its adverse effect on foam characteristics. At the same way, the beers fermented by that strain contained the lowest amounts of the volatile compounds responsible of floral and fruity flavor (esters and alcohols), the highest amounts of organic acids that increased sourness, and the lowest amount of glycerol that explained their limited fullness.

A previous study of Baiano et al. (2023) highlighted that the beer made with unmalted emmer had low overall sensory quality scores (2.5 on average) than the beers made with unmalted common and durum

wheat (>3.0). However, those results were strongly affected by the brewing procedure applied, which was similar to our BP1. Accordingly, BP1 beers had the lowest overall scores, especially those fermented by *S. cerevisiae* 17,290, the only one having scores lower than 3.00. In fact, long protein rest adversely influence the beer sensory quality (Cela et al., 2023). Conversely, the highest overall sensory score was assigned to BP2-9502 beers. According to the Pearson correlation coefficients, the overall sensory quality was positively correlated with physico-chemical characteristics such as pH ($R = 0.65$), EBC color (0.55), concentration of syringic acid (0.55) and sensory attributes such as amount (0.59) and persistence (0.57) of foam, olfactory finesse (0.73), and body (0.81). Instead, it was inversely correlate with titratable acidity (-0.56), volatile acidity (-0.69), CO_2 content (-0.67), sinapic content (-0.56), aromatic herb flavor (-0.58), saltiness (-0.59), and sourness (-0.63).

4. Conclusions

This research was aimed to optimized formulation and processing of unmalted emmer-based beers in order to contribute to scientific knowledge on the phenomena that generally affect the quality of this type of beer. It was found that both brewing procedures and yeast strains, alone and in combination, strongly affected the physical and chemical characteristics of the emmer-based beers while their sensory properties were mainly influenced by the combination of the two factors, with yeast strain having the greater weight.

The research highlights unambiguous indications regarding the brewing procedure, with BP2 allowing to obtain emmer-based beers of better overall (physical, chemical, and sensory) quality. This means that the best brewing procedures must include the use of water with a low conductivity, a brief protein rest, and no more than an hour of wort boiling. Instead, the choice of the fermentation agent requires a more in-depth evaluation. The yeasts that allowed to maximize the content of antioxidant substances – i.e. the strains isolated from Negroamaro grape must – negatively influenced the overall sensorial quality, as they produced too low (17,290) or too high (14,061) concentrations of volatile compounds which in neither case corresponded to desirable characteristics of olfactory intensity and finesse. For this reason, the yeast choice fell on *S. cerevisiae* 9502, which imparted the best overall sensory quality together with intermediate phenolic concentrations. This finding also confirms suitability of crossover fermentation in the development of high quality emmer-based white beers.

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CRedit authorship contribution statement

Maria Tufariello: Writing – review & editing, Writing – original draft, Validation, Methodology, Investigation, Formal analysis, Data curation. **Francesco Grieco:** Funding acquisition, Investigation, Methodology, Resources, Validation, Writing – review & editing. **Anna Fiore:** Formal analysis, Investigation, Methodology, Validation. **Carmela Gerardi:** Formal analysis, Investigation, Methodology, Validation. **Vittorio Capozzi:** Methodology, Validation, Writing – review & editing. **Antonietta Baiano:** Conceptualization, Data curation, Formal analysis, Funding acquisition, Investigation, Methodology, Project administration, Resources, Supervision, Validation, Visualization, Writing – original draft, Writing – review & editing.

Declaration of competing interest

The authors declare that they have no known competing financial

Table 6
Influence of brewing procedures and yeast strains on the beer sensory profiles.

