



STUDIES WITH
BLACKBERRY
AND
BLUEBERRY
IN FOOD PROCESSING

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Studies with blackberry and blueberry in food processing

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Elaboration



Support



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Acknowledgements

The book entitled **Studies with blackberry and blueberry in food processing** is the result of the action of our team of researchers aligned with the guidelines carried out during the Food Systems Summit, held in September 2021 by the UN and an FAO.

One of the common goals of the Sustainable Development Goals (SDGs) is the reduction of chronic-degenerative diseases (NCDs) and risk factors by 2030. Chronic degenerative diseases are among the main causes of disability and death in several countries, among them we highlight diabetes, hypertension, obesity, stroke, heart disease and some cancers.

The COVID-19 pandemic has compromised all the SDGs, including the reduction of CNCDs that previously represented a challenging commitment and which together with the new coronavirus pandemic and its variants may not be achieved.

Therefore, we established as an objective the development of this work, whose objective is to identify fruits with a high content of bioactive compounds and use them in different formulations and development of new products using emerging processing techniques and efficient mathematical models. For this, we are grateful for the great contribution of Dra. Vania Moda Cirino and researcher Dr. Renato Ferraz de Arruda Veiga.

Blackberries and blueberries are strategic food matrices, they are fruits with a high content of anthocyanins that act mainly in the prevention of diseases associated with oxidative stress, such as cardiovascular diseases, diabetes and neurodegenerative diseases.

As anthocyanins they represent the main group of flavonoids in these fruits and have been associated with several beneficial effects. Blueberries are known to be rich in bioactive compounds including flavonoids, phenolic acids, tannins and anthocyanins.

Blackberries, in addition to their high vitamin C content, set about 85% water, about 10% carbohydrates, rich in minerals, vitamins B and A., about 82% water, 0.4 to 0.7% protein, 0.5% fat and 0.19 to 0.25% ash content.

In relation to the extract, several studies have shown that the antioxidant in the blackberry and blueberry extract is superior in activity to other fruits. The researches carried out in each chapter of this work show the versatility of these fruits and their industrial potential in the development of new formulated products.

Thus, in each chapter we seek to determine the thermophysical properties of the mixture of the pulp of these fruits, development of new products, an example of whole yogurt, drinks based on these two pulps, among other formulations.

We hope that the work will be an effective instrument, contributing more and more to the innovative potential of strategic products for the world and increasingly showing the effort of a team with collective objectives.

The authors

Presentation

Dr. Renato Ferraz de Arruda Veiga

Administrative Director of the Scientific Research Support Foundation – FUNDAG

The United Nations Summit on Food Systems together with the International Conference on Nutrition (ICN2), in the Decade of Action on Nutrition, re-established the links between nutrition, health and healthy and sustainable food systems. Furthermore, the Food and Agriculture Organization of the United Nations (UN/FAO) affirms that food security is directly related to the improvement and investment in the modernization of nutritional agriculture and nutrition worldwide.

Sustainable agriculture and improved nutrition represent one of the main common goals through 2030 in line with several sustainable development goals (SDGs) determined by the UN. Thus, the modernization of agriculture is essential to ensure the sustainability of nutritious food production. Thus, regenerative agriculture is based on increasing crop productivity, without the need to expand agricultural areas, and the use of digital technologies will allow to optimize production processes, in addition to preserving natural resources, in food production and practices of management in soil conservation.

Brazil has the potential to be a pioneer in the transformation of sustainable agriculture, where digital technologies will allow greater interaction of production chains in food production, with quality and with less use of natural resources. Thus, food production will allow the development of new food products that, in addition to nourishing, will enable the reduction of the risk of chronic degenerative diseases. Therefore, the choice of food matrices is fundamental for product development by combining the correct nutrients in their chemical formulations.

Fruits represent the greatest natural reserves of vitamins and antioxidants, due to their richness in bioactive compounds responsible for flavor, color, odor and oxidative stability. The antioxidant activities present in these compounds have anticancer, antiinflammatory, antiallergic, antimicrobial properties, among others, promoting beneficial physiological effects, preventing or reducing the risk of numerous diseases.

Studies on fruits and their antioxidant activities have intensified around the world. In this sense, blackberries and blueberries were chosen for this work due to their relevant

chemical characteristics, high content of bioactive compounds and vitamin C, representing strategic food matrices for the new product developments.

Fruits are perishable foods due to their high amount of water, so to inhibit degradation processes it is necessary to provide some treatment conditions for conservation and handling. The choice of emerging processing techniques is efficient because they preserve the retention of thermosensitive compounds and promote enzymatic inactivation, being essential for the development of beverages, jellies, yogurts, purees, sauces, juices, dried fruits, preserving nutrients, texture, color and flavors, among other relevant possibilities.

Blackberries are rich sources of phenolic compounds, including ellagic acid, tannins, ellagitannins, quercetin, gallicidanic acid. Blueberries are known to be rich in bioactive compounds including flavonoids, phenolic acids, tannins and anthocyanins. These compounds vary according to their species, but catechins, caffeic, chlorogenic, p-coumaric and ferulic acid stand out, among many others (OKAN et al., 2018).

The blackberry tree represents a great option for diversification into small properties, as it is rustic, highly productive and presents fruits with a good balance between sugar and acidity, favorable to the production of gelled products (YAMAMOTO et al., 2013).

Blueberry (*Vaccinium spp.*) is a fruit species native to North America and Europe, where it is highly appreciated for its flavor and functional properties. The worldwide interest in cultivation is due to the high profitability and high antioxidant capacity, attributed to the great diversity of polyphenols present in fruits, mainly anthocyanins, flavonoids and cinnamic acid derivatives (CARDEÑOSA et al., 2016; LI et al., 2016).

The commercial production of blueberries occurs mainly in North America (USA and Canada), Europe (Poland, Germany) and countries in the Southern Hemisphere (Chile, Argentina, Uruguay, Australia) (OLIVEIRA et al., 2020).

In Brazil, it was introduced in the 1980s, being cultivated in approx. In Brazil, it was determined in the 1980s, being cultivated in approximately 400 ha; mainly in the states of Rio Grande do Sul, where he was chosen for the first time, and Santa Catarina, with a smaller area in São Paulo, Minas Gerais and Paraná. (OLIVEIRA et al., 2020).

Chapter 1 deals with the development of beverages with blackberry and blueberry pulp, applying the microwave-assisted pasteurization process and evaluating the influence of this processing on the total phenolic compounds, total anthocyanins and the antioxidant activity of the beverages. Microwave treatments stand out as emerging dielectric heating technologies due to rapid (relatively homogeneous) temperature rise, reduced time, zero

emission of exhaust gases or toxic waste to the environment, and high energy efficiency. Furthermore, they promote the retention of bioactive compounds and nutrients in addition to other characteristics such as flavor, color, etc.

Chapter 2 aims to regulate the concentration of maltodextrin concentration in the properties of the blackberry and blueberry mixture using the freeze-drying process. Freeze drying is considered one of the best drying methods, as it allows the maintenance of the organoleptic and nutritional properties of the food.

Chapter 3 evaluates the rheological behavior and texture profile of a blend composed of blackberry and blueberry pulp in different types of goat milk (*Capra aegagrus hircus*). Such behavior is quite complex and plays a key role in food development, processing and manufacturing.

In this chapter, I also highlight the use of milk, which according to Anvisa (1999) can be classified as a functional food, among other qualities, as it presents properties such as: (a) high levels of certain fatty acids that are beneficial to the body; (b) superior quality to cow's milk (*Bos taurus*) in terms of nutritional and therapeutic properties; and (c) better digestibility, alkalinity, high nutritional value protein content, hypoallergenicity.

Chapter 4 aims to determine the effective diffusivity, activation energy and thermodynamic properties of blackberry and blueberry pulp drying through the foam layer drying process. Knowledge of thermodynamic properties in drying is an important source of information for designing equipment, calculating the energy needed in the process, studying the properties of adsorbed water and evaluating the microstructure of foods, as well as for the study of necessary resources that occur on its surface (SANTOS et al., 2019).

On the other hand, foam layer drying consists of transforming liquid or pasty foods into stable foams that are easily rehydrated in the final process. Thus, this drying process is efficient for fruits that are food matrices related to heat, as it maintains the quality and nutritional value, in addition to sensory characteristics.

Chapter 5 aims at the development of a whole yogurt with the addition of blackberry and blueberry pulp and the evaluation of its physical-chemical, microbiological and texture characteristics. It excels in quality and safety in the development of nutritious foods with competent properties.

Chapter 6 elaborates and caveats a mixed structure containing blackberry and blueberry pulp. Emphasizes the importance of structured fruit collection of fruit puree, properly formulated to obtain nutritious products with good texture and flavor.

Finally, it is observed that this work represents the continuous effort of a relevant research team whose objective is the dissemination of strategic researches using relevant raw materials, publicly and free of charge, for several scientific areas, including agricultural engineering, especially for the healthy diet of the regional and international population.

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Chapter I

PASTEURIZATION ASSISTED BY MICROWAVE BEVERAGES PRODUCED WITH BLACKBERRY (*Morus alba*) AND BLUEBERRY (*Vaccinium sect. Cyanococcus*) PULP AND ITS EFFECT ON BIOACTIVE AND ANTIOXIDANT COMPOUNDS

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Introduction

Ready-to-beverage fruit beverages have a number of advantages, such as the combination of different aromas, flavors and nutritional benefits. The combination of fruits with antioxidant capacity can promote a synergistic effect, thus increasing the effectiveness and bioactive power. Furthermore, from an economic point of view, it guarantees the supply and sale of these fruits throughout the year (SANTOS et al., 2018). The consumption of fruit-based beverages is a common habit in several countries and, even if the intake is different compared to a serving of fruits or vegetables, the juice contains the same nutrients in different proportions (SILVA et al., 2020).

Red fruits such as grapes, cherries and blueberries are rich in anthocyanins, polyphenols with strong antioxidant activity (FREDES et al., 2020). Blueberries and blackberries are fruits with a high content of anthocyanins, they act mainly in the prevention of diseases associated with oxidative stress, such as cardiovascular disease, diabetes, and neurodegenerative diseases. These tasty wild fruits have attracted a lot of attention and exceptional interest from scientists, nutritionists, and food manufacturers and, of course, consumers, due to their scientifically reported high antioxidant capacity, resulting from their wide range of polyphenolic compounds. (RASHIDINEJAD, 2020)

However, according to González-Monroy et al. (2018), beverages based on natural fruits may present instabilities such as phase separation problems, loss of turbidity and discoloration due to the presence of enzymes. Thus, the properties of these beverages can change during their processing, therefore, finding alternative processing techniques has become a research trend (WANG et al., 2020).

Different thermal treatments have been proposed for enzymatic inactivation. Traditionally, relatively low temperatures were used to prevent unwanted changes in food products, and this required the use of a relatively long contact time to reach the desired level of inactivation. Modern industrial pasteurization processes that demand high productivity from the treatment plant, however, involve the use of high temperatures and short contact time conditions (SANTOS et al., 2020).

Microwave treatments stand out among emerging dielectric heating technologies due to rapid (relatively homogeneous) temperature increase, reduced time, zero emission of exhaust gases or toxic waste to the environment, and high energy efficiency. Short treatment times promote the retention of thermosensitive compounds and organoleptic properties such as flavor and color. sensory and microbiological (GONZÁLEZ-MONROY et al., 2018).

Thus, the present work aimed to prepare beverages with blackberry and blueberry pulp, apply the microwave-assisted pasteurization process and, finally, evaluate the influence of this thermal processing on total phenolic compounds, total anthocyanins and on the antioxidant activity of beverages.

Methodology

Raw material

The blackberries (*Morus alba*) and blueberries (*Vaccinium seita. Cyanococcus*) Figure 1 and 2, used were from the Nossa Fruta® brand, acquired in the local market in the city of Natal, Rio Grande do Norte, Brazil.



Figure 1. The blackberries (*Morus alba*) and used in the samples.



Figure 2. Blueberries (*Vaccinium seita. Cyanococcus*) and used in the samples.

Obtaining the pulp

To obtain the pulp, the fruits were processed separately with the aid of a domestic fruit processor, separating the pulp and waste fractions. The waste was discarded.

Preparation of beverages

Three beverage formulations (A1, A2 and A3) were prepared with blackberry and blueberry pulps as shown in Table 1.

Table 1. Formulations and components of elaborated beverages

Formulations	Beverage components
TO 1	Blackberry pulp
A2	Blueberry pulp
A3	Blackberry and blueberry pulp

The beverages were placed in glass containers with a capacity of 250 mL at a temperature of 25°C. After its elaboration and storage, the bottles were submitted to pasteurization.

Microwave-assisted pasteurization process

Formulations A1, A2 and A3 were submitted to microwave-assisted pasteurization process. The process scheme is represented in Figure 1.

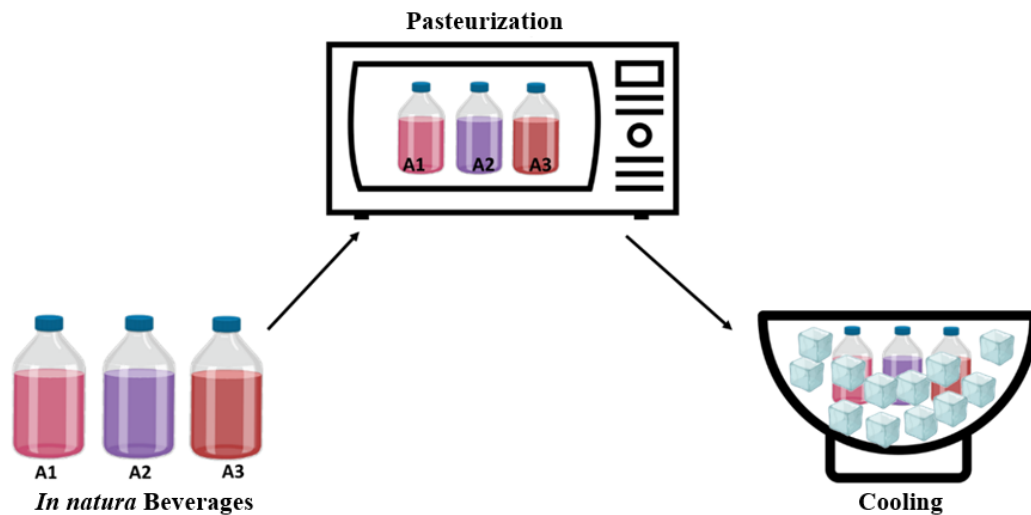


Figure 1. General scheme for microwave-assisted pasteurization of beverages made with blackberry and blueberry pulp.

