Agrociencia

EXTRACTION OF SWEET POTATO (*Ipomoea batatas* L.) ANTHOCYANINS AND THEIR APPLICATION AS A NATURAL DYE IN YOGURT

Oscar **Reyes-Morales**[1], Keisy **Peralta-Matute**[1], Beatriz **Guerrero-León**[2], Juan **Maita-Chamba**[3], Dani **Ochoa-Cervantez**[4], Percy **García**[5], Jhunior Abrahan **Marcía-Fuentes**[1*]

[1]Universidad Nacional de Agricultura. Facultad de Ciencias Tecnológicas. Catacamas, Olancho, Honduras. C. P. 16101.
[2]Universidad Nacional de Loja. Centro de Investigación, Desarrollo e Innovación en Nutrición Animal (CIDiNA). Loja, Ecuador. C. P. 110101.
[3]Universidad Nacional de Loja. Centro de Investigaciones Tropicales del Ambiente y Biodiversidad. Loja, Ecuador. C. P. 110101.
[4]Universidad Nacional de Ciencias Forestales. Siguatepeque, Comayagua, Honduras.
[5]Fortalece Consultants Inc.
* Author for correspondence: jmarcia@unag.edu.hn

Citation: Reyes-Morales O, Guerrero-León, B, Maita-Chamba J, Ochoa-Cervantez D, García P, Marcía-Fuentes JA. 2025. Extraction of sweet potato (*Ipomoea batatas* L.) anthocyanins and their application as a natural dye in yogurt. **Agrociencia.** https://doi.org/ 10.47163/agrociencia.v59i8.3424

Editor in Chief:
Dr. Fernando C. Gómez Merino

Received: February 04, 2025.
Approved: November 14, 2025.
Published in Agrociencia: November 25, 2025.

ABSTRACT

The aim of this investigation was to obtain a natural dye by ultrasound-assisted extraction from purple sweet potato (*Ipomoea batatas* L.) for its application in yogurt. The extraction parameters were optimized using a Box-Behnken design and response surface methodology in 15 treatments. Temperature, ethanol concentration, and sonication time were the variables studied in the extraction process, with anthocyanin concentration serving as the response variable, which was determined using the differential pH method. Treatment T11 was identified as the best treatment, with the following extraction conditions: a temperature of 60 °C, ethanol concentration of 60.16 %, and sonication time of 20 min. The concentration of anthocyanins fluctuated between 59.7 and 66.47 mg of cyanidin-3-glucoside L^{-1}. From T11, three yogurt samples were formulated with 0.6, 0.8, and 1 % of extract and a control without any addition (0 %). These samples were evaluated through sensory analysis with a hedonic scale of nine points with 50 panelists. The formulations with anthocyanins received favorable scores; the yogurt with 1 % dye stood out for having the highest acceptance in terms of color, although for the remaining attributes (flavor, aroma, and consistency), the yogurt with 0.8 % scored higher. In general, all samples with natural dye obtained an acceptable evaluation. Therefore, the extract can be used as a natural dye in yogurt without issue, making it a viable alternative to synthetic dyes and potentially leading to the development of functional foods due to its bioactive compound content.

Keywords: natural dye, ultrasound, sensory analysis.

INTRODUCTION

The growing interest in the intake of natural and healthy foods has driven many investigations for ingredients or bioactive compounds that benefit human health (Montero *et al*., 2020; Marcía-Fuentes *et al*., 2021; Maddela and García, 2021). In this context, the sweet potato (*Ipomoea batatas* L.) stands out for its high content of antioxidants such as anthocyanins and beta-carotene. The anthocyanins found in the skin of the purple sweet potato are known for their ability to be used as natural dyes, offering a healthy and sustainable alternative to the synthetic dyes used in the food industry (Coba-Carrera *et al*., 2019; Marcía-Fuentes *et al*., 2020; Saravia-Maldonado *et al*., 2020; Santos-Alemán *et al*., 2023a).

Anthocyanins have been shown in clinical and preclinical studies to have anti-inflammatory properties, reducing markers such as C-reactive protein, IL-6, or TNF-α, and improving endothelial function and vascular health. Additionally, they modulate the intestinal microbiota positively, stimulating beneficial bacteria such as *Bifidobacterium* and *Lactobacillus*, and favor the production of short-chain fatty acids, which are associated with better digestive and metabolic health (Fallah *et al*., 2020; Saini *et al*., 2024; Zhao *et al*., 2024).

Given the acknowledged health benefits associated with anthocyanins, the aim of this investigation was to extract anthocyanins from the purple sweet potato using ultrasound, optimizing the extraction parameters (temperature, time, and ethanol concentration) using a Box-Behnken design, for their subsequent application as a natural dye in yogurt and the evaluation of its sensory acceptability. It was proposed that the anthocyanin extract obtained with this technique can be used as a natural dye in yogurt, maintaining a favorable sensory acceptability. Its incorporation into yogurt not only provides natural color but can also improve its functional profile, favoring cardiovascular, immunological, and digestive health in consumers.

MATERIALS AND METHODS

The investigation was carried out between the months of February and September, 2024, in the facilities of the National University of Loja, in the province of Loja, Ecuador. The institution is located at an altitude of approximately 2060 m, with a mean annual temperature of 16 °C and an average rainfall of 1200 mm (INEC, 2023).