Beer acronyms	Color		Foam		Turbidity	Flavor characteristics								Gustatory characteristics				Tactile characteristics			Overall Quality	
	Foam	Liquid	Amount	Persistence		OFI	OF	Malty	Hoppy	Floral	Fruity	Spicy	Yeast	Aromatic herbs	Sweetness	Bitterness	Saltiness	Sourness	Alcoholic	Effervescence		Body/Fullness
<i>Interactive effects (Brewing Procedure × Yeast Strain)</i>																						
BP1-17,290	1.00 ± 0.00 ^a	2.50 ± 0.58 ^a	2.00 ± 0.00 ^a	2.00 ± 0.00 ^a	1.75 ± 0.50 ^a	3.00 ± 0.00 ^a	3.00 ± 0.00 ^a	2.50 ± 0.58 ^a	3.25 ± 0.50 ^{ab}	2.00 ± 0.00 ^a	2.00 ± 0.00 ^a	2.50 ± 0.58 ^a	3.25 ± 0.50 ^b	1.75 ± 0.50 ^a	1.50 ± 0.58 ^a	3.75 ± 0.50 ^c	3.25 ± 0.50 ^b	3.50 ± 0.58 ^{bc}	3.00 ± 0.00 ^a	2.50 ± 0.58 ^a	2.25 ± 0.50 ^a	2.50 ± 0.10 ^a
BP1-14,061	1.00 ± 0.00 ^a	2.25 ± 0.50 ^a	2.00 ± 0.00 ^a	2.00 ± 0.00 ^a	2.75 ± 0.96 ^b	3.00 ± 0.82 ^a	3.75 ± 0.96 ^{ab}	2.75 ± 0.96 ^{ab}	2.50 ± 0.58 ^a	3.00 ± 0.00 ^{bc}	2.50 ± 0.58 ^{ac}	2.50 ± 1.00 ^a	3.00 ± 0.00 ^{ab}	1.75 ± 0.50 ^a	2.25 ± 0.50 ^{bd}	2.75 ± 0.50 ^a	2.75 ± 0.50 ^{ab}	3.25 ± 0.50 ^{bc}	3.25 ± 0.50 ^a	3.25 ± 0.50 ^b	3.00 ± 0.00 ^{bc}	3.50 ± 0.58 ^b
BP1-9502	1.00 ± 0.00 ^a	2.75 ± 0.50 ^a	2.00 ± 0.00 ^a	2.00 ± 0.00 ^a	2.00 ± 0.00 ^a	3.25 ± 0.50 ^a	4.50 ± 0.58 ^b	3.25 ± 0.50 ^b	3.75 ± 0.50 ^b	3.50 ± 0.58 ^c	3.25 ± 0.50 ^c	2.50 ± 0.58 ^a	2.50 ± 0.58 ^a	1.75 ± 0.50 ^a	1.75 ± 0.50 ^{ab}	3.50 ± 0.58 ^{bc}	2.75 ± 0.50 ^{ab}	3.25 ± 0.50 ^{bc}	3.00 ± 0.00 ^a	3.25 ± 0.50 ^b	2.75 ± 0.50 ^{ac}	3.75 ± 0.50 ^b
BP1-9518	1.00 ± 0.00 ^a	2.25 ± 0.50 ^a	2.00 ± 0.00 ^a	2.00 ± 0.00 ^a	2.50 ± 1.29 ^b	3.25 ± 0.50 ^a	3.75 ± 0.96 ^{ab}	2.75 ± 0.50 ^{ab}	2.75 ± 0.50 ^a	2.25 ± 0.96 ^{ac}	2.75 ± 0.50 ^a	2.25 ± 0.50 ^{ab}	3.00 ± 0.00 ^{ab}	1.75 ± 0.50 ^a	2.25 ± 0.50 ^{bd}	3.00 ± 0.00 ^{ab}	3.25 ± 0.50 ^b	3.25 ± 0.50 ^{bc}	3.00 ± 0.00 ^a	3.00 ± 0.00 ^{ab}	2.50 ± 0.58 ^{ab}	3.00 ± 0.82 ^{ab}
BP2-17,290	1.00 ± 0.00 ^a	2.50 ± 0.58 ^a	2.00 ± 0.00 ^a	2.00 ± 0.00 ^a	1.75 ± 0.50 ^a	3.00 ± 0.82 ^a	3.50 ± 0.58 ^{ab}	2.50 ± 0.58 ^a	2.50 ± 0.58 ^a	2.00 ± 0.00 ^a	2.25 ± 0.50 ^{ab}	2.25 ± 0.50 ^a	3.25 ± 0.50 ^b	1.75 ± 0.50 ^a	2.00 ± 0.00 ^{bc}	3.75 ± 0.50 ^c	2.75 ± 0.50 ^{ab}	3.75 ± 0.50 ^b	3.50 ± 0.58 ^a	2.50 ± 0.58 ^a	2.50 ± 0.58 ^{ab}	2.50 ± 0.86 ^a
BP2-14,061	1.00 ± 0.00 ^a	2.50 ± 0.58 ^a	2.50 ± 0.58 ^b	2.00 ± 0.00 ^a	2.75 ± 10.96 ^b	3.50 ± 0.58 ^a	4.50 ± 0.58 ^b	3.25 ± 0.50 ^{ab}	3.25 ± 0.58 ^{ab}	2.50 ± 0.58 ^{ab}	2.75 ± 0.50 ^{ac}	2.50 ± 0.58 ^a	3.00 ± 0.00 ^{ab}	1.75 ± 0.50 ^a	2.75 ± 0.50 ^d	2.50 ± 0.58 ^a	2.75 ± 0.50 ^{ab}	3.00 ± 0.58 ^{ab}	3.00 ± 0.00 ^a	3.25 ± 0.50 ^b	3.25 ± 0.50 ^c	3.50 ± 0.58 ^b
