Influence of the drying process of Cascade hop and the dry-hopping technique on the chemical, aromatic and sensory quality of the beer

This is the submitted version (pre peer-review, preprint) of the following publication:

Published Version:
Monacci E., Baris F., Bianchi A., Vezzulli F., Pettinelli S., Lambri M., et al. (2024). Influence of the drying process of Cascade hop and the dry-hopping technique on the chemical, aromatic and sensory quality of the beer. FOOD CHEMISTRY, 460 pt 2, 1-12 [10.1016/j.foodchem.2024.140594].

Availability:
This version is available at: https://hdl.handle.net/11585/978277 since: 2024-08-19

Published:
DOI: http://doi.org/10.1016/j.foodchem.2024.140594

Food Chemistry

Influence of the drying process of Cascade hop and the dry-hopping technique on the chemical, aromatic and sensory quality of the beer

--Manuscript Draft--

Manuscript Number:	
Article Type:	Research Article (max 7,500 words)
Keywords:	Humulus lupulus L; Dehydration techniques; Dry-hopping; VOCs profile; Beer sensory quality
Corresponding Author:	Alessandro Bianchi University of Pisa, Department of Agriculture, Food and Environment ITALY
First Author:	Edoardo Monacci
Order of Authors:	Edoardo Monacci
	Federico Baris
	Alessandro Bianchi
	Fosca Vezzulli
	Stefano Pettinelli
	Milena Lambri
	Fabio Mencarelli
	Fabio Chinnici
	Chiara Sanmartin
Abstract:	Drying techniques are important to hop storage, but significantly affect the quality. Another important factor is the stage of hop addition in beer. Dry-hopping, adding hops during fermentation or conditioning, is a valid technique to enhance beer flavor. This study focuses on the impact of two drying techniques [freeze-dryer (F) and hot-stove (H)] for Cascade hop on the chemical, aromatic and sensory quality of beer, comparing beers produced without (BF and BH) and with dry-hopping (BFDH and BHDH). The dry-hopping significantly increased the bitterness index and reduced the titratable acidity. Isoamyl acetate and ethyl caprylate was high especially in BH while ethyl-n-caproate was the highest in BF. Beers with the dry-hopping had a significantly higher content in terpenes especially in BFDH. Sensory evaluation indicates varied preferences, with freeze-dried hop beers generally favored. Finally, depending on the type of beer, the different dried hops and the hopping technique can be chosen.
Suggested Reviewers:	Ombretta Marconi Università degli Studi di Perugia ombretta.marconi@unipg.it Expert in beer analysis and production
	Aldo Todaro Università degli Studi di Palermo aldo.todaro@unipa.it Beer expert
	Cristina Lasanta Universidad de Cadiz cristina.lasanta@uca.es Fermentation and beer expert

Cover Letter

Editor, *Food Chemistry*

4th March 2024
Pisa, Italy

Submission of research article for publication in *Food Chemistry*

Dear Editor,

We hereby submit our research article “**Influence of the drying process of Cascade hop and the dry-hopping technique on the chemical, aromatic and sensory quality of the beer**” by Monacci et. al., to be considered for publication.

Abstract:

Drying techniques are important to hop storage, but significantly affect the quality. Another important factor is the stage of hop addition in beer. Dry-hopping, adding hops during fermentation or conditioning, is a valid technique to enhance beer flavor. This study focuses on the impact of two drying techniques [freeze-dryer (F) and hot-stove (H)] for Cascade hop on the chemical, aromatic and sensory quality of beer, comparing beers produced without (BF and BH) and with dry-hopping (BFDH and BHDH). The dry-hopping significantly increased the bitterness index and reduced the titratable acidity. Isoamyl acetate and ethyl caprylate was high especially in BH while ethyl-n-caproate was the highest in BF. Beers with the dry-hopping had a significantly higher content in terpenes especially in BFDH. Sensory evaluation indicates varied preferences, with freeze-dried hop beers generally favored. Finally, depending on the type of beer, the different dried hops and the hopping technique can be chosen.

On behalf of my co-authors, I declare that this paper has not been published and is not being considered for publication elsewhere, and that, if accepted, the manuscript will not be published elsewhere in the same form, in English or in any other language, without the written consent of the Publisher.

Please address all correspondence to: alessandro.bianchi@phd.unipi.it
Department of Agriculture, Food and Environment – University of Pisa
Via del Borghetto 80
56124 Pisa, Italy

Yours sincerely,

Dr. Alessandro Bianchi

Highlight:

- Influence of hop drying technique and dry-hopping on beer quality.
- The drying technique affect beer aromatic profile.
- Dry-hopping influences acidity, polyphenols and antioxidant content of beer.
- Dry-hopping increases the beer aromatic characteristics.
- Depending on the beer type, the dried hop and hopping technique can be chosen.

Research papers

Influence of the drying process of Cascade hop and the dry-hopping technique on the chemical, aromatic and sensory quality of the beer

Edoardo Monacci [a], Federico Baris [b], Alessandro Bianchi [a*], Fosca Vezzulli [c], Stefano Pettinelli [a], Milena Lambri [c], Fabio Mencarelli [a], Fabio Chinnici [b] and Chiara Sanmartin [a,d]

[a] Department of Agriculture, Food and Environment, University of Pisa, Via del Borghetto 80, 56124 Pisa, Italy. e.monacci1@studenti.unipi.it (E.M.); alessandro.bianchi@phd.unipi.it (A.B.); stefano.pettinelli@phd.unipi.it (S.P.); fabio.mencarelli@unipi.it (F.M.); chiara.sanmartin@unipi.it (C.S.).

[b] Department of Agricultural and Food Sciences, Alma Mater Studiorum, University of Bologna, Viale Fanin 40, Bologna, 40127, Italy. federico.baris2@unibo.it (F.B.); fabio.chinnici@unibo.it (F.C.)

[c] Department for Sustainable Food Process, Università Cattolica del Sacro Cuore, Via Emilia Parmense 84, 29122 Piacenza, Italy. fosca.vezzulli@unicatt.it (F.V.); milena.lambri@unicatt.it (M.L.).

[d] Interdepartmental Research Centre "Nutraceuticals and Food for Health", University of Pisa, Via del Borghetto 80, 56124 Pisa, Italy.

***** Correspondence: alessandro.bianchi@phd.unipi.it

Abstract: Drying techniques are important to hop storage, but significantly affect the quality. Another important factor is the stage of hop addition in beer. Dry-hopping, adding hops during fermentation or conditioning, is a valid technique to enhance beer flavor. This study focuses on the impact of two drying techniques [freeze-dryer (F) and hot-stove (H)] for Cascade hop on the chemical, aromatic and sensory quality of beer, comparing beers produced without (BF and BH) and with dry-hopping (BFDH and BHDH). The dry-hopping significantly increased the bitterness index and reduced the titratable acidity. Isoamyl acetate and ethyl caprylate was high especially in BH while ethyl-n-caproate was the highest in BF. Beers with the dry-hopping had a significantly higher content in terpenes especially in BFDH. Sensory evaluation indicates varied preferences, with freeze-dried hop beers generally favored. Finally, depending on the type of beer, the different dried hops and the hopping technique can be chosen.

Keywords: *Humulus lupulus* L; Dehydration techniques; Dry-hopping; VOCs profile; Beer sensory quality.

1. Introduction

Hop (*Humulus lupulus L.*) is an essential raw material for beer production as it provides an increased shelf-life, bitterness, and aroma to beer (Heřmánek et al., 2018; Raut et al., 2021; Steinhaus & Schieberle, 2000). The quality of the raw material is strongly influenced by the post-harvest transformation methodologies. In fact, the hops, following harvesting, are unsuitable for use as they are in the brewing process. In order to be marketed, as well as used in the beer production process, the raw material must undergo particular conditioning and transformation methods. The water concentration inside the fresh inflorescences (moisture) is about 80 % (Monacci et al., 2023; Verzele & De Keukeleire, 1991) and in order to be marketed and used in the beer production process, the raw material must undergo a drying process (Neve, 2012; Rybacek, 2012) to reduce the water content down to 8 - 11 % (Heřmánek et al., 2018; Rybka et al., 2018).

In this regard, it is important that the matrix undergoes a drying process in the shortest time, in order to preserve its quality and increase its shelf-life (Rossini et al., 2021). The temperature at which the drying process is conducted inevitably determines the loss of chemical compounds present within the inflorescence (Rybka et al., 2018). The quantity of essential oils presents in the fresh product can be reduced by 30-40% following processing, due to the volatile of these compounds (Nance & Setzer, 2011; Rybka et al., 2018). For this reason, the control of the temperature at which the process is carried out is of extreme importance with regard to the quality of the final product for beer production (Raut et al., 2020).

Another important factor in beer process, is the stage of hop addition to beer. There is no doubt on the significant contribution of dry-hopping to the renaissance of craft beer and the subsequent boom in sales of craft beers around the world (Maye et al., 2018; Oladokun et al., 2017). Dry-hopping represents a relatively simple means of improving the beer flavor; brewers add between 2 – 12 g/L of hops in the form of cones or pellets into beer during fermentation or conditioning for periods ranging from several days to weeks (Hauser et al., 2019; Lafontaine & Shellhammer, 2019). The added hops can be left in the beer with no mixing (static dry-hopping) or with mixing, i.e. using a pump or CO_2 (dynamic dry-hopping). Perhaps one of the unintended consequences of dry-hopping is the effect this process has on perceived beer bitterness. Several studies have shown an increase in both measured analytical and perceived bitterness in dry-hopped beers (Algazzali & Shellhammer, 2016; Lafontaine & Shellhammer, 2019; Maye et al., 2016). Brewers can also add hop oil essences to beer to create specific flavor characters that mimic dry-hopped flavor in their product (Gomes et al., 2022; Klimczak et al., 2023). The increase in aroma perceived in dry-hopped beers versus conventionally hopped beers is thought to be due to elevated levels of several volatile terpene compounds, hydrocarbons and their derivatives e.g. linalool, myrcene, humulene, β-citronellol and geraniol (Klimczak et al., 2023; Oladokun et al., 2017; Takoi et al., 2010). Since these chemicals are rapidly lost through evaporation during wort boiling or fermentation, they are seldom perceived in beers that are not dry-hopped (Lafontaine & Shellhammer, 2019). Furthermore, the presence of yeast during dry-hopping adds an extra level of complexity to this process. Some researchers have reported the biotransformation of certain volatile hop compounds during and post fermentation (maturation), e.g., in the conversion of geraniol into β-citronellol (Takoi et al., 2010). This means that brewers must also decide whether to totally remove yeasts from beer before dry-hopping. The presence of yeast during dry-hopping may offer other benefits: suspended yeast can metabolize dissolved oxygen during dry-hopping, thereby protecting beer and hops volatiles from oxidation during the process (Gomes et al., 2022; Oladokun et al., 2017).

Thus, our hypothesis when we started the research project was that the effect of water removal (drying with freeze-dryer or hot-stove) could affect the quality of beer especially in term of VOCs (volatile organic compounds) when used dry-hopping.

2. Materials and Methods

2.1 Raw material

The raw material, consisted in female inflorescences of hop (*Humulus lupulus* L.) cv. Cascade, supplied by Azienda Agricola Opificio Birrario (Crespina-Lorenzana, Pisa, Italy). The quantity of inflorescences resulting, following the removal of leaves, stems and foreign material, was 25 kg. Two different water content reduction techniques were used: (i) a freeze-dryer (F), in LyoQuest lyophilizer (Azbil Telstar, S.L.U., Terrassa, Spain) two cycles, each one lasting for 24 hours, at the temperature of - 52.4 °C at a pressure of 0.072 mBar; (ii) a hot-stove (H), in Heratherm® OMS100 (Thermo Fisher Scientific, Milan, Italy) for two days at temperature of 40 °C.

For each water reduction technique, approx. 10 kg of fresh hop inflorescences were used, divided in 3 trays to place in the stove or in freeze-dryer. Chemical parameters of hop were reported based on dry matter (dm) of sample (Table 1), determined in triplicate on approximately 10 g (from each tray), drying at 105 °C until constant weight (Bianchi et al., 2024).

Table 1. Chemical parameters of hop after the 2 different drying treatments (F and H). Data are the mean (±SD) of 3 trays with hops.

Chemical parameters	Units	F	H
α-acid	(%)	7.17 ± 0.09	7.29 ± 0.08
β-acid	(%)	5.72 ± 0.05	6.73 ± 0.03
HSI	-	0.18 ± 0.02	0.23 ± 0.01
Total polyphenols	(g GAE/kg dm)	442.61 ± 3.31	339.48 ± 1.03
ABTS	(mmol TE /kg dm)	34.42 ± 0.49	22.32 ± 0.64

DPPH	(mmol TE/kg dm)	20.25 ± 0.51	13.01 ± 0.25
Chlorophyll-a	(g/kg dm)	28.86 ± 0.44	26.99 ± 0.28
Chlorophyll-b	(g/kg dm)	52.02 ± 0.63	55.71 ± 0.21
Total Chlorophyll	(g/kg dm)	80.99 ± 0.61	82.60 ± 0.29
Carotenoids	(g/kg dm)	53.02 ± 0.27	54.91 ± 0.41
Essential oil yield	(% V/w)	4.99 ± 0.09	4.64 ± 0.11

2.2 Beer production

The ingredients used for beer production (each beer-must) were: 10 kg of malt Maris Otter (Muntons, Stowmarket, United Kingdom), 10 kg of malt Pilsner (Weyermann, Bamberg, Germany), 300 g of dried hop (F and H), 15 g of dry yeast SafAle™ US-05 (Fermentis, Marquette-lez-Lille, France) and water (70 L in mash, 63.6 L in sparge). Beer was produced in spring 2023 in a stainless-steel plant Easy 100 (Polsinelli Enologia Srl, Isola del Liri, Italy) to produce 100 L of beer, following the phases described in Mastrangelo et al. (2023): (i) mash: insertion of ground malts at 35 ± 2 °C; (ii) protein rest: 55 °C for 10 minutes; (iii) mash-in: 65 °C for 50 minutes; (iv) β–glucan rest: 72 °C for 10 minutes; (v) mash-out: 78 °C for 5 minutes; (vi) Sparge with water at 78 °C for the sugar extraction from brewers grains; (vii) the must was divided into 2 tanks and boiled for 60 minutes at 95 ± 2 °C; (viii) during the boiling was added hops in 3 portions (100 g) every 20 minutes; (ix) the must was cooled (23 ± 2 °C) and the yeast was inoculated (15 g/hL).