Sweet potato (*Ipomoea batatas* L.) of the purple variety was used as the raw material, which had previously been obtained from the Loja wholesale market from local crops. For the extraction of anthocyanins and the bromatological analyses, analytical grade reagents were used: ethanol (99 %, Hayman Kimia; Witham, UK), distilled water (Puritech; Guayaquil, Ecuador), citric acid (J.T. Baker; Phillipsburg, USA), sulfuric acid (Fisher Chemical; Fair Lawn, NJ, USA), hydrochloric acid (Fisher Chemical; Fair Lawn, NJ, USA), sodium hydroxide (Thermo Scientific; Illkirch, France), petroleum ether (Merck; Darmstadt, Germany), potassium chloride (ISOLAB; Eschau, Germany), sodium acetate (Merck; Darmstadt, Germany), and acetone (Merck; Darmstadt,

Germany). A mixed cross-sectional method was used, structured into four main phases, described below.

Sample preparing and conditioning (Phase I)

The raw material was selected manually, discarding roots with signs of rot or deterioration. Subsequently, it was washed with drinkable water and disinfected in a sodium hypochlorite-based solution (100 ppm, equivalent to 2.5 ml of 4 % commercial bleach in 1 L of water). The roots were carefully peeled with a surgical scalpel and a stainless-steel knife to extract the skin, avoiding the pulp, and obtaining a yield of approximately 154 g of skin for every 1000 g of purple sweet potato. The skins were weighed using a precision scale (Mettler Toledo ME204; Columbus, OH, USA) and lyophilized at -56 °C in a vacuum for 24 h (Labotec, model 01JLGGJ12; Midrand, South Africa). The lyophilized skins were ground using a two-speed blender with 700 W of power (Oster, model BLSTBESTE; Boca Raton, FL, USA) until a fine and homogenous powder was obtained. This powder was stored in sealed containers at room temperature in the dark until it was used.

Ultrasound-assisted extraction (Phase II)

Anthocyanins were extracted following the methodology proposed by Solórzano *et al*. (2023) and Mendoza *et al*. (2023) from the skin of purple sweet potato. A Box-Behnken design was implemented with three levels and three factors (temperature, time, and ethanol concentration) to optimize the extraction, following the response surface method (Mendoza *et al*., 2023). Every treatment had three repetitions, with a total of 45 experimental units. The independent variables were time of sonication, temperature, and solvent concentration. The dependent variable was the concentration of extracted anthocyanins.

For the extraction, 15 treatments were prepared with 0.25 g of homogenized sweet potato skin powder with 60 mL of ethanol solutions at 50, 60, and 70 %, acidified at 4 % with citric acid (J.T. Baker, Phillipsburg, NJ, USA). The samples were sonicated in an ultrasonic bath (MRC Laboratory Instruments; Harlow, UK) at a frequency of 40 kHz, at temperatures of 40, 50, and 60 °C, for periods of 20, 25, and 30 min. The extract obtained was filtered using 125 mm filter paper Whatman. No. 1 to remove residual solids.

The filtrates were concentrated using a rotavaporator (Yamato Scientific Co., Tokyo, Japan) equipped with a vacuum pump (Welch, modelo 2034, Mount Prospect, IL, EE. UU.) removing ethanol at a controlled temperature of 25-30 °C to prevent degradation of anthocyanins, following the procedure described by Zapata et al. (2014). During this process, the volume was reduced from 60 mL to approximately 5 mL. Subsequently, the extracts were centrifuged using a centrifuge (Unic, model C818, Dayton, NJ, USA) at 3400 rpm for 10 min. to promote sedimentation and clarification. Finally, the concentrates were stored at 4 °C for 24 h until application.

Proximal and bromatological analysis of the sweet potato

For proximal analyses, the roots of sweet potato in optimum conditions were washed, peeled, and split into skin and complete matrix. Both parts were dried at a controlled temperature and pulverized until a homogenous particle size was obtained. Bromatological analyses were carried out using the Thiex (2009) and Association of Official Analytical Chemists (AOAC, 2019) methods.

The analyses carried out included moisture (%) determined by kiln drying loss (Binder, model FD, Tuttlingen, Germany) at 95-100 °C to constant weight (AOAC 934.01), and dry matter (%) as a supplement to moisture; raw protein (%), quantified by Kjeldahl digestion with conversion factor 6.25 (AOAC 981.10); crude fat (%), by Soxhlet extraction with petroleum ether (AOAC 920.39); crude ash (%), by mufla incineration at 600 °C to white or grey mineral residue (AOAC 942.05). Raw fibre (%) was determined by acid and alkaline digestion according to Weende (AOAC 962.09), whereas neutral detergent fibre (NDF), acid detergent fibre (ADF) and acid detergent lignin (LDA) were determined using an ANKOM system (ANKOM Technology, Macedon, NY, USA)., with filter bags according to the standardised method of Van Soest et al. (1991), validated in AOAC (2019).

Total anthocyanins determination

Quantification was performed using the differential pH method (Marcía-Fuentes *et al*., 2021) from the corrected absorbances of each treatment in triplicate. Two buffer solutions were prepared: potassium chloride 0.025 M, pH 1.0 (ISOLAB; Eschau, Germany), and 0.4 M sodium acetate, pH 4.5 (Merck; Darmstadt, Germany), stored at 4 °C and protected from the light. The pH was adjusted with 37 % HCl (Fisher Chemical; Fair Lawn, NJ, USA). The concentrated extracts were diluted in each buffer, adjusting the dilution factor to keep absorbances within 0.2–1.2. Absorbances were measured in a spectrophotometer (Shimadzu, UV-1800; Kyoto, Japan) at 520 and 700 nm, in triplicate. The differential absorbance was calculated as follows:

$$A = (A\ 520\ nm - A\ 700\ nm)_{pH1.0} - (A\ 520\ nm - A\ 700\ nm)_{pH4.5}$$

The concentrations of monomeric anthocyanins were determined using the following equation:

$$Total\ anthocyanins\ (\text{mg L}^{-1}) = \frac{A \times MW \times DF \times 1000}{\in \times 1}$$

where MW is the molecular weight of cyanidin-3-glucoside (449.2 g mol^{-1}), $\in$ is the coefficient of molar extinction (26 900 L mol^{-1} cm^{-1}), DF is the dilution factor (5), and A is the differential absorbance. The results were expressed in equivalent terms of cyanidin-3-glucoside as a reference pigment.

