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Commercial craft beers of midwest Brazil: biochemical and physicochemical properties and their relationship with its sensory profile

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Abstract

Faced with changes in eating habits, consumers currently seek to enjoy moderate consumption of healthy food products that results in sensory pleasure. Here, our study aimed to evaluate the biochemical and physicochemical properties and sensory profile of craft beers produced in the Brazilian Midwest region. Our evaluation of the beverages revealed different physicochemical characteristics such as alcohol content, soluble solids, titratable acidity in citric acid, and dry extract percentage, with values ranging between 4.06-6.13%, 5.13-10.98 °Brix, 1.71-3.45%, and 3.38-7.99%, respectively. Moreover, the average density of the beverages was 0.9906 g/cm^3. In addition, the antioxidant activity and total phenolic of the beverages ranged between 14.28-20.46 mMol of Trolox/100 mL and 74.84-108.45 mg/100 g, respectively. Phenolic compounds that are essential in human nutrition, such as epicatechin, catechin, gallic acid, p-coumaric acid, caffeic acid, ferulic acid, rutin, and kaempferol, were identified in the beverages. Regarding the sensory profile of the craft beers, the beers had an average acceptability index of 75.11% for appearance, 70.24% for aroma, 60.43% for flavor, and 73.53% for texture. Our study demonstrated a correlation between chemical composition and acceptability index of beer, indicating a preference for lighter colored, fuller, and less acidic beers.

Keywords: alcoholic beverage; flavonoids; antioxidant activity; check all that apply.

Practical Application: Currently there has been an increase in the production and consumption of craft beers. However, legislation for these products is still deficient in many respects. Thus, this work elucidated the physical and chemical characteristics and related them to the sensory aspects.

1 Introduction

Beer is a nourishing drink made from ingredients such as barley and hops, that contain magnesium, B-type vitamins, and other compounds with antioxidant potential (World Health Organization, 2014). Studies have shown that beer consumption (100 mL/60 kg/day) does not cause liver damage and the body could benefit from some biocompound present in this beverage (Wang et al., 2022). Beer is one of the oldest beverages produced and consumed in the world, having been consumed for more than three millennia and has undergone some modifications throughout history. Beer production began at the end of the 19th century, and since then the brewing industry have been generating jobs, taxes, income, and social benefits. Brazil is currently the third largest beer consumer in the world behind the United States of America and China (Capece et al., 2018; Rodhouse & Carbonero, 2019).

The brewing process, as well as the quality of the raw materials used in the production, influences the physicochemical, biochemical, and sensorial characteristics of each type of beer. For example, ale is a type of beer that is produced by top fermentation at temperatures ranging from 18 to 25 °C for a short period of time (three to five days), and owing to this process, microorganisms cannot consume all the sugar contained in the malt, giving ale a sweet as well as a complex and fruity hop flavor (Gonçalves et al., 2016). Pale ale is a type of beer that is characterized by its reddish color, bitter hop flavor, and alcoholic content owing to medium to high, which gives the beer its turbidity (Granato et al., 2011).

Craft beer is produced in limited quantities by small scaled companies, usually family businesses, with added fruits and a high amount of malt per hectoliter. The appreciation of local food products is usually attributed to greater knowledge of production methods and the association of the history of the product with the experience it provides. Consumers believe that product exclusivity is more necessary than increasing the scale of their production as done by large companies (Gellynck et al., 2012). In 2013, Brazil produced approximately 188,000 L of craft beer, and the prospect for 2023 is that the craft beer industry will form 2% of the national beer market (Serviço Brasileiro de Apoio às Micro e Pequenas Empresas, 2015). The demand for craft beer in Brazil is evolving gradually, mainly because consumers are looking for products that have a different flavor than usual, are produced in small-scale, and are exclusive to certain regions (Aquilani et al., 2015; Gellynck et al., 2012).

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In 2018, Goiás, the largest state in the Brazilian Midwest with the highest number of craft breweries, showed significant increase in the number of breweries registered with the Ministry of Agriculture, Livestock, and Supply, which is Brazil's beer regulatory organization. According to Abracerva, the Brazilian artisanal beer association, 25 craft breweries operate in Goiânia, Aparecida de Goiânia, and inland municipalities (Santana, 2018). Thus, craft beers from micro- to large-scaled breweries are increasingly becoming prevalent in the markets, specialty stores, bars, and restaurants of Brazil. Therefore, this study aimed to evaluate and establish a relationship between the biochemical and physicochemical properties and sensory profile of high-fermented pale ale craft beers produced in the state of Goiás, Brazil.

2 Material and methods

2.1 Chemical materials and reagents

Regional Indian pale ale (IPA) type craft beers were purchased from local stores in Goiânia, Brazil and named B1, B2, B3, B4, B5, and B6. For all analyses, the craft beer bottles were opened, and the beer was decarbonated under agitation in the Ultrasonic Washer SoniClean 2 (Sanders do Brasil, Santa Rita do Sapucaí, Brazil) for 15 min.

The reagents used in this study were of analytical standard; 2,2-diphenyl-1-picrylhydrazyl (DPPH), 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (Trolox), 2,2-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid, 2,6-dichlorophenolindophenol (ABTS), 2,6-ditert-butyl-4-methyl phenol, 2,4,6-tripyridyltriazine, Folin & Ciocalteu phenol reagent, and ferulic acid were purchased from Sigma Chemical Co. (St. Louis, MO, USA). Standard phenolic compounds, gallic acid, protocatechuic acid, gentisic acid, caffeic acid, p-coumaric acid, vanillic acid, ferulic acid, ellagic acid, catechin, epicatechin, rutin, quercetin, naringenin, luteolin, and kaempferol, were purchased from Sigma Chemical Co.