The dry products (300 g each) from 2 different techniques (freeze-dryer (F), hot-stove (H)) was used as aforementioned.

For the fermentation phase, the beer-must was divided into 4 (two each sample) fermenters of 20 L:

1- Beer with freeze-dried hop added during the boiling phase (BF).
2- Beer with freeze-dried hop added during the boiling phase and dry-hopping (BFDH).
3- Beer with hot-stove dried hop added during the boiling phase (BH).
4- Beer with hot-stove dried hop added during the boiling phase and dry-hopping (BHDH).

The primary fermentation lasted 16 days, 11 days at 18 ± 2°C and 5 days at 6 °C ± 1°C in cold room to allow the precipitation of the solid compounds (lagering). On the 7th day of fermentation, a static dry-hopping for 48 hours (concentration of 5 g/L) has been done in the BFDH and BHDH beer-must. Successively, the beer was bottled in 0.5 L glass bottles and capped with a crown cap. During the bottling phase, 6 g/L of commercial beet sugar were added to start the bottle secondary fermentation which lasted one month at 8 °C ± 1°C.

2.3 Chemical characterization of hop

The chlorophyll and carotenoid contents [g/kg on dry matter (dm)] were determined according to Monacci et al. (2023).

The total polyphenols (g of gallic acid equivalents (GAE)/kg dm) and the anti-radical activity (ABTS and DPPH free radical method) (mmol Trolox equivalents (TE)/kg dm) were determined as previously reported (Bianchi et al., 2023a).

The concentration of α- and β-acids (%) and the Hop Storage Index (HSI) was calculated according to previously reported (Monacci et al., 2023).

The quantification of the essential oil yield of the samples was carried out in agreement with Van Simaeys et al. (2022) utilized the hydro-distillation process and calculated as reported in the following equation:

$$Yield\ essential\ oils\ (\%\ V/w) = \frac{Volume\ hydrosol\ (mL)}{sample\ weight\ (g)} \times 100$$

2.4 Chemical analysis of beer

Beer chemical analyses were carried out on four, 0.5 L bottles, two from each fermenter vat (4 bottles each beer production), and before analyses, the beers were degassed by ultrasound. Chemical parameters: alcohol (% V/V), pH, residual sugars (g/L hexoses), titratable acidity (g/L lactic acid), volatile acidity (g/L lactic acid), total polyphenols (mg/L gallic acid), color according to the SRM

(Standard Reference Method) scale and bitterness according to the IBU (International Bitterness Unit) scale, were carried out following the official method of the American Society of Brewing Chemists as previously reported (Mastrangelo et al., 2023). Finally, the anti-radical activity of beer (mmol Trolox equivalents (TE)/L) was determined by DPPH and ABTS free radical methods as reported (Bianchi et al., 2023c).

2.5 Volatile organic compounds (VOCs)

The analysis of VOCs of beer was performed as previously reported (Mastrangelo et al., 2023). In particular, 10 mL sample of beer (2 bottles each beer production), degassed by ultrasound, and 100 μL of a 2-octanol solution at 500 mg/L was added as an internal standard. The sample was deposited on a Hypersep Retain Prep (Thermo Fisher Scientific, Milan, Italy) cartridge (60 mg), activated with 2 mL dichloromethane, 2 mL methanol and 2 mL water. The analytes were eluted with 5mL of dichloromethane, collected in sovirel on the bottom of which 2 grams of anhydrous sodium sulfate had been inserted and placed in the freezer overnight. Samples were filtered with a cellulose filter to remove sodium sulfate and concentrated to a final volume of 200 μL under a stream of N_2. The GC apparatus consists of a Trace GC ultra-gas chromatograph with a Trace DSQ with quadrupole mass detector (Thermo Fisher Scientific, Milan, Italy) and a Stabilwax DA capillary column (Restek, Bellefonte, PA, USA; 30 m, 0.25mm i.d., and 0.25 μm film thickness) and He (constant flow of 1.0 mL/min) was used as The carrier gas.

GC temperature ramp was programmed as reported by Castro-Marín et al. (2018): from 45°C (maintained for 1 minute) to 100°C (maintained for 1 minute) at 3°C/min, then to 240°C (maintained for 10 minutes) at 5°C/min. The injection was performed at 250°C in spitless mode and the volume injected is 1 µL. Analyses were done in quadruplicate (four bottles) and GC-MS parameters were obtained by using Xcalibur v 4.1 software (Thermo Fisher Scientific, Milan, Italy). Identification and the quantification of compounds (µg/L) were carried out as previously reported (Mastrangelo et al., 2023).

2.6 Sensory analysis

The beer sensory profile was evaluated by a panel of ten beer experts using QDA (qualitative discriminant analysis) method (Stone et al., 2004) at the SensoryLab of DiSTAS of Università Cattolica del Sacro Cuore compliant with the International Organization for Standardization standard (ISO 8589:2007) (Donadini et al., 2013). The samples were served randomly during the test with a rotated plan. The sensory scoresheet presented five main classes of parameters selected and slightly modified from Donaldson et al. (2012) and Donadini et al. (2013): visual, olfactory, tactile, gustatory and retro-olfactory perception. The first class was divided into: foam stability, foam compactness, turbidity, yellow straw color, golden yellow color, amber yellow color, and color pleasantness. The second class was divided into: olfactory intensity, olfactory complexity, floral, fruity, vegetable, malty and olfactory pleasantness. The third class was divided into: effervescence, body, sweet, sour, bitter, astringency, softness and balance. The last class was divided into flowers, vegetables, fruits, citrus fruits (orange, lemon, tangerin and grapefruit), tropical fruits (pineapple, banana, mango, kiwi and lychee), peach/apricot, apple/pear, yeast, malt, caramel, spicy, toasted, persistence, DMS-defects, mold-defects, cardboard-defects and retro-olfactory pleasantness. For all the samples, after the objective evaluation, the overall liking of the sample was evaluated. The intensity of each attribute was evaluated on a nine-point horizontally oriented scale anchored as ‘‘not perceived at all’’ and ‘‘extremely intense’’. Before starting the analysis, panel was calibrated around the median ± 1, calculated for each attribute, after the evaluation of an extra sample served as calibration sample. Repeatability, discrimination and collimation of the panel and panelists were evaluated thanks to the presence of an analytical replicate in the set (Vezzulli et al., 2021).

2.7 Statistical analysis

One-way ANOVA was run (CoStat, Version 6.451, CoHort Software, Pacific Grove, CA, USA) and Tukey's honestly significant difference (HSD) test with $p < 0.05$ for multiple comparison, was used for the chemical parameters.

The JMP 17 software was used to perform a principal component analysis (PCA) and a hierarchical cluster analysis (HCA) on the VOCs data as previously reported (Bianchi et al., 2023b).

Finally, sensory analysis results were processed by Big Sensory Soft 2.0 (version 2018) and elaborated in Microsoft Excel 2007, to be validated as the median values of the intensity scored by the panel to each sensory descriptor, then plotted as spider graphs.

3. Results and Discussion

3.1 Chemical characterization of beers

BH and BHDH samples have a significantly higher alcohol content than BF and BFDH samples (8.85% v/v for BH and 8.42% v/v for BHDH vs 6.34% v/v and 6.24% v/v in BF and BFDH) (Table 2) while no significant difference was found in residual sugar concentrations. Titratable acidity was higher in samples BH and BHDH and dry-hopping reduced the acidity. The observed difference between F and H samples could be due to the degree of sterilization of the matrix. The freeze-drying technique guarantees a lower depletion of the bioactive components of biological matrices, with an increase in product quality (Karam et al., 2016) but, in contrast, freeze-drying does not guarantee the complete removal of microorganisms (Yadav and Roopesh, 2020). Gram-positive bacteria have a greater ability to survive than gram-negative ones in freeze-drying (Morgan and Vesey, 2009; Wang et al., 2023). Thus the higher ethanol content of BH and BHDH could be due to by microbial competition during fermentation, consuming the same amount of sugars but affecting different metabolic processes (Romero-Rodríguez et al., 2022). Also the reduction of titratable of titratable acidity in BF and even more in BFDH can be related to the presence on hop inflorescences of bacteria, which have consumed the acids present in solution. Moreover, it is well documented in the literature that there is an increase in pH with dry-hopping, and a consequent lowering of total acidity (Hauser et al., 2019; Lafontaine & Shellhammer, 2018; Rutnik et al., 2022). The correlation can be observed if we take into consideration the concentration of acetic acid and the alcohol content in the product (Table 2). In this case, in the BF and BFDH samples there is an increase in volatile acidity, which is 0.23 g/L and 0.41 g/L respectively, and a reduction in ethanol concentration. The appearance could be determined by a microbial component on the lyophilized product, as aforementioned, that favors the formation of acetic acid from the substrates present in solution. In BH and BHDH samples the presence of acetic acid is much smaller, presenting values of 0.17 g/L and 0.14 g/L respectively.

The color of the beer is determined by the interaction between the different raw materials of the recipe, as well as by the temperatures at which the mashing phase is conducted, and by the possible enzymatic oxidation of polyphenols (Koren et al., 2020; Pahl et al., 2016; Pettinelli et al., 2022). The SRM (Table 2) values fell into the Amber category, which has an SRM of 13-16. Beers produced through the use of hops dried with a hot stove at a temperature of 40 °C (BH: 13.92; BHDH: 13.32), were slightly lighter than those with freeze-dried hops (BF 16.09; BFDH 16.15). The cause is attributable to a more marked oxidation of polyphenols in BF and BFDH samples, whose starting material had a higher content in this class of compounds (Table 1). In fact, the color of beer is conditioned by the oxidation of monomers of the flavonols class and the oligomers of proanthocyanidins (Koren et al., 2020).

Table 2. Main chemical parameters of beers. Data are the mean (± SD) of 4 bottles. Beer with freeze-dried hop added during the boiling phase (BF); Beer with freeze-dried hop added during the boiling phase and dry-hopping (BFDH); Beer with hot-stove dried hop added during the boiling phase (BH); Beer with hot-stove dried hop added during the boiling phase and dry-hopping (BHDH).

Chemical parameters	Units	BF	BFDH	BH	BHDH
Alcohol	% V/V	6.37 ± 0.11 b	6.26 ± 0.14 b	8.85 ± 0.17 a	8.42 ± 0.12 a
pH	-	4.68 ± 0.02 b	4.84 ± 0.03 a	4.50 ± 0.03 c	4.63 ± 0.04 b
Residual sugars	g/L hexoses	0.47 ± 0.07 a	0.35 ± 0.06 a	0.39 ± 0.02 a	0.42 ± 0.03 a
Titratable acidity	g/L lactic acid	1.66 ± 0.15 b	1.23 ± 0.12 c	1.97 ± 0.13 a	1.77± 0.19 ab
Volatile acidity	g/L acetic acid	0.23 ± 0.02 b	0.41 ± 0.05 a	0.17 ± 0.04 bc	0.14 ± 0.03 c
Color (SRM)	-	16.09 ± 0.52 a	16.15 ± 0.62 a	13.92 ± 0.41 b	13.32 ± 0.42 b
IBU	-	10.43 ± 0.63 b	6.26 ± 0.51 d	8.93 ± 0.27 c	16.46 ± 0.58 a
Total polyphenols	mg/L gallic acid	395 ± 25 ab	421 ± 31 a	315 ± 9 c	352 ± 21 b
ABTS	mmol TE/L	1.34 ± 0.02 b	1.60 ± 0.05 a	1.26 ± 0.03 b	1.34 ± 0.04 b
DPPH	mmol TE/L	0.82 ± 0.03 b	0.96 ± 0.02 a	0.76 ± 0.04 b	0.81 ± 0.03 b

Different letters in each row refer to significant differences (Tukey, $p < 0.05$). n.d.= not detected.

In BHDH (16.46) the dry-hopping technique significantly increased the bitterness index compared to BH (8.93). The IBU is determined by the concentration of iso-α-acids present in solution, which determine the characteristic bittering power of the product (Bocquet et al., 2018; Pahl et al., 2016; Rutnik et al., 2022). The use of this hop infusion technique tended to increase the IBU value of the final product. The appearance is related to the oxidation of α-acids with the production of humulinone, which entering into solution contributes to the increase of the IBU (Hauser et al., 2019; Salamon et al., 2022). The beers produced with freeze-dried hops had an opposite trend compared to the aforementioned ones.

In fact, the value in IBU decreased as a result of the dry-hopping technique, going from 10.43 in BF to 6.26 in BFDH. The loss in iso-α-acids is influenced by several factors, such as the absorption of these compounds by lees materials or yeast cells and their transformation which causes oxidative processes (Van Cleemput et al., 2009). Moreover, through dry-hopping a reduction of iso-α-acids is observed because they are replaced by oxidation products with less bittering power, thus IBU reduction (Maye et al., 2018). Finally, the results of total polyphenols and antioxidant power (ABTS and DDPH) show that BF and BFDH samples had higher values than BH and BHDH (Table 3). The results are affected by the quality of the starting matrix (Table 1), which had different values in total polyphenols, ABTS and DPPH, higher in BF and BFDH. The quality of the freeze-dried matrix is also influencing the antioxidant power of the finished product, especially following the dry-hopping technique (BFDH 1.60 mmol TE/L), while among the other samples, no significant differences (BH 1.26 mmol TE/L, BHDH 1.34 mmol TE/L, BF 1.34 mmol TE/L) were measured. In any case, the addition of hops following fermentation influences the amount of total polyphenols and antioxidant power. This aspect determines a greater stability of the chemical-physical characteristics of the finished product over time, increasing its shelf-life (C. W. Bamforth, 2016). Moreover, through the techniques of late hopping and dry-hopping, the extraction of α-acids in solution is favored (Oladokun et al., 2017) with the result of greater stability against microbiological contamination (Michel et al., 2020).