Incorporating the natural dye into yogurt (Phase III)

The anthocyanin extract obtained under optimal conditions was added to artisanal yogurt made with Nutri brand milk (Lácteos San Antonio; Cuenca, Ecuador) and Toni brand natural sugar-free ferment (Industrias Lácteas Toni S.A.; Guayaquil, Ecuador). Fermentation was carried out by incubating the mixture for 6 h at 38–40 °C, with 6 % of sugar as a substrate for the lactic acid bacteria (Molina *et al*., 2024). Three treatments were formulated with different dye concentrations: 0.6 (T1), 0.8 (T2), and 1 % (T3), as well as a control without dye (T4).

Sensory evaluation (Phase IV)

A descriptive sensory test was applied on 50 untrained consumers (emotional judges), who evaluated attributes of color, aroma, flavor, and consistency using a nine-point hedonic scale (1 = I dislike it extremely; 9 = I like it extremely), according to the methodology proposed by Santos-Alemán *et al*. (2023b) and Mendoza *et al*. (2023).
The experimental design of the sensory evaluation was conducted using a completely balanced block design, with treatments T1 to T4 assigned to blocks A–D. Block A corresponded to treatment T1, with 0.6 % of extract in its formulation; block B to treatment T2, with 0.8 %; block C to treatment T3, with 1 % of extract; and block D to treatment T4, which worked as a control without the addition of pigment. The average scores for each attribute were recorded for analysis.

Statistical analysis

Extraction was optimized using a polynomial regression model validated with analysis of variance (ANOVA) within the methodology (response surface methodology) using Statgraphics Centurion 18 (version 18.1.13). The assumptions of normality (Shapiro-Wilk), homoscedasticity, and independence of residuals were verified. In case of a breach, the non-parametric Kruskal-Wallis test was applied to detect significant differences between treatments. In addition, Fisher's least significant difference (LSD) method was implemented to compare means with a 95 % confidence level.

RESULTS AND DISCUSSION

Proximal analysis of the sweet potato

Results of the bromatological analyses showed that the purple sweet potato presents a high humidity content (69.09 %) and low content of ash (0.9 %), raw fiber (0.84 %), raw fat (0.17 %), and protein (1.13 %). Additionally, a considerable amount of dry matter (30.9 %) and neutral detergent fiber (NDF) (11.95 %) was recorded, along with a low content of acid detergent fiber (ADF) (0.75 %) and undetectable levels of lignin (Table 1). These results are consistent with values reported by Vidal *et al*. (2018), which backs the stability and quality of purple sweet potato as a raw material for the extraction of natural dyes (Coba-Carrera *et al*., 2019).

Table 1. Results of the proximal analysis for the fractions of purple sweet potato (*Ipomoea batatas* L.) -complete matrix and shell- expressed on a wet basis (fresh weight).

Parameter	Sweet potato (complete matrix)	Sweet potato skin
Humidity (%)	69.09	76.99
Dry matter (%)	30.9	23.0
Raw protein (%)	1.13	0.897
Raw fat (%)	0.17	0.33
Raw ash (%)	0.9	2.67
Raw fiber (%)	0.84	3.33
Neutral detergent fiber (%)	11.95	10.98
Acidic detergent fiber (%)	0.75	4.70
Acidic detergent lignin (%)	Not detected	1.54

As for the skin of purple sweet potato (Table 1), a higher content of humidity (76.99 %), ash (2.67 %), raw fiber (3.33 %), and ADF (4.7 %) was observed in comparison to the complete matrix. These results coincide with earlier investigations that point out the skin of the sweet potato as an important source of insoluble dietary fiber, with beneficial effects in digestive health (Armijos *et al*., 2020). Likewise, lignin was found in low levels (1.54 %), yet it was not quantifiable in the complete matrix. This lower presence could be related to a better digestibility of the fiber in food applications (Guarner *et al*., 2011). These results indicate that the sweet potato skin has favorable chemical features, particularly due to its fiber content, which may add functional value to the dye made from this raw material.

Concentration of anthocyanins

The results displayed significant differences in the concentrations of anthocyanins among treatments (Table 2). The optimal treatments were T11, with 62.87 mg L^{-1} under conditions of 60 % ethanol, 60 °C, and 20 min of extraction; and T9, with 61.5 mg L^{-1}, under the same ethanol percentage and extraction time, but at a temperature of 40 °C. These results coincide with the recommendations by Stoica *et al*. (2022), who suggest that the extraction of anthocyanins from red onion skin must be carried out at an ethanol concentration of 60 % to favor the extraction of anthocyanins and balance the solubility of hydrophilic molecules.

By contrast, the least efficient treatment was T4, which only reached 3.2 mg L^{-1} with 70 % of ethanol, 60 °C, and 25 min of extraction. This yield suggests that high concentrations of ethanol combined with high temperatures may reduce the degradation of anthocyanins, as pointed out by Cacace and Mazza (2003).

These findings coincide with Sadilova *et al*. (2006), who showed that anthocyanins are sensitive to temperatures, pH, and solvents such as alcohol, and degrade easily. Likewise, Patras *et al*. (2010) indicate that high temperatures affect the color and

Table 2. Concentration of anthocyanins (mg of cyanidin-3-glucoside L-1) obtained from the ultrasound-assisted extraction of 0.25 g of purple sweet potato skin (*Ipomoea batatas* L.) using the Box-Behnken design, evaluated across 15 treatments in triplicate.

