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Optimization of a craft ale-type beer enriched with cañihua malt (*Chenopodium pallidicaule*) and banana passionfruit juice (*Passiflora tripartita* var. *mollisima*)

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ABSTRACT

The global expansion of the craft beer market has driven the incorporation of native ingredients to enhance the sensory and nutritional profiles of beer. This study focused on optimizing a craft Ale-type beer enriched with Cañihua Malt (CM) and Banana Passionfruit Juice (BPJ) using a Doptimal experimental design. The aim was to evaluate how varying concentrations of these ingredients (CM: 15-25 %, BPJ: 5-15 %) influence the physicochemical, technological, and sensory attributes of beer. Results demonstrated that the malting process significantly improved the nutritional composition of cañihua, increasing fiber content (23.32 g/100 g), phenolic compounds (141.13 mg GAE/100 g), GABA (229.48 mg/100 g), and antioxidant capacity (1975.41 µmol TE/g dw). These enhancements positively affected the physicochemical properties of beer, especially foam stability and body. The addition of BPJ significantly modified the physicochemical characteristics of beer, particularly by reducing the pH and increasing the acidity. Sensory analysis showed high consumer acceptance, with positive evaluations for aroma, appearance, and body, particularly in samples containing moderate levels of CM (15-16 %) and BPJ (5-10 %). Optimization using the desirability function identified ideal concentrations of 24%-25 % CM and 5 % BPJ, achieving a balance in critical parameters such as foam stability, density, pH, and bitterness. These findings underscore the potential to combine CM and BPJ to develop a distinctive craft beer with enhanced sensory attributes and nutritional benefits.

1. Introduction

The craft beer industry has experienced remarkable global growth and is recognized for its high quality, exclusivity, and ability to incorporate novel flavors and ingredients. According to the Brewers Association, the global craft beer market was valued at USD 119.923 billion in 2023, with a projected increase to USD 186.592 billion by 2030, driven by a compound annual growth rate (CAGR) of 9.4 % between 2024 and 2030 [1]. Currently, North America leads the global market, whereas Europe has the highest growth rate

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[2]. This expansion is largely driven by consumer demand for personalized experiences, including low-alcohol beers and innovative flavors [3]. As a result, the industry has increasingly focused on experimentation, with the incorporation of native and unconventional ingredients emerging as a key strategy to meet the demands of an ever-diversifying market [4].

The diversification of ingredients in craft beer production not only offers differentiated products but also promotes the use of native crops, adding value to the market. The inclusion of indigenous and ancestral ingredients in mass-consumption products provides consumers with a unique experience that links local traditions with food innovation. Examples include beers brewed with quinoa and amaranth, which have been shown to significantly alter the sensory and technological characteristics of beer, introducing novel flavors and textures [5–7]. The use of exotic fruits in fermented beverages has also gained popularity. For instance, Montero and Fuentes [8] formulated a lager-style beer flavored with pomegranate juice, resulting in a product with a purplish-red hue, sweet and sour flavor, pleasant aroma, and smooth liquid texture. Similarly, Hlangwani et al. [9] developed a low-alcohol marula fruit beer using non-Saccharomyces yeasts, while Oliveira et al. [10] produced fermented ginger beers containing berries and cajá, further emphasizing the importance of innovation in the craft beer industry.

This study focused on two key native ingredients: cañihua malt (CM) from the Puno region and banana passionfruit juice (BPJ) from the Ancash region. Cañihua (*Chenopodium pallidicaule*), a pseudocereal native to the Andes, is recognized for its high-quality protein content, dietary fiber, vitamins, and bioactive compounds such as polyphenols, phytosterols, and betalains, which offer significant health benefits [11,12]. Germination and malting processes further enhance the nutritional and functional value of cañihua by increasing the concentration of these bioactive compounds and reducing antinutritional factors [13,14]. While malting is a well-established technique for conventional grains, its application to pseudocereals such as cañihua remains limited, making it a promising ingredient for the production of craft beers with a unique sensory profile.