2.2 Physicochemical analysis

The pH and soluble solids (SS) values were determined using a digital pH meter and refractometer, respectively. Titratable acidity (TA) was determined by titration with 0.1 M NaOH and expressed as percentage of citric acid (Association of Official Analytical Chemists, 2000).

The alcohol concentration of the craft beers was determined by the hydrometer method (Brasil, 2010). Next, distillation was performed to separate the alcohol from the craft beer samples, with subsequent quantification of the alcohol by using the value of the relative density of the distillate at 20 °C. The relative density was obtained from the ratio between the specific gravity of the distillate at 2 °C and the specific weight of water at 20 °C using a portable digital density meter DMA 35 (Anton Paar, Austria, Europe). The relative density values were used to determine alcohol concentration (Method 942.06) (Association of Official Analytical Chemists, 2005).

The dry extract of the samples was obtained by heating the craft beer samples in a water bath to evaporate the water and alcohol, and subsequently determine its final mass (Brasil, 2010).

2.3 Biochemical properties

Phenolic compounds: total quantification and identification

The total polyphenol content of the craft beers was determined using the Folin-Ciocalteu spectrophotometric method (Prior et al., 2005). Beer samples (200 µL) were mixed with 1.9 mL of Folin-Ciocalteu reagent. After 3 min, 1 mL of 20% aqueous solution of sodium carbonate (60 g/L) and 2 mL of distilled water were added. After a reaction time of 120 min in the dark at room temperature (~25 °C), absorbance was measured at 725 nm. Gallic acid was used as a standard and the results were expressed as mg of gallic acid equivalents per 100 mL of beer.

The phenolic composition was evaluated using the Thermo Scientific™ UltiMate™ 3000 HPLC System (Thermo Fisher, Waltham, MA, USA) with a reversed-phase HC-C18 (4.6 × 100 mm; 3 µm) (Agilent, Santa Clara, CA, USA), coupled to a high-resolution mass spectrometer (Q Exactive Orbitrap Mass Spectrometer; Thermo Fisher) with source heated-electrospray ionization operating in negative mode, with spray voltage 3.5 kV, sheath gas 30, auxiliary gas 10, capillary temperature 350 °C, auxiliary gas temperature 250 °C, tube lens 55, and mass range 150-700 m/z.

Each sample of lyophilized beer (2 mg) was solubilized in 1 mL of deionized water. Next, the solubilized samples were filtered through a polyester membrane filter (0.45 µm). A total of 10 µL of the extract was injected into an C18 column (4.6 × 100 mm; 3 µm) (Agilent), and the analysis was performed as follows: solution A comprised ultrapure solvent H_2O (PURELAB® Option Q; ELGA LabWater, Woodridge, IL, USA) with 0.1% formic acid; solution B comprised MeOH (HPLC grade, J.T.Baker®, Radnor, PA, USA) with 0.1% formic acid. Gradient elution was performed with 7-30% B from 0 to 10 min, 30-50% B from 10 to 15 min, 50-70% B from 15 to 18 min, 70-80% B from 18 to 20 min, 80-100% B from 20 to 23 min, and 100% B from 23 to 30 min; flow rate was 3.0 mL/min; and temperature was 20 °C. For the fragmentation study, a parallel reaction monitoring experiment was conducted with a collision energy of 30.

To identify phenolic compounds, we used a stock solution of standard phenolic compounds in methanol at a concentration of 1 mg/mL. Data were processed using Xcalibur™ software (Thermo Fisher).

Antioxidant activity

The ability to scavenge free radical of the craft beers, was measured using the DPPH assay (Yen & Chen, 1995). Samples of craft beer (0.1 mL) were mixed with 2 mL of 0.04 mmol/L DPPH in 50% methanol solution. After 10 min of incubation at room temperature (~25 °C), absorbance was measured using a UV-visible spectrophotometer (UV-5100 UV/Vis Spectrophotometer, Metash, Shanghai, China) at 515 nm. A calibration curve was prepared with Trolox solution (0.05×10^{-1} mmol/L). Data are expressed as the Trolox equivalent (TE) of antioxidant capacity per liter of beer (mmol TE/L). All measurements were performed in triplicates, and the percentage of discoloration was calculated using Equation 1.

$$\text{Discoloration}\,(\%) = \left[1\text{-}\left(\text{abs}_{\text{sample}}\text{-abs}_{\text{control}}\right)\right] \times 100 \quad (1)$$

Where $\text{abs}_{\text{sample}}$ is the absorbance of the sample, and $\text{abs}_{\text{control}}$ is the absorbance of the control.

2.4 Fourier transform infrared spectroscopy

The craft beer samples were decarbonated, lyophilized, placed on the support, and pressed by the measuring sensor. Fourier transform infrared (FTIR) spectroscopy was performed in the region of 4000-650 cm^{-1} with 10 scans and at a resolution of 4 cm^{-1} on the Frontier™ FTIR/IR spectrometer (PerkinElmer, Akron, OH, USA). The FTIR graph was generated using Origin software (version 8.1; OriginLab Corporation, Northampton, MA, USA), and the peaks were compared with those in the literature.