Treatment	Ethanol concentration (%)	Temperature (°C)	Time (min)	Anthocyanin concentration (mg L^{-1})
1	50	40	25	17.96 fg
2	70	40	25	28.13 e
3	50	60	25	11.85 h
4	70	60	25	3.20 i
5	50	50	20	15.08 g
6	70	50	20	21.09 f
7	50	50	30	26.76 e
8	70	50	30	22.59 f
9	60	40	20	61.50 a
10	60	40	30	39.75 cd
11	60	60	20	62.87 a
12	60	60	30	25.32 e
13	60	50	25	43.14 c
14	60	50	25	39.48 cd
15	60	50	25	54.73 b

Different letters in the same column indicate differences according to Tukey's test ($p \leq 0.005$).

functional activity of these molecules, whereas Zhang *et al*. (2015) report that high concentrations of ethanol may alter their molecular structure, reducing their stability and shelf life.

Analysis of variance and Pareto chart

The analysis of variance (ANOVA) of the quadratic model (Table 3) identified the time of extraction (B) and the quadratic term of ethanol (CC) as significant factors ($p = 0.0003$ and $p = 0.0000$, respectively), whereas the temperature (A) and the linear term of ethanol (C) were not significant ($p = 0.8625$ and $p = 0.1821$, respectively). The adjusted R^2 (74.69 %) indicates that the model adequately explains the variability of the experimental data. The results coincide with findings by Fernandes *et al*. (2020), who point out that the temperature, in some cases, does not have a significant influence on the efficiency of the extraction of anthocyanins when time and concentration of ethanol are controlled.

Results were verified in the Pareto chart, where time of extraction (B) and the quadratic term of the concentration of ethanol (CC) appear as the factors with the greatest influence on the concentration of anthocyanins, both of which surpass the line of reference (Figure 1A). The results of the means for time (Figure 1B) showed that short periods (20 min) maximize yield and prevent degradation caused by prolonged exposure, coinciding with reports by Tiwari *et al*. (2009). Regarding the concentration

Table 3. Analysis of variance (ANOVA) for the concentration of anthocyanins in the skin of purple sweet potato (*Ipomoea batatas* L.), considering linear and quadratic effects, as well as interactions of the independent variables.

Variables	*p*-value (concentration of anthocyanins)
Temperature	0.8625
Time	0.0003*
Ethanol concentration	0.1821
AA	0.3652
AB	0.2197
AC	0.7444
BB	0.6631
BC	0.7251
CC	0.0000*
R^2	74.69 %

*Significant to α = 0.05. AA: quadratic effect of the temperature; BB: quadratic effect of time; CC: quadratic effect of the concentration of ethanol; AB: temperature-time interaction; AC: temperature-ethanol concentration interaction; BC: time-ethanol concentration interaction.

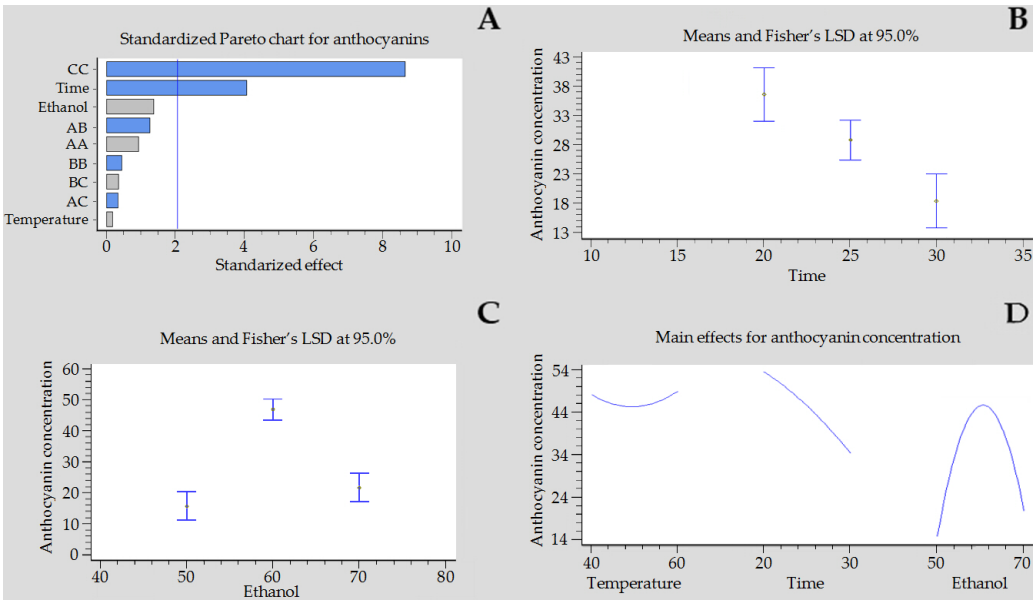

Figure 1. Graphic analysis of the factors that affect the concentration of anthocyanins (mg L^{-1}) obtained from 0.25 g of purple sweet potato (*Ipomoea batatas* L.) skin. A: Pareto chart of the concentration of anthocyanin; B: Means for time of extraction (min) + Fisher's least significant difference test (LSD); C: Means for the ethanol concentration (%) + Fisher's LSD test; D: behavior of the study variables against the concentration of anthocyanins. AA: quadratic effect of the temperature; BB: quadratic effect of time; CC: quadratic effect of the concentration of ethanol; AB: temperature-time interaction; AC: temperature-ethanol concentration interaction; BC: time-ethanol concentration interaction.

of ethanol (Figure 1C), 60 % provided the best balance between the polarity of the solvent and the stability of the anthocyanins, coinciding with descriptions by Cacace and Mazza (2003). The joint behavior of the variables (Figure 1D) indicated that the temperature keeps a stable effect in the interval between 40 and 60 °C, considered adequate to preserve the integrity of these compounds; however, prolonged exposure times may favor their degradation.