3.2 VOCs profile in beers

As in all fermented beverages, apart from ethanol and carbon dioxide which are the main products of fermentation, there are also classes of VOCs in beer which are formed as secondary products of fermentation and characterize its quality. Olaniran et al. (2017) reported what they define as “Flavour-active volatile compounds in beer”. Higher alcohols and esters represent the most important classes being the result of fermentation by yeasts. Among the esters, 1/3 is represented by ethyl acetate (Jespersen & Jakobsen, 1996) which if in low concentration provides solvent nuance. Other classes in lower concentrations but important from an aromatic point of view are terpenes, furans, ketones.

Table 3. Volatile Organic Compounds (VOCs) in beers. Data are the mean (± SD) of 4 bottles. Beer with freeze-dried hop added during the boiling phase (BF); Beer with freeze-dried hop added during the boiling phase and dry-hopping (BFDH); Beer with hot-stove dried hop added during the boiling phase (BH); Beer with hot-stove dried hop added during the boiling phase and dry-hopping (BHDH).

VOCs (μg/L)	**BF**	**BFDH**	**BH**	**BHDH**
Ethyl butyrate	71.75 ± 2.67 b	75.05 ± 2.11 b	88.85 ± 3.19 a	90.57 ± 5.87 a
Ethyl isovalerate	n.d.	n.d.	n.d.	4.18 ± 0.23 a
Isoamyl acetate	361.63 ± 21.24 b	130.87 ± 11.46 c	450.60 ± 14.03 a	478.83 ± 21.52 a
Ethyl *n*-caproate	359.37 ± 9.73 a	164.22 ± 6.91 c	263.43 ± 13.56 b	265.91 ± 11.59 b
Ethyl heptanoate	4.16 ± 0.89 c	7.39 ± 0.98 b	5.36 ± 0.57 c	10.08 ± 0.86 a
Ethyl caprylate	258.30 ± 6.17 c	238.29 ± 3.01 d	313.60 ± 3.76 b	341.92 ± 8.16 a
Ethyl decanoate	33.39 ± 2.02 c	42.58 ± 2.66 b	21.23 ± 3.51 d	55.26 ± 5.64 a
Ethyl benzoate	0.32 ± 0.09 c	2.40 ± 0.35 a	1.52 ± 0.30 b	2.40 ± 0.26 a
Diethyl succinate	8.51 ± 0.49 b	5.81 ± 0.25 d	6.59 ± 0.60 c	9.66 ± 0.22 a
Ethyl 9-decenoate	9.97 ± 1.08 c	9.62 ± 1.58 c	17.86 ± 1.37 b	23.74 ± 2.83 a
1,3-Propanediol, diacetate	21.29 ± 0.66 c	21.69 ± 0.75 c	26.65 ± 2.09 b	31.33 ± 1.48 a
Methyl salicylate	7.49 ± 0.86 c	13.56 ± 1.32 a	5.01 ± 0.62 d	8.46 ± 0.72 b

Etil-4-idrossibutanoato	21.35 ± 2.33 c	28.82 ± 1.06 b	43.56 ± 2.54 a	46.71 ± 3.37 a
2-Feniletilacetato	128.63 ± 6.25 a	33.29 ± 2.35 c	90.74 ± 7.45 b	95.20 ± 5.06 b
Ethyl 2-hydroxy-3-phenylpropanoate	8.83 ± 0.98 d	12.41 ± 1.03 c	21.00 ± 1.24 a	17.29 ± 1.79 b
Ethyl hydrogen succinate	27.24 ± 1.30 a	11.66 ± 1.02 c	15.29 ± 2.08 b	12.27 ± 1.89 bc
Ethyl vanillate	n.d.	10.66 ± 1.15 b	7.07 ± 1.36 c	14.04 ± 1.23 a
Total Esters	**1322.23 ± 57.47 b**	**808.33 ± 38.41 c**	**1382.23 ± 58.57 b**	**1507.83 ± 73.19 a**
1-Propanol	5.00 ± 0.63 c	5.45 ± 0.45 c	6.99 ± 0.62 b	11.67 ± 1.02 a
Isobutanol	212.83 ± 6.95 c	231.34 ± 4.06 b	228.02 ± 4.58 b	264.82 ± 10.36 a
trans-3-Penten-2-ol	7.28 ± 1.52 a	n.d.	9.04 ± 1.44 a	n.d.
Isoamyl alcohol	4403.08 ± 35.24 c	4411.70 ± 46.49 c	4865.19 ± 89.77 a	4601.17 ± 32.35 b
1-Pentanol	3.01 ± 2.02 a	4.97 ± 1.60 a	4.77 ± 1.98 a	4.98 ± 1.70 a
4-Methyl-2-pentanol	14.19 ± 1.67 a	14.51 ± 1.21 a	15.18 ± 1.21 a	16.07 ± 1.93 a
2-Methyl-2-buten-1-ol	4.79 ± 0.61 b	6.16 ± 0.95 a	4.82 ± 0.47 b	7.18 ± 0.52 a
3- Methyl-1-pentanolo	0.64 ± 0.25 b	0.99 ± 0.12 b	3.23 ± 0.68 a	3.68 ± 0.92 a
n-Hexanol	16.40 ± 2.01 d	25.23 ± 1.11 b	20.03 ± 1.51 cd	31.38 ± 1.83 a
3-Ethoxy-1-propanol	4.46 ± 0.84 b	4.62 ± 0.33 b	7.08 ± 0.33 a	7.87 ± 0.55 a
cis-3-Hexen-1-ol	6.91 ± 1.64 c	22.01 ± 1.56 a	4.37 ± 0.56 d	9.75 ± 0.82 b
1-Octen-3-ol	1.23 ± 0.90 b	8.82 ± 1.80 a	n.d.	6.48 ± 1.03 a
1-Heptanol	29.34 ± 1.97 b	31.19 ± 2.16 b	52.02 ± 3.16 a	53.22 ± 2.55 a
3-Methyl-2-octanol	2.57 ± 0.32 b	2.82 ± 0.24 ab	3.11 ± 0.28 a	3.26 ± 0.22 a
3-Nonanol	10.82 ± 0.86 ab	11.43 ± 1.08 a	9.59 ± 0.61 b	11.15 ± 1.00 a
2-Nonanol	7.15 ± 1.53 a	6.83 ± 1.40 a	6.72 ± 1.04 a	7.32 ± 1.85 a
2-3-Butanediol	341.27 ± 14.57 ab	315.57 ± 21.78 b	363.29 ± 11.78 a	361.10 ± 12.07 a
1-Ottanolo	12.06 ± 1.58 a	12.88 ± 2.11 a	11.90 ± 2.11 a	13.94 ± 1.54 a
Metionolo	17.23 ± 0.69 a	16.44 ± 0.77 a	17.83 ± 0.77 a	17.82 ± 0.91 a
Benzyl alcohol	17.10 ± 2.10 c	72.53 ± 3.97 a	17.64 ± 4.97 c	33.52 ± 6.85 b
Phenethyl alcohol	7740.16 ± 164.27 b	7524.38 ± 135.22 b	8645.00 ± 124.08 a	7608.98 ± 122.35 b
Total Alcohols	**12857.54 ± 242.17 c**	**12729.86 ± 228.40 c**	**14295.81 ± 251.96 a**	**13075.33 ± 202.40 b**
Acetic acid	123.59 ± 3.79 b	115.93 ± 2.11 c	133.36 ± 2.73 a	135.61 ± 1.98 a
Propanoic acid	12.72 ± 1.24 b	16.95 ± 1.02 a	6.92 ± 1.11 d	9.11 ± 1.35 c
Isobutyric acid	64.27 ± 3.74 b	58.76 ± 3.47 b	75.18 ± 4.28 a	79.57 ± 4.15 a
Butanoic acid	24.02 ± 3.00 b	19.50 ± 4.72 b	26.45 ± 2.43 a	25.84 ± 2.81 a
Pentanoic acid	123.44 ± 7.76 b	108.74 ± 10.20 b	144.51 ± 9.38 a	155.00 ± 4.63 a
Hexanoic acid	1484.71 ± 35.48 a	1403.37 ± 82.21 a	1492.86 ± 47.56 a	1507.69 ± 122.08 a
(*E*)-2-Hexenoic acid	11.52 ± 2.66 c	31.76 ± 6.21 b	16.89 ± 3.01 c	45.08 ± 2.85 a
Caprylic acid	3309.91 ± 127.12 a	2810.37 ± 65.03 b	3136.37 ± 87.24 a	2810.10 ± 53.26 b
Nonanoic acid	10.50 ± 1.26 b	12.02 ± 1.58 ab	14.23 ± 1.75 a	15.00 ± 1.23 a
Capric acid	883.68 ± 36.82 a	445.95 ± 20.19 bc	405.69 ± 21.61 c	489.13 ± 36.15 b
9-Decenoic acid	206.82 ± 9.94 b	124.55 ± 7.39 c	240.96 ± 14.84 a	210.22 ± 11.72 b
Benzoic acid	84.44 ± 4.25 bc	77.97 ± 5.26 c	115.88 ± 14.53 a	91.62 ± 7.66 b
Lauric Acid	180.00 ± 11.20 a	43.25 ± 6.24 c	81.18 ± 8.62 b	90.48 ± 7.00 b
Benzene acetic acid	220.93 ± 13.98 a	142.98 ± 14.12 b	226.53 ± 21.75 a	165.74 ± 9.01 b
Phenyl propionic acid	20.58 ± 2.49 bc	18.17 ± 1.13 c	30.41 ± 2.62 a	25.20 ± 1.83 b
Myristic acid	44.30 ± 2.08 a	14.65 ± 1.37 c	39.21 ± 3.66 a	27.23 ± 1.05 b
Palmitic acid	170.39 ± 10.73 b	99.05 ± 4.62 d	263.53 ± 15.82 a	140.55 ± 9.03 c
Stearic acid	49.44 ± 3.52 b	41.76 ± 3.74 c	118.70 ± 15.91 a	44.06 ± 4.53 bc
Total Acids	**7025.27 ± 281.05 a**	**5585.73 ± 240.62 b**	**6568.87 ± 278.84 a**	**6067.25 ± 282.32 b**
2,7-Dimethyl-2,6-octadiene	393.04 ± 19.48 a	418.71 ± 10.49 a	297.89 ± 16.26 b	321.51 ± 12.42 b
β-Myrcene	n.d.	68.95 ± 2.32 a	0.09 ± 0.04 b	45.14 ± 7.12 b
6-Methyl-5-hepten-2-one	0.95 ± 0.17 c	2.64 ± 0.19 b	1.46 ± 0.47 c	4.27 ± 0.97 a
cis-Linalool oxide	0.96 ±0.18 c	4.16 ± 0.32 a	1.98 ± 0.44 b	4.11 ± 0.28 a
trans-Linalool oxide	n.d.	1.06 ± 0.09 b	n.d.	1.98 ± 0.07 a
β-Linalool	66.86 ± 1.99 c	137.51 ± 1.32 a	73.71 ± 2.76 b	140.26 ± 2.40 a
p-Menth-2-en-1-ol	0.22 ± 0.02 b	0.41 ± 0.12 a	0.21 ± 0.03 b	0.47 ± 0.09 a
β-Caryophyllene	8.41 ± 0.77 c	16.85 ± 1.75 a	13.60 ± 0.92 ab	15.29 ± 0.78 a
Terpinene-4-ol	1.74 ± 0.44 b	4.19 ± 0.62 a	1.87 ± 0.66 b	3.62 ± 0.32 a
(*E*)-Farnesene epoxide	10.84 ± 1.14 c	21.10 ± 1.90 b	12.05 ± 0.78 c	29.19 ± 2.10 a
α-Humulene	2.36 ± 0.72 c	21.63 ± 1.89 a	3.14 ± 0.64 c	18.88 ± 1.15 b
α-Terpineol	12.74 ± 0.73 b	27.22 ± 2.44 a	13.65 ± 0.78 b	29.09 ± 1.86 a
2,6-Dimethyl-1,5,7-octatrien-3-ol	2.05 ± 0.21 b	5.22 ± 0.82 a	2.56 ± 0.78 b	7.14 ± 1.64 a

