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Jussaí (*Euterpe edulis*): a review

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Abstract

The fruit of the jussara palm (*Euterpe edulis* M.) from the Brazilian Atlantic Forest is rich in anthocyanins, mainly 3-O-glucoside cyanidin and 3-O-rutinoside cyanidin, with high antioxidant capacity which can prevent different types of diseases. The aim of this review addresses aspects of the fruit, proximate composition, bioactive compounds, main methods for determining the antioxidant activity as well as phenolic compounds, fruit processing and some products with functional properties.

Keywords: *Euterpe edulis*; *Euterpe oleracea*; antioxidant activity; health benefits.

Practical Application: The jussaí can be applied in various areas of food technology.The anthocianidins, cyanidin 3-O-glucoside and 3-O-rutinoside, present in jussaí pulp can be applied to elaborate food supplements whit high antioxidant activity.

1 Introduction

Assaí is a fruit from palm trees of the *Euterpe genus*. In the Atlantic Forest, the jussaí or jussara pulp can be obtained from jussaí fruits (*Euterpe edulis* Martius) (Figure 1) (MacFadden, 2005). In the Amazon region, assaí pulp is obtained from the assaí palm fruits (*Euterpe oleracea* Martius) and from dry land assaí tree (*Euterpe precatoria* Martius).

The fruits of *E. edulis* are fleshy, fibrous, with very abundant endosperm and not ruminated (Queiroz, 2000). The frutification often takes place between March and June (Lorenzi, 2006 apud Cerisola et al., 2007; Brito et al., 2007). Despite its wide distribution in Brazil, jussaí has the lowest consumption compared to assaí (*Euterpe oleracea*) (Brito et al., 2007). The reproductive process starts around six years after planting. Fruiting in general is abundant, and palm trees can, under favorable conditions, produce 216 to 528 bunches per hectare and 6 to 8 kg of fruit per year, which is equivalent to 8,000 and 10,000 seeds or an average of 5 kg.

The jussaí fruits have an intense purple color when ripe, due to the presence of anthocyanins (Novello, 2011). These bioactive compounds are those of greatest interest in the fruit.

Brazil is the largest producer, consumer and exporter of assaí (*E. oleraceae*) in the world. Pará is the largest national producer of fruits representing almost 90% of the Brazilian production (Table 1). The only state outside the North region that appears in official statistics is Maranhão. In the last decade, Brazilian production reached more than 100,000 tons per year, moving over 100 million reais a year. The production of jussaí from *E. edulis* in extra-Amazonian states still does not figure in IBGE statistics (Brasil, 2020).

Among the *Euterpe* species, the *E. edulis*, also known as green palm, peach palm, white palm, sweet palm and jussaí, juçara, is a dominant specie of the middle stratum in forests that cover areas of wide latitudinal variation of the Atlantic Forest biome (Veloso & Klein, 1957). This specie has been commercially exploited in the last five decades for the production of a delicacy: the heart of palm removed from the interior of the stem, the pupunha (Fantini & Guries, 2007).

E. edulis is a non-stoloniferous palm tree, with a single stem measuring 5 to 12 meters in height and 10 to 15 centimeters in diameter (Figure 1). They have alternate and pinnate leaves from 2 to 2.5 meters in length; with bunches formed by thousands of fruits measuring 10 to 15 millimeters in diameter (MacFadden, 2005).

Because has a single strain, without the formation of a tiller, it can cause plant death after cutting the heart of palm (Tsukamoto et al., 2001; MacFadden, 2005). Due to the extractive use of *E. edulis* and its high economic value as food, the natural regeneration of the plant is compromised. To reverse this situation, the stimulation of the appreciation of the consumption of jussaí pulp is started, through a process of maceration and mixing with different amounts of water, promoting a creamy liquid and a characteristic flavor like assaí (*Euterpe oleracea*) (Bicudo et al., 2014). Thus, providing the frequent use of the pulp for consumption in different types of beverages, ice cream and sweets (Schulz et al., 2016).

Since 2004, the first jussaí palm production unit has been in Santa Catarina in the southern region of Brazil that supplies the region with pulp, as well as assaí from the fruits of *E. oleracea* and *E. precatoria*. However, data on production and exports of jussaí are still outdated and scarce (Schulz et al., 2016).

Ombrophilous species, slightly hygrophilic, currently, *E. edulis* is an endangered palm species (Biodiversitas, 2011). It

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Figure 1. Palm tree (*Euterpe edulis* Martius) from Atlantic Forest, Brasil. Source: Novello (2011); Schultz (2008).

Table 1. Production of *E. oleraceae* in Brazil.