The microwave-assisted pasteurization process was carried out with open glass bottles in which they were placed in a microwave oven (Consul®) at maximum power up to a temperature of 90°C for 12 s. Immediately, the bottles were removed, closed and cooled in an ice bath (4°C) to room temperature. Rapid cooling produced a hermetic seal on the bottles, this pasteurization process was carried out according to the method proposed by González-Monroy et al. (2018).

Analysis of bioactive compounds

The formulations before and after the pasteurization process were analyzed in triplicate for the following parameters described below:

Total phenolic compounds

Total phenolic compounds were quantified by the Folin-Ciocalteu method described by Waterhouse (2006), using gallic acid as a standard. Water and methanol

were used as extraction solvents. The calculations performed for the determination of phenolic compounds were based on a standard curve with gallic acid, and readings were made in a spectrophotometer at 765 nm, with results expressed in mg/100 g of gallic acid.

Total anthocyanins

The method used to read total anthocyanins was the single pH method described by Francis (1982). The method consists of carrying out a quantitative transfer of an aliquot of the concentrated extract to a recipient and then this aliquot is diluted with an amount of Ethanol – HCl solution at 1.5 mol.L⁻¹, thus having a volume of diluted extract. The number of total anthocyanins was calculated by Equation (1).

$$Ant_{mg} = \frac{Abs_{535} \times V_{ec} \times V_{ed} \times 1000}{V_{alq} \times m \times 982} \quad (\text{Eq.1})$$

Where: Ant_{mg} is the amount of total anthocyanins expressed in mg of anthocyanins per 100 grams of sample (mg/100); Abs_{535} is the absorbance read from the diluted extract at 535 nm; V_{ec} is the volume of the concentrated extract (mL); V_{ed} is the diluted extract volume (mL); V_{alq} is the volume of the aliquot taken from the concentrated extract to make the diluted extract (mL); m is the mass of the beverage used to carry out the extraction; The value of 982 is the extinction coefficient for anthocyanins.

Determination of antioxidant activity

The antioxidant activity of DPPH• was performed according to the methodology described by Maria do Socorro et al. (2010) with adaptations. Antioxidant activity (ABTS) was determined by the method proposed by Re et al. (1999), with modifications made by Rufino et al. (2007). For both analyses, distilled water and methanol were used as extractive solvent.

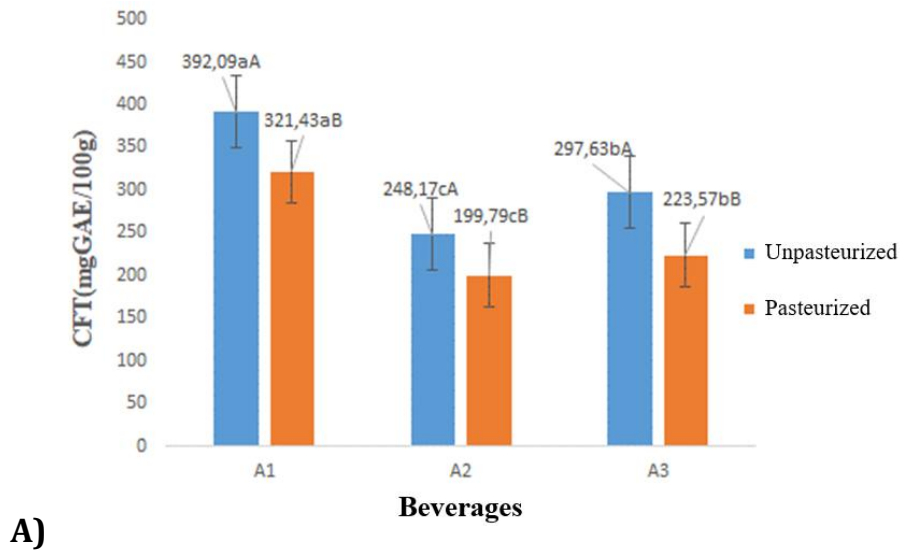
Statistical analysis

The results were subjected to statistical treatment using a completely randomized design with a comparison test of means, using the Assisat software version 7.7 beta (SILVA & AZEVEDO, 2009).

Results

Figure 2 shows the values obtained for the analysis of total phenolic compounds in beverages prepared before and after the pasteurization process. According to Rocha et al. (2011) phenolic compounds are generally associated with the adaptation mechanism and plant resistance to the environment. In Figure 2A, it is possible to observe the mean values of phenolic compounds for the aqueous extract (water), where unpasteurized beverages presented higher values than pasteurized ones, with the highest concentration obtained for formulation A1 with 392.09 mgGAE/100g. When comparing the contents obtained between unpasteurized and pasteurized formulations, statistically significant differences are observed at the 5% probability level. Pasteurized A2 formulation had a lower content of total phenolic compounds (199.79 mgGAE/100g).

In Figure 2B, it is possible to observe the mean values of phenolic compounds for the extract using methanol. Statistically, when comparing the beverages prepared before and after the pasteurization process, there were significant differences in the Tukey test applied, with the highest values obtained in the A1 formulation (631.45 mgGAE/100g) unpasteurized and (598.19 mgGAE/100g) pasteurized. However, when observing the contents of phenolic compounds by the two solvents of different polarities, it is clear that for all formulations the extract using methanol, managed to extract higher contents of these compounds.



B)

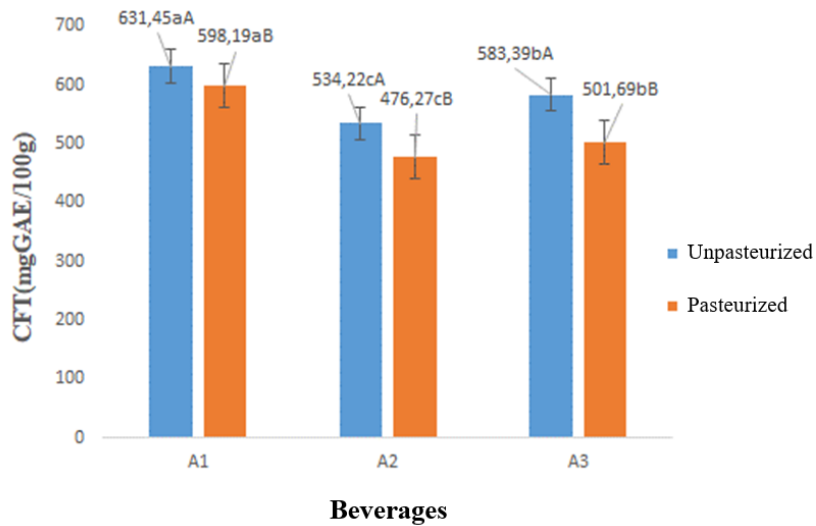


Figure 2. Total phenolic compounds of beverages prepared before and after the microwave pasteurization process, using two different solvents: A) water and B) Methanol.

Figure 3 shows the values obtained for the total anthocyanin content of beverages prepared before and after the pasteurization process.

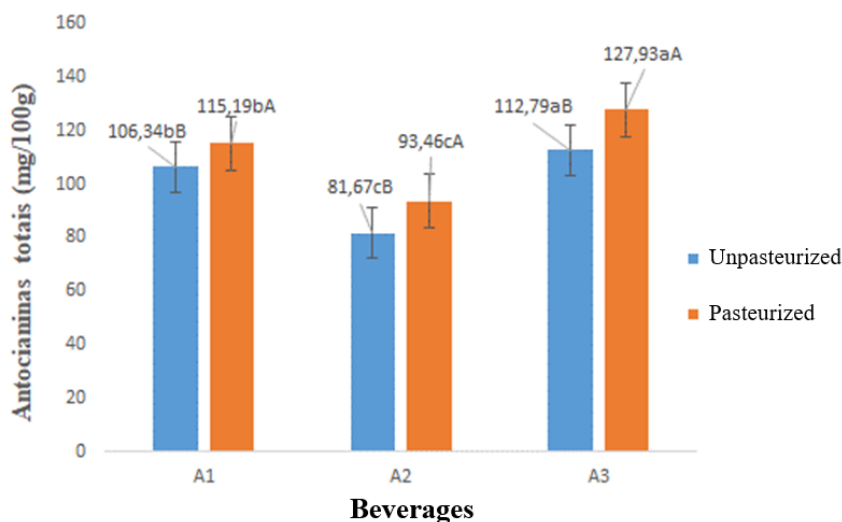


Figure 3. Total anthocyanin content of beverages prepared before and after the microwave pasteurization process.

Higher levels of anthocyanins were observed for A3 beverage, which contains blackberry and blueberry pulp in its formulation, being 112.79 mg/100g for unpasteurized beverage and 127.93 mg/100g for pasteurized beverage. It is also possible to observe that the thermal process of pasteurization promoted an increase in the concentration of anthocyanins. This increase may be related to the cleavage of covalent bonds of some bioactive compounds and the release of these molecules in the medium due to the heating of beverages.

Statistically, pasteurized and unpasteurized formulations, when compared to each other, showed significant differences at the 5% probability level. According to Freire et al. (2020) anthocyanins are pigments responsible for most of the colors in vegetables, with shades between red and blue. These compounds have shown industrial interest not only for their coloring potential, but also for their antioxidant properties.

Figure 4 shows the values obtained for antioxidant activity by the DPPH method of beverages prepared before and after the pasteruization process.

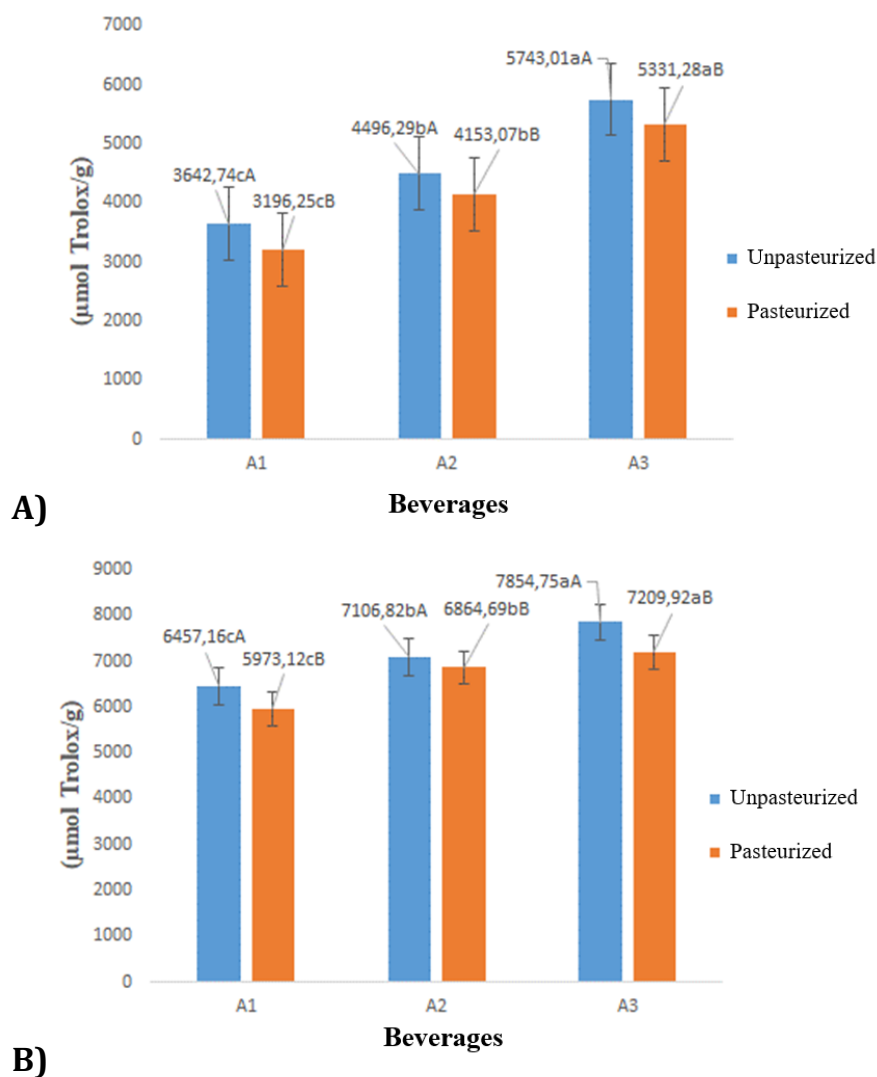


Figure 4. Antioxidant activity by the DPPH method of beverages prepared before and after the microwave pasteurization process, using two different solvents: A) water and B) Methanol.

