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Blackberry (*Rubus* spp.) chutney manufacturing aiming its antioxidant potential

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ABSTRACT

The blackberry cultivation has a great potential in the production chain of small fruits in Brazil, being a rich source of nutritious compounds and natural antioxidants. This study aimed to quantify bioactive compounds such as phenolic compounds, anthocyanins, vitamin C, etc., determine the antioxidant potential and evaluate the stability of this potential for the development of two formulations of blackberry chutney, with a 60% pulp and another with 80%. The study also presents the development of sensory analysis to evaluate acceptance and purchase intent by the tasters. A partial degradation of the evaluated compounds occurred during the drafting process of chutneys; however, this degradation was less intense in 80% chutney, which was the expected result. The sensory analysis showed good acceptability for the 80% chutney, however, the 60% chutney did not reach the average required for acceptance of 70%. Being an innovative product that does not have extensive marketing, it can be said that the blackberry chutney is a viable alternative for consumers, who are increasingly seeking healthier products, and for the food industry that is always looking for news to offer to the public.

KEYWORDS: bioactive compounds; fruit processing; sensory analysis.

INTRODUCTION

The main blackberry production regions of the world are located in North America, Europe and Asia. However, blackberry production in Brazil is growing mainly in the south and southeast regions, estimating a cultivation of 150 tons and a total area of 450 hectares. Paraná has good climatic conditions for the production of blackberries (Celant et al., 2016).

Blackberries are increasing in economic value because of their high nutritional value and benefits to physical and mental health, which can be largely attributed to their phenolic compounds, such as phenolic acids, tannins and anthocyanins (Yamashita et al., 2017). The blackberry production is concentrated in the months from November to February, and because of their high respiratory rate, the fruits have become highly perishable, creating limitations on the fresh fruit market (Mota, 2006a; Kuskoski et al., 2006).

Fruits such as berries (strawberry, blackberry, raspberry, blueberry, grape and cherry) are characterized as being small and fleshy and are consumed in many countries, primarily in natura, as juices and jams or through pulps in industrialized products. Recently, consumers' interest in these fruits has grown due to their beneficial health effects because of the antioxidant and anticancer properties and because the berries are a good source of fiber, vitamins (B, C and K), folic acid, fatty acids, polyphenols (anthocyanins) and minerals, such as Cu, Fe, I, K, Mg, and Mn (Pereira et al., 2018).

The use of fruit for the development of industrial products presents itself as a viable alternative in meeting consumer needs, seeking an innovative product quality, and providing a healthy diet rich in nutrients. The industrial use of this fruit in the form of chutney could be a new option for the use of blackberry and an alternative for consumers who crave a differentiated product in their daily lives.

In addition to products based in fruits that are normally marketed, the chutney can be cited as a differentiated product, but with the manufacturing process very similar to the fruit jelly. Chutney is an Indian sauce, spicy, consumed as a side dish to other foods, mostly meats. Chutney is the generic term for a thick sauce, comprising fruits or vegetables, sugar, spices and sometimes vinegar. The technology involved in the processing is simple, varying only in the ingredients used, cooking time and chemical characteristics of the final product (Madakadze et al., 2004). Its high sugar content has a preservative effect and vinegar addition is not always necessary, depending on the natural acidity and ripeness of the fruits used. The natural fruit acids, vinegar or those produced by fermentation, along with the high sugar content, are used to preserve Chutney after opening the bottle. According to Madakadze et al., (2004), a conservation index can be used to calculate the amounts of ingredients to be added. The same authors indicate that the conservation index allows us to "measure" the conservation power of the acid and sugar combinations, being measured as total solids. This index should not be lower than 3.6%. Thus, this study aims to use the blackberry, fruit rich in bioactive compounds such as antioxidants, to prepare chutney.

MATERIALS AND METHODS

Fruits of blackberry (*Rubus* spp.), grown in the region of Vacarias (Rio Grande do Sul, Brazil) and marketed in Shinai, in the city of Maringá (Paraná, Brazil) were used. The ingredients used in chutney formulations were also purchased from the local market in the same city. Evaluations and chutney formulations were carried out in the Food Technology laboratories in the Department of Food Engineering (DAL) and Biochemistry Food Laboratory (DQI), the State University of Maringa - UEM, Maringa, Paraná, Brazil. The reagents used in the analysis were from brand Anidrol, the existing ones in the laboratory or those purchased from the local distributor of chemicals.