Brazil, Greater Region and Federation Unit	Type of extractive product Size of plant extraction (tonnes)– 2019	
	Total	Açai (Fruit)
Brazil	..	222.706
North	..	205.116
North East	..	17.590
Rondônia State	..	1.601
Acre State	..	4.738
Amazon State	..	43.855
Pará	..	151.793
Amapá	..	3.059
Maranhão	..	17.590

produces a large amount of fruit, through rachilas with subfoliar insertion (located at the base of the palm heart) (Figure 2). Ripe fruits acquire an intense purple color and detach from the infructescence. In the ecological context, the species has a significant importance in the dense ombrophile forest as it plays na important role in the diet of vertebrate and invertebrate herbivores and can be considered a key species because its fruits are ripe in a time of general food shortage (Seoane et al., 2005).

Regarding with the commercial value, the product of greatest interest to *E. edulis* is its heart of palm, which, unlike other *Euterpe* species, is sweeter, and therefore has greater added economic value. Another characteristic of the production of this species is the amount of fruit it generates, which is lower than other *Euterpe* (Seoane et al., 2005). However, in the southeast region there are smaller scale producers (Ribeiro et al., 2011) in Serrinha, Visconde de Mauá/RJ, as a jussaí producer in the southeast region.

2 Proximal composition

The Normative Instruction, (number, 37 – continuation) defines that jussaí pulp must comply with the following characteristics, such as: total lipids between 20 to 60 g. 100 g^{-1}, proteins, at least, 6 g. 100 g^{-1} (dry basis), carbohydrates, at most, 40 g. 100 g^{-1} (dry basis), and among others. The legislation established for jussaí (*Euterpe edulis*) is classified as thick, medium or fine according to the solids content of 14%, 11%-14% and 8-11%, respectively.

Costa et al. (2000) compared the composition of jussaí pulp with those reported by Ribeiro et al. (2011) and Nascimento et al. (2008) (Table 2), finding similar results for the nutritional value, chemical and physical properties and sensory characteristics as well. The jussaí pulp presented a lipid content of 4,25 g. 100g^{-1}; carboidrates 5,66 g. 100 g^{-1}; proteins 0,36 g. 100 g^{-1} and ashes 0,41 g. 100 g^{-1}.

2.1 Antioxidant activity

Studies on its therapeutic properties have demonstrated several benefits such as anti-inflammatory, antioxidant, cardioprotective and carcinogenic properties.

Oxygen is a substance that threatens the integrity of biomolecules from biological processes, causing different types of intracellular and intramolecular damage through the production of free radicals. To reduce this damage, endogenous defense mechanisms known as antioxidants start their activities to protect the body from free radicals (oxidative stress), which is the exacerbated formation of free radicals in the body, which interact at different levels of cells causing damage, often irreversible, and when the body does not produce enough free radicals to combat the oxidative stress, it is necessary to obtain exogenous antioxidants (in the diet) (Cerqueira et al., 2007).

According to the Antioxidant Dossier (Dossiê Antioxidantes, 2016), free radicals are active atoms or molecules that have

Figure 2. Jussaí fruits (A) and bunches of jussaí palm heart. Source: Schulz et al. (2016).

Table 2. Proximate composition, pH, acidity and total soluble solids of jussaí and assaí pulps.

Parameters	Juçai pulp (g/100g)	Jussai pulp (g. 100g $^{-1}$) Ribeiro et al. (2011)	Jussai pulp ((g. 100g $^{-1}$) Nascimento et al. (2008)
Moisture	89.9 ± 0.35	88.9	89.18
Lipids	3.74 ± 0.01	4.36	4.61
Proteins	0.82 ± 0.10	0.09	0.17
Ashes	0.45 ± 0.01	0.38	0.41
Carbohydrates	5.09*	6.27	5.63
Acidity in citric acid	0.23 ± 0.02	0.19	0.19
pH	5.07	4.84	5
Total soluble solids	6.73 °Brix	3.03 °Brix	2.7 °Brix

*Calculated by difference.

an odd number of electrons in their outer orbit, for example superoxide anion, hydroxyl radical, transition metals, ozone and others. Free radicals present oxygen as reactive oxygen species, which contain superoxide radicals and the hydroxyl radical plus oxygen derivatives that do not contain odd electrons, such as hydrogen peroxide and singlet oxygen.

Due to their chemically active structure with one or more odd electrons, free radicals are highly unstable and reactive. In the body, they circulate to appropriate or donate electrons (Figure 3) and, for this reason, they damage cells, proteins and DNA (genetic material) (Dossiê Antioxidantes, 2016).

Antioxidants are substances that act against and delay damage caused by the effects of the physiological process of excess production of oxygen free radicals in animal tissue. Because the excess production of oxygen free radicals can cause cell damage (Dossiê Antioxidantes, 2016).

Thus, interest has been aroused in foods that contain antioxidant substances for the prevention of oxidative stress, the cause of many chronic diseases that cause cell damage, which can promote physiological dysfunction and cell death (Cruz, 2008).

Ribeiro et al. (2011) reported the health benefits of assaí due to its high antioxidant capacity in its composition. Therefore, the

consumption of fruits rich in antioxidants has been increasing more and more in Brazil, as well as the consumption of jussaí, which already presents studies that assess its antioxidant characteristics.