β-Citronellol	20.75 ± 1.50 c	63.88 ± 7.48 a	19.37 ± 1.11 c	38.47 ± 3.21 b
cis-Geraniol	5.49 ± 0.70 d	14.45 ± 1.31 a	8.51 ± 0.34 c	10.95 ± 0.99 b
trans-Geraniol	0.30 ± 0.03 b	0.60 ± 0.11 a	0.60 ± 0.06 a	0.67 ± 0.04 a
p-Mentha-1,8-dien-7-ol	16.01 ± 1.46 d	82.11 ± 1.46 a	37.56 ± 1.21 c	61.00 ± 2.54 b
8-Hydroxy linalool	10.33 ± 1.80 c	25.59 ± 1.98 a	13.59 ± 1.63 bc	16.50 ±1.25 b
Farnesol	21.78 ± 1.09 b	22.87 ± 1.89 b	30.89 ± 2.26 a	29.00 ± 2.20 a
Dihydro-β-ionone	12.41 ± 2.86 c	67.02 ± 3.71 a	52.87 ± 3.01 b	54.90 ± 4.25 b
Total Terpenes	**587.23 ± 35.28 c**	**1.006.18 ± 42.22 a**	**585.61 ± 29.28 c**	**832.44 ± 45.69 b**
1,1-Dimethyl-4-methylenehexahydro-1H-cyclopenta[c]furan	n.d.	3.86 ± 0.35 b	n.d.	7.03 ± 0.64 a
Furfuryl alcohol	11.11 ± 0.70 ab	10.72 ± 0.46 b	12.19 ± 1.09 a	10.84 ± 0.64 b
5-Ethyldihydro-2(3H)-furanone	2.77 ± 0.48 a	3.20 ± 0.23 a	3.23 ± 0.34 a	2.73 ± 0.45 a
2,3-Dihydro-5-hydroxy-6-methyl-4H-pyran-4-one	23.03 ± 1.11 a	21.00 ± 1.32 ab	20.50 ± 0.52 b	20.31 ± 0.93 b
2,3-Dihydro-benzofuran	31.43 ± 2.72 b	47.63 ±3.23 a	25.83 ± 2.07 c	31.36 ± 2.39 b
Total Furans	**68.34 ± 5.02 bc**	**86.41 ± 5.58 a**	**61.75 ± 4.02 c**	**72.27 ± 5.05 b**
2-Methoxy-4-vinylphenol	99.60 ± 5.17 b	108.26 ± 4.17 b	139.63 ± 2.25 a	143.52 ± 2.26 a
Acetovanillone	33.81 ± 1.98 c	34.64 ± 1.18 c	42.86 ± 1.02 b	51.74 ± 3.25 a
Methoxy-eugenol	17.25 ± 0.95 c	20.09 ± 1.01 b	21.22 ± 1.02 ab	24.80 ± 2.28 a
trans-Cinnamic acid	72.39 ± 0.99 a	69.22 ± 2.07 a	63.99 ± 1.45 b	59.19 ± 2.41 b
Tyrosol	155.29 ± 10.57 b	132.12 ± 9.09 c	208.95 ± 13.58 a	145.30 ± 11.56 bc
Total Phenols	**378.35 ± 19.66 c**	**364.34 ± 17.51 c**	**476.66 ± 19.33 a**	**424.55 ± 21.76 b**
Methyl acetoin	4.10 ± 0.38 a	3.79 ± 0.68 a	4.29 ± 0.74 a	4.52 ± 0.38 a
Acetoin	45.84 ± 4.31 a	10.27 ± 1.24 c	15.47 ± 1.23 b	17.19 ± 1.20 b
3-Hydroxy-3-methyl-2-butanone	14.03 ± 1.01 a	12.07 ± 0.67 b	13.23 ± 0.50 ab	13.56 ± 0.58 a
γ-Nonalactone	35.83 ± 0.73 b	36.42 ± 0.74 b	47.07 ± 1.22 a	45.13 ± 1.56 a
Total Ketones	**99.80 ± 6.43 a**	**62.54 ± 8.92 c**	**80.06 ± 7.71 b**	**80.41 ± 8.78 b**
n-Nonanal	2.20 ± 0.35 ab	1.97 ± 0.21 b	1.93 ±0.15 b	2.59 ± 0.24 a
Total Aldehydes	**2.20 ± 0.35 ab**	**1.97 ± 0.21 b**	**1.93 ± 0.15 b**	**2.59 ± 0.24 a**
2-Acetylpyrrole	47.98 ± 1.82 a	40.24 ± 2.22 b	39.04 ± 2.82 b	40.46 ± 1.22 b
5-Methyl-1H-pyrrole-2-carboxaldehyde	7.15 ± 0.60 b	8.68 ± 0.35 a	7.45 ± 0.93 b	7.98 ± 0.39 ab
Styrene	n.d.	n.d.	14.68 ± 2.09 a	12.62 ± 1.86 a
Total Others	**55.13 ± 2.43 b**	**48.91 ± 2.57 c**	**61.18 ± 3.76 a**	**61.06 ± 3.47 a**

Different letters in each row refer to significant differences (Tukey, $p < 0.05$). n.d.= not detected.

As regards VOCs (Table 3), during fermentation and maturation different species of secondary compounds originate, which characterize the sensory profile of beer. Esters provide beer with fruity and floral scents (Brányik et al., 2008); we found mainly isoamyl acetate especially in BH samples and very low concentration in BFDH (BH 450.60 μg/L; BHDH 478.83 μg/L; BF 361.63 μg/L; BFDH 130.87 μg/L); also ethyl caprylate was higher in BH samples while ethyl-n-caproate was the highest in BF but the lowest in BFDH. Concentrations of isoamyl acetate above 2 mg/L give fruity scents (olfactory threshold 1.2 ppm) (Olaniran et al., 2017), while ethyl caprylate and ethyl-n-caproate provide aromas of acidic apple in concentrations higher than 0.9 ppm and 0.21 ppm, respectively (Kobayashi et al., 2007); in our samples the content detected was below the olfactory perception threshold. Totally speaking, in freeze-dried hop, using the dry-hopping reduced the amount of esters while in stove hop, the dry-hopping increased the esters. Alcohols were ten-fold higher than esters and the total highest concentration was in BH (Table 3). Among the higher alcohols the most important in term of concentration were isoamyl alcohol (BH 4865.19 μg/L; BHDH 4601.17 μg/L; BF 4403.08 μg/L; BFDH 4411.70 μg/L) and 2-phenylethyl alcohol (BH 8645.00 μg/L; BHDH 7608.98 μg/L; BF 7740.16 μg/L; BFDH 7524.38 μg/L). Isoamyl alcohol if present in high concentrations negatively affects the drinkability of the product (Olaniran et al., 2017); however, the quantity detected in the test samples was below the limit at which this adverse effect is observed. 2-phenylethyl alcohol, which would give hints of rose, peppermint and orange blossom, was also below the threshold of perception (125 ppm) (Olaniran et al., 2017). The highest concentration of these two alcohols was anyway in BH sample. Caprilic acid was in the greatest amount among acids, especially where dry-hopping was not used, followed by hexanoic acid (Table 3). The total amount of acids was higher where dry-hopping was not used. These acids are from cell membrane thus coming from yeast degradation and are important in the formation of non acetic esters which play an important role for the fruity aroma. Add of dry-hopping in freeze-dried beer reduced significantly acids and consequently esters, reducing the aromatic potential. The reason of this significantly reduction especially in BF samples could be due to the presence of other microorganisms which are survived after the freeze-drying process and could have used to built cell membrane and reproducing (Romero-Rodríguez et al., 2022).

In general, hops strongly contribute to the sensory characteristics of the product, through α- and β-acids, and to the aromatic ones, depending on the concentration of terpenes (Hauser et al., 2019; Lafontaine & Shellhammer, 2018). In the class of terpenes, 20 compounds were identified (Table 3). The drying methodology determined a different influence on the aromatic component of the beer, also related to the different infusion technique, as evidenced by the hierarchical cluster shown in Figure 1. Beers produced with the dry-hopping technique, had a significantly higher contente in terpenes especially in beer from freeze-dried hops. 2,7-Dimethyl-2,6-octadiene was in significantly higher concentration than the other compounds usually characterizing hop (Mastrangelo et al., 2023; Steyer et al., 2017) but the difference between dry-hopping and not, was marked by β-linalool and β-citronellol in double concentration in BFDH and BHDH samples. The values here reported are much higher than what is present in the literature (Van Opstaele et al., 2010). Freeze-drying and dry-hopping gave the highest aconcentration of these aromatic compounds confirming the role of hops in providing terpenes which are preserved by using the least invasive drying technique. Total furans were higher in BF samples while total phenols in BH samples, regardless dry-hopping.

As shown in Figure 1, the first two principal components account for more than 90 % of the data variability (PC1 70.4 %, PC2 23.2 %), while the remaining three PCs explain the residual variance (PC3 6.4 %). Therefore, we have only reported the first 2 PCs. The PCA of Volatile Organic Compounds (VOCs) clearly distinguishes the beer without dry-hopping using the two different techniques. Specifically, they are positioned in opposite directions in the 2nd and 4th quadrants, denoted as BF and BH, respectively. Interestingly, the dry-hopped beers, regardless of the type of hops used, are clustered together in the 3rd quadrant. This observation could be attributed to the technique, wherein the second cold hopping phase may lead to the release of certain compounds (such as terpenes)or an increase in others do not present in beers without dry-hopping (BF and BH).

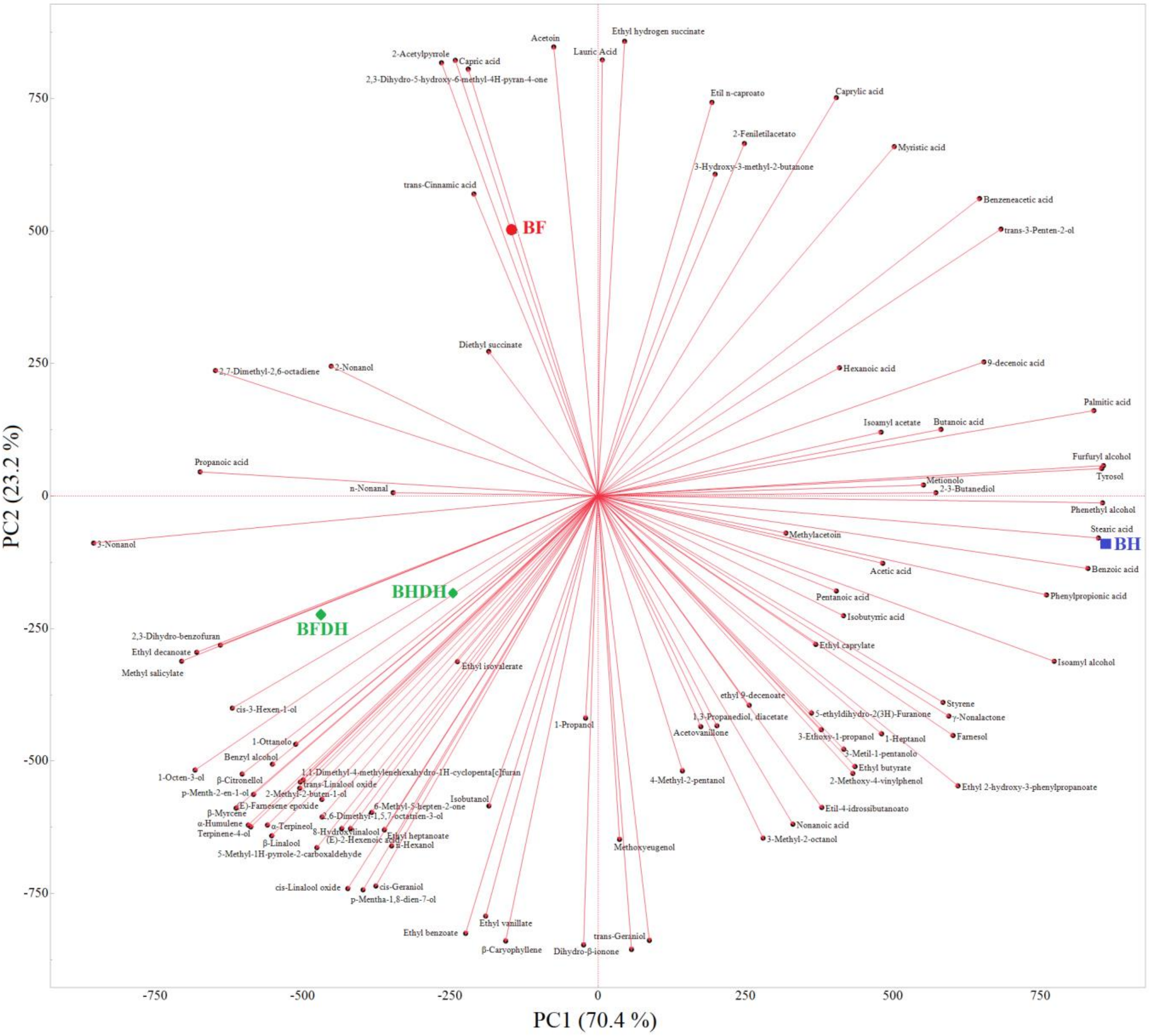


Figure 1. Biplot of Principal component analysis (PCA) of the volatile organic compounds (VOCs) of the beers. Beer with freeze-dried hop added during the boiling phase (BF); Beer with freeze-dried hop added during the boiling phase and dry-hopping (BFDH); Beer with hot-stove dried hop added during the boiling phase (BH); Beer with hot-stove dried hop added during the boiling phase and dry-hopping (BHDH).

The data of the PCA, has also confirmed by the hierarchical cluster analysis (HCA) reported in Figure 2. The HCA showed how the two beers with the dry-hopping (BHDH and BFDH) cluster together and have a profile separated to the other as confirmed the strong effect of this techniques on the aromatic profile of beers confering a geater complexity profiler to the product such as terpene compounds.

The beer without dry-hopping (BF and BH), are clearly separed in two different cluster, and this is correlated to the techniques of drying of hop (F and H) which produce different aromatic compounds in the product, probably related to the drying temperature used.