BP2-9502	1.00 ± 0.00 ^a	2.50 ± 0.58 ^a	4.75 ± 0.50 ^c	4.75 ± 0.50 ^c	2.00 ± 0.00 ^a	3.75 ± 0.50 ^a	4.50 ± 0.58 ^b	3.25 ± 0.50 ^b	3.75 ± 0.50 ^b	3.25 ± 0.50 ^c	2.25 ± 0.50 ^{ab}	2.75 ± 0.50 ^a	3.25 ± 0.50 ^b	1.50 ± 0.58 ^a	2.50 ± 0.58 ^{cd}	3.50 ± 0.58 ^{bc}	2.75 ± 0.50 ^{ab}	2.50 ± 0.00 ^a	3.25 ± 0.50 ^a	3.50 ± 0.58 ^b	3.25 ± 0.50 ^c	4.50 ± 0.58 ^c
BP2-9518	1.00 ± 0.00 ^a	2.75 ± 0.50 ^a	2.75 ± 0.50 ^b	2.75 ± 0.50 ^b	2.00 ± 0.82 ^a	3.00 ± 0.82 ^a	3.75 ± 0.82 ^a	3.00 ± 0.82 ^a	2.75 ± 0.82 ^a	2.25 ± 0.82 ^a	3.00 ± 0.82 ^a	3.00 ± 0.82 ^a	3.25 ± 0.82 ^a	1.75 ± 0.50 ^a	2.50 ± 0.58 ^{cd}	2.75 ± 0.50 ^a	2.50 ± 0.58 ^a	3.00 ± 0.00 ^{ab}	3.50 ± 0.58 ^a	3.25 ± 0.50 ^b	3.00 ± 0.00 ^{bc}	3.75 ± 0.50 ^b
<i>Significance</i>	ns	ns	*	*	*	ns	*	*	*	*	*	ns	*	ns	*	*	*	*	ns	*	*	*
<i>Single effect of Brewing Procedure</i>																						
BP1	1.00 ^a	2.44 ^a	2.00 ^a	2.00 ^a	2.25 ^a	3.13 ^a	3.75 ^a	2.81 ^a	3.06 ^a	2.69 ^a	2.63 ^a	2.44 ^a	2.94 ^a	1.75 ^a	1.94 ^a	3.25 ^a	3.00 ^b	3.31 ^b	3.06 ^a	3.00 ^a	2.63 ^a	3.19 ^a
BP2	1.00 ^a	2.56 ^a	3.00 ^b	2.88 ^b	2.13 ^a	3.31 ^a	4.06 ^a	3.00 ^a	3.06 ^a	2.50 ^a	2.56 ^a	2.63 ^a	3.19 ^a	1.69 ^a	2.44 ^b	3.13 ^a	2.69 ^a	3.06 ^a	3.31 ^a	3.13 ^a	3.00 ^a	3.56 ^b
<i>Significance</i>	ns	ns	*	*	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	*	ns	*	*	ns	ns	ns	*
<i>Single effect of Yeast Strain</i>																						
17,290	1.00 ^a	2.50 ^a	2.00 ^a	2.00 ^a	1.75 ^a	3.00 ^a	3.25 ^a	2.50 ^a	2.88 ^a	2.00 ^a	2.13 ^a	2.38 ^a	3.25 ^b	1.75 ^a	1.75 ^a	3.75 ^b	3.00 ^a	3.63 ^b	3.25 ^a	2.50 ^a	2.38 ^a	2.50 ^a
14,061	1.00 ^a	2.38 ^a	2.25 ^{ab}	2.00 ^a	2.75 ^b	3.25 ^a	4.13 ^{bc}	3.00 ^{ab}	2.8 ^a	2.75 ^b	2.63 ^{ab}	2.50 ^a	3.00 ^{ab}	1.75 ^a	2.50 ^b	2.63 ^a	2.75 ^a	3.13 ^a	3.13 ^a	3.25 ^b	3.13 ^b	3.5 ^b
9502	1.00 ^a	2.63 ^a	3.38 ^c	3.38 ^c	2.00 ^{ab}	3.50 ^a	4.50 ^c	3.25 ^b	3.75 ^b	3.38 ^c	2.75 ^b	2.63 ^a	2.88 ^a	1.63 ^a	2.13 ^{ab}	3.50 ^b	2.75 ^a	2.88 ^a	3.13 ^a	3.38 ^b	3.00 ^b	4.13 ^c
9518	1.00 ^a	2.50 ^a	2.38 ^b	2.38 ^b	2.25 ^{ab}	3.13 ^a	3.75 ^{ab}	2.88 ^{ab}	2.75 ^a	2.25 ^a	2.88 ^b	2.63 ^a	3.13 ^{ab}	1.75 ^a	2.38 ^b	2.88 ^a	2.88 ^a	3.13 ^a	3.25 ^a	3.13 ^b	2.75 ^{ab}	3.38 ^{ab}
<i>Significance</i>	ns	ns	*	*	*	ns	*	*	*	*	*	ns	*	ns	*	*	ns	*	ns	*	*	*