Currently, assaí has been indicated due to its attractive characteristics as lipids, phenolic compounds and anthocyanins, which are substances associated with a high antioxidant capacity.

Therefore, for this reason, jussaí becomes a functional food that has been gaining importance regarding its beneficial effects on health (Schultz, 2008).

On the other hand, minerals that make up the jussaí, the study by Ribeiro et al. (2011) revealed the results expressed in Table 3.

Schultz (2008) compared the antioxidant activity of assaí and jussaí finding the highest values of phenolic compounds (81%), anthocyanins (353%) and antioxidant activity (TEAC) in jussaí (Table 4) confirming the results reported by Iaderoza et al. (1992) who found greater amounts of anthocyanins in *E. edulis* fruits than in *E. oleracea*.

2.2 Functional foods

According to Pennington (2002), are foods that, in addition to providing health benefits, help to reduce risks of diseases, and known as functional foods. They can be delaying, prophylactic or auxiliary in the treatment of cardiovascular diseases, diabetes, osteoporosis, immunological diseases and even neoplasms. The beneficial components of functional foods have been termed as phytochemicals, functional compounds, or bioactive components, and are found in approximately one hundred and twenty foods.

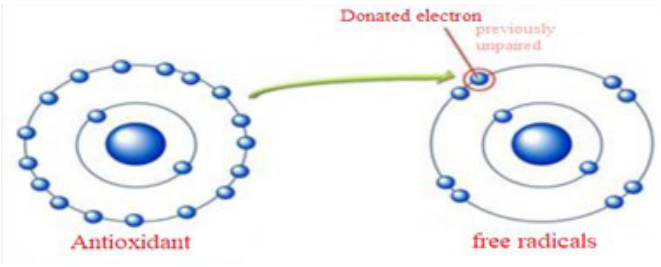

Figure 3. Antioxidant mechanism against free radicals.

Table 3. Mineral composition of jussai pulp (*E. edulis*).

Minerals	Jussaí Pulp (mg. 100 g^{-1})
Sodium	19.3 (± 6.0)
Potassium	94.8 (± 11.2)
Calcium	4.3 (± 1.0)
Iron	46.6 (± 1.5)
Phosphorus	5.2 (± 1.0)

Source: Ribeiro et al. (2011)

2.3 Bioactive compounds

Bioactive compounds (BA) are extranutritional compounds naturally present in small amounts in food and, when ingested in significant amounts, exert beneficial effects on human health (Horst et al., 2016).

Bioactive compounds have molecular structures, whose chain may have one or more functional groups, which are responsible for the therapeutic effect of natural products and organized into different groups according to their chemical similarity. They can be classified into terpenes, triterpenes, tannins, saponins, flavonoids, alkaloids, among others (Carvalho et al., 2002).

They exist in nature in large numbers, with wide variation in chemical structure, The BA are divided into several classes, with polyphenols, carotenoids and glucosinolates being the three main large groups of BA present in the usual human diet (Horst et al., 2016).

Some of them have antioxidant capacity and are associated with the protection of human health against chronic non-communicable diseases (NCDs), such as diabetes mellitus, cancer, cardiovascular diseases and neurodegenerative diseases. These diseases have their high incidence and prevalence justified by the growth of four main risk factors, which are smoking, harmful use of alcohol, physical inactivity and consumption of an unhealthy diet (Si & Liu, 2014).

The antioxidant capacity of foods varies according to their levels of vitamin C, vitamin E, carotenoids, flavonoids, anthocyanins, anthocyanidins and other phenolic compounds (Saura-Calixto & Goñi, 2006).

The jussaí from the Atlantic Forest (*E. edulis*) is rich in bioactive compounds known as anthocyanidins, and two of them are the most important, 3-O-glucoside cyanidin and 3-O-rutinoside cyanidin with high antioxidant capacity (Figure 4), representing 36, 88% and 63.12%, respectively, of the total anthocyanins (Novello, 2011).

Within the genus *Euterpe*, the fruit of *E. edulis* stands out, as it has a quantity of anthocyanins four-folder higher than *E. oleracea* (Schultz, 2008) as well as higher phenolic compounds, anthocyanins contents and antioxidant activity.

According to Iaderoza et al. (1992) and MacFadden (2005) the anthocyanin content in fresh pulps of assaí *(E. oleracea)* fruits is 336 mg.100 g^{-1} and jussaí (*E. edulis*) is 1,347 mg.100 g^{-1}. On Table 5, Ribeiro et al. (2011) reported the values found for

Table 4. Phenolic compounds, anthocyanins and antioxidant activity of *E. edulis* and *E. oleracea* (*in natura*).

