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# Impact of Hop Residue Reuse on the Chemical and Sensory Properties of Craft Beer

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**Abstract:** Hops are an important component of beer brewing, providing aromatic and bittering properties that are essential to consumer appeal. A significant amount of hop residue is generated in the dry-hop brewing process that cannot be reused due to bittering residues that disqualify them as animal feed or other products. The purpose of this research was to reuse four varieties of hop waste (Citra, Mosaic, Hallertau Blanc, and Mandarina Bavaria) through a repalletization process with the objective of integrating them into a new craft beer brewing process. Chemical properties such as the phenolic content, antioxidant capacity, and α- and β-acids were significantly reduced ($p < 0.05$) due to the reuse of the repelletized hops, leading to a decrease in the bitterness levels in all of the craft beers brewed with dry-hop residues. Finally, the sensory study conducted with non-habitual craft beer consumers revealed significant general acceptability for beers brewed with repelletized dry-hop residues (Mandarina Bavaria, Citra, and Mosaic). The reuse of hop residues for brewing presents a promising opportunity for further development in the food industry.

**Keywords:** craft beer; HPLC; aroma; flavor; bitterness



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## 1. Introduction

Beer is an ancestral alcoholic beverage widely consumed in the world [1]. Over time, the definition of beer has evolved as man's understanding of the beverage has broadened, and it is now described as a product of the transformation of barley malt and hop-based wort by yeast under controlled conditions [2]. In many countries, the craft brewing industry has experienced a rapid expansion in the number of breweries and has gained market share from the large global breweries [3]. The global beer market was valued at USD 768.55 billion in 2022 and is projected to reach a value of USD 996.49 billion by 2030. The global market is projected to grow, exhibiting a compound annual growth rate (CAGR)

of 3.30% during the forecast period [4]. The unique sensory properties of craft beers take place thanks to the incredible creative freedom of producers, who experiment with a great diversity of hops, malts, yeasts, and other unconventional materials such as fruits, honeys, and aromatic herbs in order to build consumer loyalty and differentiate themselves from their competitors [5,6].

Beer hop residues have gained attention for their potential in recovering bioactive compounds. These wastes, generated in large quantities by the brewing industry, contain a variety of valuable compounds such as polyphenols, antioxidants, and pigments [4]. The utilization of byproducts as a valuable product stream in the high-volume brewing industry has been extensively researched and applied to optimize the environmental and economic sustainability [7]. There is a growing interest to investigate the utilization of brewery byproducts among craft breweries [8]. The brewing process produces several byproducts, including spent grain, spent yeast, and hops (*Humulus lupulus* L.) [9]. Hops are primarily known as an aromatizing ingredient in beer, with the added benefits of antioxidant potential and antimicrobial properties [10]. Research on hops has generally focused on its bittering, aromatic, and preservative properties [11,12].

The most valuable hop compounds for the brewing industry are hop acids, essential oils, and flavonoids [13]. Hop acids are referred to as α- and β-acids (also known as humulones and lupulones, respectively). In their pure state, hop acids occur as pale-yellow solids. They are weak acids, poorly soluble in water, and have almost no bitter taste [14]. When hops are used at the beginning of the brewing process, their essential oils volatilize, imparting bitterness in the beer. This is ideal for beer styles that emphasize the aromas of other adjuncts, such as fruits or aromatic herbs. Conversely, when hops are added at the end of the boil or later stages of production, essential oils are retained, giving the beer a strong aromatic component from the hops. This is ideal for "hoppy beers", where the hop aroma is the main component of the sensory profile of the final product [15,16].

In order to enhance the hop aroma in beers, the brewing industry frequently uses the technique known as "dry hopping", which consists of adding additional hops during the fermentation and maturation stages. This technique makes it possible to extract the aromatic components from the solid fraction of hops using the alcohol naturally present in the beer. These hops, with distinctive and unique aroma characteristics, are grown by producers in several countries: Amarillo, Citra, Mosaic and Sorachi Ace (Washington, DC, USA); (Hallertau Blanc, Polaris and Mandarina Bavaria (Hallertau, Germany); Nelson Sauvin (Nelson, New Zealand) [17].

Dry hopping is usually performed at cold temperatures (below room temperature) to minimize the solubilization of α-acids and their isomerization to iso-α-acids. This approach allows for the extraction of volatile components without significantly increasing the bitterness of the product [17,18]. The present study aims to take advantage of the hop residue through the repelletizing process to insert it in a second process of craft beer production with sensory characteristics acceptable to consumers. Obtaining the new hop residues would not only reduce environmental pollution but also offer opportunities to develop value-added products in various industries, such as food, pharmaceuticals, and cosmetics.

## 2. Materials and Methods

### *2.1. Materials and Reagents*

For the Pale Ale-type barley malt (Bestmaltz, Bessenbach, Germany), the yeast used was US-05 (Fermentis, Aubagne, France), and the following five types of hops: Cascade, Citra, Mosaic, Hallertau Blanc, and Mandarina Bavaria (Yakima Chief Hops, Washington, DC, USA).

### 2.2. Craft Beer Brewing

The brewing process of the craft beer is shown in Figure 1. The malt grains of the Pale Ale variety (12 kg) were subjected to a milling process in a crown grinder.