2.5 Physicochemical properties

Two spectrophotometric analysis methods were used to determine the color of the beers. Color analysis was performed with a Color Quest II spectrophotometer (HunterLab, Reston, VA, USA) using the CIELAB L*a*b* system, and chroma and hue values were calculated based on the parameters a* and b* (Ahmadian-Kouchaksaraei et al., 2014). In addition, the European Brewery Convention (EBC) method 9.6) was used to read the absorbance of the of the diluted samples against a water blank on the UV-5100 UV/Vis Spectrophotometer (Metash), at 430 nm (Abs_{430nm}). The results were calculated using Equation 2, and are expressed in color units.

$$\text{EBC}\,(\text{color units}) = 25 \times \text{dilution factor} \times \text{Abs}_{430\text{nm}} \quad (2)$$

Osmolality analysis was performed using an electronic cryoscope (ITR; MK 540, Esteiro-RS, Brazil) (Musara & Pote, 2014). For the freezing point conversion, the equation $\Delta tc = Kc \times m$ was used, where Δtc is the cryoscopic descent (difference between the initial freezing temperatures of the pure solvent and the solution), Kc is the cryoscopy constant of the water (1.86 °C/mol/kg), and m is the molar concentration of the solute (osmolality expressed in mOsm/kg of solvent).

Beer stability was determined at 7 °C and 25 °C to simulate refrigeration and storage temperatures, respectively, through phase separation analysis in an analytical centrifuge (SL 701; Solab, Piracicaba, Brazil). Craft beer samples (2 g) were weighed into 2 mL microtubes and centrifuged at 14,000 rpm for 5 min. The supernatant was discarded and the mass was determined as a percentage.

2.6 Sensory profile of the craft beers

Sensory analysis protocols were approved by the Research Ethics Committee (No. 3300322). The analysis was performed in individual offices of the Sensory Analysis Laboratory of the Instituto Federal Goiano, Rio Verde, Goiás, Brazil. Among the participants, 32% were male, 68% were female, 77% were 18-25 years old, 16% were 25-35 years old, and 3% were 35-50 years old. Participants were given approximately 20 mL of each craft beer which were at a temperature of < 7 °C.

The first step of the descriptive sensory analysis was the survey for appearance, aroma, flavor, texture of the craft beers using the network method (Moskowitz, 1983). Three samples were served in three sessions to each judge, along with water and biscuits to be consumed between pairs of samples. The participants were asked to describe the differences and similarities between the pairs in the sample. In the group discussion, the participants reached a consensus on the most frequent descriptive terms used to describe the differences and similarities of the samples. These descriptive terms comprised the check-all-that-apply (CATA) analysis form.

The terms used to describe the appearance of the craft beers were: yellow, brown, cloudy, clear, foam up, no foam up, bubbles, and no bubbles. The terms used to describe the aroma of the craft beers were: yeast aroma, low yeast aroma, high fruity aroma, low fruity aroma, sweet/honey, and citric. The terms used to describe the flavor of the craft beers were: sweet, bitter, fruity presence, no fruity presence, yeast flavor, no yeast flavor, citric flavor, and no citric flavor. The terms used to describe the texture of the craft beers were: no foam up, body foam, low viscosity, high viscosity, presence of particles, and no presence of particles. For the CATA method, the samples were presented in a monadic and balanced manner to each of the 62 participants. Participants were asked to verify all the attributes they used to describe the samples. The frequency of use of each sentence was determined by counting the number of consumers that checked that term to describe each cheese sample. Cochran´s Q test was performed to identify significant differences among samples for each sentence. A Correspondence Analysis (CA) was used to analyze the association between CATA terms and beer samples using a matrix data set with 6 columns (beer samples) and 17 rowns (CATA terms).

Of the total participants, 31 evaluated the appearance, aroma, flavor, and texture of the craft beers using a 9 cm unstructured hedonic scale. The scores assigned by the participants were categorized in terms of “rejection” ($0 < \text{score} < 4.99$), “indifference” ($5.00 < \text{score} < 5.99$), and “acceptance” ($6 < \text{score} < 9$). The acceptability index is expressed as a percentage, using the average score assigned to the product × 100/maximum score assigned to the product.

3 Statistical analysis

Statistical analyses were conducted in triplicates. Analysis of variance test was used to detect significant differences between treatments, whereas means were compared using Tukey’s test with Statistica (version 6.0; StatSoft Inc., Tulsa, OK, USA). Differences were considered statistically significant at $p < 0.05$.

Principal component analysis (PCA) was performed using R software. To evaluate the results from a multidimensional point of view, we evaluated the results of the biochemical, physicochemical, and sensory analysis by PCA, a well-known method of extracting relevant information from multivariate data sets through a small number of orthogonal variables called principal components (Granato et al., 2018).

4 Results and discussion

4.1 Physicochemical and biochemical properties of craft beers

Figure 1 and Table 1 present the biochemical and physicochemical properties of the craft beers. The density varied between 0.9891 g/cm^3 (B4) and 0.9923 g/cm^3 (B6). These values were lower than the reference values for beer in the Brazilian legislation (Brasil, 2009). Sousa & Fogaça (2019) studied 12 beer samples, and all of them had a relative density lower than that present in the Brazilian legislation. Density varies according to the consumption of fermentable carbohydrates (maltose) and production of alcohol by microorganisms during the fermentation process (Walker & Walker, 2018). The titrated acidity of the beers varied between 1.71 mEq/L (B1) and 3.22 mEq/L (B3), with a significant difference between B1 and B3. Brazilian legislation does not state standards for TA; therefore, this analysis was performed for comparison and characterization of the craft beers in the present study (Taylor, 2015).