Species	Dry Matter (%)	Phenolic compounds**	Phenolic compounds **	Anthocyanins *	Anthocyanins **	TEAC (µmol. g^{-1})
***E. edulis* (n=8)**	10.1	398.6 ^{a}	40.2 ^{a}	58.5 ^{a}	5.3 ^{a}	13.6 ^{a}
***E. oleracea* (n=3)**	12.0	267.3 ^{b}	22.2 ^{b}	18.4 ^{a}	1.5 ^{b}	9.2 ^{b}

*(mg.100 $^{-1}$); **(mg. g $^{-1}$ dry basis). Souce: Schultz (2008).

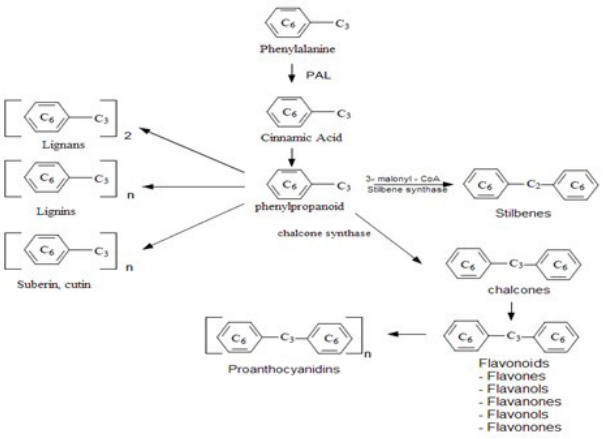

Figure 4. Molecular structure of the anthocyanidins or aglycons. Source: Novello (2011).

Table 5. Anthocyanidins (cyanidin-3-o-glucoside) mg/100 g samples of jussaí and assaí.

Pulp	Anthocyanins [a]	Monomeric Anthocyanins [b]	Anthocyanins [c]
Jussaí	235.8 ± 2.5	183.7 ± 2.5	148.63 ± 0.04
Assaí	32.32 ± 0.27	20.89 ± 0.36	60.61 ± 2.80

Source: Ribeiro et al. (2011).

anthocyanidins (cyanidin-3-o-glucoside) mg/100 g in samples of jussaí and assaí.

Borges et al. (2011) demonstrated that when obtaining five samples of assaí, from palm trees of the Euterpe edulis species, from five different regions of Santa Catarina, and evaluate their anthocyanins, they found: 72.50 ± 3.1; 14.84 ± 2.11; 207.94 ± 1.68; 79.80 ± 3.19; 409.85 ± 2.33 mg.100g-1 of fruit. These values differ between the same fruit from different regions. The extraction using methanol is the most efficient for anthocyanins. However, our objective was to seek applicability for the food industry and for pharmacotherapy, which makes the use of methanol unfeasible due to its high toxicity. Silva et al. (2013) obtained data that showed no difference for the extraction with methanol and ethanol as solvents.

Constant (2003), used 70° ethanol with HCl up to pH 2.0 in the extraction of anthocyanins in *Euterpe oleraceae*, finding an average of 103.24 (± 3.34) mg.100mL^{-1} in the pulp. Ozela et al. (1999) obtained 926.1 (crop) mg and 356.7 (off-season) mg.100g-1. Bobbio et al. (2000), obtained 50 mg of anthocyanin.100 g $^{-1}$ of the assaí fruit, equivalent to 263 mg.100 g -1 of pulp. On the other hand, Iaderoza et al. (1992), found 336 mg. 100 g-1 of assaí fruits.

Rogez (2000), on the other hand, found 44 mg of anthocyanin per 100 g of assaí fruit. The variation in these values can also be attributed to the inherent variability of the fruit.

Hassimotto et al. (2008) reported that t even small amounts absorbed by *Wistar* rats are enough to increase the antioxidant capacity in plasma. This result offers a promising perspective for the inclusion of these bioactive compounds in the human diet.

2.4 Phenolic compounds

Phenolic compounds are secondary metabolites that are synthesized by plants during natural development and in response to stress conditions. In plants, the compounds can act as phytoalexins, attractants to pollinators, as contributors to plant pigmentation, as antioxidants, protective agents against ultraviolet (UV) light among others.

In food, phenolics have characteristic of bitterness, astringency, color, flavor, odor and oxidative stability of products (Naczk & Shahidi, 2004).

These compounds occur ubiquitously in plants and are metabolic derivatives of phenylalanine and tyrosine. However, they are not uniformly distributed in plants, showing variations in the classes and subclasses (Cruz, 2008; Naczk & Shahidi, 2004).

The phenolic compounds that occur in food belong to the phenylpropanoid family and are derived from cinnamic acid. Phenylalanine ammonia-lyase (PAL) is the enzyme responsible for the metabolic pathway of phenylpropanoids, converting phenylalanine into cinnamic acid (Figure 5), and succeeding the biosynthesis of phenolics (Cruz, 2008).

Phenolic compounds present themselves as simple molecules even with a high degree of polymerization, having at least one phenol-a hydroxyl group in an aromatic ring, present in vegetables in free form or linked to sugars (glycosides) and proteins (Schultz, 2008).