In column, different letters indicate significant differences at $p < 0.05$ by LSD multiple range test. The asterisks indicate significant differences at $p < 0.05$ by LSD multiple range test. ns: not significant OFI: Overall flavor intensity. OF: Olfactory finesse.

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 data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.lwt.2024.116044>.

References

- AACC. (2012). American association of cereal chemists. Official Methods: 44-15.02 Moisture—Air-Oven Methods; 08-01.01 Ash—Basic Method; 46-30.01 Crude Protein—Combustion Method. In *AACC international approved methods analysis* (11th ed.) St. Paul, MN, USA.
- Alfeo, V., De Francesco, G., Sileoni, V., Blangiforti, S., Palmeri, R., Aerts, G., Perretti, G., & Todaro, A. (2021). Physicochemical properties, sugar profile, and non-starch polysaccharides characterization of old wheat malt landraces. *Journal of Food Composition and Analysis*, *102*, Article 103997.
- Aliakbarian, B., Casazza, A. A., & Perego, P. (2011). Valorization of olive oil solid waste using high pressure–high temperature reactor. *Food Chemistry*, *128*, 704–710.
- Anitha, S., Krishnan, S., Senthilkumar, K., & Sasirekha, V. (2020). Theoretical investigation on the structure and antioxidant activity of (+) catechin and (–) epicatechin – a comparative study. *Molecular Physics*, *118*, 17. <https://doi.org/10.1080/00268976.2020.1745917>
- Aslankooi, E., Rezaei, M. N., Vervoort, Y., Courtin, C. M., & Verstrepen, K. J. (2015). Glycerol production by fermenting yeast cells is essential for optimal bread dough fermentation. *PLoS One*, *10*, Article e0119364.
- Baiano, A., Fiore, A., la Gatta, B., Capozzi, V., De Simone, N., Gerardi, C., & Grieco, F. (2024). Unmalted cereals, oenological yeasts, and in-bottle sugar addition as synergic strategies to enhance the quality of craft beers. *Beverages*, *10*, 8. <https://doi.org/10.3390/beverages10010008>
- Baiano, A., Fiore, A., la Gatta, B., Tufariello, M., Gerardi, C., & Grieco, F. (2023). Single and interactive effects of unmalted cereals, hops, and yeasts on quality of white-inspired craft beers. *Beverages*, *9*, 9.
- Baiano, A., Mentana, A., Quinto, M., Centonze, D., Previtali, M. A., Varva, G., ... Del Nobile, M. A., & De Palma, L. (2017). Volatile composition and sensory profile of wines obtained from partially defoliated vines: The case of Nero di Troia wine. *European Food Research and Technology*, *243*, 247–261.
- Baillière, J., Laureys, D., Vermeir, P., Van Opstaele, F., De Rouck, G., De Cooman, L., Vanderputten, D., & De Clippeler, J. (2022). 10 unmalted alternative cereals and pseudocereals: A comparative analysis of their characteristics relevant to the brewing process. *Journal of Cereal Science*, *106*, Article 103482.
- Beksan, E., Schieberle, P., Robert, F., Blank, I., Fay, L. B., Schlichtherle-Cerny, H., & Hofmann, T. (2003). Synthesis and sensory characterization of novel umami-tasting glutamate glycoconjugates. *Journal of Agricultural and Food Chemistry*, *51*(18), 5428–5436.
- Belcar, J., Buczek, J., Kapusta, I., & Gorzelany, J. (2022). Quality and pro-healthy properties of belgian witbier-style beers relative to the cultivar of winter wheat and raw materials used. *Foods*, *11*(8), 1150. <https://doi.org/10.3390/foods11081150>
- Brand-Williams, W., Cuvelier, M. E., & Berset, C. (1995). Use of a free radical method to evaluate antioxidant activity. *LWT—Food Science and Technology*, *28*, 25–30.
- Bravi, E., Marconi, O., Sileoni, V., & Perretti, G. (2017). Determination of free fatty acids in beer. *Food Chemistry*, *215*, 341–346. <https://doi.org/10.1016/j.foodchem.2016.07.153>
- Cadenas, R., Caballero, I., Nimubona, D., & Blanco, C. A. (2021). Brewing with starchy adjuncts: Its influence on the sensory and nutritional properties of beer. *Foods*, *10*, 1726. <https://doi.org/10.3390/foods10081726>
