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Original Article

# Production and Physicochemical Characterization of Craft Beers with Sapodilla Fruit (*Manilkara Sapota* L.)

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## Abstract

Considering that there is still no fruit beer with sapodilla in the Brazilian market, the aim of this study was to elaborate beers using sapodilla (*Manilkara sapota* L.) pulp and evaluate its physicochemical and sensory characteristics. Three beer formulations were elaborated: beer without the addition of sapodilla pulp (beer without fruit), beer with the addition of sapodilla pulp at concentration of 8.5% (w/v) and beer with the addition of sapodilla pulp at concentration of 17% (w/v). The addition of sapodilla in the worts contributed to increase their sugar content (6.39-7.41%) compared to the wort without fruit (6.02%), and consequently, increased fruit beers alcohol content (4.60-5.60ºGL) in relation to beer without fruit (4.00ºGL). The phenolic compounds of the beers ranged from 72.00 to 107.81 mg 100 $mL^{-1}$, and the antioxidant activities of the beers ranged from 1.27 to 1.76 mM TEAC for the ABTS method and from 0.56 to 0.73 mM TEAC for the DPPH method. It was important to study the concentration of fruit pulp to be added in the formulation of beers, as the beer with 8.5% sapodilla had a better sensory acceptance than the beer with 17% sapodilla. Thus, it is possible to increase the quality of beer by adding sapodilla fruits at concentration of 8.5%, which positively impacted the flavor and aroma of the product.

**Keywords:** Fruit beer, Sapodilla fruit, Physicochemical analysis, Sensory analysis

## INTRODUCTION

Recently, there has been an increase in the production and consumption of craft beer in the Brazilian market [1,2]. The main characteristic of craft breweries is to emphasize flavor and brewing techniques and thus compete with the mass-market breweries for quality and diversity rather than low prices [3]. Consumers choose craft beers over conventional beers, as they have a variety of styles, flavors, and aromas, and are superior in quality [4].

The perceived quality of beers produced with local ingredients as fruits, herbs and spices represents an important element for the successful development of craft breweries, as it corresponds to the consumer's desire to rediscover the authenticity of local products [4,5]. The use of fruit in craft beers provides unique sensory attributes, with characteristic flavors and aromas, becoming a consumer preference factor [6].

Fruits have been used as beer adjuncts for centuries, especially in Belgian Lambic style, where cherry or raspberry is traditionally added to beers [7,8]. Currently, the fruit beer market is expanding, and nowadays consumers can find fruit beers made of strawberry, cherry, raspberry, blackberry, apricot, peach, plum, tangerine, orange, grape, and apple available in the markets [8,9].

Sapodilla (*Manilkara sapota* L.) is a tropical fruit native to Central America, and in Brazil, it is cultivated mainly in the northeastern region, due to the hot and dry climate conditions associated with irrigation. The fruit is a large ellipsoid berry, in general from 4 to 8 cm up to 15 cm wide, containing few seeds (two to five big seeds) and when fully ripened it presents high pulp yield and very sweet flavor. In the various countries where sapodilla is produced, it is mostly consumed fresh or used to produce jams, compotes, and juice [10,11]. Considering that there is still no sapodilla fruit beer in the Brazilian market, the aim of this study was to elaborate fruit beers using sapodilla pulp and evaluates its physicochemical and sensory characteristics.

## MATERIAL AND METHODS

### Materials

Sapodilla fruits were obtained in a market of local farmers of Distrito Federal (Brazil). The sapodilla fruits came from the northeastern state of Pernambuco, Brazil. The dry ale yeasts (Safale US-05) were purchased from Fermentis Ltd. (Marcq-en-Baroeul, France). Pilsen malt from Germany, Château

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Cara Gold malt from Belgium and Nugget hops from USA were purchased from a specialized store and used for the wort production.