In plants, simple phenols include phenolic acids (derived from benzoic and cinnamic acids), coumarins, flavonoids, stilbenes, condensed and hydrolysable tannins, lignans and lignins (Naczk & Shahidi, 2004) and are responsible for promoting the removal or inactivation of free radicals formed during the initiation or propagation of the oxidative process reaction through the donation of hydrogen atoms (Dossiê Antioxidantes, 2016). Thus, interrupting the chain reaction and the intermediates formed by the action of phenolic antioxidants are stable, due to the resonance of the aromatic ring present in the structure of these substances (Rocha et al., 2007).

There is a growing interest in natural antioxidants derived from plant extracts, with characteristics of secondary metabolites, as well as phenolic compounds due to their antioxidant activity.

2.5 Anthocyanins

Anthocyanins (from the Greek "anthos" meaning flower and "kianos" meaning blue) are important plant pigments, they belong to the class of phenolic compounds known as flavonoids (Kong et al., 2003). They belong to the category of secondary metabolites, present in fruits with a color spectrum that varies from red to dark blue, dark vegetables and dark grape skins (Downham & Collins, 2000). Assaí fruits have a characteristic intense purple coloration due to the high concentration of anthocyanins (Fregonesi et al., 2010).

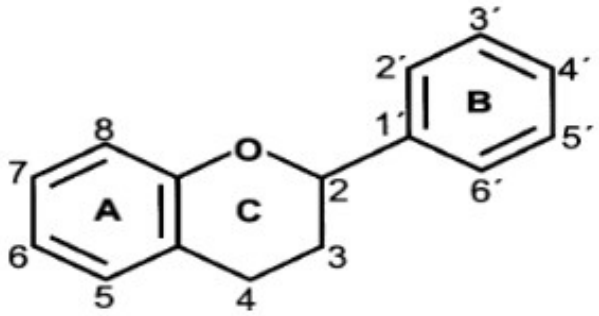

Figure 5. Flavylium (2-phenylbenzopyrylium cation). Source: Bravo (1998) apud Cruz (2008).

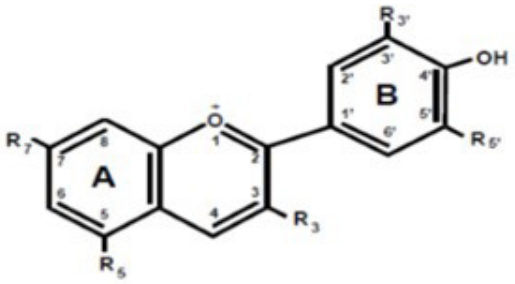

Figure 6. Metabolism of phenolic compounds in plants. Source: Cruz (2008).

Anthocyanins are flavonoid pigments belonging to the class of phenolic compounds, due to their carbon skeleton $C_6C_3C_6$ characteristics, characterized by the flavylium basic nucleus (2-phenylbenzopyrylium cation) composed of two aromatic rings joined by a three-carbon unit and condensed by an oxygen (Schultz, 2008) (Figure 6).

As they are polyphenolic compounds in the flavonoid category, they have a high redox potential, which means that they have a great capacity to donate a hydrogen to highly reactive radicals, extinguishing singlet oxygen, preventing the formation of new free radicals and are metal chelators. Its antioxidant potential depends on the number of hydroxyl groups, on the arrangements of the extension of the conjugation structure that act as electron donors and on substituents that remove electrons from the ring structure so that the formed compound has stability (Ignat et al., 2011).

In addition to their antioxidant activity, anthocyanins play an important role for plants, which is the coloring capacity of their plant products. In this way, the fruits are more attractive to pollinating animals and that facilitate the dispersion of their seeds. Together with other flavonoids, anthocyanins act as a resistance mechanism for plants against pest attacks, acting as antioxidants, phytoalexins or as antibacterial agents. The phenolic structure of anthocyanins confers antioxidant activity through the donation or transfer of electrons from hydrogen atoms. Several studies have shown the health effects due to its antioxidant properties (Martinez-Flores et al., 2002; Kuskoski et al., 2004, 2005).

The cells and tissues of the human body are constantly under attack due to the action of free radicals and reactive oxygen species (ROS), which we produce during normal oxygen metabolism or which are caused by exogenous stimuli such as ingestion of potentially inflammatory foods, stress, excessive physical activities, among others. When the production of free radicals increases and the physiological capacity that relies on the enzymes superoxide dismutase, glutathione peroxidase, catalase and antioxidant compounds such as ascorbic acid, tocopherols, and carotenoids cannot keep up with this growth, an imbalance occurs in this balance and begins to a harmful environment to various cellular structures, favoring attacks on lipids, proteins and DNA. With all these attacks, the structural and functional integrity of cell membranes, enzymes and genetic material are destabilized, facilitating the emergence of diseases that benefit from this situation, such as cancer, diabetes, atherosclerosis, among others (Tsuda et al., 1999; Prior, 2003).