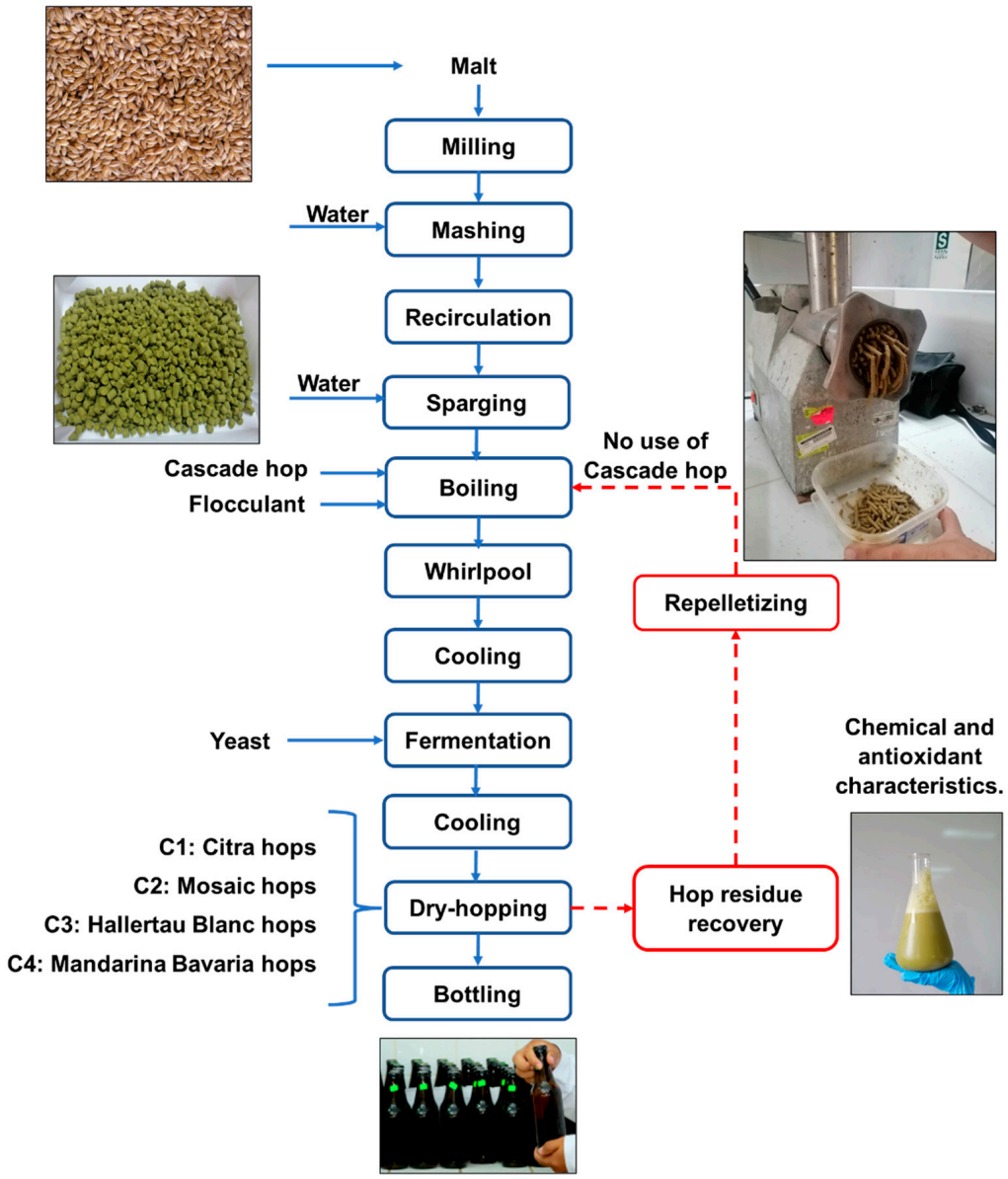


**Figure 1.** Flow chart for craft beer brewing and the reuse of residual hops.

Using a stainless-steel pot, the malt was mixed with water (40.25 L) at 70 °C and stirred vigorously for 60 min. Subsequently, the wort or mash was then collected by opening a pipe installed at the bottom of the stainless-steel pot, allowing it to flow back into the pot. The recirculation was repeated until the must had a clear, crystalline appearance with no remaining traces of grain. Next, the must was separated from the wet grain by opening the lower spout of the mash pot and draining all of the sweetened liquid into a second stainless-steel pot (boiling pot). Additionally, a batch washing was performed by adding water (30.75 L) at 70 °C to the mash pot still containing the wet grain. Then, the diluted mash (wet grain/washing water) was stirred using a stainless-steel shovel. The recirculation stage was repeated until the drained mash showed no trace of grains. The diluted mash was collected in the boiling pot through the lower spout of the mash pot. In the boiling stage, the wort collected from the previous stages was heated to a temperature

of 100 °C, at which time Cascade hop (CH) pellets were added as bittering hops. This temperature was maintained for a period of 60 min. At minute 45 of the boiling process, unflavored gelatin (20 g) was added as a flocculating agent. Once the boiling stage was finished in the boiling pot, the whirlpool was performed, where the bitter wort was stirred in a clockwise circular motion with a stainless-steel paddle for 10 min. Using a food-grade plate chiller (SS Brewtech, 1TBG-225-Glycol, San Diego, USA), the sour wort was cooled to a temperature of 25 °C. With the help of sanitary hoses connected to a stainless-steel head pump (Xinxishan, MP-15RM, Shijiazhuang, China), the fluid was conveyed to a conical fermenter (SS Brewtech, California, USA) with a maximum capacity equal to 26.5 L. The fermentation stage began with the addition of commercial yeast (SafAle US-05) and lasted for 7 days at a controlled temperature of 21 °C. After the fermentation process, the cooling system was activated by means of a glycol chiller, circulating a cooling liquid through the fermenter coil system to reduce the temperature to 7 °C for 14 days. On the seventh day of the cold conditioning stage, dry hopping was performed. This process consisted of purging the settled yeast in the fermenter cone to condition the experimental hop pellets (Citra, Mosaic, Hallertau Blanc, and Mandarina Bavaria). At the end of the 14 days, the dry-hop residues were separated and the beer obtained was collected in a stainless-steel pot, to which 250 g of blonde sugar was added for the natural carbonation process. The mixture was dissolved, and finally it was bottled using a bottle filler tube and a manual capping machine.

### 2.3. Repelletizing of Dry-Hop Residues

After 14 days of cold conditioning in the brewing process (first batch), the dry-hop waste was collected from the fermenter cone by using a sterile Erlenmeyer flask wrapped in aluminum foil to prevent the oxidation of any component of the byproduct and stored cold for preservation. A filter screen was used to separate the beer from the collected hop waste, manually squeezing out as much liquid as possible, leaving a hop paste. The residual paste was dried at room temperature for a period of 3 days until the optimum consistency for repelletizing was obtained. To obtain new pellets, the hop paste was standardized to a moisture content of 12% and fed to a pelletizer (Twothousand®, TJ22B, Shenzhen, China) with dimensions of 210 × 240 × 450 mm, nozzle diameter of 0.2 cm, and productivity of 120 kg/h. The new hop pellets were spread on a smooth surface and left to dry at room temperature for one day. The newly produced hop pellets were vacuum-packed in high-density polyethylene bags using SHIELD equipment (DZ-300/PD, Zhejiang, China), and then stored at room temperature until use for the production of new batches of beer at the brewing stage, following the usual brewing process.

### 2.4. Chemical Characterization

The moisture content was determined using an oven (POL-EKO-APARATURA®, SW115STD, Bielsko-Biała, Poland) according to AOAC 931.04 [19]. The ash was determined by incinerating organic matter at 650 °C for 3 h in a muffle (THERMOLYNE, 347034984, Waltham, MA, USA) according to AOAC 923.03 [20]. The Dumas method was used to determine the protein content according to the AOAC 990.03 method [21]. The fat content was analyzed according to Manirakiza et al. [22] in a Soxhlet fat extractor (FOSS, Soxtec TM-2043, Waltham, MA, USA) using petroleum ether (CDH Fine Chemical, Gurugram, India) as a solvent. Finally, the content of other compounds was determined by the following equation: % Carbohydrates = 100% − % moisture − % ash − % protein − % fat.

### 2.5. Determination of α- and β-Acids by HPLC

To obtain the extracts for the hop samples, 200 mg of each was used and placed in contact with 20 mL of methanol in an ultrasonic bath (Branson Ultrasonics, CPX5800H-E,

Danbury, CT, USA) for 20 min. The samples were then filtered and taken to a rotary evaporator (IKA, RV 10C S000, Staufen, Germany) at a reduced pressure and a constant temperature of 40 °C until they were dry. The samples were resuspended at 1 mg/mL with methanol, filtered through 0.2 µm syringe filters, and deposited into amber vials. An amount of 20 µL of the filtered sample was then injected into the HPLC equipment (Hitachi CM, Düsseldorf, Germany), which entered the C18 column stationary phase and with the help of the mobile phase, as follows: Solution A: water + formic acid (0.1%) and Solution B: acetonitrile with 0.1% formic acid; the flow rate was 1 mL/min and chromatograms were obtained at a wavelength of 280 nm. The run was by gradient with the following percentages: 0 min (55% A + 45% B), 2 min (55% A + 45% B), 12 min (25% A + 75% B), 17 min (5% A + 95% B), 30 min (5% A + 95% B), 35 min (10% A + 90% B), 40 min (55% A + 45% B), and 45 min (55% A + 45% B). The HPLC system consisted of a pump (CM 5160), autosampler (CM 5260), column oven (CM 5310), and diode array detector (CM 5430). A C18 column (250 mm, 5 mm, and 4 µm) was used as the stationary phase. A standard dilution of α-acids (Cohumulone and N+adhumulone) and β-acids (Colupulone and N+adlupulone) at 2 mg/mL was prepared with methanol. Sufficient aliquots were extracted for standard preparation at concentrations of 0.125, 0.25, 0.50, 1.5, and 2 mg/mL. They were then passed through a 0.2 µm syringe filter and deposited in amber vials for analysis. The samples were analyzed in triplicate (this profile was visualized in Figure 2) and the calibration curves obtained for the alpha and beta acid standards were as follows: Cohumulone ($Y = 15664541.9X + 742863.687$; $R^2 = 0.9996$), N+adhumulone ($Y= 35135534.6X +1161599.780$; $R^2 = 0.9998$), Colupulone ($Y = 10082454X + 801840.423$; $R^2 = 0.9989$), and N+adlupulone ($Y = 8617967.22X + 391922.702$; $R^2 = 0.9996$), where X is the concentration (mg/mL) and Y is the area under the curve. The results of α- and β-acids were expressed in mg/g hops.