The highest pH value was observed in B4, followed by B3 and B6. Low pH values were observed in B1, B2, and B5. Overall, the pH values were below 4.87, indicating that low pH maintains the microbiological quality of the product as per the theory of barriers (Rodhouse & Carbonero, 2019).

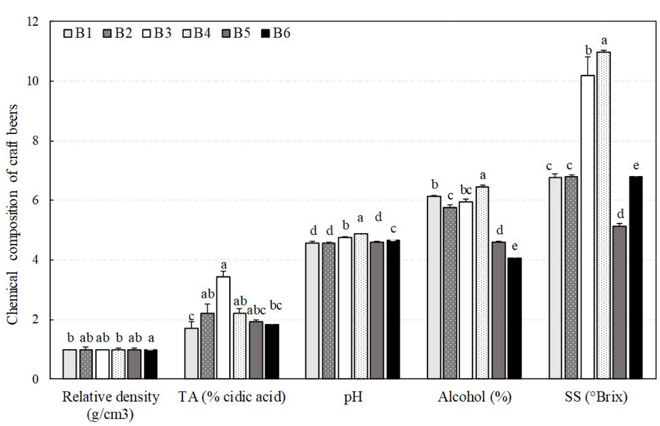

Figure 1. Biochemical and physicochemical properties of the craft beers. Chemical composition. SS – soluble solid; TA – titratable acidity. Different lowercase letters indicate a significant difference between beer samples by Tukey's test ($p < 0.05$).

Table 1. Chemical characteristics of the craft beers.

Craft beers	DE (%)	RD (g)	AC (°GL)	Label AC (°GL)
B1	4.36	0.9894	6.13	6
B2	4.58	0.9916	5.76	5.8
B3	7.99	0.9898	5.95	6.2
B4	7.48	0.9891	6.45	6.9
B5	3.33	0.9912	4.6	4.8
B6	5.26	0.9923	4.06	6.2
Brazilian legislation (Brasil, 2019)	2.0 - 7.0%.	1.007 - 1.022	> 0.5%/v	-

B4 had the highest alcohol concentration (6.45%), followed by B1, B3, B2, B5, and B6 (4.06%). Alcohol concentration of all the samples evaluated, except for B6, was 5.5-7.5% v/v, consistent with the alcohol concentration values in the BJCP for pale ale (Beer Judge Certification Program, 2016). In addition, the manufacturers of B6 declared alcohol content of 6.20% v/v on the label, which was 52% higher than the actual value detected in the analysis. The alcohol concentration determined in this study were close to those reported by Cheiran et al. (2019) for pale ale type of beer (4.8-6.3%) from different breweries.

The SS content of the craft beers ranged from 5.13 °Brix to 10.98 °Brix; B4 and B3 had high SS values, with a significant difference in SS content between B4 and B3, followed by B1, B2, B6, and B4. Different proportion and quality of the ingredients used in the production of craft beer interferes with its total SS content at the end of brewing (Olaniran et al., 2017). Moreover, intensity of the fermentation process has also been reported to interfere with the final SS content (Estela-Escalante et al., 2016).

In the present study, color of the craft beers was evaluated using the European Brewery Convention (EBC) scale and the CIELAB L* a* b* system. Table 1 presents the results of the color evaluation of the craft beers.

According to the EBC scale, B5 was light (< 20 EBC), whereas B1, B2, B3, B4, and B6 were dark (> 20 EBC). Although all the craft beers in the present study are considered pale ale, the literature has reported great color variation in craft beers. In the present study, the EBC range for the craft beer samples was 17-44 EBC units, whereas Cheiran et al. (2019) have reported an EBC ranging of 11-33 EBC units. However, the literature reports, as expected for pale ale, an EBC range of 20-30 EBC units (Benucci et al., 2021). Color analysis using the CIELAB system revealed the following: the L* value varied between 1.93-11.33 indicating darker colors (closest to zero), the hue angle showed a tendency towards yellow (close to 90°), and chroma values ranged from 3.55 to 14.02 (closest to zero), indicating neutral colors.

The apparent dry extract was calculated based on the mass obtained after the evaporation of all volatile compounds in the craft beer samples. Ale craft beer is more full-bodied due to the greater concentration of other substances than other beers or absence of a filtration phase after fermentation and maturation. The apparent dry extract in the present study varied between 4.33% and 8.9%. Dry extract values between 2.0% and 7.0% are considered within the standards of Brazilian legislation (Table 1). For B4 and B3, the dry extract values were above 7.0% (Table 2).

No significant difference between the sedimentation values of the craft beers at 7 °C and 25 °C was observed; the same was the case for the osmolality of the craft beers as well (Table 1). Although the craft beers had different apparent dry extract values, the presence of more solids did not appear to interfere with sedimentation and osmolality.

Sedimentation is the settlement of particles under gravity (Sawale et al., 2020) which can be influenced by temperature, as was observed in this study. Higher sedimentation values were observed at 7 °C (2.07-2.02) than at 25 °C (0.68-1.40). Tribst et al. (2019) have stated that prolonged refrigeration can

Table 2. Color, dry extract, sedimentation, osmolality, and acceptability index of craft beers.