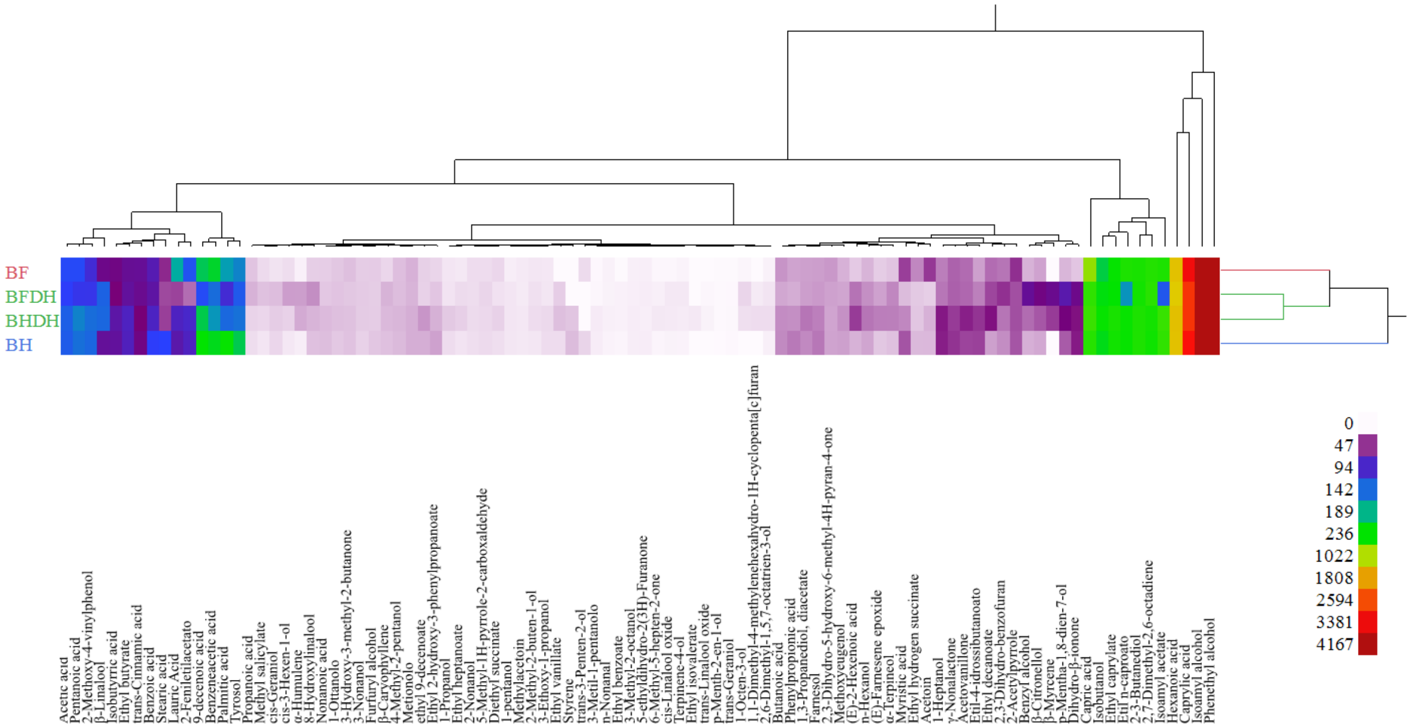


Figure 2. HCA of the volatile organic compounds (VOCs) of the beers. Beer with freeze-dried hop added during the boiling phase (BF); Beer with freeze-dried hop added during the boiling phase and dry-hopping (BFDH); Beer with hot-stove dried hop added during the boiling phase (BH); Beer with hot-stove dried hop added during the boiling phase and dry-hopping (BHDH).

3.3 Sensory evaluation

After sensory analysis data were elaborated to define sensory profile of the samples and peculiarities attributable to the different types of hopping (Figure 3a,b,c,d).

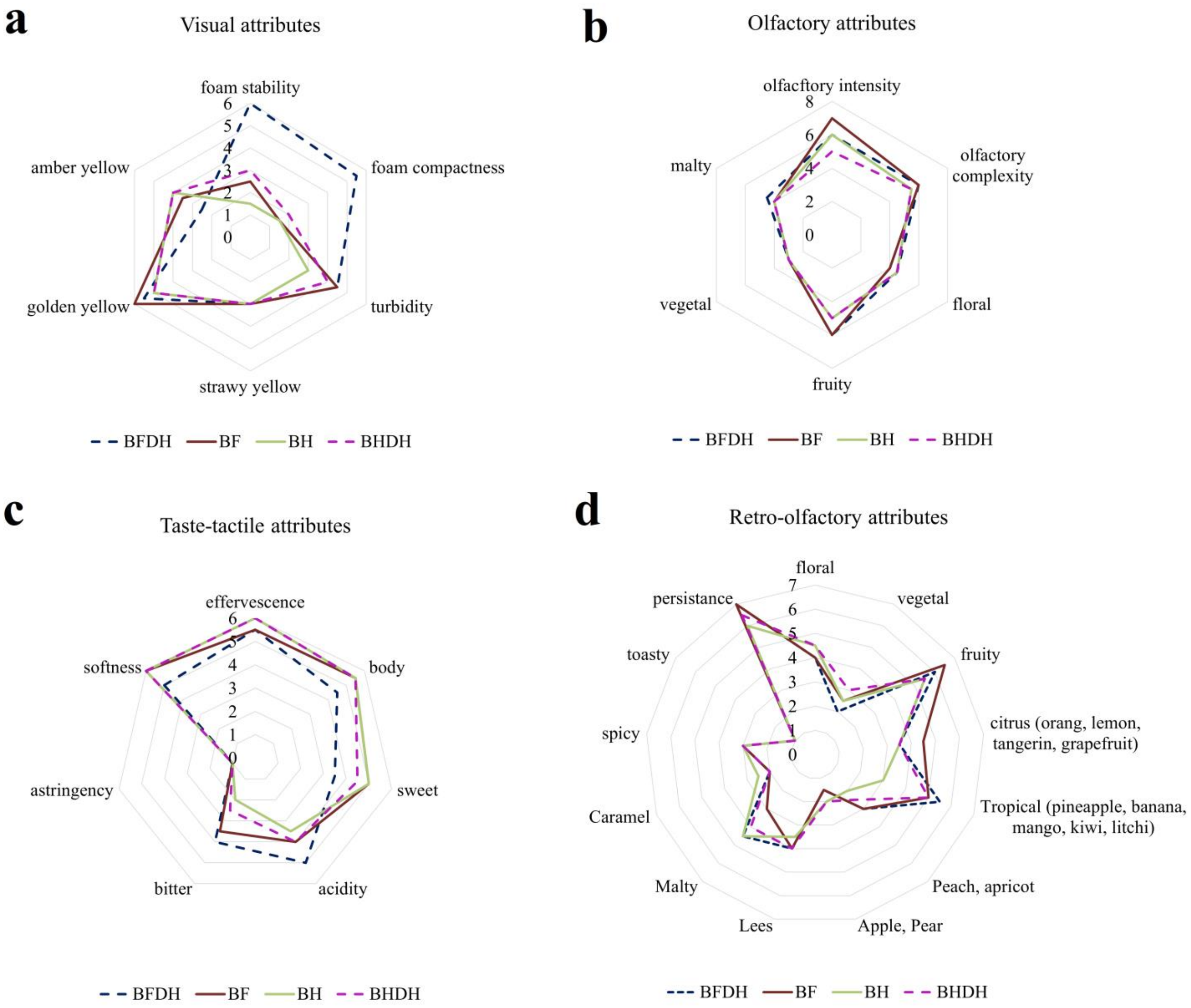


Figure 3. Sensory evaluation of beers: Visual attributes (**a**); Olfactory attributes (**b**);Taste-tactile attributes (**c**); Retro-olfactory attributes (**d**). Beer with freeze-dried hop added during the boiling phase (BF); Beer with freeze-dried hop added during the boiling phase and dry-hopping (BFDH); Beer with hot-stove dried hop added during the boiling phase (BH); Beer with hot-stove dried hop added during the boiling phase and dry-hopping (BHDH).

As general consideration it can be stated that very low scores for vegetal, apple and pear, caramel and toasty notes are common for all the samples, the same is for astringency in regard of the tactile perceptions as reported also in Medoro et al. (2016) for pilsner beer and from Carbone et al. (2021) working with the same variety of hops.

Considering BF sample, it was described as golden yellow with amber components and a present turbidity, foam was neither stable nor compact. The aroma profile at direct olfaction was the most intense and complex of the set characterized by a predominant fruity note. Together with most of the other samples, it was considered soft, medium effervescent, with a good sensation of body and sweetness. Acidity and bitterness were low, the same for the astringency. What already said for the aroma found confirmation in the retro-olfactory perception: it is persistent fruity and citrus notes prevailing upon the others.

Moving to the dry-hopped version BFDH resulted in more golden than amber, picked for foam compactness and stability and was as turbid a BF. The olfactory profile was less intense but as complex as the previous characterized by floral, fruity and malty notes. Taste wise this BFDH was the most acidic and bitter sample, this not only because of the composition but also because of lack of body, softness, and sweetness able to counterbalance these perceptions. The flavor profile was long as for BF and characterized by tropical fruits, malt, and lees scents.

The two beers produced with hot-stoved hop were the more ambered and least golden in color, with BH that was the poorest for foam stability and compactness, but also the less turbid. BHDH was comparable to BF in terms of foam and turbidity. In respect of the olfactory analysis, these two samples showed lower complexity and, particularly BHDH, also lower intensity, with a bouquet characterized by more floral than fruity notes. Considering mouth perception, hot-stove hop beers were the most effervescent and soft with the lower bitterness and, considering BH sample, the lower acidity. Considering body, softness and sweetness these two samples are aligned with BF samples. The retro-olfactive profile is less complex than those for freeze-dried hop beer, more vegetal and lees like for BHDH and more malty and caramel like for BH. Apple and pear, even if at low level, were perceived as flavors of these beers.

Moving to the liking, the least liked beer was BHDH for all the different aspects; the other three samples were generally liked evenly, with BH picking for retro-olfactory liking and balance (Figure 4).

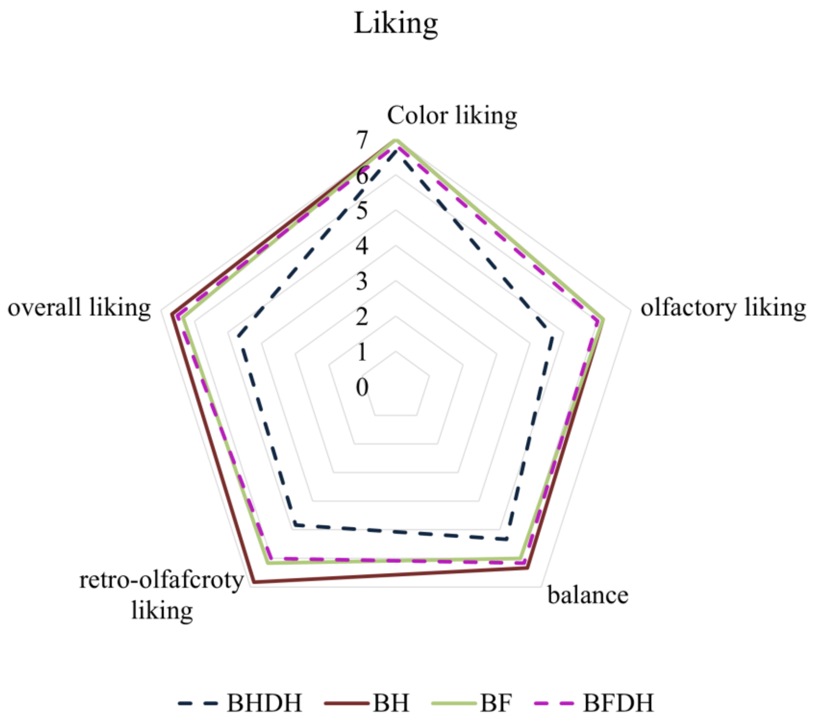


Figure 4. Sensory linking of beers. Beer with freeze-dried hop added during the boiling phase (BF); Beer with freeze-dried hop added during the boiling phase and dry-hopping (BFDH); Beer with hot-stove dried hop added during the boiling phase (BH); Beer with hot-stove dried hop added during the boiling phase and dry-hopping (BHDH).

4. Conclusions

The research activity carried out showed a clear demarcation, both aromatic and sensory, between the beers produced with different hop drying technique. Hops dried with hot stove produced beer with high ethanol and titratable acidity and low in volatile acidity. Color of the beer was lighter using hot-stove dried hops due to the polyphenol oxidation occurred in hop freeze dried. Dry-hopping has emerged as a powerful tool for brewers to modify the sensory profile and overall quality of beer. Through the addition of hops during fermentation or post-fermentation, brewers can enhance aroma, flavor, and consumer satisfaction. Beers produced with the dry-hopping technique, had a significantly higher content in terpenes especially in beer from freeze-dried hops . BFDH resulted in more golden than amber, picked for foam compactness and stability and was as turbid a BF. The olfactory profile was less intense but as complex as the previous characterized by floral, fruity and malty notes. Taste wise this BFDH was the most acidic and bitter sample. Finally, this paper highlighted the chemical changes which occurred between drying techniques and using hop-drying and we cannot say what it was better because it depends on the consumer taste. We emphasized that beers were different in chemical features, VOCs content and diversity, and in sensory analysis but none of them showed off-flavor and off-odor.

CRediT author statement

Edoardo Monacci: Formal analysis, Investigation. **Federico Baris:** Formal analysis, Investigation. **Alessandro Bianchi:** Conceptualization, Methodology, Validation, Formal analysis, Investigation, Data Curation, Writing - Original Draft, Writing - Review & Editing, Visualization. **Fosca Vezzulli**: Formal analysis, Investigation, Writing - Original Draft. **Stefano Pettinelli:** Formal analysis, Investigation. **Milena Lambri:** Methodology,Validation, Data Curation, Writing - Review & Editing. **Fabio Mencarelli:** Conceptualization, Methodology, Resources, Writing - Original Draft, Writing - Review & Editing, Visualization,. **Fabio Chinnici:** Validation, Resources, Data Curation, Writing - Original Draft, Writing - Review & Editing. **Chiara Sanmartin:** Conceptualization, Methodology, Validation, Writing - Review & Editing. Supervision. All authors have read and agreed to the published version of the manuscript.

Declaration

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

Data availability: Data will be made available on request.