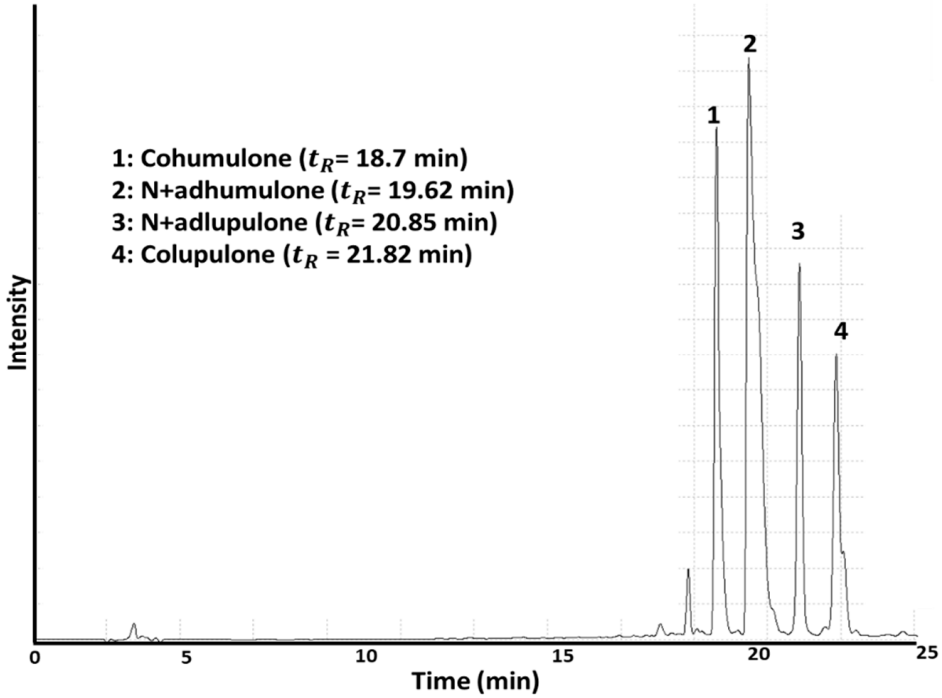


**Figure 2.** Chromatographic profile of α- and β-acids in hops; $t_R$: time retention.

### *2.6. Determination of Total Phenolic Compounds*

The total phenolic compound content of the hop extracts was determined using the Folin–Ciocalteu assay and adapted from Saura-Calixto et al. [23]. First, solutions of gallic acid ($C_7H_6O_5$) at a concentration of 450 µg/mL (0.0225 g of $C_7H_6O_5$ reagent in 50 mL of distilled water), 20% sodium carbonate ($Na_2CO_3$) (2 g of $Na_2CO_3$ in 10 mL of distilled water sonicated for 5 min), and 2N Folin–Ciocalteu reagent were prepared. From the $C_7H_6O_5$

solution (450 µg/mL), different concentrations were formulated for the construction of a calibration curve (7.2, 14.4, 21.6, 28.8, and 36 µg/mL). In Eppendorf tubes, 20, 40, 60, 60, 80, and 100 µL volumes of $C_7H_6O_5$ were added and 100 µL of Folin–Ciocalteu 2N was added (the mixture was allowed to stand for 5 min); then, 50 µL $Na_2CO_3$ (20%) was added to all of the tubes to be completed with distilled water (1080, 1060, 1040, 1020, and 1000 µL, respectively, to each tube). After 1 h of resting, 200 µL of each tube was added to a microplate well to be read at a wavelength of 730 nm on a Synergy™ H1 multimode microplate reader (BioTek Instruments, Inc., Cheadle, UK). The calibration curve was as follows: $Y = 0.0567X - 0.0664$ ($R^2 = 0.994$), where X is mg GAE/mL and Y is the absorbance.

The phenolic extracts of the hops were obtained by using 25 mL test tubes. In each test tube, 2 g of hops (with and/or without repelletizing) were mixed with 10 mL of a methanol/water solution (50/50) at a pH = 2 (2N HCl solution); then, the mixture was subjected to an ultrasonic bath for 30 min, followed by centrifugation (Sigma, model 2-16P, Osterode am Harz, Germany) at 3000 rpm for 20 min. The supernatant obtained after centrifugation was considered the phenolic extract of the hops and stored under refrigeration (T = 5 °C). Subsequently, 800 µL of the phenolic extract sample was added in an Eppendorf tube and mixed with 100 µL of Folin–Ciocalteu 2N. The mixture was then rested for 5 min. An amount of 50 µL of $Na_2CO_3$ (20%) plus 300 µL of distilled water was added. Again, the mixture was left to stand for 2 h in the dark. Finally, 200 µL of the prepared mixture was taken and the absorbance was measured in the multimodal microplate reader. The total phenolic compound results were expressed as mg of gallic acid equivalent in one gram of hops on a dry basis (mg GAE/g d.b) and were performed in triplicate for each sample.

### 2.7. Antioxidant Capacity

The antioxidant activity in the hops was determined using the method developed by Kim et al. [24] with modifications. Solutions of Trolox at a concentration of 1 mM (0.0125 g of Trolox reagent in 50 mL of 80% methanol) and DPPH at a concentration of 1 mmol (3.9 mg of DPPH reagent dissolved in 100 mL of 80% methanol) were prepared. The DPPH solution was prepared in an amber glass bottle as a protection against light and homogenized on a magnetic stirrer (VELP Scientifica®, ARE, Usmate Velate, Italy) for one hour for refrigerated storage (5 °C); this preparation was performed at the time of analysis. A calibration curve was constructed from the 1 mM Trolox solution at different concentrations (500, 400, 200, 100, 50, 25, 10, and 5 µM); then, 10 µL of each concentration was taken and 190 µL of the DPPH solution was added, allowed to stand for 10 min, and measured at 515 nm in the multimodal microplate reader Synergy™ H1 (BioTek Instruments, Inc., Cheadle, UK). The calibration curve was as follows: $Y = 0.0683X + 6.4908$ ($R^2 = 0.9965$), where X is µmol Trolox/mL and Y is % DPPH. Finally, the phenolic extracts of the hops (10 µL) were reacted with the DPPH radical (190 µL), and the same reaction procedure was followed to obtain the calibration curve, as previously described. The final antioxidant capacity concentrations (µmol Trolox/g b.s) were determined in triplicate on a dry basis.

### 2.8. Determination of International Bitterness Units (IBUs)

The bitterness unit was determined according to the ASBC method Beer-23A [25]. Beer (5 mL) was transferred into a 50 mL centrifuge tube and acidified with 3N HCl (0.5 mL). Isooctane (10 mL) was added and the mixture was shaken by hand three times before extraction on a rolled bed for 15 min. The mixture was centrifuged (Sigma, model 2-16P, Germany) at 3000 rpm twice for 5 min each time to aid the phase separation. An aliquot of the clear isooctane layer was transferred into a cuvette, and absorbance was measured with a spectrophotometer (Jasco, V-670, Tokyo, Japan) at 275 nm against a blank

of orthophosphoric acid and isooctane. The recorded absorbance was multiplied by an empirical factor of 50 to give IBU values in mg/L.