Optimization of the response variable (concentration of anthocyanins)

The optimization of the experimental design helped determine the ideal conditions to maximize the concentration of anthocyanins in the process of extraction (Table 4). The optimum treatment identified was T11, characterized by a short time, a high temperature, and an intermediate ethanol concentration. These conditions reflect the importance of a balance between time, temperature, and solvent for the efficient extraction of anthocyanins. These conditions coincide with Xue *et al*. (2021), who indicated that lower extraction times in ultrasound-assisted processes help obtain high yields, since they prevent the thermal degradation of anthocyanins.

Table 4. Optimal conditions of the anthocyanin extraction process from the skin of purple sweet potato (*Ipomoea batatas* L.), determined with the Box-Behnken design.

Variable	Optimum value
Temperature	60 °C
Time	20 min
Ethanol concentration	60.16 %
Anthocyanin concentration	60.72 mg L^{-1}

The optimal concentration obtained (60.72 mg L^{-1}), under the extraction conditions presented, validates the effectiveness of the quadratic model applied in the Box-Behnken design. This confirms its usefulness in the optimization of complex processes such as the ultrasound-assisted extraction, backing similar approaches by Tiwari *et al*. (2009) and Castañeda-Ovando *et al*. (2009) in studies on the extraction of phenolic compounds and anthocyanins.

Response surface of the Box-Behnken design

The Box-Behnken design proved to be efficient to optimize the extraction of anthocyanins from the skin of sweet potato by evaluating both the individual influence and the interactions between temperature, time, and the concentration of ethanol. This predictive tool helped identify optimal conditions, guiding potential industrial applications. Using the response surface methodology, the significance of adjusting and balancing critical parameters to promote efficient extraction was demonstrated. The most relevant response surfaces are shown and discussed below.

Surface at fixed ethanol concentration

With a constant percentage of ethanol (60 %), the concentration of anthocyanins increased considerably in combination with high temperatures (60 °C) and short times (20 min) (Figure 2). This behavior can be attributed to the greater solubility of the anthocyanins in ethanol at high temperatures, in which thermal energy facilitates the rupture of the cell walls (Castañeda-Ovando *et al*., 2009). However, prolonged times lead to a reduction in concentration, probably due to the thermal degradation of the compounds, which has also been reported by Tiwari *et al*. (2009).

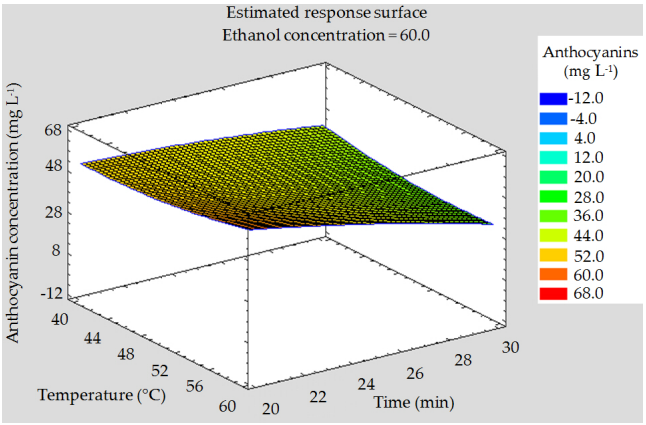

Figure 2. Response surface for the concentration of anthocyanins based on the temperature and the time of extraction with 60 % ethanol from 0.25 g of purple sweet potato (*Ipomoea batatas* L.) skin.

Surface at fixed sonication time

By keeping time constant (20 min), the concentration of anthocyanins displayed a direct dependence on the temperature and the ethanol concentration (Figure 3). The best results were obtained at 60 °C and 60 % of ethanol, thus confirming the need for a balance between temperature and solvent to avoid losses due to degradation or insufficient solvent. This finding is consistent with Cacace and Mazza (2003), who found that 60 % of ethanol offers the optimum balance between polarity and stability in the extraction of anthocyanins.

Surface at fixed temperature

When the temperature was fixed at 60 °C and the extraction time was short (20 min), the concentration of anthocyanin increased significantly (Figure 4). Slight deviations from these conditions reduced the efficiency of the process, which highlights the need for an accurate control of these variables. These findings are consistent with those reported by Xue *et al*. (2021), who also identified short time and controlled temperature as key factors in ultrasound-assisted extraction.

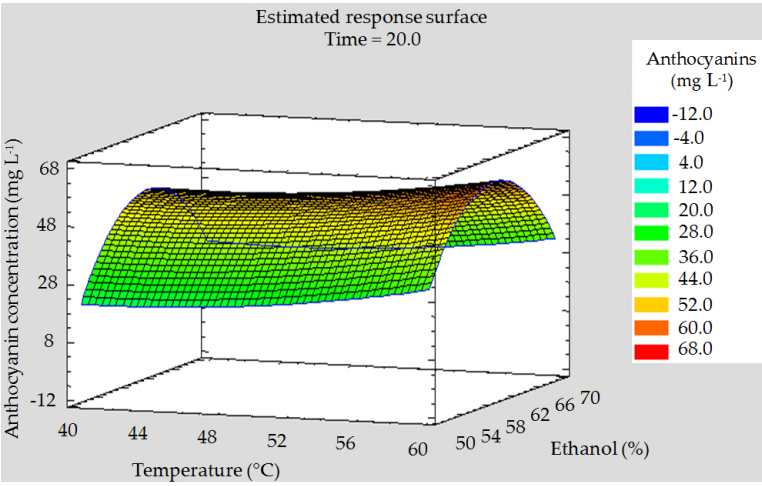

Figure 3. Response surface for the concentration of anthocyanins based on the temperature and ethanol concentration, with a fixed sonication time of 20 min from 0.25 g of purple sweet potato (*Ipomoea batatas* L.) skin.