References

Algazzali, V., & Shellhammer, T. (2016). Bitterness Intensity of Oxidized Hop Acids: Humulinones and Hulupones. *Journal of the American Society of Brewing Chemists*, *74*(1), 36–43. https://doi.org/10.1094/ASBCJ-2016-1130-01

Bamforth, C. W. (2016). *Brewing materials and processes: A practical approach to beer excellence* (Charles W Bamforth (ed.)). Academic Press, San Diego, USA.. https://doi.org/10.1016/C2013-0-13349-1

Bianchi, A., Sanmartin, C., Taglieri, I., Macaluso, M., Venturi, F., Napoli, M., Mancini, M., Fabbri, C., & Zinnai, A. (2023a). Effect of Fertilization Regime of Common Wheat (*Triticum aestivum*) on Flour Quality and Shelf-Life of PDO Tuscan Bread. *Foods*, *12*(14), 2672. https://doi.org/10.3390/foods12142672

Bianchi, A., Santini, G., Piombino, P., Pittari, E., Sanmartin, C., Moio, L., Modesti, M., Bellincontro, A., & Mencarelli, F. (2023b). Nitrogen maceration of wine grape: An alternative and sustainable technique to carbonic maceration. *Food Chemistry*, *404*, 134138. https://doi.org/10.1016/j.foodchem.2022.134138

Bianchi, A., Venturi, F., Zinnai, A., Taglieri, I., Najar, B., Macaluso, M., Merlani, G., Angelini, L. G., Tavarini, S., Clemente, C., & Sanmartin, C. (2023c). Valorization of an Old Variety of *Triticum aestivum*: A Study of Its Suitability for Breadmaking Focusing on Sensory and Nutritional Quality. *Foods*, *12*(6), 1351. https://doi.org/10.3390/foods12061351

Bianchi, A., Venturi, F., Palermo, C., Taglieri, I., Angelini, L. G., Tavarini, S., & Sanmartin, C. (2024). Primary and secondary shelf-life of bread as a function of formulation and MAP conditions : Focus on physical-chemical and sensory markers. *Food Packaging and Shelf Life*, *41*(November 2023), 101241. https://doi.org/10.1016/j.fpsl.2024.101241

Bocquet, L., Sahpaz, S., Hilbert, J. L., Rambaud, C., & Rivière, C. (2018). *Humulus lupulus* L., a very popular beer ingredient and medicinal plant: overview of its phytochemistry, its bioactivity, and its biotechnology. *Phytochemistry Reviews*, *17*(5), 1047–1090. https://doi.org/10.1007/s11101-018-9584-y

Brányik, T., Vicente, A. A., Dostálek, P., & Teixeira, J. A. (2008). A Review of Flavour Formation in Continuous Beer Fermentations. *Journal of the Institute of Brewing*, *114*(1), 3–13. https://doi.org/10.1002/j.2050-0416.2008.tb00299.x

Carbone, K., Bianchi, G., Petrozziello, M., Bonello, F., Macchioni, V., Parisse, B., De Natale, F., Alilla, R., & Cravero, M. C. (2021). Tasting the Italian Terroir through Craft Beer: Quality and Sensory Assessment of Cascade Hops Grown in Central Italy and Derived Monovarietal Beers. *Foods*, *10*(9), 2058. https://doi.org/10.3390/foods10092085

Castro-Marín, A., Buglia, A. G., Riponi, C., & Chinnici, F. (2018). Volatile and fixed composition of sulphite-free white wines obtained after fermentation in the presence of chitosan. *Lwt*, *93*(March), 174–180. https://doi.org/10.1016/j.lwt.2018.03.003

Donadini, G., Fumi, M. D., & Lambri, M. (2013). A preliminary study investigating consumer preference for cheese and beer pairings. *Food Quality and Preference*, *30*(2), 217–228. https://doi.org/10.1016/j.foodqual.2013.05.012

Donaldson, B. R., Bamforth, C. W., & Heymann, H. (2012). Sensory Descriptive Analysis and Free-Choice Profiling of Thirteen Hop Varieties as Whole Cones and after Dry Hopping of Beer. *Journal of the American Society of Brewing Chemists*, *70*(3), 176–181. https://doi.org/10.1094/ASBCJ-2012-0710-01

Gomes, F. de O., GuimarÃes, B. P., Ceola, D., & Ghesti, G. F. (2022). Advances in dry hopping for industrial brewing: a review. *Food Science and Technology*, *42*. https://doi.org/10.1590/fst.60620

Hauser, D. G., Simaeys, K. R. Van, Lafontaine, S. R., & Shellhammer, T. H. (2019). A Comparison of Single-Stage and Two-Stage Dry-Hopping Regimes. *Journal of the American Society of Brewing Chemists*, *77*(4), 251–260. https://doi.org/10.1080/03610470.2019.1668230

Heřmánek, P., Rybka, A., & Honzík, I. (2018). Determination of moisture ratio in parts of the hop cone during the drying process in belt dryer. *Agronomy Research*, *16*(3), 723–727. https://doi.org/10.15159/AR.18.076

Jespersen, L., & Jakobsen, M. (1996). Specific spoilage organisms in breweries and laboratory media for their detection. *International Journal of Food Microbiology*, *33*(1), 139–155. https://doi.org/10.1016/0168-1605(96)01154-3

Karam, M. C., Petit, J., Zimmer, D., Baudelaire Djantou, E., & Scher, J. (2016). Effects of drying and grinding in production of fruit and vegetable powders: A review. *Journal of Food Engineering*, *188*, 32–49. https://doi.org/10.1016/j.jfoodeng.2016.05.001

Klimczak, K., Cioch-Skoneczny, M., & Duda-Chodak, A. (2023). Effects of Dry-Hopping on Beer Chemistry and Sensory

Properties—A Review. *Molecules*, *28*(18), 6648. https://doi.org/10.3390/molecules28186648

Kobayashi, N., Sato, M., Fukuhara, S., Yokoi, S., Kurihara, T., Watari, J., Yokoi, T., Ohta, M., Kaku, Y., & Saito, T. (2007). Application of Shotgun DNA Microarray Technology to Gene Expression Analysis in Lager Yeast. *Journal of the American Society of Brewing Chemists*, *65*(2), 92–98. https://doi.org/10.1094/ASBCJ-2007-0319-02

Koren, D., Hegyesné Vecseri, B., Kun-Farkas, G., Urbin, Á., Nyitrai, Á., & Sipos, L. (2020). How to objectively determine the color of beer? *Journal of Food Science and Technology*, *57*(3), 1183–1189. https://doi.org/10.1007/s13197-020-04237-4

Lafontaine, S. R., & Shellhammer, T. H. (2018). Impact of static dry-hopping rate on the sensory and analytical profiles of beer. *Journal of the Institute of Brewing*, *124*(4), 434–442. https://doi.org/https://doi.org/10.1002/jib.517

Lafontaine, S. R., & Shellhammer, T. H. (2019). Investigating the factors impacting aroma, flavor, and stability in dry-hopped beers. *MBAA Technical Quarterly*, *56*(1), 13–23. https://doi.org/10.1094/TQ-56-1-0225-01

Mastrangelo, N., Bianchi, A., Pettinelli, S., Santini, G., Merlani, G., Bellincontro, A., Baris, F., Chinnici, F., & 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*(10), e20422. https://doi.org/10.1016/j.heliyon.2023.e20422

Maye, J. P., Smith, R., & Leker, J. (2016). Humulinone Formation in Hops and Hop Pellets and Its Implications for Dry Hopped Beers. *MBAA Technical Quarterly*, *53*(1), 23–27. https://doi.org/10.1094/TQ-53-1-0227-01

Maye, J. P., Smith, R., & Leker, J. (2018). Dry Hopping And Its Effects On Beer Bitterness, The IBU Test Beer Foam, and pH. *Brauwelt International*, *36*(1), 25–30. https://doi.org/10.1094/TQ-53-3-0808-01

Medoro, C., Cianciabella, M., Camilli, F., Magli, M., Gatti, E., Predieri, S., Medoro, C., Cianciabella, M., Camilli, F., Magli, M., Gatti, E., & Predieri, S. (2016). Sensory Profile of Italian Craft Beers, Beer Taster Expert versus Sensory Methods: A Comparative Study. *Food and Nutrition Sciences*, *07*(06), 454–465. https://doi.org/10.4236/fns.2016.76047

Michel, M., Cocuzza, S., Biendl, M., Peifer, F., Hans, S., Methner, Y., Pehl, F., Back, W., Jacob, F., & Hutzler, M. (2020). The impact of different hop compounds on the growth of selected beer spoilage bacteria in beer. *Journal of the Institute of Brewing*, *126*(4), 354–361. https://doi.org/10.1002/jib.624

Monacci, E., Sanmartin, C., Bianchi, A., Pettinelli, S., Taglieri, I., & Mencarelli, F. (2023). Plastic film packaging for the postharvest quality of fresh hop inflorescence (*Humulus lupulus*) cv. Cascade. *Postharvest Biology and Technology*, *206*(August), 112575. https://doi.org/10.1016/j.postharvbio.2023.112575

Morgan, C., & Vesey, G. (2009). Freeze-drying of microorganisms. In *Encyclopedia of Microbiology*. Elsevier, Amsterdam, The Netherlands. https://doi.org/10.1016/B978-012373944-5.00114-0

Nance, M. R., & Setzer, W. N. (2011). Volatile components of aroma hops (*Humulus lupulus* L.) commonly used in beer brewing.

Journal of Brewing and Distilling, *2*(April), 16–22. http://www.academicjournals.org/JBD

Neve, R. A. (2012). *Hops*. Springer Science & Business Media, New York, USA. https://doi.org/10.1007/978-94-011-3106-3

Oladokun, O., James, S., Cowley, T., Smart, K., Hort, J., & Cook, D. (2017). Dry-Hopping: The effects of temperature and hop variety on the bittering profiles and properties of resultant beers. *BrewingScience*, *70*(11–12), 187–196. https://doi.org/10.23763/BrSc17-18oladokun

Olaniran, A. O., Hiralal, L., Mokoena, M. P., & Pillay, B. (2017). Flavour-active volatile compounds in beer: production, regulation and control. *Journal of the Institute of Brewing*, *123*(1), 13–23. https://doi.org/10.1002/jib.389

Pahl, R., Meyer, B., & Biurrun, R. (2016). Chapter 6 - Wort and Wort Quality Parameters. In C. W. Bamforth (Ed.), *Brewing materials and processes : a practical approach to beer excellence* (pp. 113–121). Elsevier, Amsterdam, Netherlands,. https://doi.org/10.1016/B978-0-12-799954-8.00006-X

Pettinelli, S., Pardini, L., De Angeli, G., Bianchi, A., Najar, B., Cerreta, R., Bellincontro, A., Floridia, G., & Mencarelli, F. (2022). Innovative “Soft” Maceration Techniques in Red Grape Fermentation. *Beverages*, *8*(4), 62. https://doi.org/10.3390/beverages8040062

Raut, S., Gersdorff, G. J. E. von, Münsterer, J., Kammhuber, K., Hensel, O., & Sturm, B. (2020). Impact of Process Parameters and Bulk Properties on Quality of Dried Hops. *Processes*,*8*(11) 1507. https://doi.org/10.3390/pr8111507

Raut, S., von Gersdorff, G. J. E., Münsterer, J., Kammhuber, K., Hensel, O., & Sturm, B. (2021). Influence of pre-drying storage time on essential oil components in dried hops (*Humulus lupulus* L.). *Journal of the Science of Food and Agriculture*, *101*(6), 2247–2255. https://doi.org/10.1002/jsfa.10844

Romero-Rodríguez, R., Durán-Guerrero, E., Castro, R., Díaz, A. B., & Lasanta, C. (2022). Evaluation of the influence of the microorganisms involved in the production of beers on their sensory characteristics. *Food and Bioproducts Processing*, *135*, 33–47. https://doi.org/10.1016/j.fbp.2022.06.004

Rossini, F., Virga, G., Loreti, P., Iacuzzi, N., Ruggeri, R., & Provenzano, M. E. (2021). Hops (*Humulus lupulus* L.) as a Novel Multipurpose Crop for the Mediterranean Region of Europe: Challenges and Opportunities of Their Cultivation.*Agriculture*, *11*(6), 484. https://doi.org/10.3390/agriculture11060484

Rutnik, K., Ocvirk, M., & Košir, I. J. (2022). Impact of Hop Freshness on Dry Hopped Beer Quality.*Foods*, *11*(9). https://doi.org/10.3390/foods11091310

Rybacek, V. (2012). *Hop production* (V. Rybacek (ed.)). Elsevier Science, Amsterdam, The Netherlands.