Studies suggest that ingesting natural antioxidants increases plasma antioxidant efficiency and reduces the risk of some cancers, heart disease and stroke. (Martinez-Flores et al., 2002; Prior, 2003; Galvano et al., 2007). Other authors also corroborate these studies such as Silva et al. (2013, 2014) who concluded that the fruits of the jussara palm have several beneficial properties to health associated with the presence of bioactive compounds, mainly anthocyanins, which are natural pigments present in vegetables, being widely distributed in nature.

Anthocyanins are glycosides of polyhydroxy and polymethoxy derivatives of 2-phenylbenzopyrilium or flavyl ion. They are components of the flavonoid group. Are polar molecules, as a function of the polar hydroxyl, hydrogen, methoxyl and glycosyl groups linked to the aromatic rings, therefore, they are soluble in water. In nature, they are associated with sugar molecules and when free of these sugars they are called anthocyanidins or aglycones, which are rarely found in fresh plant material. This free form is chemically more unstable than the heterosidic one. There are more than 17 types of aglycones in nature, but only six of them have a more frequent distribution: cyanidin, delphinidin, petunidin, peonidin, pelargonidin and malvidin (Wu et al., 2006).

The most common sugars present in anthocyanins are glucose, galactose, rhamnose, xylose and arabinose. The number of sugars in the structure can vary, as can their binding position on the molecule. Therefore, the primary occurrence in nature of anthocyanins is in the glycosidic or acylglycosidic form with the respective anthocyanidin aglycone (Zuanazzi & Montanha, 2003).

Anthocyanins are reactive compounds due to their ionic state, also showing a great sensitivity to changes in temperature and pH, normally stable at pH values between 1.0 and 4.0, being in general degraded at pH above 7,0. In addition to the pH and chemical structure of the molecule, there are other factors that influence the stability of anthocyanins, such as temperature, oxygen, light, solvents, concentration, ascorbic acid, sulfur dioxide, presence of enzymes such as peroxidase, polyphenoloxidase and glycosidase, metal ions, sugars, proteins, mineral salts and other flavonoids (Rein, 2005; Schwartz et al., 2008). In foods, it

is also observed that processing and storage interfere with the stability of these compounds.

These substances are effective hydrogen donors. Its antioxidant potential is regulated by differences in chemical structure. By varying the position and chemical groups on aromatic rings, the ability to receive unpaired electrons from free radical molecules is also variable (Prior, 2003). The antioxidant capacity of anthocyanins is also correlated with the quantity and location of the hydroxyl groups and their conjugation, as well as the presence of donor electrons in the structure's ring, due to the capacity of the aromatic group to support electron mismatch (Kuskoski et al., 2004).

Despite the benefits to health, the low stability of bioactive compounds makes it difficult to process the fruits of the juçara palm, which is a bottleneck for its industrial use. Another difficulty encountered in using the fruits is due to their seasonality and high production in a short period of time, making it necessary to store them under freezing or refrigeration (Pereira, 2015).

3 Methods of determination of antioxidant activity

3.1 Iron Reduction Method (FRAP)

The FRAP (Ferric Reducing Antioxidant Power) method is based on electron transfer reactions. According to Pulido et al. (2000), this methodology was developed as an alternative for the analysis of biological fluids and aqueous solutions of pure compounds and can be applied both for studies of antioxidant activity in foods and beverages, as well as in pure substances, with results comparable to those obtained with other methodologies.

The reaction occurs through the formation of a TPTZ (2,4,6-Tris(2-pyridyl)-S-triazine) complex with Fe (III), yellowish in color. In the presence of an antioxidant, the iron present is reduced, giving rise to $[Fe(II)(TPTZ)_2]^{3+}$, which is dark in color. The reaction takes place at pH 3.6 and the maximum absorbance is 593 nm. pH values have an important effect in reducing the capacity of antioxidants. In acidic conditions, the reduction in capacity can be suppressed due to protonation with antioxidant compounds, while in basic media, dissociation of protons from phenolic compounds occurs, which can increase the capacity to reduce a sample (Huang et al., 2005).

3.2 ABTS

The indirect method of ABTS 2,2-azinobis (3-ethylbenzothiazoline-6-sulfonic acid) radical scavenging is also widely used and was first suggested by Miller et al. (1993) in testing biological samples. Like DPPH*, $ABTS^{*+}$ has excellent stability under certain analysis conditions. However, these radicals have some important differences. The DPPH* radical is ready-to-use and is soluble in organic solvents, while the $ABTS^{*+}$ needs to be generated first by chemical (such as potassium persulfate) or enzymatic reactions, and is soluble in both water and organic solvents, allowing the analysis of both hydrophilic and lipophilic samples (Arnao, 2000).

One of the method's advantages is its relative simplicity, which allows its application in routine laboratory analyses. Furthermore, it offers several absorption maxima and good solubility (Roginsky & Lissi, 2005; Kuskoski et al., 2005).