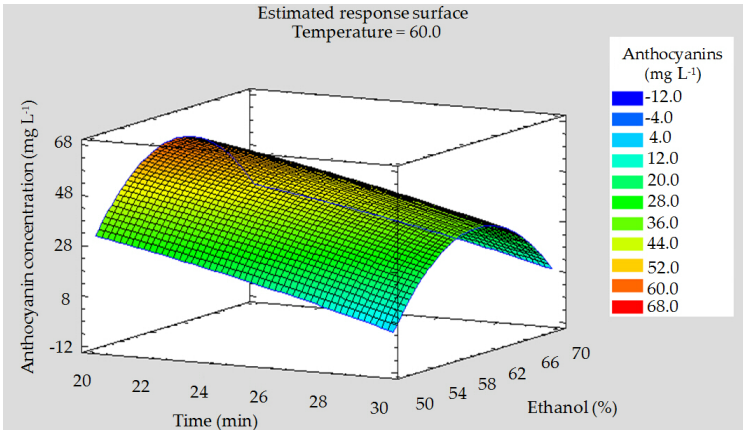

Figure 4. Response surface for the concentration of anthocyanins based on the time of extraction and concentration of ethanol, with a fixed temperature of 60 °C from 0.25 g of purple sweet potato (*Ipomoea batatas* L.) skin.

Path of steepest ascent

To further optimize the process, the path of steepest ascent was used, which projects the optimal combination of variables to maximize the concentration of anthocyanins (Table 5). A progressive increase in temperature from 50 to 70 °C favors the increase in the concentration, reaching an estimated maximum value of 87.86 mg L^{-1} at 70 °C. This behavior coincides with Chemat *et al*. (2012), who pointed out that moderate increases in temperature favor solubility and the diffusion of phenolic compounds in ultrasound-assisted extractions.

Table 5. Path of greatest ascent for the optimization of the concentration of anthocyanins in the extraction from 0.25 g of purple sweet potato (*Ipomoea batatas* L.) skin, depending on the projection of the evaluated variables.

Temperature (°C)	Time (min)	Ethanol (%)	Anthocyanin concentration prediction (mg L^{-1})
50.0	25.00	60.00	45.79
51.0	22.40	60.43	50.40
52.0	21.36	60.39	52.40
53.0	20.60	60.35	54.06
54.0	19.98	60.31	55.63
55.0	19.45	60.27	57.17
56.0	18.98	60.24	58.74
57.0	18.54	60.20	60.35
58.0	18.15	60.17	62.01
59.0	17.77	60.14	63.74
60.0	17.42	60.11	65.54
61.0	17.08	60.08	67.41
62.0	16.76	60.06	69.36
63.0	16.44	60.03	71.38
64.0	16.14	60.00	73.49
65.0	15.85	59.97	75.68
66.0	15.56	59.95	77.95
67.0	15.28	59.92	80.30
68.0	15.01	59.90	82.74
69.0	14.74	59.87	85.26
70.0	14.48	59.84	87.86

Likewise, a reduced extraction time (from 25 to 14 min) minimizes thermal degradation, allowing operation at relatively high temperatures without affecting the stability of the anthocyanins, as pointed out by Patras *et al*. (2010). During this process, the ethanol concentration was kept at around 60 %, validating the key role of this percentage to balance the solubility and stability of bioactive compounds, according to reports by Dai and Mumper (2010). The balance between a high temperature, a reduced time of extraction, and an intermediate concentration of ethanol is crucial to maximize the recovery of anthocyanins.

Sensory evaluation

The sensory evaluation helped obtain the means of the attributes evaluated, as well as the intent of purchase associated with each one of the yogurt samples, including those with natural dye and the control treatment (Table 6).

No significant differences were found in color (p = 0.5296), although T4 (yogurt without dye) had the highest score. This could be related to a preference for more natural tonalities or to samples with dye not presenting a sufficiently distinctive color.

Table 6. Results of the sensory evaluation and intent of purchase for different yogurt samples with and without natural dye, including a statistical analysis with the Kruskal-Wallis test.

Panel block	Color	Aroma	Flavor	Consistency	Intent of purchase (%)
A (T1)	6.18 ± 2.28	5.82 ± 2.30	6.02 ± 2.45	6.06 ± 2.45	60
B (T2)	6.27 ± 2.47	6.00 ± 2.32	6.43 ± 2.45	6.31 ± 2.43	60
C (T3)	6.35 ± 2.43	5.78 ± 2.21	6.18 ± 2.45	6.16 ± 2.46	66
D (T4)	6.59 ± 2.52	6.51 ± 2.16	6.94 ± 2.45	7.02 ± 2.39	82
Kruskal-Wallis test	0.5296	0.3282	0.0606	0.0526	-

*The values express a mean ± standard deviation on a hedonic nine-point scale evaluated by 50 panelists. The intent of purchase represents the percentage based on the total of the 50 evaluators willing to purchase the sample. No significant differences were observed (Kruskal-Wallis, $p > 0.05$).

Coello-García *et al*. (2000) pointed out that a greater chromatic intensity can influence the perception of the product.

The scores for aroma were similar between treatments ($p = 0.3282$). The lack of impact of the dye on aroma is consistent with studies that point out that the addition of natural pigments, such as anthocyanins, has no significant impact on the volatile compounds in charge of the aromatic profile of the product (Tian *et al*., 2022). In terms of the flavor, despite not being significant ($p = 0.0606$), T4 was the best valued, which suggests that the panelists perceived a better balance in the flavor of the yogurt without dye. According to Delwiche (2004), the perception of flavor can be influenced by visual stimuli and sensory expectations.