**Beer production**

The wort was prepared from 3.0 kg of Pilsen and 1.5 kg of Château Cara Gold type malts which were crushed to the appropriate size resulting in 4.5 kg of ground malt and mixed with 12 L of water. The mashing process was carried out in three heating steps: 20 min at 50-55°C, 20 min at 60-65°C and 25 min at 70-75°C. After, wort was filtered, and the solid part was washed using 8 L of mineral water at 78°C. Subsequently, the liquid part proceeded to the boiling step and the solid part was discarded. The wort was boiled for 60 min and during this process 30 g of hops were added. The boiled wort was separated from the hot trub and then cooled at 18°C. Prior to fermentation, the yeast (0.7 g/L) (Safale US-05) was rehydrated and added into the wort. The sapodilla pulp was also added into the wort before the fermentation. The fermentation was carried out in a fermenter with sealed lid and airlock coupled for 10 days at 20°C. After the fermentation process, the yeasts and sapodilla bagasse settled at the bottom of the fermenter and the green beer was transferred to another fermenter, and this sediment was eliminated. The green beers were kept at 5ºC for 15 days to obtain matured beers. The beers were filled in 600 mL amber glass bottles and glucose syrup (1 g/L) and a yeast suspension (0.2 g/L) were added for the carbonation process. Bottled beers were sealed and stored at room temperature for 15 days to form carbon dioxide.

**Addition of sapodilla (*Manilkara sapota* L.) pulp into the wort**

In the laboratory, the sapodilla fruits were sanitized in a solution of sodium hypochlorite (200 ppm) for 15 min and washed in running water before use. The fruits were pulped, and the seeds were discarded. The sapodilla pulp was added into the brewer's wort at concentrations of 8.5% (w/v) and 17% (w/v) and sapodilla pulp presented a soluble solid content of 25ºBrix. After the mashing process, a wort volume of 18 L was obtained with a soluble solids content of 13ºBrix. The wort was divided into 3 fermentation vessels (6 L of wort in each fermenter). Three beer formulations were elaborated: beer without the addition of sapodilla pulp (beer without fruit), beer with the addition of sapodilla pulp at concentration of 8.5% (w/v) and beer with the addition of sapodilla pulp at concentration of 17% (w/v).

**Physicochemical analyses**

The soluble solids were determined through refractometer Shimadzu and expressed in °Brix. The pH was measured using a digital potentiometer Micronal B222 and total acidity was determined by titration with 0.1 N NaOH [12]. Reducing sugars were determined using the 3,5-dinitrosalicylic acid method [13]. Total polyphenols were determined by the Folin-Denis method [14], using gallic acid as a reference compound. The in vitro antioxidant activity was performed through the evaluation of free radical capture DPPH [15] and ABTS [16]. The results were expressed as Trolox equivalents (mM TEAC) and calculated from a standard curve. The alcoholic content of the beers was determined using a Gay-Lussac alcoholometer placed directly in a volume of 250 mL of distillate at 20°C [17].

**Sensory analysis**

The sensory analysis of the beers was performed by a well-trained panel of 12 evaluators of both sexes aged between 21 and 60 years. Each evaluator received 70 mL of each beer sample at approximately 8°C, coded with three random digits. Mineral water and salt and cracker were also served to clean the palate. Flavor, aroma, appearance (color and turbidity), and global acceptance of beers were evaluated following nine-point hedonic scale (9 = like extremely, 8 = like very much, 7 = like moderately, 6 = like slightly, 5 = neither like nor dislike, 4 = dislike slightly, 3 = dislike moderately, 2 = dislike very much, 1 = dislike extremely) [18].

**Statistical analysis**

Data were expressed as mean values ± standard deviation of three independent replicates. Physicochemical and sensory analyzes results were submitted to analysis of variance (ANOVA) and the means were compared by Tukey test (Statistica 10®, Statsoft Inc). Results were considered significantly different when p values were $< 0.05$.

## RESULTS AND DISCUSSION

**Table 1** presents the physicochemical analyzes of the worts (wort without the addition of sapodilla pulp, wort with addition of sapodilla pulp at concentration of 8.5% (w/v) and wort with the addition of sapodilla pulp at concentration of 17% (w/v).

**Table 1.** Physicochemical analyses of worts.

| Analyses | Wort without fruit | Wort with sapodilla 8.5% | Wort with sapodilla 17% |
|---|---|---|---|
| pH | 6.14±0.05 [a] | 6.10±0.09 [a] | 6.04±0.07 [a] |
| Total acidity (%) | 0.21±0.00 [a] | 0.20±0.03 [a] | 0.20±0.01 [a] |
| Reducing sugars (%) | 6.02±0.02 [a] | 6.39±0.08 [b] | 7.41±0.12 [c] |
| Phenolic compounds (mg 100 mL $^{-1}$) | 146.41±5.96 [a] | 132.16 ±7.93 [a,b] | 115.91± 6.95 [b] |

Values are the means ± standard deviations of triplicate measurements. Values in the same line with different letters are statistically different ($p < 0.05$).