Craft beers	Color				Dry extract (%)	Sedimentation		Osmolality (mOsm/kg)	Acceptability index (%)			
	EBC	L*	Hue	Chroma		At 7 °C	At 25 °C		Appearance	Aroma	Flavor	Texture
B1	44.35 ± 3.80^{a}	1.93 ± 0.40^{d}	252.05 ± 6.16^{c}	3.55 ± 0.45^{e}	4.36 ± 1.10^{b}	2.55 ± 0.53^{a}	1.27 ± 0.79^{a}	155.24 ± 3.99^{a}	79.34	76.29	67.57	77.68
B2	40.44 ± 0.04^{b}	5.83 ± 0.29^{b}	257.87 ± 1.28^{b}	8.44 ± 0.53^{c}	4.58 ± 0.88^{b}	2.41 ± 0.44^{a}	1.03 ± 0.24^{a}	154.72 ± 3.57^{a}	78.02	72.64	55.67	75.20
B3	44.10 ± 0.43^{a}	11.33 ± 0.40^{a}	265.3 ± 0.36^{a}	14.02 ± 0.33^{e}	7.99 ± 1.52^{a}	2.46 ± 0.60^{a}	0.69 ± 0.10^{a}	154.02 ± 3.44^{a}	59.42	63.62	48.63	64.29
B4	38.45 ± 2.96^{b}	10.99 ± 0.78^{a}	95.77 ± 1.52^{d}	12.03 ± 1.05^{b}	8.98 ± 0.51^{a}	2.07 ± 1.03^{a}	1.36 ± 0.42^{a}	156.50 ± 1.19^{a}	74.57	67.99	73.42	85.48
B5	17.63 ± 0.02^{d}	5.50 ± 0.34^{b}	100.34 ± 1.32^{d}	4.49 ± 0.71^{e}	3.33 ± 0.54^{b}	2.15 ± 0.28^{a}	1.40 ± 0.39^{a}	155.91 ± 0.39^{a}	77.14	72.69	60.72	66.32
B6	28.75 ± 0.26^{c}	3.83 ± 0.44^{c}	268.71 ± 1.11^{a}	$5.70^{d} \pm 0.65^{d}$	5.26 ± 0.99^{b}	2.92 ± 0.82^{a}	0.68 ± 0.30^{a}	156.27 ± 1.34^{a}	82.22	68.25	56.62	72.27

Different lowercase letters indicate a significant difference between beer samples by Tukey's test ($p < 0.05$).

change the physicochemical characteristics of beers, mainly the agglomeration and sedimentation of fat and/or solids.

The mean osmolality of the craft beers was 155.44 mOsm/kg, which was 10 times lower than that reported by Feldman & Barnett (1995) for Budweiser beer (1040-1006 mOsm/kg).

Statistical differences in some biochemical and physicochemical properties, such as color, density, titratable acidity, SS content, dry extract, alcohol content, and antioxidant activity of the craft beers, were observed in the present study. These statistical differences can be attributed to the dissimilarities in the raw materials and production methods used in the manufacturing process of these craft beers (Benucci et al., 2021; Cheiran et al., 2019; Gąsior et al., 2020; Zapata et al., 2019).

Figure 2 presents the data for total phenolic compounds (TPC) present in the craft beers and the antioxidant activity of the craft beers. B2, B3, and B4 had high CFT values (average 102.97 mg/100 mL), and no significant difference in CFT values was observed among them. In addition, no significant difference in CFT values was observed between B1 (84.14 mg/100 mL) and B6 (74.84 mg/100 mL). B5 had the lowest CFT value (69.01 mg/100 mL).

B3, which was characterized by dark tonality, had the highest content of phenolic compounds, apparent extract, and TA, and this may have contributed to the lower acceptability of B3 in relation to appearance, aroma, flavor, and texture. In the present study, we noted that phenolic compounds can influence the sensory characteristics of beer by adding a bitter flavor. Moreover, phenolic compounds have antioxidant capacity, which may be the reason for the oxidative stability of beer. In addition, these antioxidants are associated with a lower risk of health problems attributed to beer consumption, such as the reduction in cardiovascular diseases (Gaetano et al., 2016).

Table 3 presents the phenolic compounds identified using high-performance liquid chromatography (HPLC). Eight phenolic compounds, catechin, epicatechin, caffeic acid, p-coumaric acid ferulic acid, gallic acid rutin, and kaempferol, were identified in the craft beer samples and evaluated in the present study. The presence of these phenolic compounds in craft beers may have important health benefits.

Catechin, epicatechin, and p-coumaric acid were detected in all the craft beer samples. Catechin has been used to evaluate the flavonoid content in beer (Breda et al., 2022). Catechin prevents many chronic diseases by inhibiting excessive oxidative stress through the activation of superoxide dismutase, glutathione peroxidase, and catalase (Fan et al., 2017). Epicatechin has been studied as a reducer in metabolic syndrome risk factors for its anti-inflammatory properties (Mechchate et al., 2021), as well as in systemic insulin resistance for its therapeutic properties (Cremonini et al., 2016). Previously, the presence of coumarins in beers (Callemien & Collin, 2009; Kumaraswamy et al., 2011) and their possible antioxidant, anti-inflammatory, hepatoprotective, and anticoagulant effects (Witaicenis et al., 2014) have been reported.

Table 3. Phenolic compounds in the craft beers identified using high-performance liquid chromatography.

Phenolic compounds	Mm	B1	B2	B3	B4	B5	B6
Catechin	290,07904	X	X	X	X	X	X
Epicatechin	290,07904	X	X	X	X	X	X
Caffeic acid	180,04226	-	-	X	X	X	X
Ferulic acid	194,05791	-	-	X	X	X	X
p-coumaric acid	164,04735	X	X	X	X	X	X
Rutin	610,15339	X	-	X	X	X	X
Galic acid	170,02153	-	-	X	X	X	-
Kaempferol	286,04774	-	-	-	-	X	X

Mm: molecular mass.