### 2.9. Sensory Analysis

The sensory analysis of the beer was performed with a set of 120 untrained panelists. The samples were previously refrigerated (T = 6.00 ± 0.50 °C). The following descriptors were analyzed: general appearance, aroma, bitterness, sweetness, color, hop flavor, turbidity, malt flavor, alcohol carbonation, and foam persistence. The panelists received a sensory primer with a continuous range of preference and/or level of acceptance, where 1 = very low/significant dislike, 5 = medium/neither like nor dislike and 7 = very high/significant like. Using a completely randomized design (CRD), the data were treated by an analysis of variance (ANOVA) to determine the significant differences in the factor, the type of craft beer. Using a Tukey test ($p < 0.05$), significant differences between treatments (types of craft beer) were determined with the use of Minitab® statistical software version 19.1.1.0.

### 2.10. Statistical Analysis

For the statistical analysis of the chemical characterization, antioxidant capacity, total phenolic compounds, and α- and β-acids in the different types of hops, as well as the bitterness level (IBU) in the craft beers brewed, a completely randomized design (CRD) and analysis of variance (ANOVA) were employed to assess the significance ($p < 0.05$) of the factor (type of hops). Tukey's test ($p < 0.05$) was used to identify the difference between treatments using Minitab® version 19.1.1.0 statistical support software.

## 3. Results and Discussion

### 3.1. Characterization of Hop Residues

Table 1 shows the comparison between the chemical characteristics of residual hop mash after brewing in the first batch (L1, L2, L3, and L4) and its repelletized hop residues (R1, R2, R3, and R4) on a dry basis. In general, the results showed that all of the residual hop pastes exhibited a significant effect of repelletizing on the moisture content in the samples ($p < 0.05$). For the other chemical characteristics, the effects were partial. For example, in the case of the carbohydrate and fat content, the effect of repelleting ($p < 0.05$) was observed only between L2 and R2. Similarly, for the protein content, the effect was observed between L1 and R1. However, for the ash content, no effect of repelleting was observed, with the exception of the L2 and R2 samples. In addition, it was noted that CH exhibited differences in the ash content compared to all other hop pastes but showed no differences in the fat content with any of them. These results were compared to the chemical composition reported in previous studies for dried hop cones, where the comparison with moisture (10%) was close, higher in protein (15%), and lower with respect to ash (8%) [26–30]. In relation to carbohydrates, the values are between 68 and 71% (Table 1), which is in agreement with Almaguer et al. [11]. According to this author, hops can contain up to 45% cellulose, monosaccharides, and pectins, in addition to approximately 7% fiber. As for the fat content, previous studies have shown that their composition includes resins, which contain α-acids, the main component responsible for the bitter taste of beer [31].

**Table 1.** Chemical characterization of the hops and repelletized residues.

| Hop | Moisture | Ashes * | Proteins * | Fat * | Carbohydrates * |
|---|---|---|---|---|---|
| CH | 7.826 ± 0.058 $^{g}$ | 3.564 ± 0.100 $^{c}$ | 20.429 ± 0.252 $^{b}$ | 6.743 ± 0.153 $^{a}$ | 69.624 ± 0.137 $^{cd}$ |
| L1 | 62.961 ± 0.002 $^{a}$ | 2.674 ± 0.066 $^{d}$ | 20.221 ± 0.250 $^{b}$ | 5.592 ± 0.156 $^{abc}$ | 71.412 ± 0.150 $^{ab}$ |
| R1 | 9.801 ± 0.002 $^{e}$ | 1.822 ± 0.002 $^{e}$ | 21.981 ± 0.268 $^{a}$ | 4.781 ± 0.156 $^{bc}$ | 71.534 ± 0.700 $^{a}$ |
| L2 | 60.296 ± 0.002 $^{c}$ | 4.025 ± 0.016 $^{a}$ | 20.354 ± 0.288 $^{b}$ | 6.888 ± 0.161 $^{a}$ | 68.733 ± 0.453 $^{d}$ |
| R2 | 6.918 ± 0.000 $^{h}$ | 4.080 ± 0.009 $^{a}$ | 21.863 ± 0.321 $^{ab}$ | 4.009 ± 1.639 $^{c}$ | 70.420± 0.418 $^{abc}$ |
| L3 | 64.587 ± 0.004 $^{b}$ | 3.800± 0.020 $^{b}$ | 21.180 ± 0.209 $^{b}$ | 6.288 ± 0.158 $^{ab}$ | 69.623 ± 0.497 $^{cd}$ |
| R3 | 8.866 ± 0.002 $^{f}$ | 4.155 ± 0.003 $^{a}$ | 20.324 ± 0.161 $^{b}$ | 6.167 ± 0.210 $^{ab}$ | 69.458 ± 0.388 $^{cd}$ |
| L4 | 59.339 ± 0.001 $^{d}$ | 1.790 ± 0.017 $^{e}$ | 21.069 ± 0.386 $^{a}$ | 6.058 ± 0.213 $^{ab}$ | 70.171 ± 0.683 $^{bc}$ |
| R4 | 7.747 ± 0.000 $^{g}$ | 3.623 ± 0.006 $^{c}$ | 20.289 ± 0.215 $^{ab}$ | 5.381 ± 0.053 $^{abc}$ | 69.927 ± 0.339 $^{cd}$ |

CH: Cascade hop; L: residual paste; R: repelletizing; 1: Mandarina Bavaria; 2: Citra; 3: Mosaic; 4: Hallertau Blanc. Different letters present significant statistical differences ($p < 0.05$). * On a dry basis.

### *3.2. Total Phenolic Compounds and Antioxidant Capacity*

Table 2 shows that the phenolic compound content of CH was higher than all hops (C1, C2, C3, and C4). The effect of the brewing process generated a significant loss of phenolic compounds on the repelletized hops R1, R2, R3, and R4 ($p < 0.05$), as the phenolic compounds of the hops are transferred to the beer. Similarly, the antioxidant capacity presented the same reduction trend, explained by the effect of the boiling temperature in the brewing process. However, the antioxidant capacity was not proportional to the content of phenolic compounds for the different hops; a striking example is shown in the content of total phenolic compounds of the Mandarina Bavaria and Citra hops (C1 < C2), since their values were different with respect to the antioxidant capacity (C1 > C2). This same behavior was observed in the residues of the repelletized hops, as the content of phenolic compounds R1, R2, R3, and R4 did not present a significant difference ($p < 0.05$). However, in the antioxidant capacity, the difference was in the order R2 > R1 > R3. Terpinc et al. [32] point out that the correlation between the content of phenolic compounds and the antioxidant capacity can be negative. It is important to note that several phenolic compounds found in hops are transferred to beer, influencing its overall flavor, particularly its fullness, astringency, and colloidal formation. The oxidation of flavonoids can impact the astringency, turbidity, and color, while low-molecular-weight phenols, such as 4-vinylsyringol, can impart off-flavors to the beer during storage [33]. The levels of phenolic compounds for the repelletized hops (Table 2) were lower than those presented by Petrón et al. [34], who determined values for beer lees (Yellow/Citra/Simcoe hop compounds) values between 5.841 and 10.339 mg GAE/g, which provided a radical scavenging activity (RSA: %) between 73.383 and 91.041% by using the DPPH method. Also, these values (R1, R2, R3, and R4) were slightly lower than by Ruiz-Ruiz et al. [35], who indicated the value of 150 mg GAE/100 g of discarded Cascade hops. The differences in the results at the level of phenolic compounds can be attributed to the technological process, since the temperature, pH, presence of microorganisms, and polar solvents can affect the polyphenols found in the beer matrix, thereby affecting their quantitative and qualitative composition [36].