- Cai, S., Han, Z., Huang, Y., Chen, Z. H., Zhang, G., & Dai, F. (2015). Genetic diversity of individual phenolic acids in barley and their correlation with barley malt quality. *Journal of Agricultural and Food Chemistry*, *63*(31), 7051–7057. <https://doi.org/10.1021/acs.jafc.5b02960>
- Capone, S., Tufariello, M., & Siciliano, P. (2013). Analytical characterisation of Negroamaro red wines by “aroma wheels”. *Food Chemistry*, *141*, 2906–2915.
- Cardoso Viana, A., Colombo Pimentel, T., Borges do Vale, R., Santos Clementino, L., Thayná, E., Ferreira, J., Magnani, M., & dos Santos Lima, M. (2021). American pale Ale craft beer: Influence of brewer’s yeast strains on the chemical composition and antioxidant capacity. *LWT—Food Science and Technology*, *152*, Article 112317.
- Cela, N., Condelli, N., Perretti, G., Di Cairano, M., Tolve, R., & Galgano, F. (2023). Gluten reduction in beer: Effect of sorghum:quinoa ratio and protein rest time on brewing parameters and consumer acceptability. *Journal of Cereal Science*, *109*, Article 103607. <https://doi.org/10.1016/j.jcs.2022.103607>
- Chen, H., Shi, Y., Wang, L., Hu, X., & Lin, X. (2023). Phenolic profile and α -glucosidase inhibitory potential of wampee (*Clausena lansium* (Lour.) Skeels) peel and pulp: In vitro digestion/in silico evaluations. *Food Research International*, *173*, Article 113274.
- Chevance, F., Guyot-Declerck, C., Dupont, J., & Collin, S. (2002). Investigation of the β -damascenone level in fresh and aged commercial beers. *Journal of Agricultural and Food Chemistry*, *50*, 3818–3821.
- CNR-ISPA. (2023). ITEM agro-food microbial culture collection. Online reference included in article URL <http://www.ispacnr.it/collezioni-microbiche>. (Accessed 22 March 2024).
- Coelho, E. M., da Silva Padilha, C. V., Miskinis, G. A., de Sá, A. G. B., Pereira, G. E., de Azevedo, L. C., & dos Santos Lima, M. (2018). Simultaneous analysis of sugars and organic acids in wine and grape juices by HPLC: Method validation and characterization of products from northeast Brazil. *Journal of Food Composition and Analysis*, *66*, 160–167.
- Coote, N., & Kirsop, B. H. (1974). The content of some organic acids in bee and other fermented media. *Journal of the Institute of Brewing*, *80*, 474–483.
- De Flaviis, R., Santarelli, V., Mutarutwa, D., Giuliani, M., & Sacchetti, G. (2022). Volatiles profile of ‘blanche’ wheat craft beer as affected by wheat origin: A chemometric study. *Food Chemistry*, *385*, Article 132696.
- Deme, G. D., Asfaw, B. T., & Gari, M. T. (2019). Evaluation of malting potential of different barley varieties. *Journal of Water Pollution & Purification Research*, *6*(3), 24–35.
- Dresel, M., Praet, T., Van Opstaele, F., Van Holle, A., Naudts, D., De Keukeleire, D., Cooman, L. De, & Aerts, G. (2015). Comparison of the analytical profiles of volatiles in single-hopped worts and beers as a function of the hop variety. *Brewing Science*, *68* (1–2), 8–28.
- European Brewery Convention. (1975). Method 9.6—Colour/Colour of beer: Spectrophotometric method. In *Analytica-EBC; schweizer brauerei-rundschau: Zürich, Switzerland*.
- Fantozzi, P., Montanari, L., Mancini, F., Gasbarrini, A., Addolorato, G., Simoncini, M., Nardini, M., Ghiselli, A., & Scaccini, C. (1998). In vitro antioxidant capacity from wort to beer. *Lebensmittel-Wissenschaft & Technologie*, *31*, 221–227.
- Fogarasi, A. L., Kun, S., Tankó, G., Stefanovits-Bányai, E., & Hegyesné-Vecseri, B. (2015). A comparative assessment of antioxidant properties, total phenolic content of einkorn, wheat, barley and their malts. *Food Chemistry*, *167*, 1–6.
- Francis, I. L., & Newton, J. L. (2005). Determining wine aroma from compositional data. *Australian Journal of Grape and Wine Research*, *11*, 114–126.
