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Article

Bioactive compounds in new food products from Amazonic fruits

Juliana E. C. Cardona [1], Marcela P. Carrillo[1], Mary Lares [2], Rafael H. Gutiérrez [2], María S. Hernández [1,*], J.Pablo Fernández-Trujillo [3]

[1] Instituto Amazónico de Investigaciones Científicas Sinchi, Avenida Vásquez Cobo 14-15, Leticia, Amazonas, Colombia; E-Mail: shernandez@sinchi.org.co

[2] Universidad Central de Venezuela, Escuela de Nutrición y Dietética, 3 floor Residencias Femeninas, Ciudad Universitaria, Caracas, Venezuela; E-Mails: marylares@hotmail.com, rafgutii@hotmail.com

[3] Universidad Politécnica de Cartagena (UPCT). Department of Agricultural and Food Engineering. Paseo Alfonso XIII, 48. ETSIA. E-30203 Cartagena (Murcia), Spain; E-Mail: juanp.fdez@upct.es

***** Author to whom correspondence should be addressed; E-Mail: shernandez@sinchi.org.co; Tel.+57-01-4442060 (ext 186)

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Abstract: Bioactive compound are extracted directly from plants, animals or microorganisms through physical, chemical or enzymatic processes, acceptable for clean process. Amazonic bioresources are abundant and contain a great amount of bioactive compounds, such as antioxidants, polyphenols and many others. Amazonic extracts from asai (*Euterpe precatoria*), copoazu (*Theobroma grandiflorum*) and cocona (*Solanum sessiliflorum*) were characterized in total polyphenols and antioxidant capacity as well as ascorbic, oxalic, citric, succinic and malic acid contents and theobromine, caffeine, epicatechin and catechin gallate, glucose, fructose and sucrose in order to preserve the nutrient and bioactive compounds. Some processing techniques were evaluated: spray drying and convective dehydration. Spry drying allows encapsulation of active ingredients and therefore effective protection of extracted metabolites compared with convective dehydration. Powders were used to evaluate some functional beverages and gums candies. Both beverages and candies showed higher antioxidant capacity when they were made from spray drying extract, confirming the success in preservation of bioactive compounds.

Keywords: antioxidant capacity; asai; beverage; bioactive ingredients; cocona; copoazu; food processing; tropical fruit

1. Introduction

The interest on functional food and cosmetics increase gradually because their bioactive compounds could relieve new and strange diseases [1-2].

Amazonian region is one of the most recognized sources of natural ingredients and bioactive compounds which have allowed its use as an additive or raw material in the formulation of standard products with certain functional properties.

Drinks have been commonly used to distribute functional ingredients in high concentrations, sports drinks or infusions, water with added vitamins, soy beverages and energy drinks are common examples [3]. In the case of gum candies are less distributed in the market, but correspond to a mass product.

The goal of this paper is to report the nutritive and bioactive quality of the ingredients obtained by spry-drying and convective dehydration from three different tropical Amazonic fruit. The antioxidant capacity in functional beverages and gum candies after powder addition was also evaluated.

2. Experimental Section

2.1. Convection dehydration

Pulps of cocona (*Solanum sessiliflorum*), asai (*Euterpe precatoria)* and copoazu (*Theobroma grandiflorum*) were obtained in the Amazon crop or wild forest. Pulp was dried by forced convection air at 55 ° C (COMEK ® dryer Colombia) with the addition of 33% of maltodextrin.

2.2. Spray drying

Asai was also subjected to spray-drying in laboratory scale equipment LabPlant ® SS-07A using an air inlet temperature of 200 °C, air outlet temperature of 190 °C, a flow power of 200 mL h-1 and a sprinkler nozzle of 1mm. Maltodextrin was also added in 33% of the total mixture.

Maltodextrin was added as adsorbent additive to achieve the increase vitreous transition temperature Tg and avoid defects agglomeration, aggregation and sedimentation at the time of reconstitution. Maltodextrin, additionally delays thermal and photo bioactive compounds degradation.