The values of consistency reflected a positive tendency for T4 ($p = 0.0526$). The dye did not perceptibly alter this attribute, indicating that it does not affect the physical structure of the yogurt. Regarding the purchase intent, the control treatment displayed the highest acceptance (82 %). Nevertheless, sample T3 (1 % dye) had a score of 66 %, which highlights an acceptable potential of the dye.

Jointly, the use of the dye displayed promising results. However, the preference for the control treatment highlights the need to adjust the concentration of the pigment in the yogurt to achieve an adequate balance between the intensity of the color and the sensory acceptance of the product. Adjustments in concentration can improve key attributes (Chemat *et al*., 2012).

CONCLUSIONS

The extraction of anthocyanins from the skin of sweet potato by ultrasound was efficient to obtain the pigment. The optimization of the process using a Box-Behnken surface response design determined that the best conditions are a temperature of 60

°C, an extraction time of 20 min, and an ethanol concentration of 60.16 %. The extract as a natural dye in yogurt displayed a good acceptance, although the control sample was the best valued. These results show the potential of the extract as a viable alternative to synthetic dyes, not only for its sensory acceptance but also for its possible health benefits thanks to its bioactive compounds.

REFERENCES

AOAC (Association of Official Analytical Chemists). 2019. Official methods of analysis of AOAC international (21st edition). Rockville, MD, USA. 700 p.

Armijos G, Villacrés E, Quelal MB, Cobeña G, Álvarez J. 2020. Evaluación físico-química y funcional de siete variedades de camote provenientes de Manabí-Ecuador. Revista Iberoamericana de Tecnología Postcosecha 21 (2).

Cacace JE, Mazza G. 2003. Optimization of extraction of anthocyanins from black currants with aqueous ethanol. Journal of Food Science 68 (1): 240–248. https://doi.org/10.1111/j.1365-2621.2003.tb14146.x

Castañeda-Ovando A, Pacheco-Hernández ML, Páez-Hernández ME, Rodríguez JA, Galán-Vidal CA. 2009. Chemical studies of anthocyanins: A review. Food Chemistry, 113 (4): 859–871. https://doi.org/10.1016/j.foodchem.2008.09.001

Chemat F, Vian MA, Cravotto G. 2012. Green extraction of natural products: Concept and principles. International Journal of Molecular Sciences 13 (7): 8615–8627. https://doi.org/10.3390/ijms13078615

Coba-Carrera RL, Apolo-Criollo LG, Segura-Mestanza JH, Brito-Moina HL. 2019. Obtención del colorante natural del Camote (*Ipomoea batatas*). Ciencia Digital 3 (3.2): 38–47. https://doi.org/10.33262/cienciadigital.v3i3.2.714

Coello-García MT, Díaz-Berciano C, Gómez-Pestaña NG. 2000. Efectos del color en la aceptabilidad, artificialidad, dulzor e intensidad del sabor de bebidas lácteas. Psicothema 12 (2): 140–144.

Dai J, Mumper RJ. 2010. Plant phenolics: Extraction, analysis and their antioxidant and anticancer properties. Molecules 15 (10): 7313–7352. https://doi.org/10.3390/molecules15107313

Delwiche J. 2004. The impact of perceptual interactions on perceived flavor. Food Quality and Preference 15 (2): 137–146. https://doi.org/10.1016/S0950-3293(03)00041-7

Fallah AA, Sarmast E, Fatehi P, Jafari T. 2020. Impact of dietary anthocyanins on systemic and vascular inflammation: Systematic review and meta-analysis on randomised clinical trials. Food and Chemical Toxicology 135: 110922. https://doi.org/10.1016/j.fct.2019.110922

Fernandes FA, Rodrigues S, de Brito ES, Tiwari BK. 2020. Ultrasound-assisted extraction of anthocyanins and phenolics from jabuticaba (*Myrciaria cauliflora*) peel: Kinetics and mathematical modeling. Journal of Food Science and Technology 57 (6): 2321–2328. https://doi.org/10.1007/s13197-020-04270-3

Guarner F, Sastre A, Requejo A, Ruíz-Roso B, Gómez-Martínez S, Esperanza-Díaz L. 2011. El libro blanco de la fibra dietética. Corporación Alimentaria Peñasanta: Siero, España.

INEC (Instituto Nacional de Estadística y Censos). 2023. Anuario meteorológico y climático del Ecuador. Quito, Ecuador. https://www.ecuadorencifras.gob.ec (Retrieved: August 2024).

Maddela NR, García LC. 2021. Innovations in biotechnology for a sustainable future. Springer: New York, NY, USA. 448 p.

Marcía-Fuentes JA, López-Salas L, Borrás-Linares I, Navarro-Alarcón M, Segura-Carretero A, Lozano-Sánchez J. 2021. Development of an innovative pressurized liquid extraction procedure by response surface methodology to recover bioactive compounds from carao tree seeds. Foods 10 (2): 398. https://doi.org/10.3390/foods10020398

Marcía-Fuentes JA, Montero-Fernández I, Zumbado-Fernández H, Lozano-Sánchez J, Santos-Alemán R, Navarro-Alarcón M, Borrás-Linares I, Saravia-Maldonado SA. 2020. Quantification of bioactive molecules, minerals and bromatological analysis in carao (*Cassia grandis*). Journal of Agricultural Science 12 (3): 88. https://doi.org/10.5539/jas.v12n3p88

Mendoza E, Marcía-Fuentes JA, Chuquilín-Goicochea RC, López J, Areche F, Ruiz J, Herrera A. 2023. Obtención de un colorante natural a partir *Tropaeolum tuberosum* Ruíz y Pavón para su aplicación en yogur. Revista Bionatura 8 (2): 38.