PROCESSING OF BLACKBERRY CHUTNEY

The ingredients and their amounts used in the preparation of formulations of blackberry chutney are presented in Table 1. The percentages of blackberry chosen were based on preliminary tests, observing the best results of antioxidants.

Table 1. Ingredients

Ingredients	60% Chutney	80% Chutney
Blackberry	60	80
Apple	40	20
Sugar**	30	30
Water	40	20
Industrial Citric Pectin***	1	1
Calcium Chloride**	0.005	0.005
Garlic***	1.7	1.7
Thyme + rosemary***	0.7	0.7
Olive oil***	1.6	1.6
Vinegar***	10.7	10.7
Salt***	1	1
Pepper***	21	21

NOTE: * Amounts expressed in percentage (%); ** Amount referring to the sum of the masses of pulp, blackberry and apple; *** Amount referring to the sum of the masses of pulps (blackberry and apple) and sugar.

For the chutney preparation, frozen fruits, at -0.5°C, obtained at the same place of the fresh fruits and sanitized, were used. To obtain the pulp, the fruit was weighed and triturated in an industrial blender (Skymsen) at 1500 rpm, followed by sieving to remove the seeds. The apple was also crushed in a blender with 40 mL of water. The obtained pulps were weighed and placed in an inox pan to begin the concentration step, along with water and ¾ the amount of refined sugar. The remaining sugar was mixed with pectin and calcium chloride and reserved.

During cooking, the total soluble solids content (TSS) was monitored with the aid of a refractometer (Biofocus AR1000C). With the onset of boiling, the remaining sugar mixed with pectin and calcium chloride was added and again the TSS content was monitored. After reaching 27ºBrix, the step of concentration was over. Then, the remaining ingredients for chutney were properly weighed and added to a pan to obtain seasoning, starting with the addition of olive oil and garlic, followed by the other ingredients. The seasoning cooking was maintained for 2 minutes after the start of the boil. As the spice was complete, it was added to chutney and the mixture was again heated. Then, the chutney was bottled hot in glass containers previously sterilized at 121 °C/15 min and cooled by immersion in cold water for 15 min.

PHYSIC-CHEMICAL ANALYSIS

The color was evaluated with Konica Minolta reflectance colorimeter. The pH was determined using digital potentiometer and the total soluble solids content with the aid of digital refractometer, the values were expressed in °Brix. The total acidity was carried out by titration method, according to the methodology described by the Food Technology Institute - ITAL (1990). Through these analyzes it was possible to determine the ratio TSS/ATT (ITAL, 1990). The moisture quantification was performed by drying at 105°C to constant weight (IAL, 2005). The ashes were quantified by incineration in muffle furnace at 550 °C (IAL, 2005). The total and reducing sugars were determined using the titration method of Lane-Eynon, with the use of Fehling Solutions A and B factored (IAL, 2005). For the protein content the methodology of Semi-Micro-Kjeldahl was used, and the lipid content was obtained by cold extraction method, according to Bligh and Dyer (1959).

Determination of ascorbic acid content was performed by titration method by Tillmans (ITAL, 1990). Neutralization titration of ascorbic acid in 2.6 diclorobenzenoindofenol solution. The extraction of samples was made with oxalic acid solution 1%. Phenolic compounds were obtained by determining the equivalent gallic acid (SAE) in the sample, from reading spectrophotometer UV/Vis. Extraction of anthocyanins was performed according to Lee and Francis methodology (1972) with some modifications. Finally, the method of the antioxidant power was made by reduction of ferric (FRAP) and conducted according to Benzie and Strain (1996).

SENSORY ANALYSIS

The sensory analysis was carried out at the State University of Maringá, Maringá (Paraná, Brazil), methodology described by Meilgaard, Civille and Carr (1991), with one hundred and twenty (120) untrained men and women with different age groups were recruited and performed acceptance testing in hedonic scale 1-9 points for evaluation of color, aroma, flavor, texture and overall acceptance separately and the intention purchase. Sensory analysis was performed with the product at room temperature, with a cracker and a glass of water. Each tester received two randomly-coded samples, a free and informed consent form, to guarantee the voluntary nature of the research, and a sensory evaluation form.