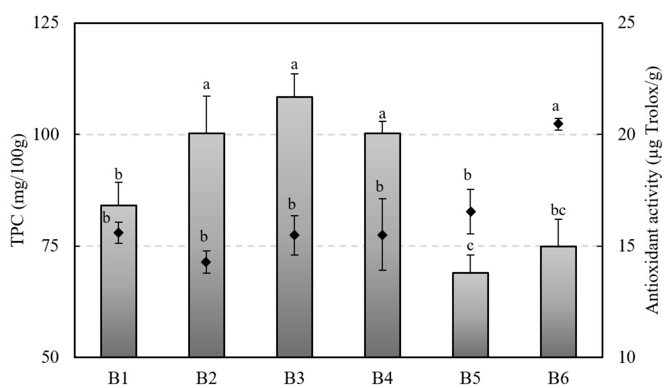

Figure 2. Total phenolic composition and antioxidant activity of the craft beers. Different lowercase letters indicate a significant difference between beer samples by Tukey's test ($p < 0.05$).

Caffeic and ferulic acids were found in B3, B4, B5, and B6. Caffeic acid is a natural phenolic compound biosynthesized by almost all plant species (Xiang et al., 2021). In a study by Kar et al. (2022) the presence of caffeic acid in the diet of hypothyroid rats inhibited lipid peroxidation, increased antioxidants, and decreased the levels of inflammatory and hepatic markers.

Ferulic acid is a simple phenolic acid commonly present in cereals and is known to have a free radical-scavenging effect (Vashistha et al., 2017). Several therapeutic properties of ferulic acid, including cardioprotective (Alam et al., 2013), anticancer (Gao et al., 2018), and neuroprotective (Elhessy et al., 2020) properties have been reported. Elhessy et al. (2020) observed that diabetic rats treated with ferulic acid showed a reduction in degenerative alterations in the cerebellum with a reduced the levels in the markers of oxidative stress.

Rutin was found in B1, B3, B4, B5, and B6, gallic acid in B3, B4, and B5, and kaempferol in B5 and B6. Rutin is a flavonoid found abundantly in citrus fruits (Elhessy et al., 2020). Studies have reported therapeutic effects of rutin on colonic inflammation, oxidative stress, and dysbiosis of the intestinal microbiota in mice (Liu et al., 2022), as well as anti-inflammatory effects of rutin on macrophages and T cells (Ganesan et al., 2021).

Gallic acid is a potent antioxidant which can scavenge free radicals such as superoxide anions, hydrogen peroxide, hydroxyl radicals, and hypochlorous acid (Kilic et al., 2019; Oyagbemi et al., 2016). Kaempferol has been reported for its role in cancer prevention, as well as to have several pharmacological properties, which include antimicrobial, anti-inflammatory, antioxidant, antitumor, cardioprotective, neuroprotective, and antidiabetic properties. Mechanisms of action of gallic acid includes apoptosis, cell cycle arrest in the G2/M phase, ownregulation of epithelial-mesenchymal transition related markers and phosphoinositide 3-kinase/protein kinase B signaling pathways (Imran et al., 2019).

All of the compounds detected in the craft beers in the present study have been previously reported in lager style craft beer, Pilsen, Märzenbier, non-alcoholic beer, IPA, and Weiss (Cheiran et al., 2019; Quifer-Rada et al., 2015). Caffeic acid derivatives found B3, B4, B5, and B6, and p-coumaric acid derivatives found in all the samples of craft beers are normally used to categorize craft beers as IPA, lager, and Weiss (Cheiran et al., 2019).

The FTIR spectrum analysis (Table 4) revealed the presence of different functional chemical groups in the craft beer samples. Bands with similar characteristics as CO, $CHCH_3$, C=C, CH, and OH groups were found in all the craft beer samples. The C=C and C-H groups are associated with the presence of phenolic compounds, whereas CO group is associated with the presence of alcoholic compounds; this composition may vary according to the beer style and fermentation process (Gordon et al., 2018).

In the FTIR spectrum of B6, CH and C=C bands showed the highest intensity (90.03%), and this may be associated with the high antioxidant activity of B6 (20.46 μg Trolox/g). Beer is a source of compounds with antioxidant potential, and antioxidant composition in beer depends not only on the raw materials but also on the techniques used in the production of beer (Jurková et al., 2012). In beer, malt and hops are the main sources of phenolic compounds, which are antioxidants (Benucci et al., 2021), and polyphenols, which impart characteristic sensory attributes to these products in terms of flavor, aroma, bitterness, and color (Collin et al., 2013).

The absorption spectra, 1260-1000 cm^{-1}, corresponds to CO bonds that are present in alcohols and indicates the presence of primary, secondary, or tertiary alcohols. In dark beers, lager and pale ale, the absorption bands observed between 1080 cm^{-1} and 1040 cm^{-1} are due to CO bonds of primary and/or secondary alcohols (Biancolillo et al., 2014; Gordon et al., 2018; Grassi et al., 2014). In the present study, absorption bands for C=O groups appeared between 1016 cm^{-1} and 1026 cm^{-1}, and this band was intense in B2, B3, B4, and B6, which presented an average of 5.55% of alcohol content.

For B3 and B4, SS content was 10.20 °Brix and 10.98 °Brix, respectively, and bitterness was 61% IBU and 66% IBU (as indicated by the manufacturer on the label), respectively. These characteristics may be associated with the C=O group identified in the study. These bands are characteristic of esters that are formed from lactic and acetic acids produced by bacteria and yeast (Colomer et al., 2019) or by the enzymatic reaction between acyl-CoA and alcohol during the fermentation process (Humia et al., 2020). The presence of esters is usually associated with the aroma of flowers, fruits, and herbs in foods (Egea et al., 2014), and has been shown to be essential for craft beers (Bettenhausen et al., 2018).