**Table 2.** Characterization of the phenolic compounds and antioxidant capacity in hops.

| Hop | Total Phenolic Compounds (mg GAE/100 g) * | Antioxidant Capacity (μmol Trolox/g) * |
|---|---|---|
| CH | 229.333 ± 0.751 $^{a}$ | 496.467 ± 2.060 $^{ab}$ |
| C1 | 216.023 ± 4.240 $^{b}$ | 504.963 ± 17.589 $^{a}$ |
| R1 | 121.583 ± 1.178 $^{c}$ | 286.269 ± 24.125 $^{c}$ |
| C2 | 227.135 ± 1.373 $^{a}$ | 463.669 ± 7.699 $^{a}$ |
| R2 | 123.802 ± 4.685 $^{c}$ | 343.669 ± 19.558 $^{b}$ |
| C3 | 221.288 ± 2.603 $^{ab}$ | 326.449 ± 14.713 $^{bc}$ |
| R3 | 121.940 ± 5.517 $^{c}$ | 148.509 ± 10.090 $^{d}$ |
| C4 | 214.792 ± 4.215 $^{b}$ | 348.835 ± 9.588 $^{b}$ |
| R4 | 118.138 ± 1.319 $^{c}$ | 328.171 ± 10.754 $^{bc}$ |

CH: Cascade hop; C: dry-hop control; R: repelletizing; 1: Mandarina Bavaria; 2: Citra; 3: Mosaic; 4: Hallertau Blanc. Different letters present significant statistical differences ($p < 0.05$). * On a dry basis.

### *3.3. IBU Values in Craft Beers and Content of α- and β-Total Acids in Hop Residues*

Beer bitterness was evaluated by calculating the IBU values (Table 3). As expected, all of the beers formulated with the dry-hop and repelletizing processes had a higher bitterness value than the beer obtained with the CH. The effect of the repelletizing process significantly influenced the IBU values of the dry hop ($p < 0.05$), while the IBU values of the beers obtained with C1 and C2 dry hops presented higher and significant ($p < 0.05$) values than the beers obtained with their residual repelletized hops. The beer produced by C1 and C2 presented the highest IBU values, which indicated that the hops derived by Mandarina Bavaria and Citra confer a higher bitterness to the final product. The IBU values for CH, C3, C4, R1, R2, R3, and R4 were within the range of bitterness observed in 34 commercial lager beers (8–36 mg/L) from different countries (Australia, Belgium, Cuba, Czech Republic, Denmark, France, Germany, Hungary, Italy, Netherlands, Poland, Peru, Romania, South Africa, Turkey, the UK, and the USA) according to the study conducted by Oladokun et al. [37].

**Table 3.** International Bitterness Unit values in beer.

| Beer | IBU (mg/L) |
|---|---|
| CH | 17.290 ± 4.079 $^{d}$ |
| C1 | 41.197 ± 5.799 $^{a}$ |
| C2 | 40.588 ± 3.727 $^{ab}$ |
| C3 | 27.427 ± 1.106 $^{cd}$ |
| C4 | 29.255 ± 3.499 $^{bc}$ |
| R1 | 28.342 ± 2.921 $^{cd}$ |
| R2 | 18.095 ± 5.031 $^{cd}$ |
| R3 | 23.998 ± 3.935 $^{cd}$ |
| R4 | 26.407 ± 5.212 $^{cd}$ |

CH: Cascade hop; C: dry-hop control; R: repelletizing; 1: Mandarina Bavaria; 2: Citra; 3: Mosaic; 4: Hallertau Blanc. Different letters present significant statistical differences ($p < 0.05$).

Table 4 presents the α- and β-acid content for the different hop treatments in craft brewing, where CH corresponds to the Cascade hops or the starting hops for conventional brewing, C symbolizes the hops used in the dry-hop method to obtain craft beer, L corresponds to the residues of the hops used in the dry-hop method, and R is the repelletized residues that are finally used in the new craft brewing process. First of all, it can be observed that the cohumulone content in the CH was higher for all of the dry-hop hops, even in the residual and repelletized hops. Likewise, for the N+adhumulone content, the dry-hop C1 hop was superior to all of the treatments (including the residual and repelletized hops). Schindler et al. [38] indicate that the α-acid contents for the hops on a dry basis should be

between 5 and 7%, as shown in Table 4 (α-acids = Cohumulone + N+adhumulone), and it can be seen that this was met for CH (5.3%), C2 (5.2%), and C4 (5.7%); however, for C1 (20%) and C3 (11%), this was not met. Another important finding was that N+adhumulone presented the majority α-acid in the dry-hop hops (control, residual, and repelletized), though this did not occur in CH. The effect of the treatments throughout the brewing process led to a significant reduction in α-acids due to the dry-hopping treatment. This reduction was not caused by the hop boiling, as the hop addition was performed cold. The reduction in cohumulone and N+adhumulone in the dry hops can be attributed to a combination of factors, such as the adsorption on the yeast and other solids, the chemical interactions with the beer compounds, microbial degradation, changes in the solubility and stability, oxidation, and environmental conditions such as the pH. These elements contribute to the complexity of the chemical and biological processes in beer, influencing the final composition of the product [16,39]. It should be noted that the addition of these hops aims to modify the bitterness (cohumulone), flavor, and aroma profile (N+adhumulone) of the final product [40]. The occurrence of bitter and aromatic characteristics in beer is strongly related to the chemical composition of the hops, the amount added, the type of hops, and the timing of hop dosing to the wort [41]. Conventionally, beer bitterness is achieved by adding hops to the hot wort at the beginning of the boil. The main reason for adding hops at this stage is to facilitate the thermal conversion of hop bitter acids (α-acids) into flavorful bitter compounds (iso-α-acids) and water-soluble bitterness. The yield of iso-α-acid increases with the boiling time, while most of the volatile compounds are lost through evaporation [15]. Iso-α-acids were not quantified in this study, as evidence suggests that conventional hopping (boiling) results in very low isomerization yields. Their utilization in cold hopping would be more complex. For example, at the end of boiling, less than 35–40% of the α-acids are typically transformed into iso-α-acids [42].

**Table 4.** Total α-acids and β-acids in the different types of hops (g/g).

| Hop | α-Acids * | | β-Acids * | |
|---|---|---|---|---|
| | Cohumulone | N+adhumulone | Colupulone | N+adlupulone |
| CH | $0.052 \pm 0.000^{a}$ | $0.001 \pm 0.000^{i}$ | $0.077 \pm 0.002^{a}$ | $0.084 \pm 0.000^{a}$ |
| C1 | $0.033 \pm 0.001^{a}$ | $0.167 \pm 0.001^{a}$ | $0.057 \pm 0.001^{b}$ | $0.067 \pm 0.002^{b}$ |
| C2 | $0.024 \pm 0.001^{f}$ | $0.028 \pm 0.000^{d}$ | $0.019 \pm 0.000^{f}$ | $0.039 \pm 0.001^{f}$ |
| C3 | $0.049 \pm 0.002^{b}$ | $0.062 \pm 0.001^{b}$ | $0.025 \pm 0.001^{e}$ | $0.047 \pm 0.003^{e}$ |
| C4 | $0.027 \pm 0.001^{e}$ | $0.030 \pm 0.002^{d}$ | $0.031 \pm 0.001^{c}$ | $0.062 \pm 0.001^{c}$ |
| L1 | $0.018 \pm 0.001^{g}$ | $0.024 \pm 0.000^{f}$ | $0.025 \pm 0.000^{e}$ | $0.034 \pm 0.001^{g}$ |
| L2 | $0.011 \pm 0.001^{h}$ | $0.029 \pm 0.002^{de}$ | $0.006 \pm 0.000^{g}$ | $0.012 \pm 0.000^{h}$ |
| L3 | $0.030 \pm 0.001^{d}$ | $0.035 \pm 0.001^{c}$ | $0.029 \pm 0.001^{d}$ | $0.031 \pm 0.001^{g}$ |
| L4 | $0.023 \pm 0.001^{f}$ | $0.026 \pm 0.001^{ef}$ | $0.026 \pm 0.002^{e}$ | $0.055 \pm 0.001^{d}$ |
| R1 | $0.009 \pm 0.001^{h}$ | $0.013 \pm 0.001^{g}$ | $0.001 \pm 0.001^{h}$ | $0.002 \pm 0.001^{i}$ |
| R2 | $0.011 \pm 0.001^{h}$ | $0.011 \pm 0.001^{g}$ | $0.000 \pm 0.000^{h}$ | $0.000 \pm 0.000^{i}$ |
| R3 | $0.023 \pm 0.001^{f}$ | $0.000 \pm 0.000^{i}$ | $0.001 \pm 0.001^{h}$ | $0.002 \pm 0.001^{i}$ |
| R4 | $0.005 \pm 0.001^{i}$ | $0.004 \pm 0.001^{h}$ | $0.000 \pm 0.001^{h}$ | $0.000 \pm 0.000^{i}$ |

CH: Cascade hop; C: dry-hop control; L: residual paste; R: repelletizing; 1: Mandarina Bavaria; 2: Citra; 3: Mosaic; 4: Hallertau Blanc. Different letters present significant statistical differences ($p < 0.05$). * On a dry basis.