Banana passionfruit (*Passiflora tripartita* var. *mollissima*), known as "*tumbo serrano*", is an Andean native fruit characterized by its exotic flavor and high nutritional value, particularly its elevated vitamin C and antioxidant content. Its pulp, which has a low pH, is well-suited for fermented products due to its freshness and acidity—qualities that complement the organoleptic properties of an Aletype beer [15]. This fruit has demonstrated versatility in the production of fermented products such as probiotic yogurts [16], functional beverages [17], nectars [18], and jams [19], where it has positively influenced both sensory and nutritional profiles. Incorporating banana passionfruit into craft beer production could enhance its nutritional value and add complexity and freshness to its flavor profile, creating a unique sensory experience for consumers.

To leverage these properties, this study proposes the development of a craft beer that retains the characteristics of a conventional beer while offering a differentiated sensory and nutritional profile. The aim of this research was to optimize the formulation of a craft Ale-type beer enriched with CM and BPJ, evaluating how different combinations of these ingredients influence the physicochemical, technological, and sensory attributes of the final product. This study seeks to contribute to the diversification of the craft beer market while promoting the use of native resources and the development of products with a distinct local identity.

2. Materials and methods

2.1. Raw materials

The raw materials used in this study included Pilsen malt (PM) (32 Best Malz), Simcoe hop pellets (13.5 % alpha acid w/w) from the United States, and *Saccharomyces cerevisiae* SafAle US-05 yeast (Lima, Peru). Cañihua grains (var. Illpa-INIA) were sourced from the Agrarian Experimental Station of the *Instituto Nacional de Innovación Agraria* in Puno, Perú. Banana passionfruit (*Passiflora tripartita* var. *mollissima*) (Fig. 1a–c) was harvested from Musho village, Yungay, Áncash, Peru (9°08′22″S 77°44′42″W).

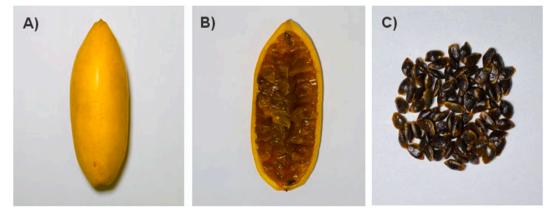


Fig. 1. Banana Passionfruit whole: fruit (A), longitudinal cut (B), and seeds (C).

2.2. Production of banana passionfruit juice (BPJ)

Banana passionfruit were washed with running water, disinfected in a 200 mg/L sodium hypochlorite solution for 15 min, and rinsed with deionized water. The physicochemical characterization of the fruits included measuring the equatorial diameter of 15 randomly selected fruits using a digital Vernier caliper (IP54, China). To obtain BPJ, the pulp was processed using a multipurpose pulper and refiner (Electronica Venetta, PAS/EV, Italy). Finally, the BPJ was stored in a cool dark place at refrigeration temperatures (4–7 °C) until craft beer brewing.

2.3. Characterization of the raw materials

Prior to malting, the proximate composition of raw cañihua grains was determined using AACCI methods [20], including moisture content (method 44–01.01), protein (method 46–13.01, with a nitrogen-to-protein conversion factor of 5.7), ether extract (method 30–25.01), dietary fiber (method 32–05.01), and ash (method 08–01.01). The digestible carbohydrate content was calculated by subtracting the values of protein, ether extract, dietary fiber, and ash from the sample's initial dry weight. Additionally, total starch and phytic acid contents were measured using enzymatic kits K-TSTA-100A and K-PHYT (Megazyme, Wicklow, Ireland), respectively.

Total soluble phenolic compounds (TSP) were quantified using the Folin-Ciocalteu method, as described by Pico et al. [21]. Gamma-aminobutyric acid (GABA) was quantified using reverse-phase high-performance liquid chromatography (RP-HPLC) in an Agilent 1200 separation module (Agilent, Santa Clara, CA, USA) equipped with a G1314B diode array detector (DAD) and a Zorbax Eclipse Plus C18 stationary phase column (4.6×150 mm, 3 μ m), following Cáceres et al. [22]. Antioxidant activity was measured using the oxygen radical absorbance capacity (ORAC) method, as described by Dávalos et al. [23]. All results were expressed in grams per 100 g of dry weight (g/100 g dw). The germination rate was determined using Valenzuela's method [24].