Rybka, A., Krofta, K., Heřmánek, P., Honzík, I., & Pokorný, J. (2018). Effect of drying temperature on the content and composition of hop oils. *Plant, Soil and Environment*, *64*(10), 512–516. https://doi.org/10.17221/482/2018-PSE

Salamon, R. V, Dabija, A., Ferencz, Á., Tankó, G., Ciocan, M. E., & Codină, G. G. (2022). The Effect of Dry Hopping Efficiency on β-Myrcene Dissolution into Beer.*Plants*, *11*(8), 1043. https://doi.org/10.3390/plants11081043

Steinhaus, M., & Schieberle, P. (2000). Comparison of the Most Odor-Active Compounds in Fresh and Dried Hop Cones (*Humulus lupulus* L. Variety Spalter Select) Based on GC−Olfactometry and Odor Dilution Techniques. *Journal of Agricultural and Food Chemistry*, *48*(5), 1776–1783. https://doi.org/10.1021/jf990514l

Steyer, D., Tristram, P., Clayeux, C., Heitz, F., & Laugel, B. (2017). Yeast strains and hop varieties synergy on beer volatile compounds. *BrewingScience*, *70*(9–10), 131–141. https://doi.org/10.23763/BrSc17-13Steyer

Stone, H., Sidel, J., Oliver, S., Woolsey, A., & Singleton, R. C. (2004). Sensory Evaluation by Quantitative Descriptive Analysis. In M. C. Gacula (Ed.), *Descriptive Sensory Analysis in Practice* (pp. 23–34). John Wiley & Sons, Ltd, Hoboken, New Jersey, USA. https://doi.org/10.1002/9780470385036.ch1c

Takoi, K., Koie, K., Itoga, Y., Katayama, Y., Shimase, M., Nakayama, Y., & Watari, J. (2010). Biotransformation of Hop-Derived Monoterpene Alcohols by Lager Yeast and Their Contribution to the Flavor of Hopped Beer. *Journal of Agricultural and Food Chemistry*, *58*(8), 5050–5058. https://doi.org/10.1021/jf1000524

Van Cleemput, M., Cattoor, K., De Bosscher, K., Haegeman, G., De Keukeleire, D., & Heyerick, A. (2009). Hop (*Humulus lupulus*)-Derived Bitter Acids as Multipotent Bioactive Compounds. *Journal of Natural Products*, *72*(6), 1220–1230. https://doi.org/10.1021/np800740m

Van Opstaele, F., De Rouck, G., De Clippeleer, J., Aerts, G., & De Cooman, L. (2010). Analytical and Sensory Assessment of Hoppy Aroma and Bitterness of Conventionally Hopped and Advanced Hopped Pilsner Beers. *Journal of the Institute of Brewing*, *116*(4), 445–458. https://doi.org/10.1002/j.2050-0416.2010.tb00796.x

Van Simaeys, K. R., Féchir, M., Gallagher, A., Stokholm, A., Weaver, G., & Shellhammer, T. H. (2022). Examining Chemical and Sensory Differences of New American Aroma Hops Grown in the Willamette Valley, Oregon. *Journal of the American Society of Brewing Chemists*, *80*(4), 370–378. https://doi.org/10.1080/03610470.2021.1968271

Verzele, M., & De Keukeleire, D. (1991). *Chemistry and Analysis of Hop and Beer Bitter Acids* (Issue 27). Elsevier Science, Amsterdam, The Netherlands. https://books.google.it/books?id=YQkhAQAAMAAJ

Vezzulli, F., Bertuzzi, T., Rastelli, S., Mulazzi, A., & Lambri, M. (2021). Sensory profile of Italian Espresso brewed Arabica Specialty Coffee under three roasting profiles with chemical and safety insight on roasted beans. *International Journal of Food Science & Technology*, *56*(12), 6765–6776. https://doi.org/10.1111/ijfs.15380

Wang, D., Zhang, M., Ju, R., Mujumdar, A. S., & Yu, D. (2023). Novel drying techniques for controlling microbial contamination in fresh food: A review. *Drying Technology*, *41*(2), 172–189. https://doi.org/10.1080/07373937.2022.2080704

Yadav, B., & Roopesh, M. S. (2020). In-package atmospheric cold plasma inactivation of Salmonella in freeze-dried pet foods: Effect of inoculum population, water activity, and storage. *Innovative Food Science & Emerging Technologies*, *66*, 102543. https://doi.org/10.1016/j.ifset.2020.102543

Declaration of interests

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

☐The authors declare the following financial interests/personal relationships which may be considered as potential competing interests:

Graphical Abstract

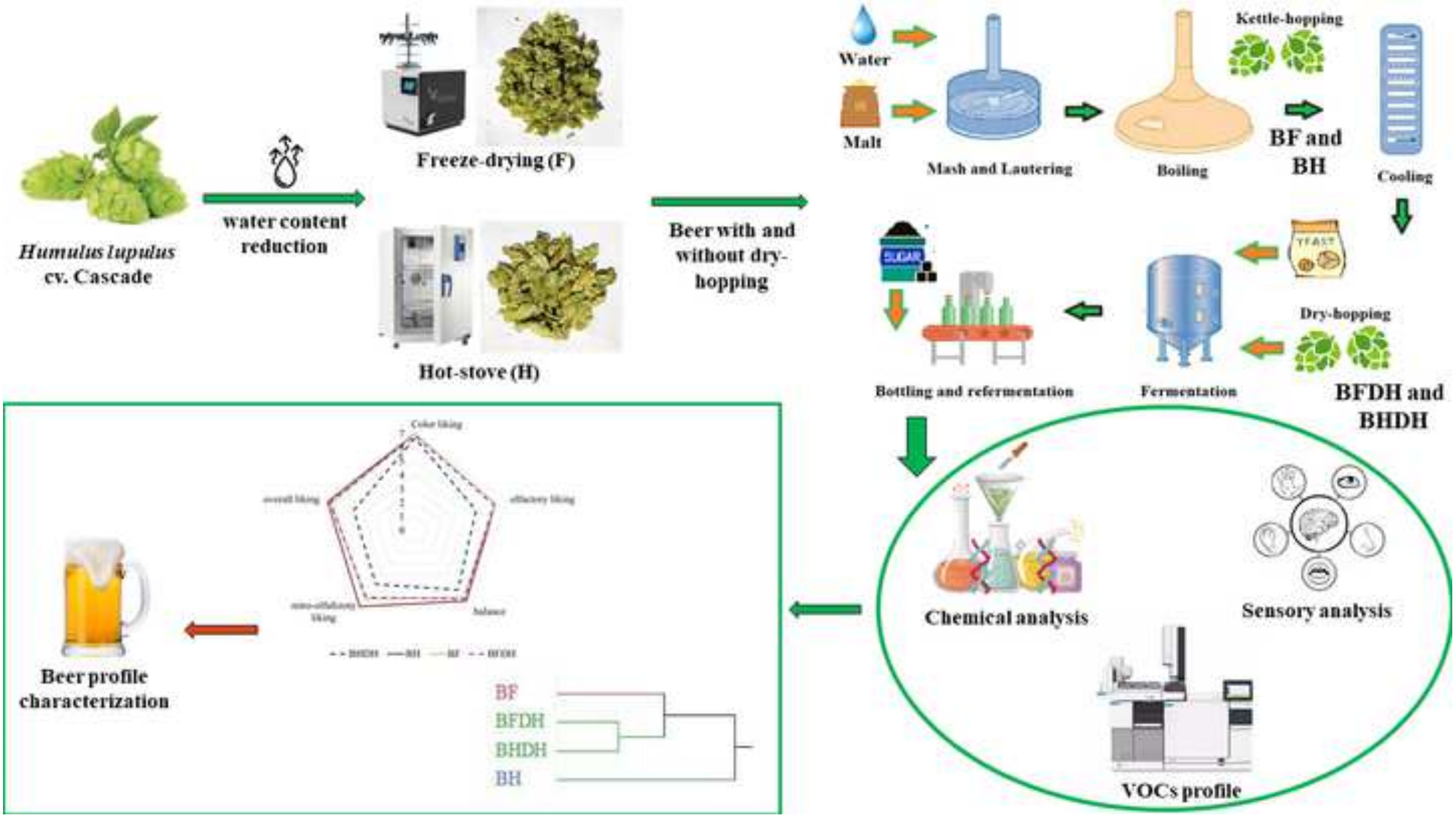


Figure 1

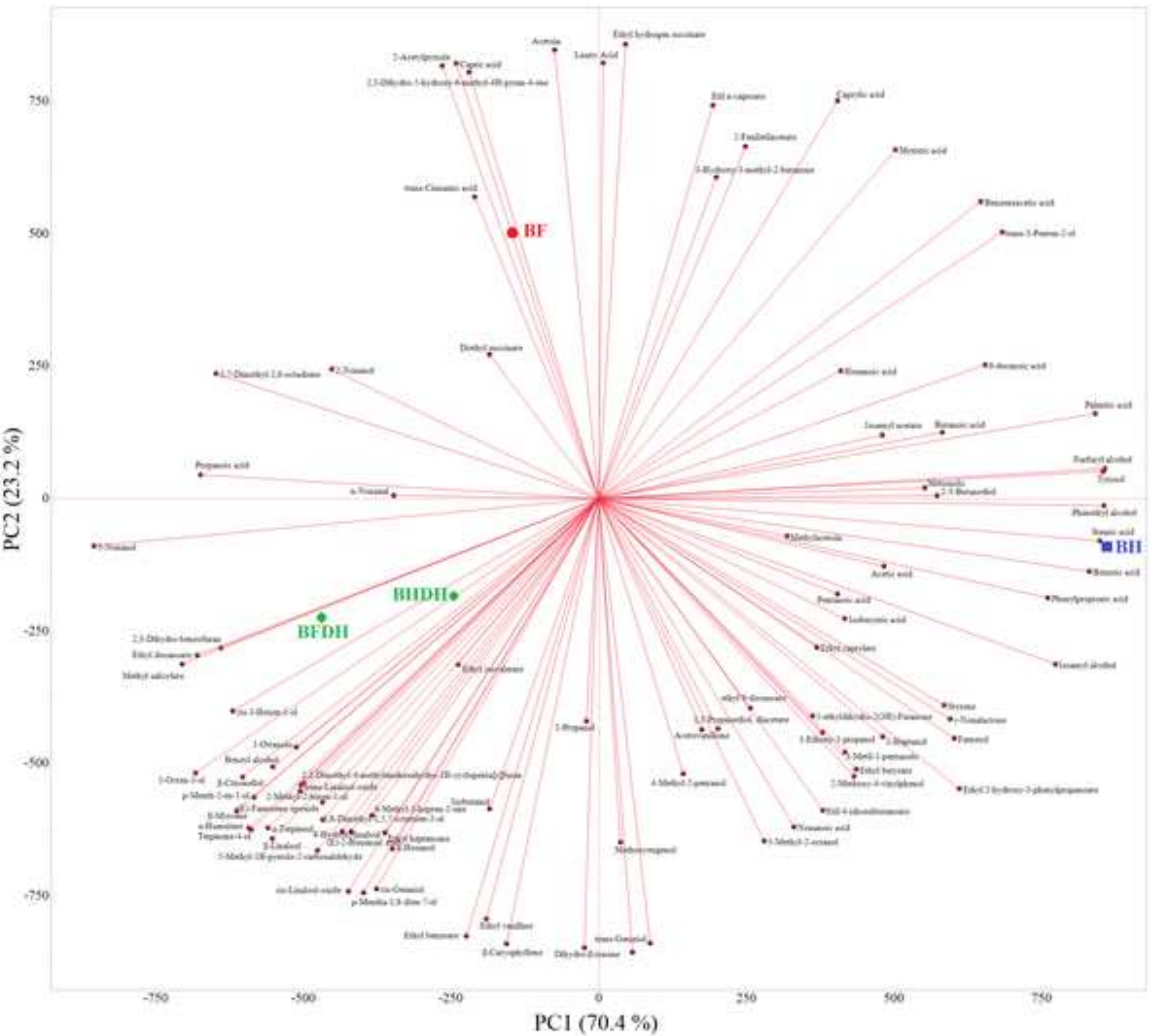

PC2 (23.2 %)
PC1 (70.4 %)
750
500
250
0
-250
-500
-750
-750
-500
-250
0
250
500
750
BF
BH
BHDH
BFDH
2-Acetylpyrrole
Capric acid
2,3-Dihydro-3-hydroxy-6-methyl-4H-pyran-4-one
Acetoin
Lauric Acid
Ethyl hydrogen succinate
Ethyl n-caproate
Caprylic acid
3-Hydroxy-3-methyl-2-butanone
Myristic acid
trans-Cinnamic acid
Benzeneacetic acid
trans-3-Penten-2-ol
Diethyl succinate
1,7-Dimethyl-2,8-octadiene
2-Nonanol
Propanoic acid
n-Nonanal
Palmitic acid
Isoamyl acetate
Butanoic acid
Furfuryl alcohol
Eugenol
2,3-Butanediol
Phenethyl alcohol
Sorbic acid
Acetic acid
Phenylpropanoic acid
Isobutyric acid
Ethyl caprylate
Isoamyl alcohol
2,3-Dihydro-benzofuran
Ethyl decanoate
Methyl salicylate
Ethyl succinate
cis-3-Hexen-1-ol
1-Octen-3-ol
Benzyl alcohol
β-Citronellol
p-Menth-1-en-1-ol
trans-Linalool oxide
2-Methyl-2-hepten-1-ol
(E)-Farnesene epoxide
β-Myrcene
α-Humulene
Terpinen-4-ol
α-Terpineol
β-Linalool
6-Methyl-5-hepten-2-one
Isoborneol
Ethyl heptanoate
1-Hexanol
cis-Linalool oxide
cis-Geraniol
Ethyl vanillate
Ethyl benzoate
β-Caryophyllene
Dihydro-β-ionone
trans-Geraniol
Methoxyeugenol
2-Propanol
Acetovanillone
1,3-Propanediol, diacetate
4-Methyl-2-pentanol
Styrene
γ-Nonalactone
Farnesol
2-Heptanol
3-Methyl-1-pentanol
Ethyl butyrate
2-Methoxy-4-vinylphenol
Nonanoic acid
3-Methyl-2-octanol