Regarding the quantification of β-acids, it was observed that the N+adlupulone content was higher than the colupulone content in all of the hop samples (CH, dry-hop, and residual samples). However, in the repelletized hops, the concentration of β-acids was significantly lower, likely due to the same effect observed in the α-acids. The β-acid content for the CH and dry hops was in the range of 9–10%, as indicated by Liu et al. [43]. The β-acids in the hops are important for beer quality because they act as antioxidants,

protecting flavor and color during storage [44]. Additionally, they influence foam formation and stability by interacting with other compounds in beer, and possess antimicrobial properties that help prevent the growth of undesirable microorganisms [45,46]. Although they do not directly affect bitterness, they can modify the flavor and aroma profile during fermentation and aging, generally improving the quality and durability of the product.

### *3.4. Sensory Attributes Evaluation*

The sensory analysis (Table 5) provided interesting results on the attributes evaluated. In terms of the overall acceptability, it was observed that craft beers brewed with repelletized residues of the four types of hops studied were better rated by consumers. For example, beers R1 and R2 showed a significantly higher overall acceptability than their respective controls, C1 and C2 ($p < 0.05$). In contrast, beers R3 and R4 showed no significant differences compared to C3 and C4. These results indicate that beers brewed with repelletized hop residues may very well be accepted and consumed by a public that is not necessarily in the usual market for the consumption of craft beers, especially bitter beers. This preference for beers brewed with repelletized hop residues by the panelists is explained by a low preference for bitterness and hop flavor, and a significant preference ($p < 0.05$) for sweetness. Multiple studies have analyzed the relationship between bitterness perception, preference, and food consumption, finding that people with greater sensitivity to bitterness generally exhibit less liking for bitter products [47–49]. Furthermore, it has been observed that more adventurous tasters (judges) tend to rate bitter fruits and vegetables more favorably than less adventurous tasters (consumers), who typically assign lower ratings to these foods. [50]. Associations between bitter taste and the consumption of bitter alcoholic beverages do not always follow a linear pattern [51]. Some findings suggest that higher perceived bitterness may correlate positively with intake [52,53]. These findings suggest the need for further research to explore the moderating influence of personality traits on the potential relationships between bitterness perception, liking, and the consumption of bitter products [54]. The results did not show significant differences regarding the aroma, color, turbidity, carbonation, foam, alcohol, and overall malt flavor ($p < 0.05$). Based on the type of panelists used in this sensory analysis, the final product is well suited for this target audience within the brewery market, offering promising potential for both consumption and sales.

**Table 5.** Sensory characteristics of craft beer brewed with hop residues.

| Characteristics | Beer | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | **CH** | **C1** | **C2** | **C3** | **C4** | **R1** | **R2** | **R3** | **R4** |
| General acceptability | $6.87 \pm 1.45^{a}$ | $4.26 \pm 1.91^{d}$ | $4.42 \pm 2.03^{d}$ | $5.98 \pm 1.72^{ab}$ | $5.86 \pm 1.87^{bc}$ | $6.84 \pm 1.78^{a}$ | $6.55 \pm 1.36^{ab}$ | $6.49 \pm 0.99^{ab}$ | $5.02 \pm 1.06^{cd}$ |
| Aroma | $6.53 \pm 1.34^{a}$ | $5.26 \pm 1.87^{b}$ | $4.94 \pm 1.88^{b}$ | $5.84 \pm 1.93^{ab}$ | $5.68 \pm 1.93^{ab}$ | $5.35 \pm 2.00^{b}$ | $5.55 \pm 1.38^{b}$ | $5.65 \pm 1.21^{ab}$ | $5.43 \pm 1.18^{b}$ |
| Color | $6.76 \pm 1.41^{a}$ | $5.98 \pm 1.14^{abcd}$ | $5.39 \pm 1.45^{d}$ | $6.26 \pm 1.67^{abc}$ | $6.44 \pm 1.30^{ab}$ | $6.29 \pm 1.97^{abc}$ | $5.76 \pm 1.55^{bcd}$ | $5.79 \pm 0.89^{bcd}$ | $5.52 \pm 1.21^{cd}$ |
| Turbidity | $5.45 \pm 1.72^{abc}$ | $4.97 \pm 1.57^{bc}$ | $4.96 \pm 1.59^{bc}$ | $5.78 \pm 1.69^{ab}$ | $6.01 \pm 1.90^{a}$ | $5.46 \pm 2.38^{abc}$ | $5.67 \pm 1.67^{abc}$ | $5.98 \pm 1.41^{a}$ | $4.60 \pm 1.51^{c}$ |
| Carbonation | $5.99 \pm 1.65^{a}$ | $5.77 \pm 1.74^{ab}$ | $5.08 \pm 1.85^{abcd}$ | $5.34 \pm 1.90^{abcd}$ | $4.89 \pm 1.79^{bcd}$ | $4.73 \pm 1.73^{d}$ | $5.69 \pm 1.66^{abc}$ | $5.64 \pm 1.31^{abcd}$ | $4.78 \pm 1.21^{cd}$ |
| Foam | $6.30 \pm 1.67^{a}$ | $5.70 \pm 1.59^{ab}$ | $5.11 \pm 1.93^{b}$ | $5.10 \pm 1.77^{b}$ | $5.05 \pm 1.54^{b}$ | $5.15 \pm 1.24^{b}$ | $5.56 \pm 1.60^{ab}$ | $5.67 \pm 0.99^{ab}$ | $5.04 \pm 1.00^{b}$ |
| Bitterness | $5.56 \pm 1.47^{bcd}$ | $6.08 \pm 2.23^{ab}$ | $6.68 \pm 1.67^{a}$ | $5.62 \pm 1.73^{bcd}$ | $5.55 \pm 1.59^{bcd}$ | $5.16 \pm 1.40^{cd}$ | $5.67 \pm 1.29^{bcd}$ | $5.92 \pm 1.31^{abc}$ | $4.98 \pm 1.26^{d}$ |
| Sweetness | $4.64 \pm 1.40^{ab}$ | $3.23 \pm 1.68^{d}$ | $3.40 \pm 1.67^{cd}$ | $4.18 \pm 1.74^{bc}$ | $3.81 \pm 1.67^{bcd}$ | $5.10 \pm 1.32^{a}$ | $4.87 \pm 1.38^{ab}$ | $5.18 \pm 1.40^{a}$ | $4.13 \pm 1.17^{bc}$ |
| Hops flavor | $5.71 \pm 1.49^{ab}$ | $6.01 \pm 1.87^{a}$ | $5.46 \pm 1.95^{ab}$ | $5.56 \pm 1.70^{ab}$ | $6.15 \pm 1.64^{a}$ | $4.79 \pm 2.01^{b}$ | $5.41 \pm 1.33^{ab}$ | $5.36 \pm 1.24^{ab}$ | $5.07 \pm 1.03^{b}$ |
| Malt flavor | $5.41 \pm 1.66^{bc}$ | $5.86 \pm 1.60^{ab}$ | $5.31 \pm 1.87^{bc}$ | $5.63 \pm 1.60^{abc}$ | $5.07 \pm 1.70^{bc}$ | $5.17 \pm 1.37^{bc}$ | $5.91 \pm 1.55^{a}$ | $5.46 \pm 0.89^{bc}$ | $4.83 \pm 1.06^{c}$ |
| Alcohol | $5.02 \pm 1.84^{ab}$ | $5.04 \pm 1.97^{ab}$ | $5.10 \pm 1.93^{ab}$ | $5.33 \pm 1.83^{ab}$ | $4.94 \pm 1.29^{ab}$ | $5.63 \pm 2.46^{a}$ | $5.53 \pm 1.73^{ab}$ | $5.58 \pm 1.49^{a}$ | $4.51 \pm 1.22^{b}$ |