Molina C, Campos-García SK, Marcía-Fuentes JA, Ore-Areche F, Yadav A, Santos-Alemán R. 2024. Effects of capulin (*C. xalapensis*) on the microbiological, physicochemical and sensory properties of yogurt. Dairy 5 (3): 515–525. https://doi.org/10.3390/dairy5030039

Montero IF, Saravia SA, Santos RA, dos Santos RC, Marcía-Fuentes JA, da Costa HN. 2020. Nutrients in Amazonian fruit pulps with functional and pharmacological interest. African Journal of Pharmacy and Pharmacology 14 (5): 118–127. https://doi.org/10.5897/ajpp2020.5136

Patras A, Brunton NP, O'Donnell C, Tiwari BK. 2010. Effect of thermal processing on anthocyanin stability in foods; mechanisms and kinetics of degradation. Trends in Food Science and Technology 21 (1): 3–11. https://doi.org/10.1016/j.tifs.2009.07.004

Sadilova E, Stintzing FC, Carle R. 2006. Thermal degradation of acylated and nonacylated anthocyanins. Journal of Food Science 71 (8): C504–C512. https://doi.org/10.1111/j.1750-3841.2006.00148.x

Saini RK, Khan MI, Shang X, Kumar V, Kumari V, Kesarwani A, Ko EY. 2024. Dietary sources, stabilization, health benefits, and industrial application of anthocyanins—a review. Foods 13 (8). https://doi.org/10.3390/foods13081227

Santos-Alemán R, Marcía-Fuentes JA, Montero-Fernández I, King J, Pournaki S, Hoskin R, Moncada M. 2023b. Novel liquor-based hot sauce: Physicochemical attributes, volatile compounds, sensory evaluation, consumer perception, emotions, and purchase intent. Foods 12 (2): 369. https://doi.org/10.3390/foods12020369

Santos-Alemán R, Marcía-Fuentes JA, Page R, Kazemzadeh Pournaki S, Martín-Vertedor D, Manrique-Fernández V, Aryana K. 2023a. Effects of yogurt with carao (*Cassia grandis*) on intestinal barrier dysfunction, α-glycosidase activity, lipase activity, hypoglycemic effect, and antioxidant activity. Fermentation 9 (6): 566. https://doi.org/10.3390/fermentation9060566

Saravia-Maldonado SA, Santos-Alemán RS, Marcía-Fuentes JA, da Conceição MCF. 2020. Determination of total phenolic compounds, antioxidant activity and nutrients in Brazil nuts (*Bertholletia excelsa* H. B. K). Journal of Medicinal Plants Research 14 (8): 373–376. https://doi.org/10.5897/jmpr2020.6953

Solórzano L, Marcía-Fuentes JA, Chuquilín-Goicochea RC, Areche F, Herrera A, Ruíz-Cardona JM. 2023. Evaluación de la pulpa de macha macha (*Vaccinium floribundum* Kunth), en el desarrollo de una bebida isotónica. Revista Bionatura 8 (4): 38. https://doi.org/10.21931/rb/2023.08.04.38

Stoica F, Constantin OE, Stănciuc N, Aprodu I, Bahrim GE, Râpeanu G. 2022. Optimization of the parameters influencing the antioxidant activity and concentration of anthocyanins

extracted from red onion skins using a central composite design. Inventions 7 (4): 89. https://doi.org/10.3390/inventions7040089

Thiex N. 2009. Evaluation of analytical methods for the determination of moisture, crude protein, crude fat, and crude fiber in distillers dried grains with solubles. Journal of AOAC International 92 (1): 61–73. https://doi.org/10.1093/jaoac/92.1.61

Tian XZ, Wang X, Ban C, Luo QY, Li JX, Lu Q. 2022. Effect of purple corn anthocyanin on antioxidant activity, volatile compound and sensory property in milk during storage and light prevention. Frontiers in Nutrition 9: 862689. https://doi.org/10.3389/fnut.2022.862689

Tiwari BK, O'donnell CP, Patras A, Brunton N, Cullen PJ. 2009. Stability of anthocyanins and ascorbic acid in sonicated strawberry juice during storage. European Food Research and Technology 228 (5): 717–724. https://doi.org/10.1007/s00217-008-0982-z

van Soest PJ, Robertson JB, Lewis BA. 1991. Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. Journal of Dairy Science 74 (10): 3583–3597. https://doi.org/10.3168/jds.S0022-0302(91)78551-2

Vidal AR, Zaucedo-Zuñiga AL, Ramos-García ML. 2018. Propiedades nutrimentales del camote (*Ipomoea batatas* L.) y sus beneficios en la salud humana. Revista Iberoamericana de Tecnología Postcosecha 19 (2).

Xue H, Tan J, Li Q, Cai X, Tang J. 2021. Optimization ultrasound-assisted extraction of anthocyanins from cranberry using response surface methodology coupled with genetic algorithm and identification anthocyanins with HPLC-MS2. Journal of Food Processing and Preservation 45 (7): e15378. https://doi.org/10.1111/jfpp.15378

Zapata LM, Heredia AM, Quinteros CF, Malleret AD, Clemente GC. 2014. Optimización de la extracción de antocianinas de arándanos. Ciencia, Docencia y Tecnología 25 (49): 166–192.

Zhang B, Liu R, He FZ, Duan CQ. 2015. Copigmentation of malvidin-3-O-glucoside with five hydroxybenzoic acids in red wine model solutions: Experimental and theoretical investigations. Food Chemistry 170: 226–233. https://doi.org/10.1016/j.foodchem.2014.08.026

Zhao Y, Wang L, Huang Y, Evans PC, Little PJ, Tian X, Weng J, Xu S. 2024. Anthocyanins in vascular health and disease: mechanisms of action and therapeutic potential. Journal of Cardiovascular Pharmacology 84 (3): 289–302. https://doi.org/10.1097/FJC.0000000000001602