- Fritsch, H. T., & Schieberle, P. (2005). Identification based on quantitative measurements and aroma recombination of the character impact odorants in a Bavarian Pilsner-type beer. *Journal of Agricultural and Food Chemistry*, *53*(19), 7544–7551.
- Gandolpho, B. C. G., Almeida, A. R., Gandolpho, G. M., Freitas, D. Z., Gasparini, O. C., Machado, M. H., & Barreto, P. L. M. (2021). Optimization of brewing waste’s (trub) phenolic compounds extraction by ultrasound assisted using response surface methodology. *Química Nova*, *44*, 478–483.
- García-Carpintero, E. G., Sánchez-Palomo, E., Gallego, M. G., & González-Viñas, M. A. (2011). Volatile and sensory characterization of red wines from cv. Moravia Agria minority grape variety cultivated in La Mancha region over five consecutive vintages. *Food Research International*, *44*, 1549–1560.
- García-Carpintero, E. G., Sánchez-Palomo, E., Gómez Gallego, M. A., & González-Viñas, M. A. (2012). Characterization of impact odorants and sensory profile of Bobal red wines from Spain’s La Mancha region. *Flavour and Fragrance Journal*, *27*, 60–68.
- Gómez-Míguez, M. J., Cacho, J. F., Ferreira, V., Vicario, I. M., & Heredia, F. J. (2007). Volatile components of Zalema white wines. *Food Chemistry*, *100*, 1464–1473.
- Gugino, I. M., Alfeo, V., Ashkezary, M. R., Marconi, O., Pirrone, A., Francesca, N., ... Todaro, A. (2024). Maiorca wheat malt: A comprehensive analysis of physicochemical properties, volatile compounds, and sensory evaluation in brewing process and final product quality. *Food Chemistry*, *435*, Article 137517.
- Hiralal, L., Olaniran, A. O., & Pillay, B. (2014). Aroma-active ester profile of ale beer produced under different fermentation and nutritional conditions. *Journal of Bioscience and Bioengineering*, *117*(1), 57–64.
- Hirst, M. B., & Richter, C. L. (2016). Review of aroma formation through metabolic pathways of *Saccharomyces cerevisiae* in beverage fermentations. *American Journal of Enology and Viticulture*, *67*, 361–370.
- Iorizzo, M., Letizia, F., Albanese, G., Coppola, F., Gambuti, A., Testa, B., Aversano, R., Forino, M., & Coppola, R. (2021). Potential for lager beer production from *saccharomyces cerevisiae* strains isolated from the vineyard environment. *Processes*, *9*, 1628. <https://doi.org/10.3390/pr9091628>
- Katarína, F., Katarína, M., Katarína, D., Ivan, Š., & Fedor, M. (2014). Influence of yeast strain on aromatic profile of Gewürztraminer wine. *LWT—Food Science and Technology*, *59*, 256–262.
- Kollmannsberger, H., Biendl, M., & Nitz, S. (2006). Occurrence of glycosidically bound flavour compounds in hops, hop products and beer. *Monatsschrift für Brauwissenschaft*, *5*(6), 83–89.
- Langos, D., Granvogel, M., & Schieberle, P. (2013). Characterization of the key aroma compounds in two Bavarian wheat beers by means of the sensomics approach. *Journal of Agricultural and Food Chemistry*, *61*, 11303–11311.
- Law 1354. (1962). *Disciplina igienica della produzione e del commercio della birra* (p. 234). Gazzetta Ufficiale della Repubblica Italiana.
- Leitao, C., Marchioni, E., Bergaentzle, M., Zhao, M., Didierjean, L., Taidi, B., & Ennahar, S. (2012). Effects of processing steps on the phenolic content and antioxidant activity of beer. *Journal of Agricultural and Food Chemistry*, *59*, 1249–1255.
- Li, G., & Liu, F. (2015). Changes in organic acids during beer fermentation. *Journal of the American Society of Brewing Chemists*, *73*(3), 275–279. <https://doi.org/10.1094/ASBCJ-2015-0509-01>
- Luo, Y., Li, S., & Ho, C. T. (2021). Key aspects of Amadori rearrangement products as future food additives. *Molecules*, *26*(14), 4314.
- Marconi, O., Mayer, H., Chiacchieroni, F., Ricci, E., Perretti, G., & Fantozzi, P. (2013). The influence of glumes on malting and brewing of hulled wheats. *Journal of the American Society of Brewing Chemists*, *71*, 41–48.