The worts presented pH from 6.04 to 6.14 and acidity from 0.20 to 0.21%. The addition of sapodilla pulp did not increase the acidity of the worts, as sapodilla is a low acid fruit. Oliveira [10] reported that during the ripening of sapodilla, acidity decreased from 0.31 to 0.16%. The acidity of sapodilla is very low when compared to other acidic fruits such as acerola (1.80%) [17]. Martinez [9] showed similar results, where the wort with the addition of a lower concentration of persimmon juice (50%) had a pH of 4.58, while the wort with the addition of a higher concentration of persimmon juice (75%) had a higher pH of 5.73. On the contrary, Kawa-Rygielska [19], reported that the wort without fruit had a pH of 5.31, while the wort with addition of red Cornelian cherries had a lower pH of 3.55.

The values of reducing sugars in the worts of this study ranged from 6.02 to 7.41%. The reducing sugars represent the main fermentable sugars (maltose and glucose) of the brewer's wort [20]. The addition of sapodilla pulp in the worts contributed to the increase the sugar content in relation to the wort without fruit. According to Oliveira [21], sapodilla is a fruit with a high content of sugars in its composition (11.2%). Kawa-Rygielska [19] also observed an increase in the concentration of sugars comparing the wort without fruit (7.97%) and the wort with addition of red Cornelian cherries (8.22%). Nunes [22] reported that the wort with the addition of 30% of cocoa pulp had 8.20% of fermentable sugars (glucose, fructose, and maltose).

The contents of phenolic compounds in the worts ranged from 115.91 to 146.41 mg 100 mL $^{-1}$. In the brewer's wort, about 70 to 80% of the phenolic compounds come from malt and 20 to 30% come from hops [23]. The addition of sapodilla pulp did not increase the phenolic compounds content of the worts, which is an unusual result, as tropical fruits are generally rich in phenolic compounds [24]. Oliveira [10] reported that the ripening of sapodilla resulted in a decrease in total phenolic content from 2091.00 mg 100 g $^{-1}$ to 67.00 mg 100 g $^{-1}$.

**Table 2** presents the results of the physicochemical analyzes of the three beers of this study (beer without fruit, beer with addition of sapodilla pulp at concentration of 8.5% and beer with the addition of sapodilla pulp at concentration of 17%).

**Table 2.** Physicochemical analyses of the beers.

| Analyses | Beer without fruit | Beer with sapodilla 8.5% | Beer with sapodilla 17% |
|---|---|---|---|
| pH | 4.59±0.08 $^{a}$ | 4.20±0.01 $^{b}$ | 4.15±0.01 $^{b}$ |
| Total acidity (%) | 0.26±0.01$^{a}$ | 0.31±0.02$^{b}$ | 0.34±0.04$^{b}$ |
| Soluble Solids (ºBrix) | 6.25±0.01$^{a}$ | 6.00±0.01$^{b}$ | 6.00±0.01$^{b}$ |
| Reducing sugars (%) | 2.15±0.01 $^{a}$ | 1.95±0.07 $^{a}$ | 1.60±0.05 $^{b}$ |
| Alcohol content (ºGL) | 4.00±0.01$^{a}$ | 4.60±0.01$^{b}$ | 5.60±0.01$^{c}$ |
| Phenolic compounds (mg 100 mL $^{-1}$) | 107.81±2.20 $^{a}$ | 96.00±4.59 $^{b}$ | 72.00±5.90 $^{c}$ |
| ABTS (mM TEAC) | 1.76±3.83 $^{a}$ | 1.34±4.27 $^{b}$ | 1.27±5.60 $^{c}$ |
| DPPH (mM TEAC) | 0.73±2.76 $^{a}$ | 0.70±2.17 $^{a}$ | 0.56±2.34 $^{b}$ |

Values are the means ± standard deviations of triplicate measurements. Values in the same line with different letters are statistically different ($p < 0.05$).