Sample B6 showed greater intensity in the C-H and C=C bands (90.03%), which may be related to the fact that this sample showed high antioxidant activity (20.46 μg Trolox/g). Beer is a source of compounds with antioxidant potential, and this composition depends not only on the raw materials, but also on the technology used in beer production (Jurková et al., 2012) such as phenolic compounds that are antioxidants from malt and hops (Benucci et al., 2021) and they contribute characteristic sensory attributes to these products, including taste, aroma, bitterness and color (Collin et al., 2013).

Table 4. Number of waves (cm^{-1}) and band designations displayed for the craft beer samples.

Chemical groups	B1	B2	B3	B4	B5	B6
	Number of waves					
s (C-O)	1016,44	1026,44	1026,44	1026,44	1016,44	1026,43
m-s (C-H-CH_3)	1364,14	1374,13	1374,13	1374,13	1385,24	1364,14
w-m (C=C)	1616,30	1616,30	1627,41	1627,41	1616,30	1616,30
w (C=O)	-	-	1732,94	1742,94	-	-
m-s (C-H)	2912,68	2912,68	2912,68	2933,79	2912,68	2933,79
L (O-H)	3270,38	3280,38	3291,49	3291,49	3291,49	3291,49

Where s = strong, m = mean, w = weak, L = wide.

4.2 Sensory profile of craft beers

CATA

In the present study, CATA method was used to evaluate the sensory profile of the craft beers. Aggregate participant CATA data can yield useful sensory profiles of food products. One reason for this is the linear relationship between CATA attribute citation rates and the attribute intensities (Jaeger et al., 2020).

The nonparametric Cochran's Q test revealed that the results for the attributes for aroma (little fermented/yeast, sweet/honey, citrus aroma), flavor (bitter sweet, fruity/fruit flavor, little fruity, yeast or fermented flavor, weak yeast flavor, and citrus flavor), and texture (presence of residues/particles and absence of residues) obtained by CATA were not significant and were therefore excluded from the CA (Table 5).

The results of the Cochran's Q test also demonstrated the discriminatory capacity of the participants, who were able to verify the differences between the craft beer samples for the 15 remaining attributes. Sensory analysis revealed an association between the craft beer samples and the sensory attributes in CATA, which explains approximately 78% of the original information in two dimensions (Figure 3). The first and second dimensions represent 26.6% and 51.5% of the total variability, respectively.

The greatest correlation between attributes was observed in B3, B4, and B6. Cloudy appearance was associated with higher SS content (10.2, 10.9, and 6.75 °Brix) and higher dry extract values (7.99, 7.48, and 5.26%) of B3, B4, and B5 than other craft beers.

B2 and B4 were classified to have a light appearance, and B5 was classified to have the lightest appearance of all the craft beer samples (17.63 EBC units), with a strong fruity aroma. Regarding appearance, B1 was associated as beer without foam and bubbles, and regarding texture it was associated as beer with high viscosity. Differences in beer viscosities have been linked to the quality of ingredients used in brewing, considering that high viscosity can still hinder steps such as filtration in beer processing (Rosa & Lannes, 2022).

Sensory acceptability

As expected, the sensory acceptance of the craft beers by attributes was satisfactory, with an average acceptability index

Table 5. Frequency of choice of each attribute of craft beers as a result of the Cochran Q test for CATA.

Attributes		B1	B2	B3	B4	B5	B6	p valor
Appearance	Yellow color	23	22	2	6	19	3	< 0.00000
	Brown color	0	0	18	17	4	18	< 0.00008
	Cloudy	13	5	11	16	11	16	< 0.02637
	Clear	9	17	6	6	12	7	< 0.01309
	Foam up	2	17	16	11	13	13	< 0.00008
	No foam up	19	6	6	11	9	9	< 0.00458
	Bubbles	3	14	13	10	10	10	< 0.04859
	No bubbles	19	9	9	12	13	11	< 0.13042
Aroma	Yeast aroma	7	12	16	15	11	8	< 0.24896
	Low yeast aroma	7	14	8	11	12	12	< 1.69692
	High fruity aroma	6	16	13	4	14	11	< 0.01456
	Low fruity aroma	19	5	18	9	9	11	< 0.00013
	Sweet/honey	13	14	13	10	12	15	< 7.36318
	Citric	8	9	10	12	11	6	< 5.04178
Flavor	Sweet	7	6	6	8	4	5	< 8.19985
	Bitter	16	16	17	15	18	18	< 8.99107
	Fruity presence	8	10	10	9	12	7	< 7.19250
	No fruity presence	16	12	13	11	11	14	< 6.01546
	Yeast flavor	15	11	14	13	13	13	< 8.95636
	No Yeast flavor	7	13	7	10	8	9	< 4.46367
	Citric flavor	11	11	8	12	14	14	< 4.62140
	No citric flavor	12	12	14	9	9	9	< 5.05952
Texture	No foam up	7	8	12	8	20	11	< 0.00403
	Body foam	16	15	11	15	4	2	< 0.01463
	Low viscosity	10	10	10	15	14	19	< 0.27513
	High viscosity	12	12	13	8	9	4	< 0.65621
	Presence of particles	3	2	4	7	2	2	< 1.29375
	No presence of particles	20	21	19	16	21	10	< 2.06767