In Figure 4A, it is possible to observe the mean values of antioxidant activity by the DPPH method for the aqueous extract (water). High values of antioxidant activity were obtained for unpasteurized beverages, with 3642.74 $\mu\text{mol Trolox/g}$ (A1), 4496.29 $\mu\text{mol Trolox/g}$ (A2) and 5743.01 $\mu\text{mol Trolox/g}$ (A3), the process of pasteurization reduced its values to 3196.25 $\mu\text{mol Trolox/g}$ (A1), 4153.07 $\mu\text{mol Trolox/g}$ (A2) and 5331.28 $\mu\text{mol Trolox/g}$ (A3), thus being the A3 formulation with the highest antioxidant capacity. From a statistical point of view, pasteurized and unpasteurized beverages presented significant differences between them. In Figure 4B, it can be seen the mean values of antioxidant activity by the DPPH method for the

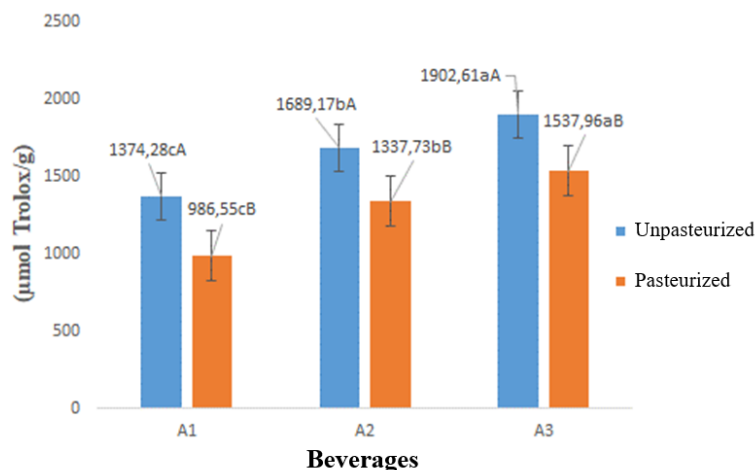
extract using methanol, the values obtained were higher than those obtained for the aqueous extract, due to its polarity. The thermal process of pasteurization promoted a reduction in the antioxidant activity for, this degradation being statistically significant ($p > 0.05$).

According to Magalhães et al. (2008), the antioxidant action of a compound is directly related to the bioactive components present and depends on the chemical structure and concentrations of these phytochemicals in the food. Figure 5 shows the values obtained for antioxidant activity by the ABTS method of beverages prepared before and after the pasteurization process.

The antioxidant activity (aqueous extract) of unpasteurized beverages (Figure 5A) due to the ABTS radical capture capacity, obtained values lower than those obtained by the DPPH method (Figure 4A), in which the values ranged from 1374.28 to 1902.61 $\mu\text{mol Trolox/g}$. The thermal process promoted a statistically significant reduction, in which the values ranged from 986.55 to 1537.96 $\mu\text{mol Trolox/g}$. Lameiro et al. (2020) in their studies with blackberry and blueberry pulp diluted in 50% of mineral water, obtained antioxidant activity by the ABTS method of 896.07 and 1056.74 $\mu\text{mol Trolox/g}$.

Regarding the antioxidant activity using the methanol extract (Figure 5B), the values were higher when compared to water for the same ABTS radical scavenging capacity. Values differed significantly ($p > 0.05$) between pasteurized (1267.18 to 1824.06 $\mu\text{mol Trolox/g}$) and unpasteurized (1562.54 to 2310.19 $\mu\text{mol Trolox/g}$) beverages. This same reduction behavior after the application of heat treatment was also previously reported by Santos et al. (2020) in studies of pasteurization with Mandacaru pulp and peel and by Almeida et al. (2020a) in studies of pasteurization with juju fruit pulp.

A)



B)

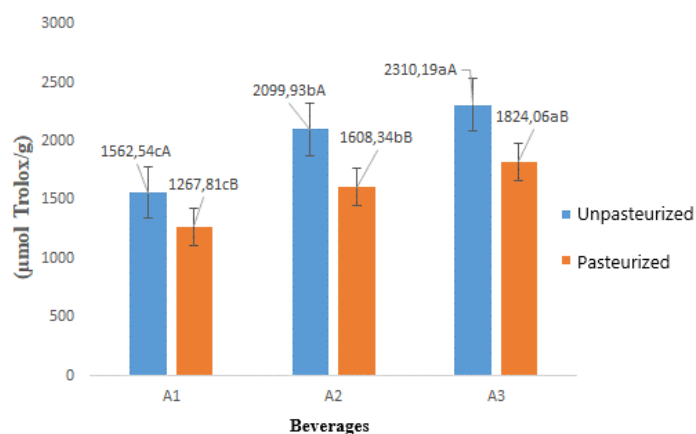


Figure 5. Antioxidant activity by the ABTS method of beverages prepared before and after the microwave pasteurization process, using two different solvents: A) water and B) Methanol.

According to Almeida et al. (2020b), the solubility in a certain solvent is a peculiar characteristic of the phytochemical, which justifies the inexistence of a universal extraction procedure due to the structural diversity and sensitivity of the phenolic compounds to the extraction conditions.

Conclusion

Through the results obtained, it can be concluded that:

Beverages made with blackberry pulp and blueberries can be pasteurized in microwaves with a relatively short time of 12s;

The total phenolic compounds and the antioxidant activity of the beverages were degraded as a result of the thermal process;

There was a concentration of total anthocyanins due to the pasteurization process for all analyzed beverage formulations;

Methanol as extraction solvent showed higher extraction values;

Higher antioxidant activity values were obtained by the DPPH method.

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Chapter II

PHYSICAL PROPERTIES OF THE BLEND OF BLACKBERRY (*Morus alba*) AND LYOPHILLED BLUEBERRY (*Vaccinium sect. Cyanococcus*) IN DIFFERENT FORMULATIONS

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Introduction

Blackberries are rich sources of phenolic compounds, including ellagic acid, tannins, ellagitannins, quercetin, gallic acid, anthocyanins and cyanidins, which have the potential to reduce the risk of chronic diseases such as cardiovascular disease, diabetes and certain cancers. In addition to fresh consumption around the world, blackberries can also be processed into a variety of food products, including puree, sauce, juice, jelly, canning and dried blackberry (TOMAS et al., 2019).

Blueberries belong to the genus *Vaccinium*, a widespread genus with more than 200 species of perennial and deciduous woody plants that range in size from dwarf shrubs to trees. Recently, blueberries have received a lot of attention due to their very positive effect on the human body. Blueberry fruits are characterized by their biological activities. They are important sources of polyphenolic compounds and flavonoids, mainly anthocyanins, and have high antioxidant activity (ZHANG et al., 2020).

Fruits are perishable foods, therefore, to inhibit the degradation processes, it is necessary to obtain some treatment conditions for conservation and manipulation. To minimize this degradation of biological products or food, several preservation techniques are used, one of them is the reduction of water content through the freeze-drying process (IBIPIANA et al., 2018; FONGIN et al., 2019).

Freeze drying is considered to be one of the best drying methods, as it allows the maintenance of the organoleptic and nutritional properties of foods. The method consists of freezing the product followed by dehydration, which occurs through the sublimation process, providing a reduction in the water content and consequently minimizing the occurrence of most reactions that cause product degradation (ALMEIDA et al., 2020).

The addition of substances known as carrier agents, encapsulants or drying aids influence the properties and stability of fruit powders. The physical properties of powders can influence handling and storage conditions and are important parameters that determine how the processor or consumer will handle the powdered product (ALVES et al., 2020). Maltodextrin is the carrier agent commonly used in spray drying,

due to its low hygroscopicity, high solubility in cold water and low cost (FERRARI et al., 2012).

In this context, the present work aimed to evaluate the influence of maltodextrin concentration on the physical properties of blackberry and freeze-dried blueberry blend.

Methodology

Raw material

The blackberries (*Morus alba*) and blueberries (*Vaccinium sect. Cyanococcus*) figure 1, used were from the Nossa Fruta® brand and maltodextrin from the Integral Médico® brand, both purchased in the local market of the city of Natal, Rio Grande do Norte, Brazil.



Figure 1. The blackberries (*Morus alba*) and blueberries (*Vaccinium sect. Cyanococcus*) used in the samples.

Preparation of formulations

Five formulations were prepared with blackberry, blueberry and maltodextrin pulps as shown in Table 1.

Table 1. Blackberry and blueberry blend formulations

Formulations	Maltodextrin (%)
A1	0
A2	10
A3	15
A4	20
A5	25

Lyophilization of formulations

The elaborated formulations were frozen in a freezer at -18 °C for 48 h and taken to the bench lyophilizer for a period of 72 h. After lyophilization, the formulations were placed in laminated packages until the moment of analysis. In Figure 2, a summarized flowchart of the main steps for obtaining lyophilized powders can be seen.



Figure 2. General flowchart for obtaining lyophilized powders.

Physical properties

In triplicate, the following physical properties of the prepared lyophilized formulations were determined:

Water content

The water content was determined by drying in an oven at 105°C until constant weight (BRASIL, 2008).

Water Activity

Water activity (aw) was determined using the Decagon® Aqualab CX-2T device at 25°C.

Bulk density

Bulk density was determined with the aid of a 10 mL beaker previously weighed and then filled with flour, and determined through Equation 1 (TONON et al., 2009).

$$\rho_{ap} = \frac{m}{V} \quad (\text{Eq.1})$$

Where: ρ_{ap} is the apparent density (g cm⁻³), m is the mass and V is the occupied volume.

Compacted density

The compacted density was determined from the assembly used in the apparent density, subjecting the beating of the beaker filled with the sample to 50 times on the

bench, from a pre-established height of 2.5 cm, calculating the ratio through of Equation 2 (TONON et al., 2009).

$$\rho_c = \frac{m}{V_c} \quad (\text{Eq.2})$$

Where: ρ_c is the compacted density (g cm^{-3}), m is the mass and V_c is the volume occupied after compaction.

Hausner Factor

The Hausner Factor (FH), which assesses the cohesiveness of the flour, was determined based on the methodology of Wells (1988), and calculated according to Equation 3.

$$FH = \frac{\rho_c}{\rho_{ap}} \quad (\text{Eq.3})$$

Where: ρ_c is the tapped density (g cm^{-3}) and ρ_{ap} is the apparent density (g cm^{-3}).

Carr Index

The Carr index (CI), which comprises the fluidity index, was determined using the Wells (1988) methodology, and calculated according to Equation 4.

$$IC = \frac{\rho_c - \rho_{ap}}{\rho_{ap}} \times 100\% \quad (\text{Eq.4})$$

Where: ρ_c is the tapped density (g cm^{-3}) and ρ_{ap} is the apparent density (g cm^{-3}).

Water absorption capacity

The water absorption capacity was determined according to Beuchat (1977), in which about 10 ml of water (distilled) were added to 1 g of the sample in centrifuge tubes. The suspension was homogenized for 30 seconds and then left to rest for 30 minutes. Afterwards, the tubes were closed and centrifuged for 15 minutes at 1000 rpm. The outer walls of the tubes were dried and then weighed. The mass of water absorbed was expressed as a percentage (%).

Hygroscopicity

Hygroscopicity was measured according to the method described by Jaya et al. (2006). Briefly, 5 g of freeze-dried powder was placed in a pre-weighed aluminum dish. Then the dish was placed in a desiccator under 81% RH and 25°C temperature for 7 days.

Statistical analysis

The results were subjected to statistical treatment using a completely randomized design with a comparison test of means, using the Assistat software version 7.7 beta (SILVA & AZEVEDO, 2009).

Results

In Table 2, it is possible to observe the mean values of the water content obtained for the blackberry and blueberry blend formulations lyophilized with different concentrations of maltodextrin.

Table 2. Water content of lyophilized blackberry and blueberry powders with different concentrations of maltodextrin

Maltodextrin concentration	Water content (%)
0	17.12 ± 0.12a
10	12.05 ± 0.10b
15	9.71 ± 0.08c
20	7.78 ± 0.21d
25	6.42 ± 0.06e

Note: Equal letters in the same column do not differ significantly from each other by Tukey's test at the 5% level.

The water content of the freeze-dried blend was reduced when the concentration of maltodextrin was increased by up to 25%. The lowest content (6.42%) was obtained for the blend with 25% maltodextrin. Statistically, the values when compared to each other showed significant differences at the 5% probability level.

Andrade et al. (2021) when determining the water content of lyophilized araçá-boi pulps with different concentrations of maltodextrin (14, 21 and 28%) obtained values of 7.63, 7.88 and 7.53%, respectively. Silva et al. (2016) obtained a water content of 27.01% for a freeze-dried pineapple and acerola blend.

In Table 3, it is possible to observe the mean values of water activity obtained for the blackberry and blueberry blend formulations lyophilized with different concentrations of maltodextrin. According to Staudt et al. (2013), water activity is one of the most important parameters in the study of food, as it is related to the amount of water available for physical, chemical, biochemical reactions and microbiological growth. It is a factor that directly influences the characteristics of foods and their stability, indicating whether there is a possibility of microbial growth.

Table 3. Water activity of freeze-dried blackberry and blueberry powders with different concentrations of maltodextrin

Maltodextrin concentration	Water activity
0	0.291 ± 0.00a
10	0.262 ± 0.01a
15	0.246 ± 0.01a
20	0.230 ± 0.02a
25	0.228 ± 0.01a

Note: Equal letters in the same column do not differ significantly from each other by Tukey's test at the 5% level.

Regarding water activity, the values did not show statistically significant differences ($p>0.05$) and ranged from 0.291 to 0.228, showing a reduction when there was an increase in maltodextrin. Araújo et al. (2021) in their studies with freeze-dried jambo pulp, obtained water activity of 0.259. Seerangurayar et al. (2017) when using gum arabic at different concentrations as a carrier in date powders, obtained water activity values ranging between 0.14 and 0.34. In Table 4, it is possible to observe the mean values of the bulk density for the blackberry and blueberry blend formulations freeze-dried with different concentrations of maltodextrin.