CH: Cascade hops; C: dry-hop control; R: repelletizing; 1: Mandarina Bavaria; 2: Citra; 3: Mosaic; 4: Hallertau Blanc. Different letters present in the same line significant statistical differences ($p < 0.05$).

## 4. Conclusions

The findings of this study demonstrate that the dry-hop residues obtained from the craft brewing can be effectively reused by subjecting the residues to a repelletization process (at room temperature). The contents of phenolic compounds, the antioxidant capacity, and α- and β-acids were significantly reduced ($p < 0.05$) in the residual hops compared to the starting hops due to the effects of the thermal process in the second brewing batch. However, the use of the residual hops for brewing generated significant sensory acceptability even surpassing the beer brewed with the starting hops, which exhibited higher bitterness values (IBUs). The use of an untrained sensory panel demonstrated that beers brewed with Mandarina Bavaria, Citra, and Mosaic hop residues have great potential for application in the brewing industry. Further studies are recommended to identify specific sensory profiles with trained panels (judges) on the beers obtained and specific treatments to optimize the residual dry-hop extraction and maximize its use. In addition, it is essential to conduct studies among consumers to evaluate the acceptability of the beers developed in this study in comparison with low bitterness beers such as Pilsen-type beers.

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## References

1. Marrucci, L.; Daddi, T.; Iraldo, F. Identifying the most sustainable beer packaging through a Life Cycle Assessment. *Sci. Total Environ.* **2024**, *948*, 174941. [CrossRef]
2. Mosher, M.; Trantham, K. *Brewing Science: A Multidisciplinary Approach*; Springer: New York, NY, USA, 2016. [CrossRef]
3. Murray, D.W.; O'Neill, M.A. Craft beer: Penetrating a niche market. *Br. Food J.* **2012**, *114*, 899–909. [CrossRef]
4. Salanță, L.C.; Fărcaş, A.C.; Borșa, A.; Pop, C.R. Current strategies for the management of valuable compounds from hops waste for a circular economy. *Food Chem. X* **2023**, *19*, 100876. [CrossRef]
5. Baiano, A. Craft beer: An overview. *Compr. Rev. Food Sci. Food Saf.* **2020**, *20*, 1829–1856. [CrossRef] [PubMed]
6. Villacreces, S.; Blanco, C.A.; Caballero, I. Developments and characteristics of craft beer production processes. *Food Biosci.* **2021**, *45*, 101495. [CrossRef]
7. Huige, N. Brewery by-products and effluents. In *Handbook of Brewing*, 2nd ed.; Priest, F.G., Stewart, G.G., Eds.; Taylor & Francis Group: Boca Raton, FL, USA, 2006; pp. 656–707.
8. Kerby, C.; Vriesekoop, F. An Overview of the Utilisation of Brewery By-Products as Generated by British Craft Breweries. *Beverages* **2017**, *3*, 24. [CrossRef]
9. Dos Santos Mathias, T.R.; De Mello Pedro, P.M.; Eliana, F.C.S. Solid wastes in brewing process: A review. *J. Brew. Distill.* **2014**, *5*, 1–9. [CrossRef]

10. Duarte, L.M.; Aredes, R.S.; Amorim, T.L.; De Carvalho Marques, F.F.; De Oliveira, M.A.L. Determination of α- and β-acids in hops by liquid chromatography or electromigration techniques: A critical review. *Food Chem.* **2022**, *397*, 133671. [CrossRef] [PubMed]
11. Almaguer, C.; Schönberger, C.; Gastl, M.; Arendt, E.K.; Becker, T. Humulus lupulus-a story that begs to be told. A review. *J. Inst. Brew.* **2014**, *120*, 289–314. [CrossRef]
12. Cvijetić, I.; Bigović, M.; Ristivojević, P.; Vitorović-Todorović, M.; Zloh, M.; Milojković-Opsenica, D. DFT study of the radical scavenging activity of isoxanthohumol, humulones (α-acids), and iso-α-acids from beer. *Struct. Chem.* **2021**, *32*, 2051–2059. [CrossRef]
13. Bartmańska, A.; Wałecka-Zacharska, E.; Tronina, T.; Popłoński, J.; Sordon, S.; Brzezowska, E.; Bania, J.; Huszcza, E. Antimicrobial Properties of Spent Hops Extracts, Flavonoids Isolated Therefrom, and Their Derivatives. *Molecules* **2018**, *23*, 2059. [CrossRef]
14. Anderson, H.E.; Santos, I.C.; Hildenbrand, Z.L.; Schug, K.A. A review of the analytical methods used for beer ingredient and finished product analysis and quality control. *Anal. Chim. Acta* **2019**, *1085*, 1–20. [CrossRef]
15. Rettberg, N.; Biendl, M.; Garbe, L. Hop Aroma and Hoppy Beer Flavor: Chemical Backgrounds and Analytical Tools—A Review. *J. Am. Soc. Brew. Chem.* **2018**, *76*, 1–20. [CrossRef]
16. Titus, B.M.; Lerno, L.A.; Beaver, J.W.; Byrnes, N.K.; Heymann, H.; Oberholster, A. Impact of Dry Hopping on Beer Flavor Stability. *Foods* **2021**, *10*, 1264. [CrossRef]
17. De Keukeleire, D. Fundamentals of beer and hop chemistry. *Quim. Nova* **2000**, *23*, 108–112. [CrossRef]
18. De Oliveira Gomes, F.; Guimarães, B.P.; Ceola, D.; Ghesti, G.F. Advances in dry hopping for industrial brewing: A review. *Food Sci. Technol.* **2021**, *42*, ctaAR60620. [CrossRef]
19. AOAC. *Official Method 931.04 Moisture in Cacao Products Gravimetric Method*; AOAC: Rockville, MD, USA, 1931.
20. AOAC. *Method 923.03, Ash of Flour, Official Methods of Analysis*, 16th ed.; Adobe® Software® and E-DOC/CJS; AOAC: Rockville, MD, USA, 1997.
21. AOAC. *Official Methods of Analysis. Dumas Method (990.03)*, 15th ed.; AOAC: Washington, DC, USA, 2005.
22. Manirakiza, P.; Covaci, A.; Schepens, P. Comparative Study on Total Lipid Determination using Soxhlet, Roese-Gottlieb, Bligh & Dyer, and Modified Bligh & Dyer Extraction Methods. *J. Food Compos. Anal.* **2001**, *14*, 93–100. [CrossRef]
23. Saura-Calixto, F.; Serrano, J.; Goñi, I. Intake and bioaccessibility of total polyphenols in a whole diet. *Food Chem.* **2006**, *101*, 492–501. [CrossRef]
24. Kim, D.; Lee, K.W.; Lee, H.J.; Lee, C.Y. Vitamin C Equivalent Antioxidant Capacity (VCEAC) of Phenolic Phytochemicals. *J. Agric. Food Chem.* **2002**, *50*, 3713–3717. [CrossRef] [PubMed]
25. ASBC. *Method of Analysis. American Society of Brewing Chemists*; Beer Bitterness, Beer-23A; The Society: St. Paul, MN, USA, 2011.
26. Roberts, T.R.; Wilson, R.J.H. Hops. In *Handbook of Brewing*, 2nd ed.; Priest, F.J., Stewart, G.G., Eds.; Taylor & Francis: Boca Raton, FL, USA, 2006; pp. 177–280.
27. Stevens, R. The Chemistry of Hop Constituents. *Chem. Rev.* **1967**, *67*, 19–71. [CrossRef]
28. Verzele, M. 100 years of hop chemistry and its relevance to brewing. *J. Inst. Brew.* **1986**, *92*, 32–48. [CrossRef]
29. Palamand, S.R.; Aldenhoff, J.M. Bitter tasting compounds of beer. Chemistry and taste properties of some hop resin compounds. *J. Agric. Food Chem.* **1973**, *21*, 535–543. [CrossRef]
30. Hough, J.S.; Briggs, D.E.; Stevens, R.; Young, T.W. *Malting and Brewing Science Volume 2: Hopped Wort and Beer*, 2nd ed.; Chapman Hall: New York, NY, USA, 1982.
31. Verzele, M.; De Keukeleire, D. *Chemistry and Analysis of Hop and Beer Bitter Acids*; Elsevier: Amsterdam, The Netherlands, 1991.
32. Terpinc, P.; Čeh, B.; Ulrih, N.P.; Abramovič, H. Studies of the correlation between antioxidant properties and the total phenolic content of different oil cake extracts. *Ind. Crops Prod.* **2012**, *39*, 210–217. [CrossRef]
33. Collin, S.; Jerkovic, V.; Bröhan, M.; Callemien, D. Natural Products. In *Polyphenols and Beer Quality*; Ramawat, K., Mérillon, J.M., Eds.; Springer: Berlin/Heidelberg, Germany, 2013.
34. Petrón, M.; Andrés, A.; Esteban, G.; Timón, M. Study of antioxidant activity and phenolic compounds of extracts obtained from different craft beer by-products. *J. Cereal Sci.* **2021**, *98*, 103162. [CrossRef]
35. Ruiz-Ruiz, J.C.; Del Carmen Esapadas Aldana, G.; Cruz, A.I.C.; Segura-Campos, M.R. *Antioxidant Activity of Polyphenols Extracted from Hop Used in Craft Beer*; Elsevier: Amsterdam, The Netherlands, 2019; pp. 283–310. [CrossRef]
36. Ambra, R.; Pastore, G.; Lucchetti, S. The Role of Bioactive Phenolic Compounds on the Impact of Beer on Health. *Molecules* **2021**, *26*, 486. [CrossRef]
37. Oladokun, O.; Tarrega, A.; James, S.; Smart, K.; Hort, J.; Cook, D. The impact of hop bitter acid and polyphenol profiles on the perceived bitterness of beer. *Food Chem.* **2016**, *205*, 212–220. [CrossRef] [PubMed]
38. Schindler, R.; Sharrett, Z.; Perri, M.J.; Lares, M. Quantification of α-Acids in Fresh Hops by Reverse-Phase High-Performance Liquid Chromatography. *ACS Omega* **2019**, *4*, 3565–3570. [CrossRef]
39. Bruner, J.; Marcus, A.; Fox, G. Dry-Hop Creep Potential of Various Saccharomyces Yeast Species and Strains. *Fermentation* **2021**, *7*, 66. [CrossRef]