3.3 DPPH

The DPPH (2,2-diphenyl-1-picrylhydrazine) free radical scavenging method was first suggested in the mid-1950s, originally to discover hydrogen donors in natural products and later to determine the antioxidant potential in individual phenolics and foods (Roginsky & Lissi, 2005). Currently, it is one of the most used methods to evaluate the antioxidant activity.

The absorbance decay found by the remaining amount of DPPH radical varies significantly with different types and concentrations of antioxidants. Currently, the EC50 technique is widely used, which was proposed to determine the anti-radical efficiency through the amount of antioxidant needed to drop by 50% the initial concentration of the DPPH radical and the time needed to achieve stability in its concentration (Sanchez-Moreno et al., 1998). This methodology is limited by the fact that DPPH radicals interact with other radicals (Alkyl), and the time response curve to reach steady state is not linear, with different antioxidant/DPPH ratios*

3.4 ORAC

The ORAC method, different from DPPH and ABTS, is based on hydrogen atom transfer reactions (Pazinatto, 2008), being more relevant because it uses a source of radicals that are more similar to the biological system.

The following constituents participate in the ORAC test reaction: peroxyl radicals; compound AAPH [2,2′-azobis(2-methylpropionamidine) dihydrochloride] that promotes the generation of peroxyls in the reaction through their spontaneous decomposition above a certain temperature; and fluorescein, which is a photostable and thermostable fluorescent marker, which has its fluorescence reduced by peroxyl (Ribeiro et al., 2011) by having its chemical structure degraded (Pereira, 2009).

Radical thermal generators lead to constant fluxes of peroxyl radicals in saturated air solutions, causing competition between the antioxidant and the oxidant for the radicals with inhibition or delay of the oxidant substrate (Pazinatto, 2008).

The antioxidant, when donating hydrogen atoms, inhibits the loss of fluorescence intensity, and the inhibition is proportional to the antioxidant activity. Although the standard used to construct the curve is Trolox, unlike DPPH and ABTS, the decay does not follow a first-order kinetics (linear with time), but a second-order kinetics with time, so the quantification is performed by through the technique of the area under the decay curve (AUC) (Pereira, 2009; Ribeiro et al., 2011).

Like ABTS, ORAC can be applied to determine the antioxidant capacity of lipophilic and hydrophilic compounds. When compared to other methods, ORAC has the advantages of suffering less interference from colored compounds present in samples such as fruits and vegetables, as it is based on fluorescence rather than absorbance. Furthermore, the assay reflects more the real conditions of the biological system by use pro-oxidant peroxyl

or hydroxyl radicals (Pereira, 2009), abundant radicals and those most responsible for oxidative damage (Ribeiro et al., 2011).

3.5 Total phenolic compounds

The total content of phenolic compounds is determined by spectrophotometry using the Folin-Ciocalteu method. The method is based on the reduction of phosphotungstic (H3PW12O40) and phosphomolybdic ($H_3PMo_{12}O_{40}$) acids, present in the Folin-Ciocalteu reagent, with the phenolics present in the sample when reacting, forming tungsten oxide (W_8O_{23}) and molybdenum oxide (Mo_8O_{23}) in an alkaline medium and showing phosphotungstic blue. The phosphotungstic blue formed is quantified by the absorbance of the solution in the visible region (760 nm), which is proportional to the number of aromatic phenolic groups and compared with the quantification of gallic acid, used as the standard. Thus, when applying gallic acid to the calibration curve, it is possible to correlate the color intensity with the concentration of phenols present in the sample, representing the result in gallic acid equivalent (GAE) (Cruz, 2008).

3.6 Total anthocyanins

One of the methodologies for the quantification of total anthocyanins is the pH difference performance which evaluates the quantification of total and monomeric anthocyanins as a function of the differentiated spectral behavior of monomeric compared to polymeric in two buffer systems: chloride potassium pH 1.0 (0.025M) and sodium acetate pH 4.5 (0.4M) (Cruz, 2008).

Anthocyanin pigments undergo reversible structural transformations with the change in pH by absorbance spectra. Anthocyanins are color-enhanced at pH 1.0 and have no absorbance at 700 nm, in which only interfering degradation compounds are absorbed. However, the difference between the reading found at λmax and at 700 nm of the pH 1.0 solution is proportional to the concentration of total anthocyanins. While at pH 4.5 only the monomeric anthocyanins will be present, which is colorless. Hence, the difference between the readings between the two wavelengths at pH 1.0 is subtracted by the difference between the readings at the two wavelengths at pH 4.5, resulting in monomeric anthocyanins (Cruz, 2008).

3.7 Anthocyanidins

High Performance Liquid Chromatography (HPLC) is the newest and most important member of a family of separation techniques (Collins et al., 1993). HPLC is currently widely used, both for separation and quantification of phenolic compounds. Various supports and mobile phases are available for analysis of anthocyanins, pro-anthocyanins, flavonones, flavonols, flavones and phenolic acids. The development of reversed-phase columns has significantly contributed to the successful use of HPLC in the separation of different classes of phenolic compounds (Shahidi & Naczk, 1995).