Table 4. Bulk density of lyophilized blackberry and blueberry powders with different concentrations of maltodextrin

Maltodextrin concentration	Bulk density (g cm⁻³)
0	0.451 ± 0.01a
10	0.402 ± 0.02b
15	0.371 ± 0.04c
20	0.336 ± 0.01d
25	0.308 ± 0.03e

Note: Equal letters in the same column do not differ significantly from each other by Tukey's test at the 5% level.

Bulk density values were significantly influenced ($p>0.05$) with increased maltodextrin concentration. The highest value being 0.451 g cm⁻³ for the blend with 0% maltodextrin and 0.336 g cm⁻³ for the blend with 25% maltodextrin.

Martins et al. (2019) when preparing freeze-dried blends with banana pulp and peel, they obtained bulk density ranging from 0.56 to 0.58 g cm⁻³.

According to Cavalcante et al. (2017) the low bulk density values are associated with the low water content of the obtained powder. Table 5 shows the mean values of tap density for the blackberry and blueberry blend formulations freeze-dried with different concentrations of maltodextrin.

Table 5. Compacted density of freeze-dried blackberry and blueberry powders with different concentrations of maltodextrin

Maltodextrin concentration	Compacted density (g cm⁻³)
0	0.562 ± 0.00a
10	0.539 ± 0.01b
15	0.508 ± 0.04c
20	0.465 ± 0.01d
25	0.427 ± 0.02e

Note: Equal letters in the same column do not differ significantly from each other by Tukey's test at the 5% level.

The compacted density presented values higher than those obtained for bulk density, due to the smaller volume occupied by the powder. However, it followed the same behavior of the apparent density, that is, it reduced its values as the maltodextrin concentration increased. Values ranged from 0.562 to 0.427 gcm⁻³ and were statistically different when compared to each other ($p>0.05$).

Martins et al. (2019) when preparing freeze-dried blends with banana pulp and peel, they obtained compacted density ranging from 0.77 to 0.80 g cm⁻³. Table 6 shows the mean values of the Carr index obtained for the blackberry and blueberry blend formulations lyophilized with different concentrations of maltodextrin.

Table 6. Carr index of lyophilized blackberry and blueberry powders with different concentrations of maltodextrin

Maltodextrin concentration	Carr Index (%)
0	24.61 ± 0.09d
10	34.08 ± 0.11c
15	36.93 ± 0.21b
20	38.39 ± 0.01a
25	38.64 ± 0.08a

Note: Equal letters in the same column do not differ significantly from each other by Tukey's test at the 5% level.

The values of the Carr index showed a tendency to increase when there was an increase in the concentration of maltodextrin. The values obtained were between 24.61 and 38.64%, the formulations with 20 and 25% of maltodextrin did not differ significantly ($p > 0.05$). According to Aziz et al. (2018) and Alves et al. (2020), Carr index (CI) greater than 26% indicate high compressibility of the particles, being little fluid when packaged and stored. Zea et al. (2013), studying freeze-dried powders of pitaya, guava and the mixture of pitaya and guava powder, both with 10% maltodextrin, observed a Carr index of 34.87, 27.19 and 37, 29%, respectively.

Table 7 shows the average Hausner factor values obtained for the blackberry and blueberry blend formulations lyophilized with different concentrations of maltodextrin. According to Santos et al. (2021) the Hausner factor assesses the cohesion of the powders.

Table 7. Hausner factor of blackberry and blueberry powders freeze-dried with different concentrations of maltodextrin

Maltodextrin concentration	Hausner factor (FH)
0	1.25 ± 0.00d
10	1.34 ± 0.00b
15	1.37 ± 0.00a
20	1.38 ± 0.01a
25	1.29 ± 0.00c

Note: Equal letters in the same column do not differ significantly from each other by Tukey's test at the 5% level.

The Hausner Factor (FH) values of the blends with 15 and 20% maltodextrin did not present statistically significant differences at the 5% probability level. According to Santhalakshmy et al. (2015), powders with FH of less than 1.2 are classified as having low cohesion, FH between 1.2 and 1.4 of intermediate cohesiveness and $FH > 1.4$, high cohesion. Thus, the freeze-dried blends in their different maltodextrin concentrations were classified as having intermediate cohesion. Alves et al. (2020) when preparing freeze-dried red pitaya pulp with different carrier agents (maltodextrin, gum arabic, dextrin) obtained Hausner factor values ranging from 1.29 to 1.75.

In Table 8, it is possible to observe the mean values of the water absorption capacity obtained for the blackberry and blueberry blend formulations lyophilized with different concentrations of maltodextrin. According to Asokapandian et al. (2016) the powder reconstitution properties in an aqueous phase are one of the best quality indicators of the powder product.

Table 8. Water absorption capacity of blackberry and blueberry powders freeze-dried with different concentrations of maltodextrin

Maltodextrin concentration	Absorption capacity in water (%)
0	81.72 ± 0.13e
10	86.10 ± 0.19d
15	89.07 ± 0.09c
20	90.89 ± 0.34b
25	94.15 ± 0.27a

Note: Equal letters in the same column do not differ significantly from each other by Tukey's test at the 5% level.

The mean values obtained for water absorption capacity showed statistically significant differences when compared to each other ($p > 0.05$). The observed behavior was of increase in values when there was an increase in the concentration of maltodextrin, ranging from 81.72 to 94.15%. Table 9 shows the mean hygroscopicity values obtained for the blackberry and blueberry blend formulations lyophilized with different concentrations of maltodextrin.

Table 9. Hygroscopicity of lyophilized blackberry and blueberry powders with different concentrations of maltodextrin

Maltodextrin concentration	Hygroscopicity (%)
0	9.78 ± 0.57a
10	8.56 ± 0.32b
15	7.01 ± 0.23c
20	5.97 ± 0.14d
25	4.25 ± 0.29e

Note: Equal letters in the same column do not differ significantly from each other by Tukey's test at the 5% level.

The hygroscopicity of the formulations showed values ranging from 4.25 to 9.78% with statistically significant differences ($p > 0.05$), the lowest values being for blends with higher concentrations of maltodextrin. According to Tonon et al. (2009) the greater the amount of added maltodextrin, the lower the hygroscopicity of the powders, as maltodextrin presents this behavior due to its low hygroscopicity. Araújo et al. (2021) in their studies with lyophilized jambo pulp, obtained hygroscopicity of 9.38%. Alves et al. (2020) when preparing lyophilized red pitaya pulp with different carrier agents (maltodextrin, gum arabic, dextrin) obtained hygroscopicity values ranging from 6.81 to 10.37%.

Conclusion

Through the results obtained, it can be concluded that:

The freeze-drying process was viable for the conservation of the elaborated blends, as they presented low values of water content and water activity;

All physical parameters were influenced by the increased concentration of maltodextrin;

The elaborated powders were classified as having intermediate cohesion and presented high water absorption capacity and low hygroscopicity.

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Chapter III

RHEOLOGICAL PARAMETERS AND TEXTURE PROFILE OF A BLACKBERRY (*Morus alba*) AND BLUEBERRY (*Vaccinium sect. Cyanococcus*) BLEND IN DIFFERENT CONCENTRATIONS OF GOAT'S MILK

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Introduction

In the juice, pulp and fruit blends production industry, there are several processing steps in which the product is submitted, and it is important to know the rheological parameters of the products, as these data, compiled in the form of equations, are used in the calculations projects for sizing the piping and equipment suitable for processing to avoid wasting energy (DEUS et al., 2019).

In addition, the rheological properties of the formulated formulations are also of fundamental importance for modeling the drying process, as these properties can determine the flow behavior, which must be known for the design of the feeding system, as well as for understanding the distribution of material within the dryer. The study of rheological behavior consists of applying a force to the sample to be investigated and measuring its deformation or applying a deformation and measuring its resistance to flow. Lack of knowledge of data may lead industries to apply, in the processing of these products, conditions similar to those used for whole juices, which may lead to errors in the development of the product and the process (FEITOSA et al., 2018; BORGES et al., 2017).

However, another important parameter to be evaluated is the texture profile, which, according to Alcântara et al. (2019), the determination of the texture profile is carried out through the use of a texturometer, which is an equipment that enables the analysis of numerous rheological parameters that simulate existing conditions during the tasting process, and constitute an important attribute of food quality together with appearance and taste.

In this context, this study aimed to evaluate the rheological behavior and texture profile of a blend composed of blackberry and blueberry pulp in different concentrations of goat milk (0.15 and 30%).

Methodology

The blackberries (*Morus alba*) and blueberries (*Vaccinium sect. Cyanococcus*) figure 1, used were from the Nossa Fruta® brand and powdered goat milk from the

Caprilat® brand, both purchased in the local market in the city of Natal, Rio Grande do North, Brazil.



Figure 1. Blueberries (*Vaccinium sect. Cyanococcus*) and blackberries (*Morus alba*) used in the samples.

Preparation of formulations

To prepare the formulations, it was initially necessary to process the fruits to obtain the pulp. The fruits were processed separately with the aid of a domestic fruit processor, separating the pulp and residue fractions. The waste was discarded. The pulp obtained was added with powdered goat milk at concentrations of 0, 15 and 30%, after addition the mixture (50% blackberry pulp and 50% blueberry pulp) was homogenized with the aid of a domestic blender.

In Figure 2, a summarized flowchart of the main steps carried out in the study can be seen.



Figure 2. General flowchart of the main steps carried out in this study.

Rheological behavior

To determine the rheological study of the elaborated pulp and goat milk blend, a Brookfield model DV II + Pro viscometer was used to read the apparent viscosity and torque percentage values of each formulation at the different concentrations of goat milk in powder (0, 15 and 30%) and temperature of 25°C (measured with the aid of a thermostat, at ambient atmospheric pressure) at different speeds of rotation. To transform torque readings into rheological measurements, the methodology proposed by Mitschka (1982) was used.

With shear stress data as a function of strain rate, rheograms were drawn and the rheological models of Ostwald-de-Waele (Equation 1), Herschel-Bulkley (Equation 2) and Mizrahi e Berk (Equation 3) were adjusted using the Statistica version 7.0 program.

$$\tau = k \gamma^n \quad (\text{Eq.1})$$

$$\tau - \tau_{oH} = K_H \gamma^{nH} \quad (\text{Eq.2})$$

$$\tau^{0,5} = k_{oc} + k_c \gamma^{\eta 1} \quad (\text{Eq.3})$$

To evaluate the adjustment of the rheological models to the experimental data, the values of the coefficients of determination (R^2) were calculated and the Chi-square test was applied (Equation 4).

$$\chi^2 = \sum (X_{obs} - X_{pre})^2 \quad (\text{Eq.4})$$

Where: χ^2 is the chi-square function; X_{obs} is the experimental value; X_{pre} is the value predicted by the model.

Determination of texture profile

The three formulations were submitted to texture profile analysis (TPA) using a universal texturometer model TA-XT plus -Textura Analyzer from the manufacturer Stable Micro Systems, equipped with the Exponent Stable Micro Systems software, using the P-36R probe, to obtaining the attributes of firmness, consistency, cohesiveness and viscosity index.

Statistical analysis

The results of the instrumental texture profile were subjected to statistical treatment using a completely randomized design with a comparison test of means, using the Assisat software version 7.7 beta (SILVA & AZEVEDO, 2009).

Results

Table 1 shows the values obtained for the parameters of the Ostwald-de-Waele rheological model adjusted to the rheograms of the blend of blackberry and blueberry pulp with different concentrations of powdered goat milk.

Table 1. Parameters, coefficients of determination (R^2), chi-square (χ^2) values of the Ostwald–de-Waele rheological model adjusted to the rheograms of the blend of blackberry and blueberry pulp with different concentrations of powdered goat milk

Model	Goat milk (%)	Parameters			R^2	χ^2
		K (Pa.s) ⁿ	n	-		
Ostwald-de-Waele	0	8,821	0.4345	-	98.66	0.02331
	15	12,553	0.2793	-	97.86	0.00273
	30	16,044	0.2101	-	94.80	0.00104

According to Almeida et al. (2020) K is the consistency index and indicates the degree of resistance of the fluid to flow. It is observed that it showed a tendency to increase when there was an increase in the concentration of goat milk. The values of n

were less than 1 ($n < 1$), showing a reduction for higher concentrations of goat milk. According to Sousa et al. (2017) the fluid behavior index (n) presents values lower than unity ($n < 1$), characterizing, therefore, the fluids as non-Newtonian and pseudoplastic. According to Costa et al. (2021) the pseudoplastic fluid has its apparent viscosity gradually decreased as the shear stress increases and, therefore, its viscosity cannot be expressed by a single value.

It is also observed in Table 1 that the coefficient of determination (R^2) was greater than 94% and the chi-square values ranged from 0.00104 to 0.02331, however, the adjustment of the Ostwald-de-Waele model is not satisfactory for the experimental dataset. Figure 3 shows the rheogram of the blend of blackberry and blueberry pulp with different concentrations of powdered goat milk at temperatures of 25°C, with the curves fitted with the Ostwald-de-Waele model.

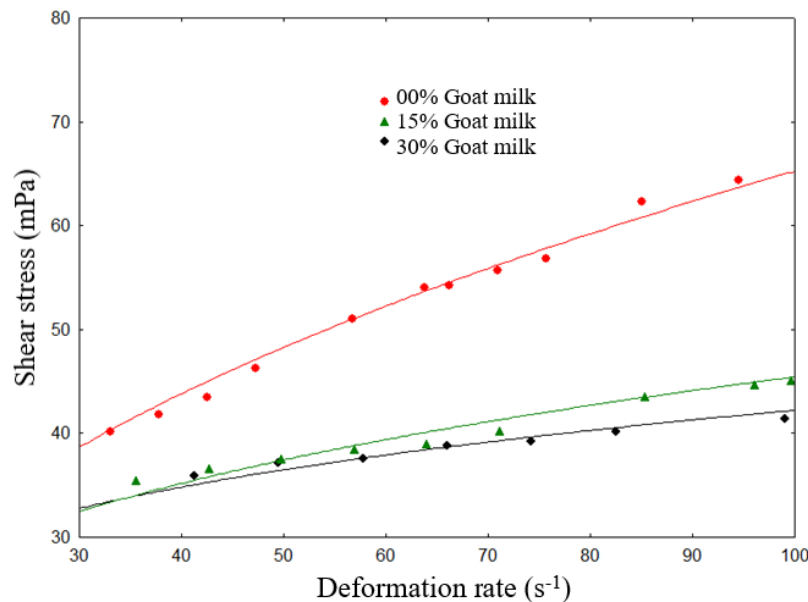


Figure 3. Rheogram of the blend of blackberry and blueberry pulp with different concentrations of powdered goat milk at a temperature of 25°C with the Ostwald-de-Waele model adjusted to the experimental data.