- Martins, N., Barros, L., & Ferreira, I. C. F. R. (2016). In vivo antioxidant activity of phenolic compounds: Facts and gaps. *Trends in Food Science & Technology*, *48*, 1–12. <https://doi.org/10.1016/j.tifs.2015.11.008>
- Mastrangelo, N., Bianchi, A., Pettinelli, S., Santini, G., Merlani, G., Bellincontro, A., ... Mencarelli, F. (2023). Novelty of Italian Grape Ale (IGA) beer: Influence of the addition of Gamay macerated grape must or dehydrated Aleatico grape pomace on the aromatic profile. *Heliyon*, *9*, Article e20422.
- Muñoz-Insa, A., Gastl, M., & Becker, T. (2015). Use of polyphenol-rich hop products to reduce sunstruck flavor in beer. *Journal of the American Society of Brewing Chemists*, *73*(3), 228–235.
- Oliveira de Araújo Melo, C., Cidália Vieira, T., Duarte Gigonzac, M. A., Soares Fortes, J., Moreira Duarte, S. S., da Cruz, A. D., & Silva, D. d. M. E. (2020). Evaluation of polymorphisms in repair and detoxification genes in alcohol drinkers and non-drinkers using capillary electrophoresis. *Electrophoresis*, *41*, 254–258. <https://doi.org/10.1002/elps.201900193>
- Palombi, L., Tufariello, M., Durante, M., Fiore, A., Baiano, A., & Grieco, F. (2023). Assessment of the impact of unmalted cereals, hops, and yeast strains on volatolomic and olfactory profiles of blanche craft beers: A chemometric approach. *Food Chemistry*, *416*, Article 135783.
- Paszkot, J., Gasiński, A., & Kawa-Rygielska, J. (2023). Evaluation of volatile compound profiles and sensory properties of dark and pale beers fermented by different strains of brewing yeast. *Scientific Reports*, *13*, 6725.
- Peixoto, J. A. B., Álvarez-Rivera, G., Alves, R. C., Costa, A. S. G., Machado, S., Cifuentes, A., Ibáñez, E., & Oliveira, M. B. P. P. (2021). Comprehensive phenolic and free amino acid analysis of rosemary infusions: influence on the antioxidant potential. *Antioxidants*, *10*(3), 500. <https://doi.org/10.3390/antiox10030500>. PMID: 33807074; PMCID: PMC8004834.
- Pietrafesa, R., Siesto, G., Tufariello, M., Palombi, L., Baiano, A., Gerardi, C., Braghieri, A., Genovese, F., Grieco, F., & Capece, A. (2023). A multivariate approach to explore the volatolomic and sensory profiles of craft Italian Grape Ale beers produced with novel *Saccharomyces cerevisiae* strains. *Frontiers in Microbiology*, *14*, Article 1234884.
- Pires, E. J., Teixeira, J. A., Brányik, T., & Vicente, A. A. (2014). Yeast: The soul of beer's aroma—a review of flavour-active esters and higher alcohols produced by the brewing yeast. *Applied Microbiology and Biotechnology*, *98*, 1937–1949.
- Rodrigues, J. E. A., Erny, G. L., Barros, A. S., Esteves, V. I., Brandão, T., Ferreira, A. A., Cabrita, E., & Gil, A. M. (2010). Quantification of organic acids in beer by nuclear magnetic resonance (NMR)-based methods. *Analytica Chimica Acta*, *674*(2), 166–175. <https://doi.org/10.1016/j.aca.2010.06.029>
- Rossi, S., Sileoni, V., Perretti, G., & Marconi, O. (2014). Characterization of the volatile profiles of beer using headspace solid-phase microextraction and gas chromatography–mass spectrometry. *Journal of the Science of Food and Agriculture*, *94*, 919–928.
- Sánchez-Palomo, E., Gómez García-Carpintero, E., Alonso-Villegas, R., & González-Viñas, M. A. (2010). Characterization of aroma compounds of Verdejo white wines from the La Mancha region by odour odour activity values. *Flavour/Flavour and fragrance Fragrance journal*, *25*, 456–462.
- Schwarz, K. J., Boitz Inken, L., & Methner, F. J. (2012). Release of phenolic acids and amino acids during mashing dependent on temperature, pH, time, and raw materials. *Journal of the American Society of Brewing Chemists*, *70*(4), 290–295. <https://doi.org/10.1094/ASBCJ-2012-1011-02>
- Selecký, R., Šmogrovičová, D., & Sulo, P. (2008). Beer with reduced ethanol content produced using *Saccharomyces cerevisiae* yeasts deficient in various tricarboxylic acid cycle enzymes. *Journal of the Institute of Brewing*, *114*, 97–101, 2008.
- Siesto, G., Pietrafesa, R., Tufariello, M., Gerardi, C., Grieco, F., & Capece, A. (2023). Application of microbial cross-over for the production of Italian grape ale (IGA), a fruit beer obtained by grape must addition. *Food Bioscience*, *52*, Article 102487.