The physicochemical composition of BPJ was analyzed following AOAC methods [25]: moisture content (method 925.10), protein (method 981.10), ash (method 923.03), fat (method 920.39), dietary fiber (method 985.29), and carbohydrate content calculated by difference. The instrumental color of BPJ was assessed using a Minolta CR-310 colorimeter (Osaka, Japan), and results were obtained in triplicate in the CIE-Lab* color space. pH was determined using AOAC methods [25], and titratable acidity was measured using IRAM 14520 standards, expressed as a percentage of citric acid [26].

2.4. Production of cañihua malt (CM)

The cañihua malting process was carried out following the optimized germination parameters described by Abderrahim et al. [13] with minor modifications. The process included four stages: steeping, germination, kilning, and de-rooting. Initially, the grains (Fig. 2a) were washed and disinfected in a 0.1 % sodium hypochlorite solution for 30 min. They were then rinsed with running water until reaching a neutral pH and steeped in distilled water at a 1:5 grain-to-water ratio for 6 h at 25 °C in darkness.

For germination, the hydrated grains were placed in plastic trays covered with damp paper towels and maintained in a pilot-scale germination chamber (Maquilak, Lima, Peru). Germination was conducted for 72 h at 20 $^{\circ}$ C to enhance bioactive compounds and antioxidant capacity. When the radicles reached 7–10 mm in length, germination was stopped, and the germinated grains (Fig. 2b) were dried by air convection in an oven (Poleko, Poland) at 45 $^{\circ}$ C for 24 h until reaching a moisture content of 5–8%. The radicles were then manually removed to obtain malt. The resulting CM (Fig. 2c) was ground in a blade mill (Brabender, Duisburg, Germany) and sieved to obtain 0.25-mm particles. The same characterization analyses performed for raw cañihua (Section 2.1) were applied to the malted grains. The CM was stored in vacuum-sealed plastic bags at -20 $^{\circ}$ C in darkness.

2.5. Experimental design: D-optimal

A D-optimal design with three independent variables was used to optimize the formulation: percentage of PM (X_1) , CM (X_2) , and BPJ (X_3) . Variable ranges were established based on preliminary tests to ensure acceptable sensory and physical characteristics. CM



Fig. 2. Raw (A), germinated (B), and malted (C) cañihua grains.

was limited to 7%–25 % to avoid excessive bitterness, whereas BPJ was set at 5%–15 % to enhance fruity notes without compromising foam stability. The PM range was adjusted to 70%–80 % to balance the formulation. The Design Expert software (version 7.0) generated 13 experimental treatments (Table 1).

2.6. Production of ale-type craft beer

The beer production process is detailed in Fig. 3. First, the ingredients were received and weighed. The malt grains were measured according to the PM and CM proportions specified in the experimental design (Table 1), followed by milling and wort preparation, which involved a grain-to-water ratio of 1:4. The brewing process began by heating water to 76 °C. At this point, the grain mixture was immersed in a nylon bag, causing the temperature to decrease to 65°C-68 °C. This temperature range was maintained for 90 min to allow the enzymatic conversion (saccharification) of starches into fermentable sugars. During this stage, the water was continuously stirred to ensure a uniform heat distribution and enzymatic activity. The temperature was then raised to 76 °C for 10 min to inactivate the enzymes and ensure complete starch conversion. The wort was subsequently filtered and recirculated to clarify and remove suspended particles (*Insert* Fig. 3 here).

Before boiling, hops were added, and after 1 h of boiling at $100\,^{\circ}$ C, BPJ was introduced. After the boiling stage, the wort was rapidly cooled to $15-21\,^{\circ}$ C. The wort density was then measured from 1.044 to $1.052\,$ g/cm³. The cooled wort was inoculated with *Saccharomyces cerevisiae* yeast and fermented at $16\,^{\circ}$ C for 5 days. Following fermentation, the final product density $(1.010-1.015\,$ g/cm³) was recorded. The beer was then matured at $0-4\,^{\circ}$ C for 3 days using lagering techniques commonly employed to improve clarity and minimize chill haze. Both stages were carried out in a food-grade polyethylene fermenter with a $30\,$ L capacity, equipped with a bottling spout and an airlock lid (Vintners Best, model 30FT-Peru).