STATISTICAL ANALYSIS

All the physical and chemical determinations were performed in triplicate and results, including the sensory ones, were assessed by analysis of variance

(ANOVA) and Tukey test, both at the 5% level of significance, using Excel 2010 and Sisvar 2.0 statistical program.

RESULTS AND DISCUSSION

The results of the characterization of blackberry fruits and chutneys color are shown in Table 2. The color parameters indicate the brightness (L*) and chromaticity of the sample (+a* direction for the red, -a* direction to the green, +b* direction for the yellow and -b* direction for blue). The chroma (C*) expresses the saturation or intensity of color, while the hue angle (h).

Table 1. Color analysis

Parameters	Pulp	60% Chutney	80% Chutney
L	17.02±0.42^{a}	22.26±2.36^{b}	26.88±1.13^{c}
a	6.58±1.88^{a}	15.55±2.26^{b}	22.73±0.31^{c}
b	1.78±0.62^{a}	5.76±0.69^{b}	9.24±0.58^{c}
c	6.81±1.87^{a}	22.73±1.91^{b}	24.39±0.70^{c}
h	14.87±1.43^{a}	25.86±1.01^{b}	22.23±0.79^{c}

NOTE: Equal letters in the same line do not differ by Tukey test at 5% significance level.

The color of the fruit and its derived products is an important parameter for producers and consumers, as it indicates whether or not the fruit has ideal conditions for marketing and consumption and, in general, consumers have a preference for strong and bright colored fruits. Tosun et al. (2008) explains that the brightness (L) decreases with the ripening of the blackberry fruit, making the color of the fruit darker. The chroma indicates the color intensity, ie the higher values indicate the samples with more vivid colors. The Bramble purple color is related to the presence of a large amount of phenolic compounds and the nearest h values of 0 indicate fruits tending to red, while values closer to 90 indicated a tendency to yellow.

Table 2 shows a significant difference between all color parameters for pulp and both chutneys formulations. It can be seen that the products brightness increases as the percentage of fruit becomes larger. At the same time, by increasing the percentage of fruit, red color (+) and chroma (C) are more evidenced in 80% chutney.

The results of the determinations of pH, total soluble solids, moisture, ash, titratable acidity and ratio are presented in Table 3 below. The consistency of chutney is the result of a set of factors such as sugar concentration, acidity and concentration of pectin. In the case of acidity, pH optimum to provide this consistency is close to 3.2. It can be noted that the blackberry pulp has a pH close to the aforementioned, which allowed achieve consistency Chutney without acidification.

From the table it can be observed that the pulp has an acid pH and it remained in the same pattern for both blackberry chutney formulations, ie, no significant difference ($p = 0.05$) between values. This characteristic can be considered favorable for conservation of the product, preventing the proliferation of microorganisms and allowing a long shelf life. Moreover, the high water activity of chutneys can damage the product for making this a suitable food for growth of these organisms.

Table 2. Physic-Chemical Analysis

Analysis	Pulp	60% chutney	80% chutney
Ph	3.41±0.05^{a}	3.32±0.03^{a}	3.34±0.18^{a}
Total Soluble Solids	7.75±0.25^{a}	27.93±0.11^{b}	33.27±0.25^{c}
Moisture	89.61±0.84^{a}	65.15±3.50^{b}	59.72±0.69^{b}
Ash	3.125±3.34^{a}	0.935±1.84^{b}	0.806±0.77^{b}
Titratable acidity	1.44±0.02^{a}	1.32±0.014^{b}	1.45±0.02^{a}
Ratio	5.38±0.10^{a}	21.16±0.16^{c}	22.94±0.29^{c}

NOTE: Soluble solids expressed in °Brix; Moisture and ash expressed in percentage (%); Titratable acidity expressed in g citric acid/100 g of pulp; Ratio is expressed by SST/ATT; Equal letters in the same line do not differ by Tukey test at 5% significance level.

As there is no specific legislation for chutney, it is hard to state what should be the mean values for the total soluble solids that the product must reach at the end of the process, as there is, for example, for gels. For blackberry pulp, Hirsch (2012) found values between 7.3 to 10.2 °Brix, coinciding with this work.

Studies by Mota (2006b) have humidity values of blackberry pulp between 90.47% and 91.7%. Chim (2008) shows values between 87% to 88.3%, so the values found are within the expected. Mota (2006b) also finds values for total soluble solids of 7.6% to 10.37%, while studies in Greece showed values between 9.6% to 11.5% (Pantelidis et al., 2007) for different blackberry cultivars. Considering these figures, it can be said that the soluble solids match those found in the literature.