- Šimić, G., Horvat, D., Dvojković, K., Abičić, I., Vuletić, M. V., Tucak, M., & Lalić, A. (2017). Evaluation of total phenolic content and antioxidant activity of malting and hullless barley grain and malt extracts. *Czech Journal of Food Sciences*, *35*(1), 73–78.
- Singleton, V. L., & Rossi, J. A. (1965). Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. *American Journal of Enology and Viticulture*, *16*, 144–158.
- Swiegers, J. H., Bartowsky, E. J., Henschke, P. A., & Pretorius, I. (2005). Yeast and bacterial modulation of wine aroma and flavour/flavour. *Australian Journal of Grape and Wine Research*, *11*, 139–173.
- Tamura, H., Boonbumrung, S., Yoshizawa, T., & Varanyanond, W. (2001). The volatile constituents in the peel and pulp of a green Thai mango, Khieo Sawoei cultivar (*Mangifera indica* L.). *Food Science and Technology Research*, *7*, 72–77.
- Tristezza, M., Fantastico, L., Vetrano, C., Bleve, G., Corallo, D., Mita, G., & Grieco, F. (2014). Molecular and technological characterization of *Saccharomyces cerevisiae* strains isolated from natural fermentation of Susumaniello grape must in Apulia, Southern Italy. *International Journal of Microbiology*, Article 897428.
- Tufariello, M., Chiriatti, M. A., Grieco, F., Perrotta, C., Capone, S., Rampino, P., Tristezza, M., & Mita, G. (2014). Influence of autochthonous *Saccharomyces cerevisiae* strains on volatile profile of Negroamaro wines. *LWT—Food Science and Technology*, *58*, 35–48.
- Tufariello, M., Maiorano, G., Rampino, P., Spano, G., Grieco, F., Perrotta, C., Capozzi, V., & Grieco, F. (2019). Selection of an autochthonous yeast starter culture for industrial production of Primitivo “Gioia del Colle” PDO/DOC in Apulia (Southern Italy). *LWT—Food Science and Technology*, *99*, 188–196.
- Vrinceanu, C. R., Bărbulescu, I. D., Matei, F., Begea, M., Tudor, V., Frîncu, M., Ionuț, & Teodorescu, R. I. (2022). Review: Actual approaches for the craft beer fermentations. *Series B, Horticulture*, *LXVI*(1), 944–949.
- Wannenmacher, J., Gast, M., & Becker, T. (2018). Phenolic substances in beer: Structural diversity, reactive potential and relevance for brewing process and beer quality. *Comprehensive Reviews in Food Science and Food Safety*, *17*, 953–988.
- Xu, Y. Q., Zou, C., Gao, Y., Chen, J. X., Wang, F., Chen, G. S., & Yin, J. F. (2017). Effect of the type of brewing water on the chemical composition, sensory quality and antioxidant capacity of Chinese teas. *Food Chemistry*, *236*, 142–151.
- Yamamoto, S., Bamba, T., Sano, A., Kodama, Y., Imamura, M., Obata, A., & Fukusaki, E. (2012). Metabolite profiling of soy sauce using gas chromatography with time-of-flight mass spectrometry and analysis of correlation with quantitative descriptive analysis. *Journal of Bioscience and Bioengineering*, *114*(2), 170–175.
- Yamauchi, Y., Okamoto, T., Murayama, H., Nagara, A., Kashihara, T., Yoshida, M., & Nakaniship, K. (1995). Rapid fermentation of beer using an immobilized yeast multistage bioreactor system: Balance control of extract and amino acid uptake. *Applied Biochemistry and Biotechnology*, *53*, 245–259.
- Yang, D., & Gao, X. (2021). Research progress on the antioxidant biological activity of beer and strategy for applications. *Trends in Food Science & Technology*, *110*, 754–764. <https://doi.org/10.1016/j.tifs.2021.02.048>
- Yorke, J., Cook, D., & Ford, R. (2021). Brewing with unmalted cereal adjuncts: Sensory and analytical impacts on beer quality. *Beverages*, *7*, 4. <https://doi.org/10.3390/beverages7010004>
- Zhao, X., Procopio, S., & Becker, T. (2015). Flavor impacts of glycerol in the processing of yeast fermented beverages: A review. *Journal of Food Science and Technology*, *52*(12), 7588–7598. <https://doi.org/10.1007/s13197-015-1977-y>
- Zrčková, M., Capouchová, I., Paznocht, L., Eliášová, M., Dvořák, P., Konvalina, P., ... Bečková, L. (2019). Variation of the total content of polyphenols and phenolic acids in einkorn, emmer, spelt and common wheat grain as a function of genotype, wheat species and crop year. *Plant, Soil and Environment*, *65*(5), 260–266.