Carbonation was achieved by adding 6 g/L of sugar to the product, which was then stored at 16 $^{\circ}$ C for two weeks to complete carbonation. Finally, the beer was bottled and refrigerated at 4–6 $^{\circ}$ C until further analysis.

2.7. Physicochemical characterization of beer

Beer sample density was measured before and after fermentation using a triple-scale hydrometer at 20 °C (Stevenson, Scotland), following the ASBC Beer-2 method [27], which allowed the calculation of sugar conversion into alcohol. Soluble solids were determined using a portable digital refractometer (OPTi, UK) to precisely measure residual sugar content in the samples. The pH was measured using a digital pH meter (Thermo Scientific Orion Star A211), and titratable acidity was determined by titration with 0.1 M NaOH in degassed samples, with results expressed as a percentage of lactic acid (equivalent weight of 0.09 g/meq) [25].

The alcohol content was calculated based on the difference between the initial and final densities after fermentation, quantifying the conversion of sugars into ethanol. Finally, turbidity was assessed according to Pascual's protocol [28] using a turbidimeter (Orion™ AQ4500, USA) calibrated with AC45ST standard patterns (0 and 10 NTU), with results reported in Nephelometric Turbidity Units (NTU).

2.8. Technological characterization of beer

The beer color was determined using the standard spectrophotometric method of Analytica-EBC (European Brewery Convention), as specified in NTP 213.027:2016 [29]. This involved measuring the absorbance of degassed and clarified beer using a UV–VIS spectrophotometer (Único SQ2800) at 430 nm in 1-cm quartz cuvettes. The absorbance value was multiplied by 25 to obtain the color in EBC units.

Foam stability was evaluated following the method described by Romero et al. [30]. In this method, 40 mL of beer was poured into a beaker and manually shaken to generate foam. Foam stability was determined by recording the initial liquid volume (Vi), remaining

 Table 1

 D-optimal experimental design of craft beers.

Experiments	X ₁ %	X ₂ %	X ₃ %		
	Pilsen malt (PM)	Cañihua malt (CM)	Banana passionfruit juice (BPJ)		
E1	70	21	9		
E2	75	15	10		
E3	74	16	10		
E4	75	15	10		
E5	80	13	7		
E6	70	15	15		
E7	75	15	10		
E8	76	19	5		
E9	70	15	15		
E10	70	25	5		
E11	80	13	7		
E12	78	7	15		
E13	75	11	14		

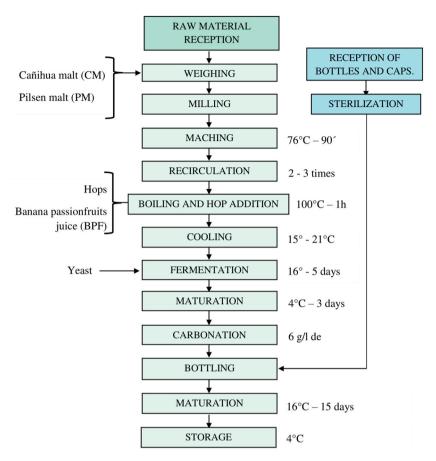


Fig. 3. Flowchart of the production process of the Ale-type craft beers.

liquid volume (VL), total volume (VT), and foam volume (VE).

Bitterness was assessed as described by Martin et al. [31], which measures the concentration of alpha acids in hops by spectrophotometry at 275 nm. The results were expressed in International Bitterness Units (IBU), calculated by extracting and quantifying alpha acids.

2.9. Sensory analysis

The sensory analysis was approved by the National University of Santa (UNS) under project contract N° 027-2020-DAL-UNS (Supplementary material 1). The study was conducted at the Institute of Technological and Agroindustrial Research - UNS, involving 120 consumers aged 18–60 years. All participants provided informed consent before participation.