The maintenance of acidity of the fruit is important because it ensures taste and odor to the product (Cecchi, 2003), so blackberries evaluated in this study have shown good raw materials for the manufacture chutney with 1.44g of titratable acidity citric acid/100 g of pulp. Table 4 shows the results for determination of sugars, proteins, lipids and vitamin C.

Table 3. Physic-Chemical Analysis

Analysis	Pulp	60% chutney	80% chutney
Reducing Sugar	3.92±0.15^{ab}	4.64±0.37^{a}	2.14±0.01^{b}
Total Sugar	4.64±0.11^{a}	15.23±0.95^{b}	13.75±0.48^{b}
Protein	0.87±0.01^{a}	0.58±0.01^{b}	1.31±0.01^{c}
Lipids	0.48±0.005^{a}	0.36±0.001^{b}	1.85±0.02^{c}
Vitamin C	1.74±0.75^{a}	6.66±1.34^{b}	5.38±0.77^{ab}

NOTE: Reducing sugars, total sugars, proteins and lipids are expressed in percentage (%); Vitamin C is expressed in mg ascorbic acid/100 g of pulp; Equal letters in the same line do not differ by Tukey test at 5% significance level.

The sugar content is an important parameter used as reference for classification of pulps for the production of gel and therefore interferes significantly with the formulation of chutneys. Analyzing the averages for the sugars, there is a significant difference between the chutneys only for gear units. The pulp presents itself slightly lower than those found in the literature. Araújo (2009) shows values of 6.86% for reducing sugars and 7.03% for total sugars.

It is possible to observe a significant difference ($p \geq 0.05$) to the levels of lipids and proteins between the two chutney formulations, and 80% chutney presents higher amount of both components in its composition. In the analysis of pulp, Araújo (2009) cites a protein value of 0.51% and 0.45% lipids, which are slightly similar values to those found.

The quantification of the vitamin C is hampered by the instability of the compound, and typically is through quantification of reducing agents (Fontannaz et al., 2006). In chutney formulations, vitamin C was given the highest form, with no significant difference ($p \geq 0.05$) between samples.

The results of determination of the levels of anthocyanins, phenolics and antioxidant potential of blackberry are shown in Table 5.

Table 4. Chemical Analysis

Analysis	Pulp	60% chutney	80% chutney
Anthocyanins	144.71 ± 35^{a}	24.95 ± 2.88^{b}	22.95 ± 7.62^{b}
Phenolics compounds	1487.50 ± 366^{a}	1385.26 ± 47^{a}	1461.36 ± 109^{a}
Antioxidants	1693 ± 64^{a}	575 ± 13^{b}	752 ± 22^{b}

NOTE: Anthocyanins expressed in mg/100g. Phenolic compounds expressed as mg gallic acid equivalent (mgGAE/100 g). Antioxidant expressed in mg trolox/100 g; Equal letters in the same line do not differ by Tukey test at 5% significance level.

The Jacques study (2010) also presents an analysis of the content of individual phenolic compounds made by high-performance liquid chromatography (HPLC). In this, gallic acid was the predominant phenolic acid considered among the phenolic compounds with an amount of 1938.70 mg gallic acid/100 g fruit. According to these data, it was identified in this work significant amount of phenolic compounds for the blackberry pulp and to both chutneys formulations, the 80% chutney showed a higher amount than the 60% chutney, since it presents a higher percentage of fruit.

The content of anthocyanins found in blackberry was 144.71 mgEAG/100 g of fruit, very similar to that found by Chim (2008) of 137.59 mgEAG/100 g of fruit. Studies have shown that its contents can vary from 74.4 mgEAG/100 g of fruit in unripe fruit to 317 mgEAG/100 g of fruit in overripe fruits (Siriwoharn & Wrolstad, 2004). Besides the maturity, anthocyanins show variability depending on different cultivars and planting location.

Several factors influenced both chutneys formulations to have low of anthocyanins values, for example, prolonged exposure to light and oxygen during processing, heating the pulp and especially the use of frozen blackberry pulp. Anthocyanins are pigments responsible for the coloration of blackberry and therefore the addition of apple pulp and other ingredients also favored in this coloring reduction, lowering the content of anthocyanin compound 24.95 mgEAG/100 g in 60% chutney and 22.95 mgEAG/100 g for 80% chutney.