During alcoholic fermentation, there is an increase in the acidity of beers (0.26-0.34%) in relation to worts (0.20-0.21%), due to the formation of organic acids by the yeasts [25]. And even though sapodilla is a low acid fruit that initially did not increase the acidity of the worts, its addition contributed to increase the acidity of fruit beers (0.31-0.34%) in relation to beer without fruit (0.26 %). Moreover, the pH of beers with sapodilla (4.15-4.20) was lower than the pH of beer without fruit (4.59). Martinez [9] reported similar results and observed that the acidity of beer formulation with higher percentage of persimmon juice increased compared to beer without fruit (from 0.60% in malt beer to 1.49% in 25:75% wort-persimmon juice beer). Kawa-Rygielska [19] also observed a decrease in pH from 4.59 in beer without fruit to 3.56 in beer with red Cornelian cherries. The acidity of fruit beer is affected by both the type and quantity of fruits added to wort as well as by the fermentation process which decreases the pH of beer [9,26].

Sugars in worts (6.02-7.41%) were reduced in beers (1.60-2.15%), showing that yeasts consumed these sugars to produce ethanol. Martinez [9] showed that the initial glucose concentration (6.07%) of the wort with 75% persimmon juice was reduced to 0.23% after 3 days of fermentation. Kawa-Rygielska [19] reported a concentration of 2.54% glucose and maltose in beer with red Cornelian cherries. The soluble solids values of the beers in this study ranged from 6.00-6.25ºBrix. Some fruit beers showed a decrease from 13.00-12.00°Brix in worts to 5.92-4.50°Brix in beers [6,9,21,27]. The decrease in Brix values is caused by consumption of sugars due to their utilization by fermenting yeast [26].

The addition of sapodilla in beers increased their alcohol content (4.60-5.60ºGL) in relation to beer without fruit (4.00ºGL). Martinez [9] reported similar results and ethanol

content of the beers depended on the initial reducing sugar concentration in their formulations and, therefore, those containing more persimmon juice contained more alcohol (100:0, 75:25, 50:50 and 25:75% wort-persimmon beers contained 2.91, 3.64, 4.72 and 5.63% ethanol, respectively). Zapata [28] reported that the addition of quince to beer slightly increased its alcohol content. The beer without fruit was the one with the lowest alcohol concentration (5.27%), while the beers with quince fruits had alcohol concentrations of 5.51-5.62%. Nunes [22] also reported that compared with the beer produced without fruit, an increase in ethanol production was observed with the addition of cocoa pulp as an adjunct in fruit beer that presented 5.95-6.02% of ethanol. Thus, the addition of fruits rich in sugars has the potential to increase the alcohol content of fruit beers.

The phenolic compounds of the beers ranged from 72.00 to 107.81 mg 100 $mL^{-1}$ and these values were higher than the values reported by Koren [29], where dark beer available in Hungarian retail presented 42.79 mg 100 $mL^{-1}$ of phenolic compounds. Granato [30] analyzed 11 brands of Brazilian Ale beers and the values of phenolic compounds ranged from 28.01 to 52.59 mg 100 $mL^{-1}$. The beer without fruit in this study showed a higher value of phenolic compounds (107.81 mg 100 $mL^{-1}$) than the beers with sapodilla (72.00-96.00 mg 100 $mL^{-1}$). A similar result was observed by Martinez [9], where the beers formulations showed a decline in the values of phenolic compounds with the increase of the concentration of persimmon juice, so the beer without fruit showed a value of phenolic compounds of 55.52 mg 100 $mL^{-1}$ and the beers with 25 and 75% of persimmon juice showed phenolic compounds values of 30.88 mg 100 $mL^{-1}$ and 29.03 mg 100 $mL^{-1}$, respectively. Both sapodilla and persimmon are not fruits rich in polyphenols and therefore did not contribute to the increase of these in beers.

The antioxidant activities of the beers in this study ranged from 1.27 to 1.76 mM TEAC for the ABTS method and from 0.56 to 0.73 mM TEAC for the DPPH method. Zapata [28], found antioxidant activity of 0.72-0.73 mM TEAC by the ABTS method in beers with quince fruits. Deng [31] reported values of 0.18-0.76 mM TEAC by the DPPH method in omija fruit beers. The antioxidant activities of beers with sapodilla were lower than those of beer without fruit. These results were already expected, as there is a positive correlation between the content of phenolic compounds in beer and its antioxidant capacity [29]. Oliveira [10] reported a drastic decrease in antioxidant activity during sapodilla ripening (from 0.42 to 0.01 mM TEAC by the ABTS method). Martinez [9] also reported that the antioxidant activity of beer without fruit was significantly higher than that of beers with persimmon juice.