In the second stage, the Check-All-That-Apply (CATA) method was employed, in which participants selected all applicable attributes for each sample. Thirty-seven attributes were presented and categorized into five groups: appearance (opaque, bright, transparent, foamy, low foam, bubbly), color (yellow, golden, caramel, brown, light, medium, intense), aroma (fruity, herbal, roasted grains, spicy, floral, citrus), taste (toasted cereal, malt, bitter, sour, citrus, sweet, fruity, herbal, refreshing, caramel, light, intense), and mouthfeel (viscous, watery, astringent, fizzy, foamy, delicate). These attributes were defined through a three-session focus group with 15 trained panelists. In each session, panelists were presented with 3–4 samples and asked to freely list the attributes they perceived in terms of appearance, color, aroma, taste, and mouthfeel. The final attribute list was refined by grouping similar terms to eliminate redundancies and standardize terminology.

2.10. Statistical analysis

All nutritional, physicochemical, and technological characterization analyses were conducted in triplicate. The statistical analysis of the experimental data was performed using Design Expert software (version 7.0) under a D-optimal design. The relationship between response variables and factors was evaluated using response surface methodology (RSM). The quality of fit of the mathematical models was assessed using the coefficient of determination (R^2), with a minimum threshold of 0.7 set to ensure predictive reliability. When significant differences were identified (p < 0.05), multiple comparisons among samples were performed using Tukey's test (Statgraphics Centurion, version 16.1).

For the acceptability test data, analysis of variance (ANOVA) was performed. When significant differences were observed (p < 0.05), Tukey's test was used for sample comparison. For the CATA test data, Cochran's Q test was used to identify discriminating terms (p \leq 0.10). Significant terms were further analyzed through correspondence analysis using Hellinger distance, complemented by principal coordinates analysis and penalty analysis to evaluate the relationship between CATA attributes and overall acceptance. The analyses were performed using XLSTAT software (ver 2023.5, ADDINSOFT, New York, USA).

3. Results and discussion

3.1. Physicochemical characteristics of the raw materials

Banana passionfruit exhibited an elliptical shape, with an average diameter of 38.3 ± 0.25 mm and a weight range of 70.46 ± 4.77 to 90.56 ± 9.73 g, consistent with previous studies [19,34].

The extracted BPJ had a pH of 3.21 ± 0.09 , an acidity of 2.45 ± 0.24 %, and a soluble solids content of $12.43\pm1.23^\circ$ Brix. These values are consistent with those reported by Chañi-Paucar et al. [15], showing a pulp pH of 3.2 and acidity of 2.64 %. However, other studies, such as that by Rojas-Iparraguirre [35], documented slightly higher acidity (0.85 %) and lower pH (3.10) with a Brix value of 4° . Similarly, De Florio [36] reported higher acidity (1.65 %) and a pH range between 3.55 and 3.78, along with soluble solids at 12° Brix. These variations can be attributed to factors such as fruit maturity, variety, and cultivation conditions, which significantly influence physicochemical properties. The low pH of BPJ may affect the acidity balance of beer formulations. Preliminary tests were conducted to define an appropriate concentration range for the experimental design to prevent interference with the desired sensory profile.

The proximate composition of BPJ revealed a water content of 85.21 ± 5.98 %, carbohydrates at 8.46 ± 0.08 %, protein at 1.32 ± 0.13 %, fat at 0.78 ± 0.04 %, dietary fiber at 3.45 ± 0.26 %, and ash content below 1 %. This composition is typical of fruit juices, exhibiting lower fiber and protein content than whole fruit. These values are consistent with those reported by Santos et al. [37], documenting 80 % water, 2 % protein, and 0.7 % fat. Rojas [38] similarly observed lower protein and fiber content than our findings, linking these differences to fruit variety, cultivation location, and maturity stage.