Figure 2

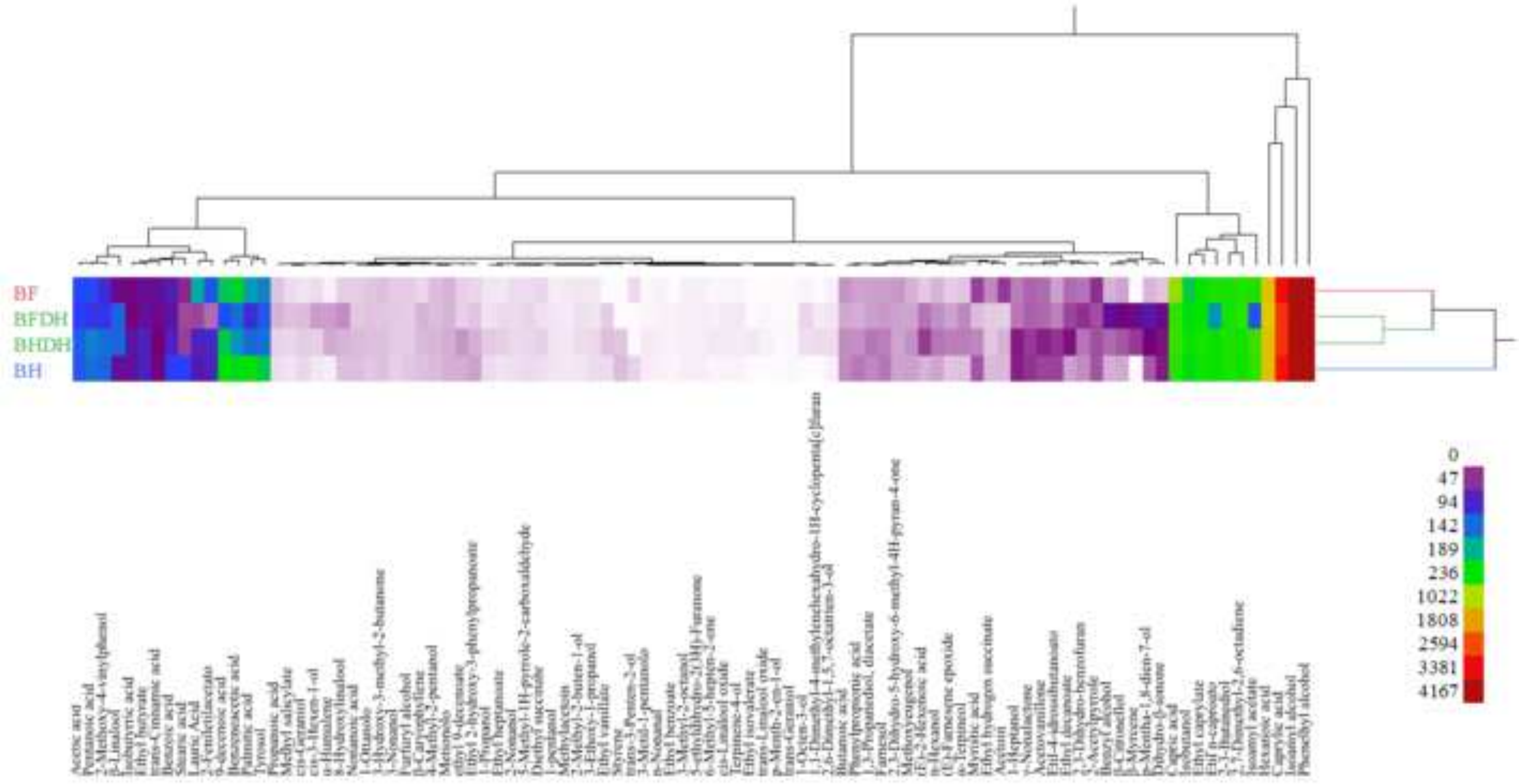
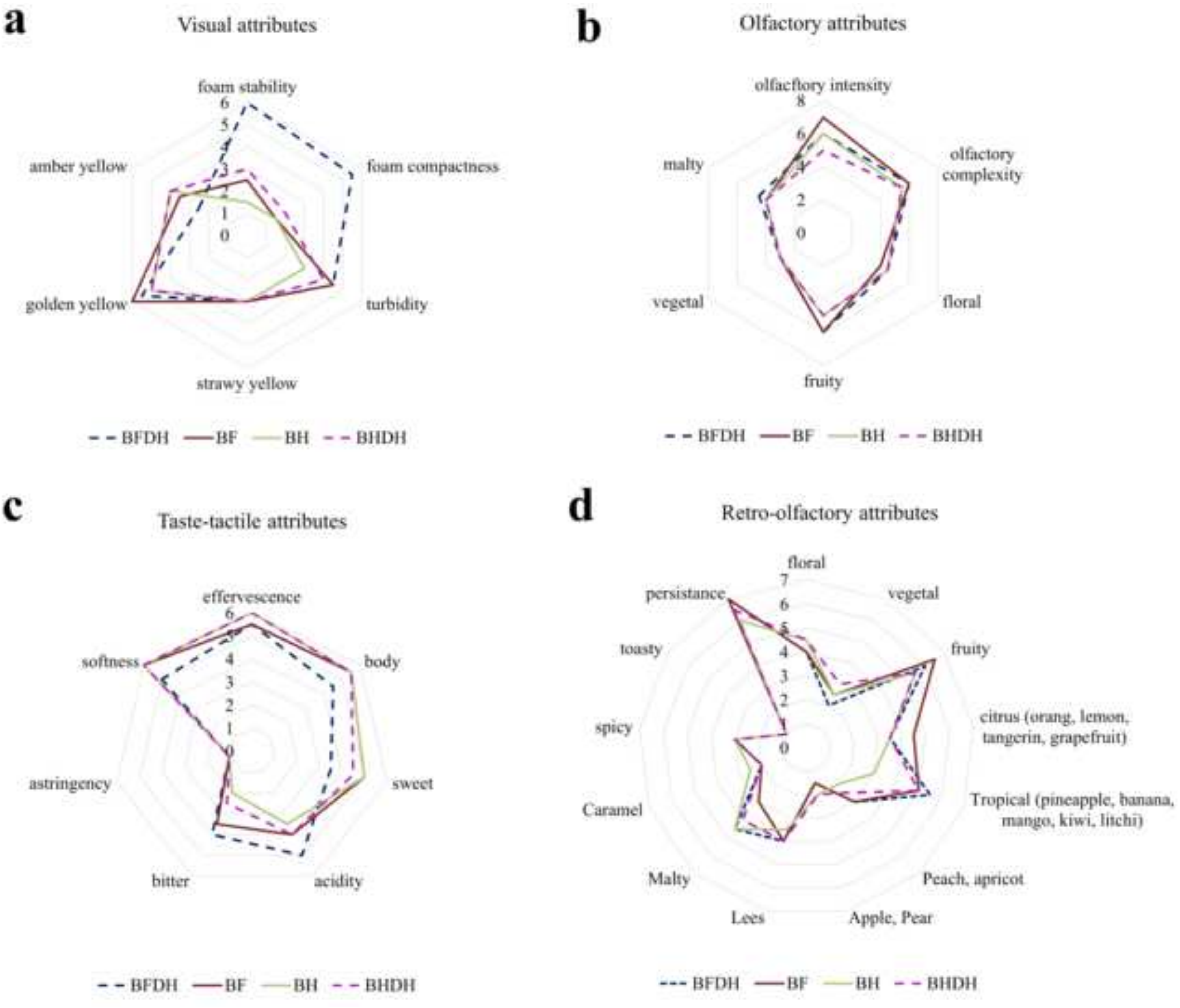
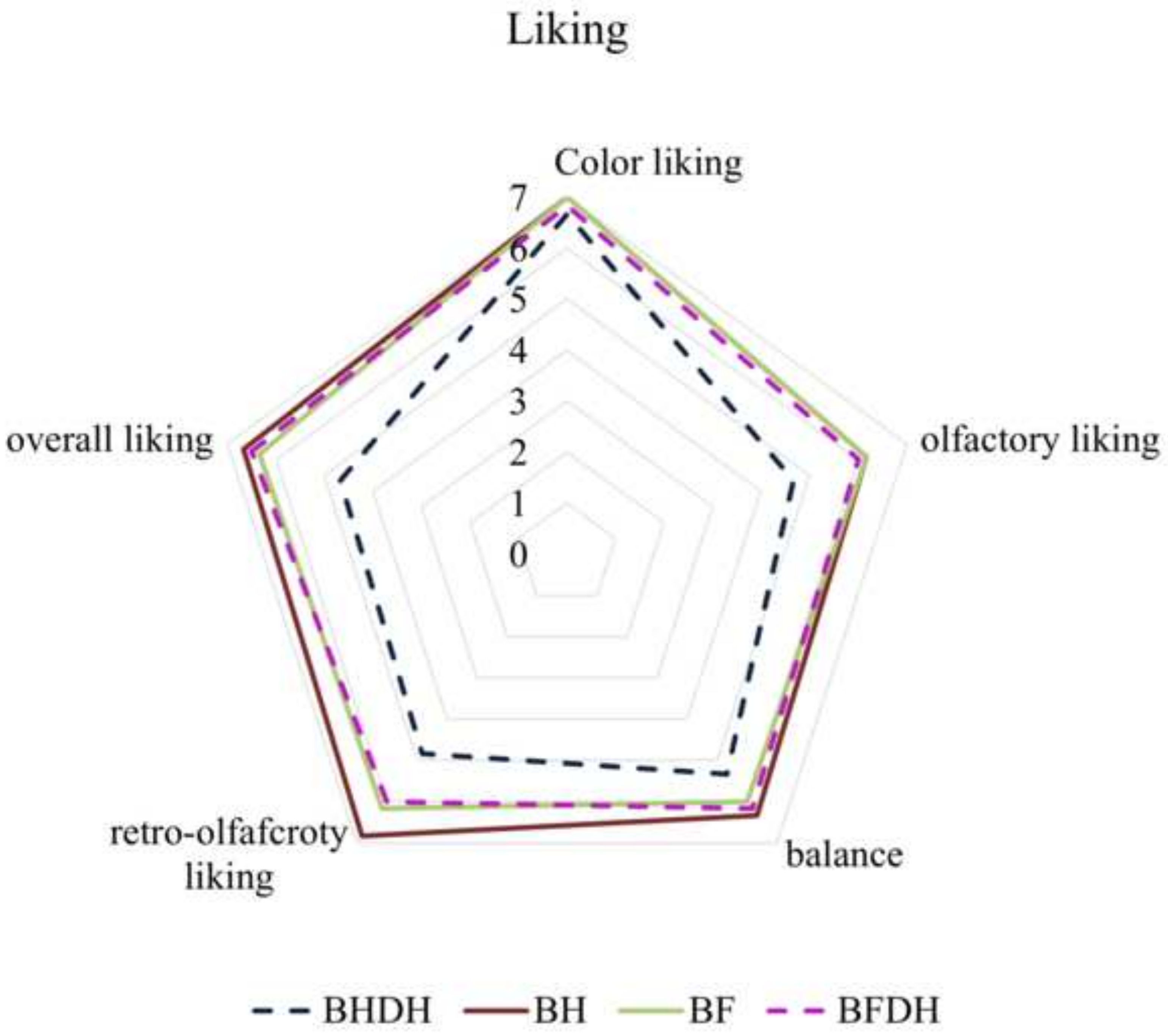

BF
BFDH
BHDH
BH
0
47
94
142
189
236
1022
1808
2594
3381
4167
Acetic acid
Pentanoic acid
2-Methoxy-4-vinylphenol
β-Linalool
Isobutyric acid
Ethyl butyrate
trans-Cinnamic acid
Benzoic acid
Stearic acid
Lauric Acid
2-Feniletilacetato
9-decenoic acid
Benzeneacetic acid
Palmitic acid
Tyrosol
Propanoic acid
Methyl salicylate
cis-Geraniol
cis-3-Hexen-1-ol
α-Humulene
8-Hydroxylinalool
Nonanoic acid
1-Ottanolo
1-Hydroxy-3-methyl-2-butanone
1-Nonanol
Furfuryl alcohol
β-Caryophyllene
4-Methyl-2-pentanol
Methionol
ethyl 9-decenoate
Ethyl 2-hydroxy-3-phenylpropanoate
1-Propanol
Ethyl heptanoate
2-Nonanol
5-Methyl-1H-pyrrole-2-carboxaldehyde
Diethyl succinate
1-pentanol
Methylacetoin
2-Methyl-2-buten-1-ol
3-Ethoxy-1-propanol
Ethyl vanillate
Styrene
trans-3-Penten-2-ol
3-Metil-1-pentanolo
n-Nonanal
Ethyl benzoate
3-Methyl-2-octanol
5-ethyldihydro-2(3H)-Furanone
6-Methyl-5-hepten-2-one
cis-Linalool oxide
Terpinen-4-ol
Ethyl isovalerate
trans-Linalool oxide
p-Menth-2-en-1-ol
trans-Geraniol
1-Octen-3-ol
1,1-Dimethyl-1,4-methylenehexahydro-1H-cyclopenta[c]furan
2,6-Dimethyl-1,5,7-octatrien-3-ol
Butanoic acid
Phenylpropionic acid
1,3-Propanediol, diacetate
Farnesol
2,3-Dihydro-5-hydroxy-6-methyl-4H-pyran-4-one
Methoxyeugenol
(E)-2-Hexenoic acid
n-Hexanol
(E)-Farnesene epoxide
α-Terpineol
Myristic acid
Ethyl hydrogen succinate
Acetoin
1-Heptanol
γ-Nonalactone
Acetovanillone
Etil-4-idrossibutanoato
Ethyl decanoate
2,3-Dihydro-benzofuran
2-Acetylpyrrole
Benzyl alcohol
β-Citronellol
β-Myrcene
p-Mentha-1,8-dien-7-ol
Dihydro-β-ionone
Caproic acid
Isobutanol
Ethyl caprylate
Etil n-caproato
2,3-Butanediol
3,7-Dimethyl-1,3,6-octadiene
Isoamyl acetate
Hexanoic acid
Caprylic acid
Isoamyl alcohol
Phenethyl alcohol

Figure 3


a
Visual attributes
foam stability
foam compactness
turbidity
strawy yellow
golden yellow
amber yellow
0
1
2
3
4
5
6
BFDH
BF
BH
BHDH
b
Olfactory attributes
olfacftory intensity
olfactory complexity
floral
fruity
vegetal
malty
0
2
4
6
8
BFDH
BF
BH
BHDH
c
Taste-tactile attributes
effervescence
body
sweet
acidity
bitter
astringency
softness
0
1
2
3
4
5
6
BFDH
BF
BH
BHDH
d
Retro-olfactory attributes
floral
vegetal
fruity
citrus (orang, lemon, tangerin, grapefruit)
Tropical (pineapple, banana, mango, kiwi, litchi)
Peach, apricot
Apple, Pear
Lees
Malty
Caramel
spicy
toasty
persistance
0
1
2
3
4
5
6
7
BFDH
BF
BH
BHDH


Liking
Color liking
olfactory liking
balance
retro-olfafcroty liking
overall liking
7
6
5
4
3
2
1
0
BHDH
BH
BF
BFDH