40. Castro, R.; Díaz, A.B.; Durán-Guerrero, E.; Lasanta, C. Influence of different fermentation conditions on the analytical and sensory properties of craft beers: Hopping, fermentation temperature and yeast strain. *J. Food Compos. Anal.* **2021**, *106*, 104278. [CrossRef]
41. Castorena-García, J.H.; Juárez-Pérez, V.; Cano-Hernández, M.; Santiago-Santiago, V.; López-Mejía, O.A. Caracterización Físico-química de Cerveza Artesanal don Adjunto de Maíz Azul y Derivados de Caña de Azúcar. *Concienc. Tecnol.* **2020**, *60*, 94465715001. Available online: https://www.redalyc.org/articulo.oa?id=94465715001 (accessed on 17 October 2024).
42. Jaskula-Goiris, B.; Aerts, G.; De Cooman, L. Hop α-acids isomerisation and utilisation: An experimental review. *Cerevisia* **2010**, *35*, 57–70. [CrossRef]
43. Liu, Y.; Lu, N.; Tang, J. Synthesis, characterization, crystal structure, and antioxidant activity of hexahydro-β-acids. *J. Mol. Struct.* **2018**, *1175*, 721–727. [CrossRef]
44. Formato, A.; Gallo, M.; Ianniello, D.; Montesano, D.; Naviglio, D. Supercritical fluid extraction of α- and β-acids from hops compared to cyclically pressurized solid–liquid extraction. *J. Supercrit. Fluids* **2013**, *84*, 113–120. [CrossRef]
45. Simpson, W.; Smith, A. Factors affecting antibacterial activity of hop compounds and their derivatives. *J. Appl. Bacteriol.* **1992**, *72*, 327–334. [CrossRef]
46. Mizobuchi, S.; Sato, Y. Antifungal activities of hop bitter resins and related compounds. *Agric. Biol. Chem.* **1985**, *49*, 399–403. [CrossRef]
47. Bell, K.I.; Tepper, B.J. Short-term vegetable intake by young children classified by 6-n-propylthoiuracil bitter-taste phenotypey. *Am. J. Clin. Nutr.* **2006**, *84*, 245–251. [CrossRef]
48. Dinehart, M.; Hayes, J.; Bartoshuk, L.; Lanier, S.; Duffy, V. Bitter taste markers explain variability in vegetable sweetness, bitterness, and intake. *Physiol. Behav.* **2005**, *87*, 304–313. [CrossRef] [PubMed]
49. Duffy, E.; Hayes, J.; Feeney, E. Understanding taste and texture perception to enhance vegetable acceptance. *Proc. Nutr. Soc.* **2017**, *76*, E67. [CrossRef]
50. Ullrich, N.V.; Touger-Decker, R.; O'Sullivan-Maillet, J.; Tepper, B.J. PROP taster status and self-perceived food adventurousness influence food preferences. *J. Am. Diet. Assoc.* **2004**, *104*, 543–549. [CrossRef] [PubMed]
51. Thibodeau, M.; Bajec, M.; Pickering, G. Orosensory responsiveness and alcohol behaviour. *Physiol. Behav.* **2017**, *177*, 91–98. [CrossRef]
52. Lipchock, S.V.; Spielman, A.I.; Mennella, J.A.; Mansfield, C.J.; Hwang, L.; Douglas, J.E.; Reed, D.R. Caffeine Bitterness is Related to Daily Caffeine Intake and Bitter Receptor mRNA Abundance in Human Taste Tissue. *Perception* **2017**, *46*, 245–256. [CrossRef] [PubMed]
53. Ong, J.; Hwang, L.; Zhong, V.W.; An, J.; Gharahkhani, P.; Breslin, P.A.S.; Wright, M.J.; Lawlor, D.A.; Whitfield, J.; MacGregor, S.; et al. Understanding the role of bitter taste perception in coffee, tea and alcohol consumption through Mendelian randomization. *Sci. Rep.* **2018**, *8*, 16414. [CrossRef]
54. Higgins, M.J.; Bakke, A.J.; Hayes, J.E. Personality traits and bitterness perception influence the liking and intake of pale ale style beers. *Food Qual. Prefer.* **2020**, *86*, 103994. [CrossRef]