The color analysis of BPJ showed instrumental values of lightness (L) at $48.57 \pm 1.85^*$, chroma a (red-green component) at $36.67 \pm 2.10^*$, and chroma b (yellow-blue component) at $56.19 \pm 4.25^*$, indicating a strong tendency toward red tones, likely due to its acidity and anthocyanin content—compounds responsible for the red coloration in many fruits [39]. The L values suggest intermediate brightness*, whereas the b values indicate a shift toward orange*, which may be attributed to the presence of carotenoids [16]. Additionally, BPJ is rich in vitamin C and other volatile compounds, which contribute to its chromatic and sensory characteristics [40].

3.1.1. Cañihua malt (CM)

The malting process, which involved germination to activate enzymes and convert starches into fermentable sugars, followed by controlled drying to halt growth, resulted in significant modifications in the nutritional composition of cañihua (Table 2). During germination, cañihua grains demonstrated a 98 ± 0.13 % germination capacity within 72 h, indicating high germinative energy compared to other pseudocereals. For example, Salamon et al. [41] reported germination energies of 78 % in oats and 67 % in

 Table 2

 Physicochemical properties of Cañihua (Chenopodium pallidicaule) variety Illpa-INIA before and after malting.

Properties	Raw cañihua	Cañihua malt (CM)	
Fat (g/100 g dw)	6.87		
Ash (g/100 g dw)	3.07**	2.52	
Total Dietary Fiber (g/100 g dw)	8.98	23.32**	
Protein (g/100 g dw)	22.90**	20.12	
Carbohydrates (g/100 g dw)	67.16	70.51**	
TSPC (mg GAE/100g)	97.50	141.13**	
GABA (g/100 g dw)	27.05	229.48**	
ORAC (µmol TE/g dw)	1326.64	1975.41**	
PA (g/100 g dw)	1.30**	0.93	

(**) denote statistical differences between samples (ANOVA, Bonferroni post hoc test, $p \le 0.05$). Abbreviations: dw, dry weight; GABA, γ -aminobutyric acid; GAE, gallic acid equivalents; TDF: total dietary fiber; ND: not detected; ORAC, oxygen radical absorbance capacity; PA, phytic acid; TSPC, total soluble phenolic compounds; TE, Trolox equivalents; RC, raw cañihua flour.

Table 3 Physicochemical and technological characteristics of Ale-type craft beers.

Experiment Physicoc pH	sicochemical Characteristics					Technological Characteristics				
	pН	Soluble Solids (Brix)	Acidity (g/ml)	Absolute Density (g/ ml)	Viscosity (mPa.s)	Alcohol Content (%)	Color (EBC)	Turbidity (NTU)	Foam Capacity (% E)	Bitterness (IBU)
Control	4.26 ^c	4.0 ^d	0.293 ^a	1.018 ^b	1.571 ^f	5.07 ^{bc}	16.5 ^d	56.8 ^h	52.5 ^{cd}	16 ^b
E1	4.26 ^c	4.0 ^d	0.297^{a}	1.016^{b}	1.425 ^{cd}	5.16 ^{bcd}	$11.7^{\rm b}$	11.9 ^a	58.0 ^{de}	15 ^a
E2	4.12 ^a	3.5 ^c	0.396 ^{cdef}	1.014^{b}	1.378^{b}	4.94 ^a	14.8 ^{cd}	12.1 ^a	60.0 ^{de}	16 ^b
E3	4.27^{c}	4.3 ^e	0.387 ^{cde}	$1.017^{\rm b}$	1.472 ^e	5.37 ^{abcd}	13.3 ^{bc}	23.3 ^d	12.5 ^a	15 ^a
E4	4.10^{a}	3.5°	$0.392^{\rm cde}$	1.014 ^b	1.387 ^b	5.33 ^{bcd}	15.0 ^{cd}	11.0^{a}	98.0 ^{gh}	16^{b}
E5	4.16 ^{ab}	3.3^{b}	0.324^{ab}	1.013^{b}	1.445 ^{de}	5.62	14.9 ^{cd}	30.7 ^e	85.0 ^{fg}	16^{b}
E6	4.09 ^a	3.0^{a}	0.387^{cd}	1.001 ^a	1.317 ^a	5.48 ^{abcd}	11.4^{b}	24.9 ^d	57.5 ^{de}	16 ^b
E7	4.10^{a}	3.5 ^c	0.360^{bc}	1.014^{b}	1.385 ^b	5.73	33.4 ^g	15.6 ^b	27.5 ^{ab}	16 ^b
E8	4.28 ^c	5.3 ^g	0.315 ^{ab}	1.021^{b}	1.473 ^e	5.23 ^{bcd}	14.4 ^c	16.2 ^b	$110.0^{\rm h}$	16 ^b
E9	4.14 ^{ab}	3.0^{a}	0.441 ^f	1.001 ^a	$1.377^{\rm b}$	5.53	29.9 ^f	96.5 ^f	12.5 ^a	16 ^b
E10	4.22 ^{bc}	4.3 ^e	0.324^{ab}	1.018^{b}	1.455 ^{de}	5.83 ^{bcd}	8.1 ^a	17.2 ^{bc}	75.0 ^{ef}	15 ^a
E11	4.13 ^a	$3.3^{\rm b}$	0.306^{a}	1.013^{b}	1.469 ^e	5.90 ^{de}	20.8 ^e	37.9 ⁱ	42.5 ^{bcd}	16 ^b
E12	4.29 ^c	4.5 ^f	0.423 ^{def}	$1.018^{\rm b}$	1.397^{bc}	6.19^{e}	8.8 ^a	18.6 ^c	37.5 ^{bc}	16^{b}
E13	4.43 ^d	4.5 ^f	0.432^{ef}	1.018^{b}	1.455 ^{de}	5.32 ^{bcd}	8.6 ^a	45.2 ^g	32.5^{b}	16 ^b