2.3. Organic acids and sugars

Sugars and organic acids were measured following the methods previously reported by Hernández *et al.* [4]

2.4. DPPH antioxidant capacity

Antioxidant capacity of natural ingredients was measured by measuring the radical scavenging properties of 2,2-diphenyl-1-picrylhydrazyl (DPPH) using the methodology reported by Rufino *et al.*

[5] for asai dry pulp. About 50 mg of each ingredient were subjected to extraction with 1 mL of a mixture methanol: water acidified (pH = 1) with constant stirring for one hour in vortex and after this time the sample was centrifuged at 15000 rpm for 15 min. The liquid phase was withdrawn and the solid phase was subjected to a second extraction with 1 mL of acetone: water with constant stirring for one hour in vortex thereupon the sample was centrifuged at 15000 rpm for 15 min. Both liquid phases were pooled and brought to a volume of 2 mL. The extraction was immediately stored at -18°C for the shortest time possible before spectrophotometric analysis.

Quantification was performed by mixed with 40 µL of sample, 1960 µL of methanol solution of DPPH (60 µM). The absorbance at 515 nm was recorded every 20 seconds while the reaction was conducted in the cell to achieve equilibrium. The absorbance at equilibrium was compared with the calibration curve of DPPH absorbance to calculate the amount of free radicals in the reaction. The absorbance at equilibrium was transformed into trolox equivalents by using a standard trolox calibration curve prepared from concentrations between 0.5 and 1 mM). Trolox was used as a reference antioxidant.

2.5. Total Polyphenols

Total polyphenols were measured by the method adapted from Ainsworth & Gillespie [6] and Cicco *et al.* [7]. A sample of 100 mg dry pulp was mixed with 1 mL 70% methanol using a vortex and extracted for 1 h with constant agitation. The mixture was centrifuged at 15000 rpm for 15 min and the liquid phase was removed for analysis. An aliquot of the extraction solution (100 µL) was mixed with 200 µL of Folin - Ciocalteu (diluted 1:10 in distilled water) for 2 min. Then 800 µL of Na_2CO_3 700 mM was added and the reaction was allowed to proceed in the dark and at room temperature for 2 hours. 100 µl were analyzed after that the reaction spectrophotometrically at 710 nm and absorbance was collated with the calibration curve obtained by the reaction of Folin-Ciocalteu with different concentrations of gallic acid (15 - 150 ppm). Results were expressed in grams of gallic acid equivalents (GAE) per 100 g of sample.

2.6. Alcaloids

To quantified theobromine, caffeine, epicatechin and catechin gallate, , a sample of 100 mg of each dry fruit ingredient was extracted with water (90°C) at constant mixing during 15 minutes, and then centrifugated (15000 rpm for 15 minutes). Supernatant was filtrated trough PTFE filter (0,45 µm), and analyzed by HPLC (Agilent Technologies ® 1200 series), with a Bio Rad RP318 column (Bio-Rad Laboratories®, California – USA) at 35°C, using DAD and UV-VIS detectors, and acetonitrile:formic acid (5% aqueous solution)(1:9) as liquid phase at 1mL/min flux. Metabolites were quantified trough comparing area and retention time of certified standards of each compound (Sigma – Aldrich ®).

2.7. Instant drink formulation

The powder mixture was formulated to be reconstituted from asaí ingredient obtained by spray drying as the main source of polyphenols in the instant drink, while copoazu ingredient was added as

source of sweetness, sourness and flavor besides of epicatechine, pulverized sucrose as edulcorant and trisodic phosphate as ant agglomerate.

2.8. Gum candies

Gums were formulated from asai ingredient, water, gelatin (260 bloom), sucrose, glucose, citric acid and sodium benzoate (Table 6).

3. Results and Discussion

The formulated powders from asai, copoazu and cocona ingredients contained 33% of maltodextrine that improved powder solubility (1:10 ingredient-water), preserved the bioactive compounds and prevented the agglomeration process during storage. Maltodextrine inclusions above 30% did not exhibit agglomeration avoid because vitreous transition temperature (T_g) of these powders (around 55°C; [1]) was not attained.

3.1. Organic acids and sugars

Citric acid contents in copoazú and cocona ingredients conferred acidity useful in beverages production as well as antioxidants with technological functionality. Asai ingredient did not exhibit any organic acid detectable content but its anthocyanin content makes it a very important bioactive source (Table 1).

The three carbohydrates in formulated asai, cocona and copoazu powders were sucrose, glucose and fructose (Table 2). Copoazu showed the highest carbohydrate contents among the three powders (Table 2). High sucrose and fructose content gives a remarkable sweetness and caloric power to the beverages prepared. This can be used in food products elaboration such as energy drinks and others as gum candies.