The tests carried out in this study showed that blackberry extract has high values for antioxidant capacity (1693 mg of trolox.100g^{-1} fruit). Silva (2006) points values between 989 and 1203 mg of trolox.100g^{-1} fruit, slightly lower than the values found. The antioxidant capacity of blackberry pulp are superior to the reported grape pulps (161.5 mg of trolox.100g^{-1} fruit), açai (163 mg trolox.100g^{-1} fruit), mango (224.7 mg of trolox.100g^{-1} fruit) and guava (120 mg of trolox.100g^{-1} fruit), and compatible reported to the acerola pulp (1198.9 mg trolox.100g^{-1} fruit) (Jacques, 2009), demonstrating the blackberry consists of a rich source of antioxidants, which is demonstrated by its high content of phenolic and anthocyanin compounds.

Chim (2008) also conducted a study on the characterization of bioactive compounds in blackberry (*Rubus* sp.) And its stability in the process and conventional jellies storage and light. In this study, the conventional jelly showed 884 mg of 1-trolox.100 g fruit storage for 2 months and 796 mg of 1-trolox.100 g fruit to 6 months. What proves that the freezing of the product and the pulp and processing derived directly affects the antioxidant capacity.

Table 6 shows the mean assigned to chutneys by the tasters for the taste attributes, color, flavor, texture and overall acceptability.

Table 5. Sensory Analysis Results

Parameters	60% chutney	80% chutney
Flavor	5.56±2.42[a]	5.90±2.29[a]
Color	7.30±1.60[a]	7.40±1.53[a]
Aroma	5.94±2.03[a]	6.15±1.93[a]
Texture	6.76±1.75[a]	6.81±1.85[a]
Overall acceptability	6.08±2.03[a]	6.31±1.96[a]

NOTE: Equal letters in the same line do not differ by Tukey test at 5% significance level.

By observing the mean values found for each sensory parameter and relating both formulations chutney blackberry, you can check that there was no significant difference in any of the evaluated attributes. It can be seen that 80% chutney obtained a larger amount of notes in the higher hedonic terms, while 60% chutney scales are located in lowest terms. This result is mainly due to the greater percentage of fruit added in the formulation of 80%, which pleased the tasters. Many tasters have reported that the sweet taste of chutney is intensified when the percentage of fruit added is lower.

CONCLUSIONS

The fruit of the blackberry tree has good potential for development of chutney. The process reduces the anthocyanin content and antioxidant capacity, but the product can be considered as a source of these compounds due to the high levels observed in the finished product. Sensory analysis showed the acceptability of 80% chutney by the tasters. Therefore, the blackberry chutney is feasible for consumers seeking healthy alternatives to insert in the diet, and for traders seeking new products to offer to the public.

Fabricação de chutney de amora-preta visando seu potencial antioxidante

RESUMO

O cultivo de amora-preta apresenta alto potencial na cadeia produtiva de pequenas frutas no Brasil, sendo uma fonte rica em compostos nutritivos e de antioxidantes naturais. O estudo objetivou quantificar compostos bioativos como compostos fenólicos, antocianinas, vitamina C, etc., determinar o potencial antioxidante e avaliar a estabilidade deste potencial durante a elaboração de duas formulações de chutney de amora-preta, uma com 60% de polpa e a outra com 80%. O estudo também apresenta o desenvolvimento da análise sensorial para a avaliação da aceitação e intenção de compra pelos provadores. Durante o processo de elaboração dos chutneys ocorreu uma degradação parcial dos os compostos avaliados, no entanto, esta degradação foi menos intensa na elaboração do chutney de 80%, que foi o resultado esperado. A análise sensorial apresentou boa aceitabilidade para o chutney de 80%, porém, o chutney de 60% não atingiu a média necessária para aceitação de 70%. Por ser um produto inovador que não possui vasta comercialização, pode-se afirmar que o chutney de amora-preta é uma alternativa viável para os consumidores, que buscam cada vez mais produtos saudáveis, e para a indústria alimentícia que está sempre procurando novidades para oferecer ao público.

PALAVRAS-CHAVE: compostos bioativos; processamento de frutas; análise sensorial.

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