Moderate consumption of beer can have a positive effect on human health. Beers are rich in vitamin B, minerals, and biologically active components. In beers, folic acid is found in relatively high concentrations. The most common polyphenols of beers are simple phenols, phenolic acids, benzoic acid derivatives, cinnamic acids, flavanones, flavanols, flavonols, isoflavones and flavones. These polyphenols are antioxidants and protect against oxidative stress caused by reactive oxygen and nitrogen species. Thus, drinking beer has been reported to increase plasma antioxidant activities and positively affect plasma lipid levels in humans. In addition, moderate beer consumption has been reported to exert protective effects on cardiovascular risk factor [8,29].

The sensory evaluation scores for different beers are shown in **Table 3**. The formulations preferred by the evaluators were beer without fruit (score of 7.50 for global acceptance) and beer with sapodilla 8.5% (score of 7.13 for global acceptance). Other studies also reported a good acceptability of fruit beers. Santos [6] reported a score of 7.60 for overall acceptance of the soursop fruit beer. Nunes [22] obtained a score of 7.16 for overall impression of the cocoa fruit beer. Additionally, Martinez [9] reported a global acceptance of 7.10 for the beer formulated with 25% of persimmon juice.

**Table 3.** Sensory analysis of the beers.

| Attributes | Beer without fruit | Beer with sapodilla 8.5% | Beer with sapodilla 17% |
|---|---|---|---|
| Flavor | 7.37 [a] | 6.75 [a] | 4.75 [b] |
| Aroma | 6.87 [a] | 6.87 [a] | 5.63 [b] |
| Appearance (color and turbidity) | 7.87 [a] | 6.00 [b] | 4.12 [c] |
| Overall acceptance | 7.50 [a] | 7.13 [a] | 4.63 [b] |

Values are expressed as means. Values in the same line with different letters are statistically different ($p < 0.05$).

The fact that beer without fruit achieves the highest global acceptance score is not an unexpected result, as this beer represents the standard expected by the consumer. Although, the averages of the attributes flavor, aroma, and global acceptance of the beer with 8.5% sapodilla pulp were all above 6.5 points and statistically there was no difference from the beer without fruit, indicating that the evaluators had a good receptivity to this product. The appearance attribute of the beer with sapodilla 8.5% had the lowest score, probably due to the higher turbidity compared to the beer without fruit. However, the beer with 17% sapodilla pulp did not have a good sensory acceptance and obtained the lowest score of 4.63 for global acceptance, probably because the excess of fruit pulp incorporated in the formulation impaired its sensory attributes, which received much lower scores compared to the 8.5% sapodilla beer.

Our study showed that it is important to carry out tests to define the best concentration of fruit in the beer formulation, to obtain a good sensory acceptance of the product. Other studies also described tests where the concentration of fruit in the beer formulations varied. Castro Marin [32] added

different amounts of juice (5, 10 and 20%) from Lambrusco red grapes to Lager wort, before primary fermentation. Contrary to 5 and 10%, the addition of 20% of red grapes juice significantly impacted the olfactory and gustative descriptors, being described as more astringent, fruity, and acidic beer. Zapata [28] added quince fruits at different percentages (5, 10 and 15%) to a standard Ale beer. After sensory analysis, the evaluators decided that the addition of a 10% of quince fruit to the beer was the best formulation, providing proper beer flavor, but getting the plus of getting quince key notes and no off-flavors. Kyung-Mi [7] also tested different concentrations of peach (3, 5 and 8%) in beer formulations and based on the overall acceptability score, the optimum proportion of peach fruits to be added was 5%.

## CONCLUSIONS

The production of craft beer with sapodilla was successful. It was important to study the amount of fruit pulp to be added in the formulation of beers, as the beer with 8.5% sapodilla had a good sensory acceptance, while the beer with 17% sapodilla was not well accepted. Due to the high concentration of sugars present in the fruit, beers with sapodilla had higher alcohol content when compared to beer without fruit. Thus, it is possible to increase the quality of beer by adding sapodilla fruits, which positively impacted the flavor and aroma of the product.

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## DECLARATION OF COMPETING INTEREST

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

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