It can be seen that, the lowest values for the shear stress are observed in the highest concentrations of goat milk (30%) at a given strain rate. Table 2 shows the values obtained for the parameters of the Mizrahi e Berk rheological model adjusted to the rheograms of the blend of blackberry and blueberry pulp with different concentrations of powdered goat milk.

Table 2. Parameters, coefficients of determination (R^2), chi-square (χ^2) values of the Mizrahi e Berk rheological model adjusted to the rheograms of the blueberry and blackberry pulp blend with different concentrations of powdered goat milk

Model	Goat milk (%)	Parameters			R^2	χ^2
		K_{OM} (Pa)	n_M	K_M (Pa.s) ⁿ		
Mizrahi e Berk	0	24.729	0.897	0.680	99.43	0.000581
	15	29.528	0.967	0.179	99.55	0.011655
	30	34.595	0.987	0.008	99.11	0.000602

For the parameter n_M of the Mizrahi e Berk model, the same behavior of the model shown in Table 1 was observed, that is, values less than 1. However, the values tended to increase as the concentration of milk increased. goat. The initial shear stress (K_{OM} and τ_{0H}) is a finite stress necessary for the fluid to begin to flow (ALMEIDA et al., 2020). The K_{OM} values were influenced by the concentration of goat milk ranging from 24,729 to 35,595 Pa when the milk concentration ranged from 0 to 30%.

Regarding the statistical parameters, it is observed in Table 2 that the coefficient of determination (R^2) was greater than 99% and the chi-square values ranged from 0.000581 to 0.011655, indicating a good fit of the model to the data experimental tests. Figure 4 shows the rheogram of the blend of blackberry and blueberry pulp with different concentrations of powdered goat milk at temperatures of 25°C, with the curves fitted using the Mizrahi e Berk model.

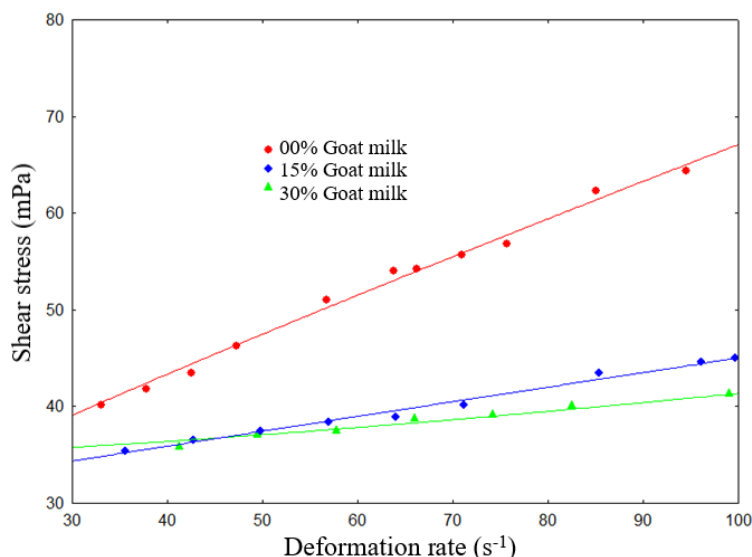


Figure 4. Rheogram of the blend of blackberry and blueberry pulp with different concentrations of powdered goat milk at a temperature of 25°C with the Mizrahi e Berk model adjusted to the experimental data.

It is observed through Figure 4 that higher shear stress values were obtained for the blend that did not contain goat milk in its formulation and the formulations with 15 and 30% goat milk showed lower values, in addition to a cross between curves, indicating according to Barros et al. (2020) that increasing the concentration of milk from 15 to 30% had little influence on the position of the curves. Table 3 shows the values obtained for the parameters of the Herschel-Bulkley rheological model adjusted to the rheograms of the blend of blackberry and blueberry pulp with different concentrations of powdered goat milk.

Table 3. Parameters, coefficients of determination (R^2), chi-square (χ^2) values of the Herschel-Bulkley rheological model adjusted to the rheograms of the blend of blackberry and blueberry pulp with different concentrations of powdered goat milk

Model	Goat milk (%)	Parameters			R^2	χ^2
		K_H (Pa.s) ⁿ	n_H	τ_{OH} (Pa)		
Herschel-Bulkley	0	0.6800	0.897	24.729	99.44	0.00006
	15	0.179	0.967	29.528	99.55	0.00012
	30	0.0089	0.987	34.594	99.11	0.00011

The nH values ranged from 0.897 to 0.987 being influenced by the increase in the concentration of goat milk. According to Cruz et al. (2021), the lower the value of η , the lower the apparent viscosity with the increase in the shear rate and, therefore, the lower the fluid's resistance to flow. The coefficient of determination (R^2) was greater than 99% and the chi-square values were the lowest among the three fitted models, ranging from 0.000006 to 0.00012, therefore, the Herschel-Bulkley model was chosen as the best fit model to the experimental data. Silva et al. (2012), analyzed the rheological behavior of mixed cajá and mango drinks, added with inulin and fructooligosaccharides and found that the Herschel-Bulkley model was the best fit for the study, with coefficients of determination greater than 0.93.

Figure 5 shows the rheogram of the blend of blackberry and blueberry pulp with different concentrations of powdered goat milk at temperatures of 25°C, with the curves fitted with the Herschel-Bulkley model.

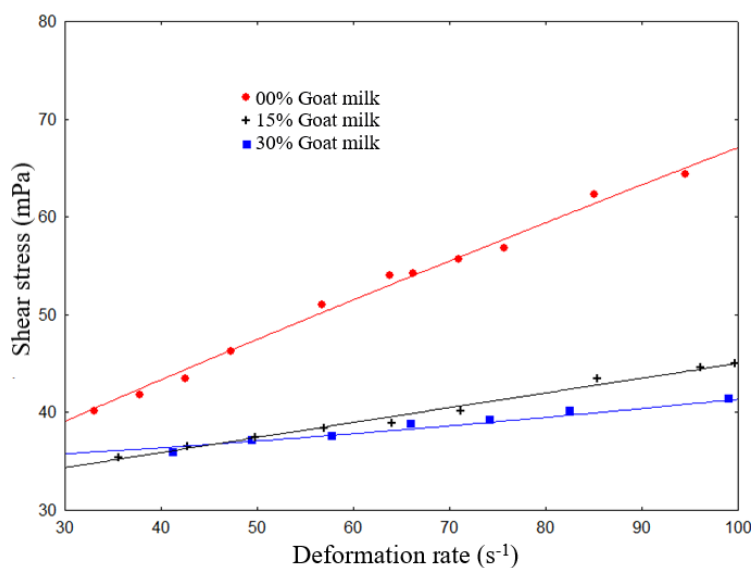


Figure 5. Rheogram of the blend of blackberry and blueberry pulp with different concentrations of powdered goat milk at a temperature of 25°C with the Herschel-Bulkley model adjusted to experimental data.

The best model fitted to the experimental data is shown in the Figure above. The blend of blackberry and blueberry pulp with different concentrations of powdered goat milk (0, 15 and 30%) is a non-Newtonian fluid. For Silva et al. (2019), non-

Newtonian fluids have a complex behavior, which must be researched and understood so that its applicability gradually increases, as its characteristics are important for technological advancement in various sectors, such as the oil, pharmaceutical, cosmetics industry, among others.

In Table 4, it is possible to observe the mean values obtained for the parameter of firmness of the blend of blackberry and blueberry pulp with different concentrations of powdered goat milk.

Table 4. Firmness values obtained for the blend of blackberry and blueberry pulp with different concentrations of powdered goat milk at a temperature of 25°C

Formulation (%)	Firmness (N)
0	0.552a
15	0,411b
30	0.407b

Note: Means followed by the same letter in the same column do not differ significantly from each other ($p>0.05$).

Firmness values ranged from 0.552 to 0.407 N, showing no significant reduction when there was an increase in the concentration of powdered goat milk from 15 to 30%. According to Santos et al. (2020) firmness is the force necessary to reach a certain deformation in the food. Table 5 shows the mean values obtained for the consistency parameter of the blend of blackberry and blueberry pulp with different concentrations of powdered goat milk.

Table 5. Consistency values obtained for the blend of blackberry and blueberry pulp with different concentrations of powdered goat milk at a temperature of 25°C

Formulation (%)	Consistency (Nos)
0	12,696a
15	9,456b
30	9,105b

Note: Means followed by the same letter in the same column do not differ significantly from each other ($p>0.05$).

The consistency of the elaborated fluids showed a reduction in their values when there was an increase in the concentration of goat milk, with values ranging from 12.696 to 9.105 N.s. Statistically the values obtained when compared to each other, only the formulations with 15 and 30% did not show significant differences ($p>0.05$). According to Mousavi et al. (2018), consistency is a parameter that strongly influences the acceptability of the food. This parameter represents the union of the product molecules until a deformation occurs due to the application of external forces. Table 6 shows the mean values obtained for the cohesiveness parameter of the blend of blackberry and blueberry pulp with different concentrations of powdered goat milk.

Table 6. Cohesiveness values obtained for the blend of blackberry and blueberry pulp with different concentrations of powdered goat milk at a temperature of 25°C

Formulation (%)	Cohesiveness (N)
0	0.417a
15	0.304b
30	0.286c

Note: Means followed by the same letter in the same column do not differ significantly from each other ($p>0.05$).

Regarding the cohesiveness parameter, it can be observed that the values obtained were less than 0.5 N, ranging from 0.286 to 0.417 N, the highest value being obtained for the blend that did not contain goat milk in its formulation (0%). Statistically all formulations showed significant differences at the 5% probability level. According to Mantovani et al. (2012), cohesiveness allows the assessment of the product's resistance to dissolving during the taster's tasting. Table 7 shows the mean values obtained for the viscosity index of the blend of blackberry and blueberry pulp with different concentrations of powdered goat milk.

Table 7. Viscosity index values obtained for the blend of blackberry and blueberry pulp with different concentrations of powdered goat milk at a temperature of 25°C

Formulation (%)	Viscosity index (Ns)
0	1,338a
15	0,823b
30	0.762c

Note: Means followed by the same letter in the same column do not differ significantly from each other ($p>0.05$).

Viscosity index values tended to decrease when there was an increase in milk concentration. The indices obtained were significantly different ($p>0.05$) and presented values ranging from 0.762 N.s (blend with 30%) to 1.338 N.s (blend with 0%). According to Salem et al. (2020) the viscosity index measures how much the fluid can vary its viscosity as a function of temperature.

Conclusion

Through the results obtained, it can be concluded that:

The blend of blackberry and blueberry in different concentrations of goat milk was classified as non-Newtonian and pseudoplastic;

The Mizrahi e Berk model was the best fit to the experimental data with a coefficient of determination (R^2) greater than 99% and low chi-square values;

The increase in goat milk concentration from 15 to 30% did not significantly influence firmness values;

The consistency, cohesiveness and viscosity index showed a reduction in their values when there was an increase in the percentage of milk in the blends formulation.

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Chapter IV

DETERMINATION OF EFFECTIVE DIFFUSIVITY, ACTIVATION ENERGY AND THERMODYNAMIC PROPERTIES OF BLACKBERRY (*Morus alba*) AND BLUEBERRY (*Vaccinium sect. Cyanococcus*) PULP DRYING IN FOAM MAT

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Introduction

Fruits have great nutritional importance for human nutrition, which are recommended due to the availability of carbohydrates, minerals, fiber, carotenoids, vitamins C, phenolic substances and antioxidant action, responsible for the prevention of various diseases when consumed regularly (RIGUETO et al., 2018).

Most of these fruits deteriorate in a few days, harming their commercialization, especially in the case of long distances. The deterioration occurs mainly due to the high water content present in the fruits, therefore, it is necessary to apply conservation techniques (SANTOS et al., 2013).

Conservation techniques, such as drying in a foam layer, are often used to reduce the water content and water activity of the product, the main parameters responsible for deterioration caused by the action of deteriorating microorganisms and the occurrence of reactions, whether enzymatic or oxidative (SOUZA et al., 2019).

Foam bed drying has gained a lot of attention because it is an effective technique and because it does not present major problems associated with traditional dehydration methods, including poor rehydration characteristics, unfavorable sensory profile, and long drying time (AZIZPOUR et al., 2016).

Foam bed drying is efficient for biological materials such as fruits, which are heat-sensitive food matrices, as it maintains quality and nutritional value, in addition to sensory characteristics. The method consists of transforming a liquid or pasty food into a stable foam by incorporating air and subsequently forming bubbles in the process. With this, a porous and easy rehydration end product is obtained, with lower operating cost (MELO et al., 2013)

In addition, the powder properties such as fluidity, cohesion, rehydration and hygroscopicity are improved by the foam mat drying method (NG & SULAIMAN, 2018). Some researchers have already used foam bed drying for food products such as: acerola (SILVA et al., 2020); raspberry (OZCELIK et al., 2019); strawberry (VIMERCANTI et al., 2019); sleeve (CHAUX-GUTIÉRREZ et al., 2017; WILSON et al., 2012); banana (NAKNAEN et al., 2016).