This technique was pioneered in anthocyanin analysis and spread with the advent of the reverse phase column, when it became widely used since then. The most widely used support is silica chemically bonded to octyl and octadecyl groups (Strack & Wray, 1989).

Reversed phase liquid chromatography is, of course, the most widely used method for separating anthocyanins. Since different methods of analysis have been developed for a specific need, it is impossible to describe a simple standard procedure. However, certain conditions are commonly followed. Detection is normally carried out using a diode beam in the UV/Visible region, which allows obtaining the full absorption spectrum of the extract "on-line". The UV/Visible spectrum can give information about the nature of the aglycone, type and number of sugars and possible acylation (Hong & Wrolstad, 1990 cited by Costa et al., 2000). Simple UV/Visible detectors can selectively determine anthocyanins in the region of maximum visible absorption, between 520 nm and 546 nm, where no other phenolic compound has absorption. C18 columns are the most used, but the use of polystyrene columns has also been reported.

The separation of several phenolic compounds, with similar structures, constituents of the anthocyanin extract is more effective when using gradient elution with methanol and/or acetonitrile acidified with formic, acetic or trifluoroacetic acid (Costa et al., 2000).

The polarity of anthocyanidins is perhaps the factor that most influences the retention time in the column. Usually the order of elution is initially delphinidin, followed by cyanidin, petunidin, pelargonidin, peonidin, and finally malvidin. The retention, therefore, decreases with increasing polarity which, in the present case, means an increase in the number of hydroxyl groups in the flavillium nucleus. The presence of sugars increases the retention of anthocyanins, with diglycosides generally eluting before monoglycosides. Acylation also increases the retention time of anthocyanins when compared to their non-acylated counterparts (Costa et al., 2000).

HPLC has been widely used for the qualitative analysis of anthocyanins and several methodologies have been applied. Normally, to characterize the anthocyanin composition, it is customary to use patterns obtained from sources whose composition is already known and compare their spectrum and retention times with the results obtained from the extract of the new source chromatographed under identical conditions. Furthermore, it is customary to employ other chromatographic techniques in conjunction with HPLC, either for pre-purification, for the preparation and isolation of standards, or to confirm the identity of certain anthocyanins by means of the respective Rf. HPLC is hardly used as the only technique in the analysis of anthocyanins, either qualitatively or quantitatively (Gao & Mazza, 1995; Andersen, 1987; Bakker & Timberlake, 1997).

For the purpose of determining the chemical structure of unexamined anthocyanin fractions, it is often necessary to use HPLC in conjunction with mass spectrometry and nuclear magnetic resonance spectroscopy, since the use of chromatographic techniques and specific chemical reactions is not sufficient for identify more complex structures. Some authors cite the use of such techniques together (Cameira-dos-Santos et al., 1996; Torskangerpoll et al., 1999; Slimestad & Andersen, 1998). It is observed that the existing methodologies for HPLC are varied and

that they fundamentally depend on the case studied. Normally, when evaluating a new extract, some existing methodology for a similar situation is tested, making the necessary adaptations, such as changing the elution gradient, flow, solvent system, among others.

Fiorini (1995) used preparative HPLC for purification of anthocyanins from extracts obtained from strawberry, elderberry, eggplant and radish. The chromatographic conditions were optimized for each case, in order to achieve efficient purifications in less time. Two Spherisorb columns (reverse phase) of different diameters were used for analytical and preparative HPLC. The authors concluded that preparative HPLC can be an efficient method to isolate pure anthocyanins, from monoglycosides to more complex structures such as acylated di- and tri-glycosides.

3.8 Processing

Freezing is characterized by a reduction in the temperature of the food, with the formation of ice crystals, increasing its shelf life by reducing water activity. When freezing, the temperature is kept below -18 ºC, which allows the product to be preserved for months or years. Cooling, on the other hand, corresponds to a reduction in the temperature of the food between -1 and 8 °C, in order to reduce enzymatic and microbiological activities, thus prolonging its shelf life for days or weeks. Both conservation methods can be used by jussara processing industries. In Figure 7 it can be observed the flowchart of the jussaí process.

3.9 Products

From the pulp of jussaí, several products can be prepared in the form of food supplements, such as lyophilized form or nanoemulsions where their applications in industry can change macroscopic characteristics such as texture, flavor, other sensory attributes, color strength, processing and stability in sheflife (Quintanilla-Carvajal et al., 2010), among other products with high antioxidant activity.

Figure 7. Processing flowchart of jussaí pulp. Source: MacFadden (2005).

4 Conclusions

Being used as part of the diet by different consumers in Brazil, and exported to other countries, the jussaí have high antioxidante activit. Promoting nutritional benefits, for its functional characteristics preventing many kinds of diseases. In addition, the jusara palm is industrialized as "palmito pupunha", the heart of palm with high comercial value. On the other hand, many kinds of jussaí supplements can be obtained by different processes as funcional products including in the dried form and nanemulsions as well.

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