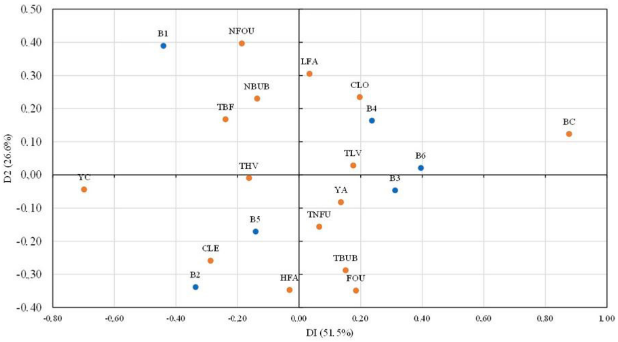

Figure 3. Representation of samples (B1, B2, B3, B4, B5, and B6) and the attributes (see footnote) of check-all-that-apply data after correspondence analysis.Apparence: YC – yellow color; BC – brown color; CLO – cloudy; CLE – clear; FOU – foam up; NFOU – no foam up; TBUB – bubbles; NBUB – no bubbles. Aroma: YA – yeast aroma; HFA – high fruity aroma; LFA – low fruity aroma. Texture: TNFU – no foam up; TBF – body foam; TLV – low viscosity; THV –high viscosity.

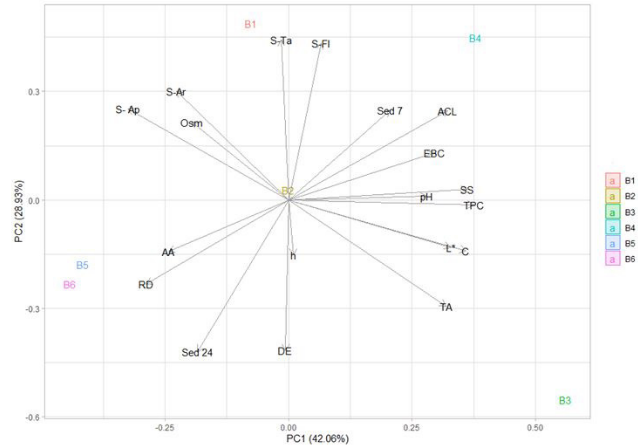

Figure 4. Score chart of the principal components calculated from the results obtained from the biochemical, physicochemical, and sensory analysis of the craft beers. RD – relative density; TA – titratable acidity; pH – hydrogen potential; ACL – alcohol; SS – soluble solids; SED 24 – sedimentation at 24 °C; SED 7 – sedimentation at 7 °C; DE – dry extract; EBC – EBC color; L* – luminosity; h – hue; C – chroma; OSM – osmolality; AA – antioxidant activity; and TPC – total phenolic compounds. Sensory acceptability: S-AP – appearance; S-AR – aroma; S-TA – flavor; and S-FI – texture.

of 75.11% for appearance, 70.24% for aroma, 60.43% for flavor, and 73.53% for texture (Table 1). The appearance and texture attributes obtained a higher acceptability index, reaching 82% and 85% for B6 and B4, respectively. Higher acceptance, with regard to texture and flavor, was noted for B4 (Table 1), which may be associated with the higher SS content (10.98 °Brix) and dry extract (7.48%) in B4 than other craft beers (Figure 1).

A lower acceptability index for flavor was noted for B3 than other craft beers, which may be associated with the high content of total phenolic compounds (108.45 mg/100 mL) and titratable acidity (3.45%) in B3. The presence of phenolic compounds can result in an astringent and bitter taste (Maye et al., 2016; Oladokun et al., 2017).

4.3 Principal component analysis

Physicochemical properties, such as relative density, titratable acidity, pH, alcohol content, SS content, sedimentation at 7 °C and 25 °C, dry extract, osmolality, TPC, antioxidant activity, EBC, L* value, hue, chroma, and the acceptability index of craft beers for appearance, aroma, flavor, and texture were determined using PCA (Figure 4). In this multivariate analysis technique, it is possible to analyze the interrelationships between variables by combining the original coordinates to create a new space with metrics capable of grouping samples according to similarities that cannot be perceived in the original space. The first main component (PC 1) explained 42.06% of the total variability contained in the original variables, and the second main component (PC 2) explained 28.93%, which was 70.99%. in total.

PCA grouped the craft beer samples with similar biochemical, physicochemical, and sensory properties. The resulting groups were sufficiently isolated from each other (Figure 4). Samples B3 and B4 showed higher TPC (108.45 and 100.23%) of soluble solids (~10 °Brix), alcohol (> 5%), acidity (3.45 and 2.22%) and average pH of 4.8.

B5 and B6 showed higher antioxidant activity (16.54 and 20.46 µg of Trolox/g, respectively) than other craft beers. With regard to the acceptability index, B6 and B2 showed greater acceptance (82% and 78%, respectively) than other craft beers. B3 showed the highest titratable acidity as opposed to B1 and B4, which showed greater acceptance (67.57% and 73.42%, respectively), demonstrating an inverse relationship of flavor and titratable acidity.

5 Conclusion

Our study provides insights into the profile of biochemical and physichochemical properties and sensory profile of the craft beers, which can be useful for further research on the sensory profile and health promoting properties of beer. The craft beers showed variations in their physicochemical properties, phenolic compositions, and alcohol contents. Our study of craft beers revealed greater acceptability for less acidic, lighter colored, and fuller-bodied beers.

Conflict of interest

The authors declare no conflict of interest.

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