3.2. Alcaloids, polyphenols and DPPH antioxidant capacity

Purine alkaloid content was below detectable levels except caffeine and theobromine in cocona (0.97±0.05 and 0.138±0.14 mg/100 g pulp, respectively) and cannot be considered a significant source of such type of metabolites. On the other hand, catechin gallate was not detected and epicatechin exhibited remarkable levels in cocona (1.31±0.11 mg/100g dw) and copoazu (3.85 mg/100g dw). The former level in copoazu, is enough to consider it as a functional source to prevent cardiovascular disease and other illness due to the inhibition of LDL oxidation in the human physiology [8]

Dry powder obtained from asai pulp showed the highest total polyphenol content compared with cocona and/or copoazu (Table 2). By comparing our results with those obtained by Schauss *et al.* [9] and Bichara & Rogez [10], our *Euterpe precatoria* powder ingredient exhibited higher amounts of total polyphenol or antioxidant capacity (Tables 2 and 3) compared to those obtained in fresh fruits of *Euterpe oleraceae* that have been harvested in Brazil.

3.2. Instant drink and gum candies

Asai and copoazu dry powder ingredient were processed by convection drying and spray drying technique. Spray drying allows the encapsulation of the bioactive compounds and lead to a homogeny powder with a reduced particle size and a high antioxidant capacity [11] (Table 4).

Sensory tests performed showed the consumer acceptance to fruit flavor in a reconstitution in of the powder between 13% and 15% of the beverage, which corresponds to 30 g per 200 mL (one portion) of the beverage (Table 5). Asai gum candies exhibited also good texture and attractive flavor in the 23% of the product.

4. Conclusions

Nutritional value and stability in Amazon ingredients is very remarkable. Although spray drying encapsulated and preserved bioactive compounds (particularly anthocyanins) of asaí fruit, convection drying is also a viable strategy to produce functional ingredients from neglected fruits..

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Conflict of Interest

The authors declare no conflict of interest.

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TABLES AND FIGURES

Table 1. Organic acids content (mean±SE, n=3; all in mg/100 g of dry weight) in cocona and copoazu powders. Nd = non detectable amounts.

Fruit powder	Oxalic	Citric	Malic	Succinic
Cocona	41.6 ± 2.7	6579.2 ± 42.1	164 ± 11	356 ± 23
Copoazu	nd	8770.8 ± 17.3	537 ± 10	933 ± 41

Table 2. Carbohydrate and total polyphenol contents (mean±SE, n=3; in mg/100 g of dry weight) in asai, cocona and copoazu powders obtained by convective dehydration.

Fruit powder	Sucrose	Glucose	Fructose	Total polyphenols
Asai	3438 ± 47	12202 ± 15	411 ± 43	876± 10
Cocona	14617 ± 93	4541 ± 29	2631 ± 17	54±11
Copoazu	22013 ± 35	12024 ± 27	11156 ± 27	32±4

Table 3. Antioxidant capacity (mean±SE, n=3) in asai, cocona and copoazu powders.

Fruit powder	**Antioxidant capacity** (g trolox / g powder)	**DPPH antioxidant capacity** (g DPPH / g powder)
Asai	0.499 ± 0.03	1.958 ± 0.10
Cocona	0.044 ± 0.00	0.176 ± 0.01
Copoazu	0.046 ± 0.004	0.181 ± 0.02

Tabla 4. Antioxidant capacity (mean±SE, n=3) in asai and copoazú beverages elaborated with fruit powders.

Fruit and ingredient process of the beverage	**Antioxidant capacity** (g trolox / g sample)	**DPPH antioxidant capacity** (g DPPH / g sample)
Asai – Convection drying – No filter	0.5± 0.03	2 ± 0.1
Asai - Convection drying– filter	0.1 ± 0.01	0.3± 0.02
Asai – Spray drying– Filter	0.01 ± 0.0	0.01±0.0
Copoazu – convection drying – No filter	0.05± 0.0	0.2 ± 0.0

Table 5. Instant drink balance matter from asai and /or copoazu powder ingredient base.

Component	**Dry ingredients**	**Reconstituted Product**	**Equivalent in one portion (200 mL) of the beverage**
Asai ingredient	65%	15%	30 g
Copoazu ingredient	22%	5%	10 g
Sucrose powder	13%	3%	6 g
Trisodium phosphate	1%	0.2%	0.4 g
Drinkable water		76.8%	153.6 g

Table 6. Candies gums balance matter from asai powder ingredient base.

Ingredient	**Ingredient content % (w/w)**
Gelatine	7%
Water	34%
Powder	23%
Sucrose	23%
Glucose	11%
Starch	1%
Citric acid	1%
Sodium benzoate	1000 ppm