Knowledge of thermodynamic properties in drying is an important source of information for designing equipment, calculating the energy required in the process, studying the properties of adsorbed water and evaluating the microstructure of foods, as well as for the study of physical phenomena that occur on their surface (SANTOS et al., 2019).

The enthalpy variation measures the energy difference in the interaction of water molecules with the product during the process. The entropy variation is associated with the degree of disorder in the system, it is a state function that tends to increase in an isolated system. Furthermore, entropy may be related to the binding or repulsion of forces in the system, and to the product-water spatial arrangement.

Gibbs free energy is the energy needed to transfer the water molecules present in the product to the air, that is, it is the work performed by the system to carry out the sorption process (SOUZA et al., 2020).

In this context, the present study aims to determine the effective diffusivity, activation energy and thermodynamic properties of blackberry and blueberry pulp drying through the foam layer drying process.

Methodology

Raw material

The blackberries (*Morus alba*) and blueberries (*Vaccinium sect. Cyanococcus*) figure 1, used were from the Nossa Fruta® brand and powdered albumin from the Naturovos® brand, both were purchased in the local market in the city of Natal, Rio Grande do Norte, Brazil.



Figure 1. Blueberries (*Vaccinium sect. Cyanococcus*) and blackberries (*Morus alba*) used in the samples.

Preparation of mixtures

The fruits were processed separately with the aid of a domestic fruit processed. The pulp obtained was used to prepare the formulations shown in Table 1.

Table 1. Formulations designed to obtain the foam

Formulations	Ingredients
P1	Blackberry pulp + powdered albumin
P2	Blueberry pulp + powdered albumin
P3	Blackberry pulp + blueberry pulp + powdered albumin

For each formulation P1 (100% blackberry pulp), P2 (100% blueberry pulp) and P3 (50% blackberry pulp and 50% blueberry pulp) were added 8% powdered albumin. The mixtures were homogenized in a domestic mixer for 15 minutes, using its maximum speed. The foams obtained were subjected to the drying process.

Drying of foams

The formulations (P1, P2 and P3) were dried at temperatures of 50, 60 and 70°C, in a fixed air circulation oven with a speed of 1.5 m s⁻¹. The foams were evenly placed on aluminum trays forming a 5 mm thin layer. Moisture loss was recorded

using a digital scale with an accuracy of 0.001g. The drying process was continued until the constant mass reading was registered.

Humidity Ratio Calculation

The moisture ratio of the drying process was calculated according to Equation 1.

$$X^*(t) = \frac{X(t) - X_{eq}}{X_i - X_{eq}} \quad (\text{Eq.1})$$

Where: X^* is the moisture ratio (dimensionless), X_{eq} is the equilibrium moisture content (dry basis), $X(t)$ is the actual moisture content of the sample at the moment t (dry basis) and X_i is the initial moisture content (dry basis).

Calculation of diffusivity

The effective diffusivities (Def) of the formulations (P1, P2 and P3) at different drying temperatures (50, 60 and 70°C) were determined using the diffusion equation (Equation 2) for rectangular coordinate systems (CRANK, 1975). In calculating Def, the analytical solution for Fick's second law of diffusion was applied in the form of an infinite series (Equation 3).

$$\frac{\partial X}{\partial t} = \frac{\partial}{\partial x} \left(Def \frac{\partial X}{\partial x} \right) + \frac{\partial}{\partial y} \left(Def \frac{\partial Y}{\partial y} \right) + \frac{\partial}{\partial z} \left(Def \frac{\partial Z}{\partial z} \right) \quad (\text{Eq.2})$$

$$X^* = \frac{8}{\pi^2} \sum_{n=0}^{\infty} \frac{1}{(2n+1)^2} \exp \left[- (2n+1)^2 \pi^2 \frac{Def}{L^2} t \right] \quad (\text{Eq.3})$$

Where: X^* is the dimensionless moisture ratio; n is the number of terms; Def is the effective diffusivity ($\text{m}^2 \text{min}^{-1}$); L is the wall thickness (m); t is the time (min).

Calculation of activation energy

The relationship between diffusivity and drying temperatures was described using the Arrhenius Equation (Equation 4).

$$D_{ef} = D_0 \exp\left(-\frac{E_a}{R(T + 273.15)}\right) \quad (\text{Eq.4})$$

Where: D_0 is a constant called the pre-exponential factor; R is the universal constant of ideal gases ($8,314 \text{ J mol.K}^{-1}$). This adjustment makes it possible to determine the activation energy (E_a).

Calculation of thermodynamic properties

The thermodynamic properties of enthalpy (Equation 5), entropy (Equation 6) and Gibbs free energy (Equation 7) related to the drying process were determined by the method described by the universal gas constant (SILVA et al., 2016; SANTOS et al., 2019).

$$\Delta H = E_a - R(T + 273.15) \quad (\text{Eq.5})$$

$$\Delta S = R \left[\ln D_0 - \ln\left(\frac{k_b}{h_p}\right) - \ln(T + 273.15) \right] \quad (\text{Eq.6})$$

$$\Delta G = \Delta H - (T + 273.15)\Delta S \quad (\text{Eq.7})$$

Where: ΔH is specific enthalpy (J mol^{-1}); E_a is the activation energy; ΔS is specific entropy (J mol.K^{-1}); ΔG is the Gibbs free energy (J mol^{-1}); k_b is the Boltzmann constant ($1.38 \times 10^{-23} \text{ J K}^{-1}$); h_p is Planck's constant ($6,626 \times 10^{-34} \text{ J s}^{-1}$); T is the temperature ($^{\circ}\text{C}$).

By linearizing the Arrhenius Equation, it was possible to obtain the $\ln D_0$ coefficient of the entropy expression (ΔS), in which the following expression was used (Equation 8).

$$\ln D = \ln D_0 - \frac{Ea}{R} \cdot \frac{1}{(T + 273.15)} \quad (\text{Eq.8})$$

Results

Table 2 shows the values obtained for the effective diffusivity of the foam layer drying process of the formulations made with blackberry pulp, blueberry pulp and powdered albumin.

Table 2. Effective diffusivity (D_{ef}) of the foam layer drying process of the formulated formulations

T (°C)	$D_{ef}(\text{m}^2 \text{min}^{-1})$		
	P1	P2	P3
50	$2,21 \times 10^{-6}$	$2,54 \times 10^{-6}$	$2,74 \times 10^{-6}$
60	$3,46 \times 10^{-6}$	$3,18 \times 10^{-6}$	$3,92 \times 10^{-6}$
70	$4,52 \times 10^{-6}$	$4,05 \times 10^{-6}$	$5,12 \times 10^{-6}$

Note: P1 (100% blackberry pulp); P2 (100% blueberry pulp); P3 (50% blackberry pulp and 50% blueberry pulp).

The magnitudes of the effective diffusion coefficients ranged from 2.21 to 4.52 x 10⁻⁶ m² min⁻¹ (formulation P1), from 2.54 to 4.05 x 10⁻⁶ m² min⁻¹ (formulation P2) and from 2.74 to 5.12 x 10⁻⁶ m² min⁻¹ (formulation P3). It can also be observed that for all formulations the effective diffusion coefficients increased their values when there were larger temperature gradients, indicating a greater speed of water outflow of the product.

Baptestini et al. (2015) when carrying out the drying process in a foam layer of soursop pulp with 7.43% albumin at temperatures ranging from 40 to 80 °C, they obtained diffusivity values ranging from 4.12 to 17.66 x 10⁻¹⁰ m² s⁻¹. Silva et al. (2021) in their studies of drying in a foam layer of acerola pulp, obtained diffusivity

values ranging from 1.3967 to $5.4528 \times 10^{-5} \text{ m}^2 \text{ min}^{-1}$ for a temperature range of 50 to 70°C .

According to Almeida et al. (2020) the effective diffusivity indicates the speed at which water can be transferred from the interior to the surface. Usually, this coefficient is used due to its complexity, in addition to limited information about the movement of water inside the food during the drying process.

Table 3 shows the values obtained for the activation energy of the drying process in a foam layer of the formulations made with blackberry pulp, blueberry pulp and powdered albumin.

Table 3. Activation energy (E_a) of the foam layer drying process of the formulated formulations

Formulations	Activation energy (kJ mol^{-1})	R^2
P1	31.29	0.9918
P2	21.72	0.9993
P3	28.15	0.9978

Note: P1 (100% blackberry pulp); P2 (100% blueberry pulp); P3 (50% blackberry pulp and 50% blueberry pulp).

The activation energy for drying in a foam layer of the formulation ranged from 21.72 to $31.29 \text{ kJ mol}^{-1}$, with the formulation P2 (100% blueberry pulp) having the lowest value obtained. Freitas et al. (2018) in their studies of drying in a foam layer with cajá pulp, obtained an activation energy of $54.983 \text{ kJ mol}^{-1}$.

According to Santos et al. (2021), the activation energy corresponds to the barrier that must be overcome so that the diffusion process can be triggered, constituting the minimum energy necessary for the water molecules to initiate the movement from the inside to the outside of the product. In drying processes, the lower the activation energy, the greater the diffusivity of water, that is, the lower the energy needed to transform water into steam.

Table 4 shows the values obtained for enthalpy (H) from the foam layer drying process of the formulations made with blackberry pulp, blueberry pulp and powdered albumin.

The enthalpy variation values were reduced inversely proportional to the temperature increase, with small percentage variations, but gradually and consistently. Ranging from 28.69 to 28.53 kJ mol⁻¹ (P1), 19.11 to 18.95 kJ mol⁻¹ (P2) and from 25.53 to 25.38 kJ mol⁻¹ (P3). Morais et al. (2019) in their studies of drying with bacaba pulp, obtained enthalpy ranging from 34.4054 to 34.2391 kJ mol⁻¹ for a temperature range ranging from 40 to 60°C.

Table 4. Enthalpy (H) of the foam layer drying process of the elaborated formulations

ΔH (kJ mol ⁻¹)			
T (°C)	P1	P2	P3
50	28,69	19,11	25,54
60	28,61	19,03	25,46
70	28,53	18,95	25,38

Note: P1 (100% blackberry pulp); P2 (100% blueberry pulp); P3 (50% blackberry pulp and 50% blueberry pulp).

According to Araújo et al. (2017) and Santos et al. (2021), with the increase in temperature and consequent increase in the partial pressure of water vapor inside the material, there is a reduction in the enthalpy of vaporization of free water, so that in the final balance, with the increase in the temperature of the drying air, there is a reduction in enthalpy.

Table 5 shows the values obtained for entropy (S) from the foam layer drying process of the formulations made with blackberry pulp, blueberry pulp and powdered albumin. According to Goneli et al. (2010), entropy is a thermodynamic property often described as the level of disorder.

Table 5. Entropy (S) of the foam layer drying process of the formulated formulations

ΔS (J mol K ⁻¹)			
T (°C)	P1	P2	P3
50	-256,31	-266,13	-264,50
60	-256,57	-266,40	-264,76
70	-256,82	-266,65	-265,02

Note: P1 (100% blackberry pulp); P2 (100% blueberry pulp); P3 (50% blackberry pulp and 50% blueberry pulp).

The entropy (ΔS) of formulation P1 ranged from -256.82 (70°C) to -256.31 kJ mol⁻¹ K⁻¹ (50°C), of formulation P2 ranged from -266.65 (70°C)) to -266.13 kJ mol⁻¹ K⁻¹ (50°C), and formulation P3 ranged from -265.02 (70 °C) to -264.50 kJ mol⁻¹ K⁻¹ (50°C)), and there was a reduction in this thermodynamic parameter with the increase in temperature, related to the reduction in moisture content during the dehydration process, which hindered the movement of water molecules. Alves et al. (2021) when determining the thermodynamic properties of the drying process of baru almond flours, obtained entropy values ranging from -229.16 to -229.64 kJ mol⁻¹ K⁻¹ with temperature ranging from 60 to 80°C.

Table 6 shows the values obtained for Gibbs free energy (G) from the foam layer drying process of the formulations made with blackberry pulp, blueberry pulp and powdered albumin. Gibbs free energy can be defined as a measure of system activity in the adsorption/desorption process and provides a better understanding of the thermodynamic driving forces that influence reactions (NADI & TZEMPELIKOS, 2018).

Table 6. Gibbs free energy (G) of the foam layer drying process of the formulated formulations

ΔG (kJ mol ⁻¹)			
T (°C)	P1	P2	P3
50	108,95	102,45	108,37
60	111,52	105,20	111,02
70	114,09	107,78	113,67

Note: P1 (100% blackberry pulp); P2 (100% blueberry pulp); P3 (50% blackberry pulp and 50% blueberry pulp).

The variation of Gibbs free energy showed positive values, indicating that the process is non-spontaneous, therefore, it is necessary to supply thermal energy for it to occur. It is also possible to observe in Table 6, that the G values increased when the drying temperature increased.

Formulation P1 with 100% blackberry pulp presented values ranging from 108.95 to 114.09 kJ mol⁻¹, formulation P2 with 100% blueberry pulp presented values ranging from 102.45 to 107.78 kJ mol⁻¹ and formulation P3 with 50% blackberry pulp and 50% blueberry pulp, presented values ranging from 108.37 to 113.67 kJ mol⁻¹.

Conclusion

Through the results obtained, it can be concluded that:

The effective diffusion coefficients were in the range of 10⁻⁶, being influenced by the increase in drying temperature. Formulation P3 showed the highest values for all temperatures;

The activation energy of the process was less than 32 kJ mol⁻¹, the lowest value being obtained for formulation P2;

Thermodynamic properties were affected by drying temperature;

The drying process was characterized as being non-spontaneous endergonic, requiring energy to perform.

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Chapter V

DEVELOPMENT AND CHARACTERIZATION OF WHOLESALE YOGHURT FLAVORED WITH BLACKBERRY (*Morus alba*) AND BLUEBERRY (*Vaccinium sect. Cyanococcus*) PULP

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Introduction

The dairy industry is constantly expanding in Brazil. It is estimated that, in the last 20 years, there has been an increase of about 100% in the per capita consumption of yogurt, which is considered to be a tasty product with high nutritional value (GONÇALVES et al., 2018).

Yogurt is defined as a fermented milk produced by cultivating *Streptococcus thermophilus* and *Lactobacillus delbrueckii* spp. *bulgaricus*. The fermentation process causes the breakdown of organic substances, transforming them into smaller compounds, and consequently enables the production of a more digestible, stable, flavored food with better nutritional value. When compared to milk, yogurt is found to be more nutritious and an excellent source of protein, calcium, phosphorus, riboflavin, thiamine, vitamin B12, folate, niacin, magnesium, and zinc (BARROS et al., 2020).

According to Neres et al. (2015), the market has a vast diversification of yogurts, due to its production from pasteurized milk added with dairy culture and the possibility of being enriched with proteins, vitamins and minerals, alone or through the addition of fruit in the form of pieces, syrups and jellies, thus giving the final product a characteristic flavor and aroma. The group of foods made from milk is an important source to obtain nutrients that are so necessary for the proper functioning of the human body (PAIVA et al., 2015).

Therefore, the increase in yogurt consumption can be attributed to the growing concern of people with ingesting natural products and the benefits that food brings to the human body. There is also a tendency to add fruits and vegetables to dairy foods with the aim of improving the nutritional and sensory value of the final product, since fruits and vegetables are rich in bioactive compounds, such as vitamins, antioxidant compounds and fiber (BARBOSA & GALLINA, 2017).

Fruits represent the largest natural reserves of nutrients and antioxidants, such as vitamins A, C and E and phenolic compounds, which are responsible for flavor, color, odor and oxidative stability. Such bioactive compounds modulate metabolic processes, act directly on cellular activities, with their antioxidant, anticancer, anti-inflammatory, antimicrobial and anti-allergic properties, causing beneficial physiological effects,

preventing or reducing the risk of diseases such as type 2 diabetes and cardiovascular diseases (BERNARDI et al., 2019; DONNO et al., 2019).

Red fruits, especially blackberries and blueberries, deserve special attention, as they are strategic food matrices, as they act in the prevention of chronic diseases due to the high concentration of anthocyanins. As anthocyanins, in addition to being responsible for the blue color, like blueberries, they are the main group of flavonoids in these fruits and have been associated with several beneficial effects. Blueberries are known to be rich in bioactive compounds including flavonoids, phenolic acids, tannins and anthocyanins. These compounds vary according to the species, but stand out as catechins, caffeic, chlorogenic, p-coumaric and ferulic acid, among many others (OKAN et al., 2018).

Blackberries, in addition to their high vitamin C content, contain about 85% water, about 10% carbohydrates, rich in minerals, vitamins B and A., about 82% water, 0.4 to 0.7% protein, 0.5% fat and 0.19 to 0.25% ash content (SOUSA et al., 2017).

In this context, the search for healthier foods and aiming to expand the consumption of foods with functional properties, the present study aims to develop whole yogurt added with blackberry and blueberry pulp and evaluate its physicochemical, microbiological and texture characteristics.

Methodology

Raw material

The blackberries (*Morus alba*) and blueberries (*Vaccinium sect. Cyanococcus*) figure 1, used were from the Nossa Fruta® brand and whole milk powder from the Italcac® brand, both purchased in the local market in the city of Natal, Rio Grande do Norte, Brazil.



Figure 1. Blueberries (*Vaccinium sect. Cyanococcus*) and blackberries (*Morus alba*) used in the samples.

Preparation of whole yogurt

To prepare the yogurts, the inoculum was initially prepared. To prepare the inoculum, Italac® whole milk powder, reconstituted as described on its packaging, was used. It was sterilized at 80°C for 15 minutes, cooled to $45 \pm 1^\circ\text{C}$. After preparing the inoculum, the commercial culture of *Streptococcus salivarius* sub sp. thermophilus, incubating it at $45 \pm 1^\circ\text{C}$ for 4 hours (until the desired percentage of acidity is reached). After this fermentation period, the inoculum was kept refrigerated for later use.

Four yoghurts were prepared, as shown in Table 1. For each formulation, the incubation procedure described above was carried out.

Table 1. Formulations for the preparation of wholegrain yoghurts added with blackberry and blueberry pulp

Formulations	Ingredients
TO 1	Natural yogurt
A2	Natural yogurt + blackberry pulp
A3	Natural yogurt + blueberry pulp
A4	Natural yoghurt + blackberry pulp + blueberry pulp

Each formulated formulation was placed in previously sterilized plastic packages.

Physicochemical characterization

All formulations developed were characterized according to the following parameters:

(1) moisture content was determined by drying in an oven at 105°C to constant weight (BRASIL, 2008);

(2) Water activity (A_w) was determined using the Decagon® Aqualab CX-2T device at 25°C;

(3) ash content was determined by muffle incineration (BRASIL, 2008);

(4) pH was determined through direct reading on the digital pH meter (BRASIL, 2008);

(5) Total titratable acidity determined by titration with sodium hydroxide (BRASIL, 2008).

Microbiological analysis

After production, all formulations were analyzed for the following parameters: Coliforms at 35 and 45°C, *Staphylococcus* spp, and *Salmonella* sp. following the methodology described by Paiva et al. (2015).

Determination of firmness

All formulated formulations were submitted to firmness analysis. This analysis was performed using a universal texturometer, model TA-XTplusC Texture Analyzer, from the manufacturer Stable Micro Systems, equipped with the Exponent Stable Micro Systems software, using the P-36R probe.

Statistical analysis

The experimental data were analyzed in triplicate and the results were submitted to the analysis of variance of a single factor (ANOVA) of 5% probability and the significant qualitative responses were submitted to the Tukey test, adopting the same level of 5% of significance. For the development of statistical analysis, the software Assisat 7.7 (SILVA & AZEVEDO, 2016) was used.

Results

Table 2 shows the values obtained for the moisture content of whole-grain yoghurts made with blackberry and blueberry pulp.

Table 2. Results obtained for the moisture content of prepared whole yogurt formulations

Formulation	Moisture content (%)
TO 1	78.12±0.10c
A2	81.23±0.15b
A3	82.01±0.12a
A4	81.98±0.09ab

Note: Equal letters in the same column do not differ significantly from each other by Tukey's test at the 5% level.

In the analysis of moisture content, values ranging from 78.12% to 82.01% are observed, the highest percentage being obtained for formulation added with blueberry

pulp (A3). When the formulations were compared with each other, the mean value obtained for formulation A4 did not show statistically significant differences between formulations A3 and A2 at the 5% probability level.

Although Brazilian legislation does not have defined standards for the moisture content of yoghurts, the findings of our study are similar to those reported in the literature by Barros et al. (2020) who obtained values ranging from 81.69 to 86.10% for yogurts made with the addition of pumpkin jelly and corn starch and by Antunes et al. (2015) who obtained a moisture content of 77.76 to 83.29% for semi-skimmed yoghurts added with whey protein concentrate.

Table 3 shows the values obtained for the water activity of whole yogurts made with blackberry and blueberry pulp.

Table 3. Results obtained for water activity of prepared whole yogurt formulations

Formulation	Water activity
TO 1	0.934±0.01b
A2	0.956 ± 0.02a
A3	0.963±0.00a
A4	0.961±0.03y

Note: Equal letters in the same column do not differ significantly from each other by Tukey's test at the 5% level.

All formulations had values above 0.9 with respect to the water activity parameter, which varied from 0.934 to 0.961. Statistically, only the control formulation (A1), which did not contain in its formulation the addition of blackberry and blueberry pulp, had the lowest water activity value and showed significant differences when compared to the others ($p>0.05$). Barros et al. (2020) obtained water activity values ranging from 0.947 to 0.989 for yogurts made with the addition of pumpkin jelly and corn starch. Table 4 shows the values obtained for the ash content of whole yogurts made with blackberry and blueberry pulp.

Table 4. Results obtained for the ash content of prepared whole yogurt formulations

Formulation	Ashes (%)
TO 1	0.51±0.17d
A2	0.82±0.21b
A3	0.72±0.19c
A4	1.16±0.26a

Note: Equal letters in the same column do not differ significantly from each other by Tukey's test at the 5% level.

The ash content of a food represents the amount of minerals contained in it, where the values found ranged from 0.51 to 1.16%, differing significantly from each other in the Tukey test ($p>0.05$).

Values close to those of the present study were reported by Abreu et al. (2019) when preparing probiotic yogurts from goat's milk added with guava pulp, they observed ash content of 0.70% and by Silva et al. (2020) for yoghurt added with tamarind pulp (1.17%).

Table 5 shows the values obtained for the pH of whole-wheat yoghurts made with blackberry and blueberry pulp.

Table 5. Results obtained for the pH of the prepared whole yogurt formulations

Formulation	pH
TO 1	4.38±0.01a
A2	4.09±0.00c
A3	4.15±0.01b
A4	3.96±0.02d

Note: Equal letters in the same column do not differ significantly from each other by Tukey's test at the 5% level.

Regarding the values obtained for the pH of the control whole yogurt (A1) and with the addition of blackberry and blueberry pulp (A2, A3 and A4), values below 4.4 were observed, being characterized as slightly acidic.

According to Araújo et al. (2021) pH below 4.5, as obtained in this research, is essential for the conservation of the product. Statistically all formulations showed

statistically significant differences when compared to each other ($p > 0.05$). The formulation with the addition of blackberry and blueberry pulp (A4) with the lowest value for this parameter was 3.96.

Araújo et al. (2021) when preparing flavored yogurt with jambo pulp obtained a pH of 4.4. Modesto Júnior et al. (2016), when evaluating the pH of Greek-style yoghurts made with buffalo milk and different concentrations of sour sour syrup, they obtained a pH variation from 3.63 to 4.13.

Table 6 shows the values obtained for the total titratable acidity of whole-grain yogurts made with blackberry and blueberry pulp.

Table 6. Results obtained for total titratable acidity of prepared whole yogurt formulations

Formulation	Acidity (%)
TO 1	0.59±0.02d
A2	0.92±0.01b
A3	0.85±0.01c
A4	1.09±0.03y

Note: Equal letters in the same column do not differ significantly from each other by Tukey's test at the 5% level.

As expected, the A4 formulation had a higher average than the elaborated formulations, with a value of 1.09%, since the pH obtained previously was 3.96 and the A1 formulation had a lower mean than the elaborated formulations, with a value 0.59%, since the pH obtained previously was 4.38.

Neres et al. (2015) in their studies with yogurt added with pineapple pulp and husk flour, obtained values ranging from 0.56% to 0.70%. Table 7 shows the results obtained for the microbiological evaluation (coliforms 35 and 45°C, *Staphyococcus spp.* and *Salmonella sp.*) of whole yogurts made with blackberry and blueberry pulp.

Table 7. Results obtained for the microbiological evaluation of the prepared whole yogurt formulations

Parameters	Formulations			
	TO 1	A2	A3	A4
Coliforms 35°C	Absent	Absent	Absent	Absent
Coliforms 45°C	Absent	Absent	Absent	Absent
<i>Staphyococcus</i> spp	Absent	Absent	Absent	Absent
<i>Salmonella</i> sp.	Absent	Absent	Absent	Absent

All the elaborated formulations are within the standards established by Resolution No. 12 of ANVISA (BRASIL, 2001). With absence of positive tubes for total coliforms (35°C) and thermotolerant coliforms (45°C), as well as absence of typical colonies of *Staphyococcus* spp and *Salmonella* sp.

Paiva et al. (2015) when developing yogurts with pineapple pulp and honey, they also obtained an absence of total coliforms and thermotolerant coliforms in all formulations.

Gonçalves et al. (2018) when preparing yogurts with cashew jelly (*Spondias mombin* l.) added with probiotics, they also observed an absence of *Salmonella* sp. in all formulations.

Silva et al. (2020) when preparing Greek yogurt with the addition of tamarind pulp, they also observed an absence for all analyzed microorganisms.

According to Cohen et al. (2011), microbial contamination in food can occur during any stage of its production, becoming a risk to the population, when they cause Foodborne Diseases (FDAs), however, microbial proliferation is often associated with the absence or inadequate handling , as well as insufficient thermal treatment (GONÇALVES et al., 2018).

Table 8 shows the values obtained for the firmness parameter of whole-wheat yoghurts made with blackberry and blueberry pulp. According to Egea et al. (2019) the texture and drying of yogurt are two factors that strongly influence the acceptance of yogurt, and there is a strong preference by consumers for homogeneous yogurts, smooth, with a smooth texture, viscous body, without syneresis and with such consistency that can eat it with the spoon.

Table 8. Results obtained firmness of the prepared whole yogurt formulations

Formulation	Firmness (N)
TO 1	0.81±0.05y
A2	0.66 ± 0.02b
A3	0.62±0.01b
A4	0.53±0.04c

Note: Equal letters in the same column do not differ significantly from each other by Tukey's test at the 5% level.

Regarding the values obtained for the firmness attribute, a variation between 0.53 to 0.81 N was observed between the elaborated formulations. Formulations A2 and A3 did not show statistically significant differences ($p>0.05$). The formulation with blackberry and blueberry pulp (A4) showed the lowest firmness value (0.53 N). According to Almeida et al. (2019), once the product presents a lower firmness, consequently its consistency will be lower.

Conclusion

Through the results obtained, it can be concluded that:

Whole-wheat yoghurt made with blackberry and blueberry pulp can be considered a great option for consumption of dairy products;

High moisture contents (>78%) and high water activity values (>0.9) were obtained;

The low pH values were adequate for the conservation of the product;

The formulations were microbiologically safe for consumption, with the absence of all microorganisms;

The formulation with the addition of the two pulps showed lower firmness.

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Chapter VI

PREPARATION AND CHARACTERIZATION OF A MIXED BLACKBERRY (*Morus alba*) AND BLUEBERRY (*Vaccinium sect. Cyanococcus*) PULP STRUCTURE

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Introduction

Fruit consumption has high acceptability; however, this consumption is also associated with a high degree of perishability. Thus, due to seasonal periods, it is necessary to apply technologies that can provide a better use of these raw materials. The use of new processes that can add value and increase the shelf life of products generated from fruit, together with the diversity of research in the agro-industrial area, has been awakening a more innovative profile in the food market (ALBUQUERQUE et al., 2021).

The food industry is looking for innovations that can favor the exploitation and increase of the market niche for relatively well-known foods, such as blackberry and blueberry pulp. In addition, it tries, at all times, to develop methods that can preserve food for a longer period of time, maintaining its sensory and nutritional characteristics in the best possible way, since, in order to take the fruits to other locations, in a safe way for consumption, the use of adequate technologies is necessary (SILVA et al., 2005).

Evaporation consists of concentrating a liquid through boiling, which occurs exactly at the boiling point. The main purpose of evaporation is to increase the concentration of the number of total solids to reduce water activity (a_w), the concentration inhibits enzymatic activity and microbial growth, providing a longer shelf life of the food. Free water on the product surface is more easily removed by evaporation, where it moves by gaseous diffusion to the surrounding air (KERR, 2019).

Evaporation, as a procedure for eliminating water by boiling, requires a heating medium that transmits the heat required for the change of state (sensible heat and latent heat of evaporation). In the food industry, saturated water vapor (primary steam) is normally used as a heating fluid, which condenses, yielding its latent heat to the product that evaporates. It is, therefore, the exchange of latent heat (from condensation and evaporation). Evaporation takes place in heat exchangers (JESUS et al., 2016). Among the processing techniques, the structuring of fruit pulp represents an innovation in the food area.

In the North American market, products such as structured fruits are already consolidated. Brazil, on the contrary, still has a large and promising market to be explored in relation to such types of products, mainly due to the great richness of flavors found throughout its territory in relation to tropical fruits. (CARVALHO et al., 2011)

Fruit structured products are products obtained from fruit purees, properly formulated to obtain nutritious products with good texture and flavor. Hydrocolloids are used, responsible for reducing the moisture of the food and structuring the pulp, through gelatinization, providing a pleasant texture and appearance to the final product. The products after being structured are subjected to drying, showing good stability (CARVALHO et al., 2011).

In this context, the present study aims to develop and characterize a mixed structure containing blackberry and blueberry pulp.

Methodology

Raw material

The blackberries (*Morus alba*) and blueberries (*Vaccinium sect. Cyanococcus*) used were from the Nossa Fruta® brand and maltodextrin from the Integral Médico® brand, both purchased in the local market of the city of Natal, Rio Grande do Norte, Brazil.



Figure 1. Blackberries (*Morus alba*) and Blueberries (*Vaccinium sect. Cyanococcus*) and used in the samples.

Obtaining the pulp

The blackberries (*Morus alba*) and blueberries (*Vaccinium sect. Cyanococcus*) used were from the Nossa Fruta® brand, both purchased in the local market in the city of Natal, Rio Grande do Norte, Brazil. To obtain the pulp, the fruits were processed separately with the aid of a domestic fruit processor, separating the pulp and waste fractions. The waste was discarded.



Figure 2. Fruit Processor Mix Philco 700 with Speed Control used in the samples.

Elaboration of the structured

The flowchart for obtaining the blackberry and blueberry structured is described in Figure 1.

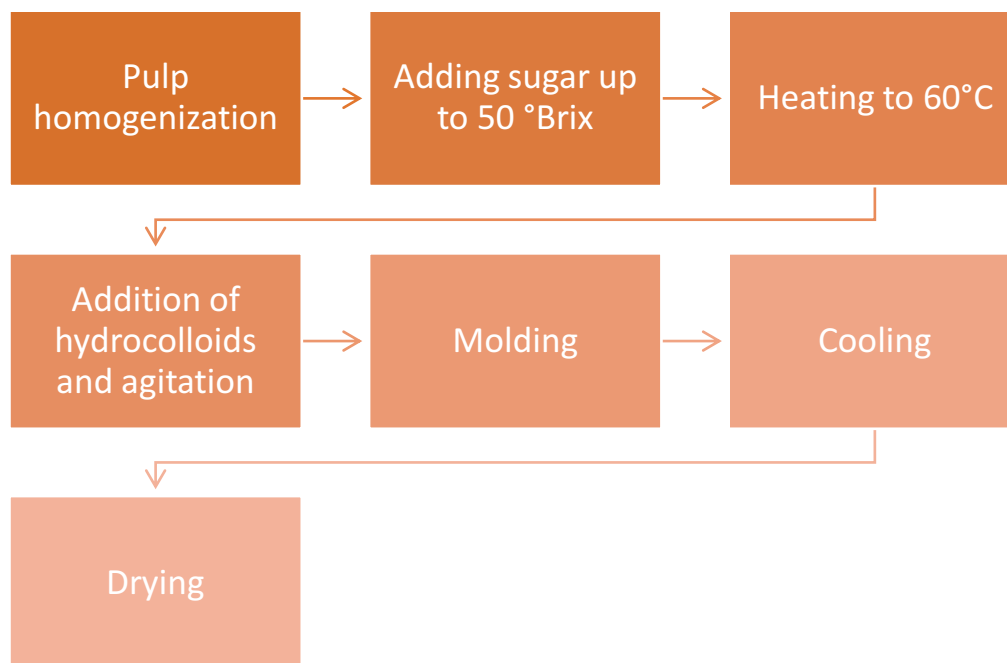


Figure 1. Flowchart for the processing of mixed structured with blackberry and blueberry pulp.

The scaffold was processed with 50% blackberry pulp and 50% blueberry pulp (Ratio 1:1). The processing of structured items was carried out according to the methodology proposed by Grizotto et al. (2005) and Carvalho et al. (2011), described below.

The pulps were homogenized in a mixer, with agitation intensity of 20,500 rpm, for about 5 minutes. Then, glycerol was added and, depending on the soluble solids content obtained, the amount of sucrose sufficient to raise the soluble solids content to 50 °Brix was calculated.

Then, the mixture was heated to 60°C, adding calcium phosphate and hydrocolloids (pectin, alginate and gelatin) under vigorous stirring, using a propeller mixer for about 5 minutes. For the molding of the structured, glass containers were used, which were kept under refrigeration (10°C/24 h) to complete the gelation. After that, the structures were cut with the aid of a stainless-steel cutter and then subjected to drying in an oven with air circulation at 45°C, for a period of 5 hours.

Characterization of the structure

The structured elaborated was characterized in triplicate regarding the following parameters:

Proximate composition

(1) moisture content was determined by drying in an oven at 105°C to constant weight (BRASIL, 2008).

(2) ash content was determined by muffle incineration (BRASIL, 2008);

(3) total protein content was quantified by the Micro-Kjeldahl method, which consisted in the determination of total nitrogen according to the methodology described by BRASIL (2008).

(4) lipid content was quantified by the modified method of Blig and Dyer (1959).

(5) total carbohydrate content was calculated by difference to obtain 100% of the total composition (FAO, 2003).

Physicochemical characterization

(1) Water activity (aw) was determined using the Decagon® Aqualab CX-2T device at 25°C.

(2) pH was determined through direct reading on the digital pH meter (BRASIL, 2008).

(3) Total titratable acidity determined by titration with sodium hydroxide (BRASIL, 2008).

Bioactive compounds

(1) The content of total phenolic compounds was quantified using aqueous extract from the Folin-Ciocalteu method described by Waterhouse (2006), using gallic acid as standard.

(2) The total anthocyanin content was determined following the unique pH method described by Francis (1982).

Microbiological analysis

After production, the following parameters were analyzed: *E. coli* and *Salmonella* sp. following the methodology described by Paiva et al. (2015).

Determination of firmness

This analysis was performed using a universal texturometer, model TA-XTplusC Texture Analyzer, from the manufacturer Stable Micro Systems, equipped with the Exponent Stable Micro Systems software, using the P-36R probe.

Results

Table 1 shows the results obtained for the proximate composition of the mixed structure, composed of 50% blackberry pulp and 50% blueberry pulp. According to Lameiro et al. (2011), nutritional information on products and foods is important for the population to acquire knowledge and can then make choices and consume nutrients in a balanced way according to the recommended daily intake.

Table 1. Proximate composition of the mixed structure of blackberry and blueberry Pulp

Parameters (g/100g)	Mixed structured
Moisture	27.31 ± 0.09
Ash	0.74 ± 0.12
Lipids	0.66 ± 0.20
Proteins	0.92 ± 0.16
Carbohydrates	70.37 ± 0.25

The moisture content of the mixed structure with blackberry and blueberry pulp was 27.31%, this value is considered relatively low, since the structures were subjected to a drying step. The low moisture value is important to ensure the conservation and integrity of the structure obtained (FONSECA et al., 2021). Higher moisture values (30.32%) were obtained by Carvalho et al. (2014) in their studies with mixed structures of umbu and passion fruit. Ash content was 0.74%. Carvalho et al. (2011) obtained ash content of 1.61% for structured with cajá and papaya pulp.

Regarding the levels of lipids and proteins in the present study, we obtained 0.66 and 0.92%, respectively. Values close to those of the present study were reported by Silva et al. (2009), who in their studies with structured cajá and guava pulp, obtained lipid contents of 0.34 and 0.45%, respectively. And for the protein content, in the same study, the authors reported 0.67% for the structured cajá and 0.39% for the structured guava. Fonseca et al. (2021) obtained protein content ranging from 0.77 to 0.96% for jabuticaba structured.

The carbohydrate content was 70.37%, which may represent a good caloric source in human food. Table 2 shows the results obtained for the physicochemical characteristics of the mixed structure. According to Santos et al. (2020), the water activity indicates the amount of water available to facilitate the occurrence of biochemical transformations or for the growth of microbial cells in food.

Table 2. Physicochemical parameters of the mixed structure of blackberry and blueberry Pulp

Parameters	Mixed structured
Water activity	0.801 ± 0.02
pH	3.90 ± 0.01
Acidity (% citric acid)	0.89 ± 0.12

The mixed structure with blackberry and blueberry pulp presented a water activity of 0.801. Similar values were also obtained by Jesus et al. (2016) when elaborating structures with fruit pulps, they obtained water activity of 0.8 for structures of umbu and melon, 0.6 for structures of umbu and pomegranate and 0.8 for structures of umbu and melon. Oliveira et al. (2008) reported umbu structures with

water activity values between 0.80 and 0.93, but the structures were not subjected to the drying process.

Regarding the pH, a value of 3.90 was obtained. Grizotto et al. (2005) in their studies with concentrated papaya pulp structured, evaluated the influence of different concentrations of pectin, glycerol and alginate and obtained pH values ranging from 3.39 to 3.82.

A low acidity value for the structure (0.89% citric acid) was determined. Silva et al. (2009), in their studies with structured cajá and guava pulp, obtained acidity values ranging from 0.69 to 0.80% of citric acid. Table 3 shows the results obtained for the total phenolic compounds and total anthocyanins of the mixed structure, composed of 50% blackberry pulp and 50% blueberry pulp.

Table 3. Bioactive compounds of the mixed structure of blackberry and blueberry Pulp

Parameters	Mixed structured
Total phenolic compounds (mgGAE/100g)	294.12 ± 2.18
Total anthocyanins (mg/100g)	90.48 ± 1.06

High levels of total phenolic compounds (294.12 mgGAE/100g) were quantified for the mixed structure. This high value is due to blackberry and blueberry pulps being rich sources of bioactive compounds. The anthocyanin content obtained in this study was 90.48 mg/100g. Higher values for these same parameters were reported by Lameiro et al. (2019) for fresh pulps, being 141.02 mg/100g for blackberry pulp and 156.03 mg/100g for blueberry pulp.

Table 4 shows the result obtained for the physical parameters of firmness of the mixed structure. According to Almeida et al. (2020), firmness is the strength necessary to reach a certain deformation in the food.

Table 4. Firmness of the mixed structure of blackberry and blueberry Pulp

Parameter	Mixed structured
Firmness (N)	52.14 ± 1.27

The mixed structure composed of blackberry and blueberry pulp (1:1) showed a firmness of 52.04 N. Carvalho et al. (2011), when elaborating structures from a mix of cajá and papaya pulps with different concentrations of alginate, pectin and gelatin, they obtained firmness values ranging from 9 to 1103.20N. In Table 5, the results obtained from the microbiological analyzes of the mixed structure of blackberry pulp and blueberry pulp are presented.

Table 5. Microbiological analyzes of the mixed structure of blackberry and blueberry Pulp

Parameters	Mixed structured
<i>E. coli</i>	Absence
<i>Salmonell a sp.</i>	Absence

Based on the results obtained (Table 5), the absence of the microorganisms *E. coli* and *Salmonella sp.* in the elaborated structure, indicating adequate and safe hygiene conditions. Fonseca et al. (2021) when preparing structures with jabuticaba pulp concentrate, they observed that they remained microbiologically stable and no microbial growth was observed, in accordance with the legislation.

Conclusion

Through the results obtained, it can be concluded that:

The use of 50% blackberry pulp and 50% blueberry pulp was viable for the preparation of the mixed structure.

The additional drying step reduced the moisture content of the structure.

Nutritional information indicated that structured food can be considered a good source of calories in human food.

The structured ones presented high values of total phenolic compounds and total anthocyanins.

And finally, the elaborated product was considered microbiologically safe for human consumption.

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