^{*}Values within a row with different letters are significantly different through Tukey's test (p < 0.05).

buckwheat over a longer period (120 h), highlighting the superior efficiency of cañihua germination.

Malting reduced both ash and protein content in CM. The decrease in ash content can be explained by the leaching of minerals during steeping and their migration to the radicle during germination, which is subsequently removed during de-rooting [42]. These findings are consistent with those of Luna [43], showing similar reductions in two cañihua accessions. However, Chauhan et al. [44] reported increases in ash content in other malted grains, suggesting that these variations depend on grain type and malting conditions.

Regarding protein content reduction, the results were consistent with studies on other pseudocereals [45]. During germination, proteins undergo enzymatic degradation to nourish the radicle, contributing to the observed decline [46]. Antezana et al. [47] documented similar reductions in quinoa protein content, with protein levels decreasing by up to 22.5 % in some varieties. Additionally, the removal of radicles during de-rooting contributes to protein loss.

Conversely, total dietary fiber content increased by 260 % after malting, a beneficial modification for wort filtration during mashing, as it facilitates the separation of wort from the grains. However, excessive fiber content can negatively affect the extraction efficiency of fermentable sugars [14]. The available carbohydrate content remained at 70.51 ± 1.47 g/100 g in CM, ensuring sufficient conversion into maltose and maltotriose, along with other fermentable sugars and dextrins metabolized by yeast during fermentation to produce alcohol and carbon dioxide—key components of beer [48].

Regarding bioactive compounds, the phenolic content increased by 44 % compared with its initial pre-malting level, reaching 141.13 ± 4.85 mg GAE/100 g, whereas GABA content increased by 202 %. These findings are consistent with those of previous studies reporting the enrichment of phenolic compounds and GABA in germinated grains [13,49]. The activation of glutamate decarboxylase (GAD) during germination facilitates the conversion of glutamate into GABA, explaining the observed increase in GABA concentration [50].

Malting also enhanced the antioxidant capacity of cañihua by 49 %, consistent with findings on malted quinoa [51]. Pseudocereal grains are known for their high antioxidant content, which is largely attributed to their phenolic compound concentrations and cultivation conditions [52]. The antioxidant potential of malt is crucial for beer preservation during storage, as it contributes to oxidative stability [53]. Traditionally, beer has a low antioxidant and nutritional value. However, recent studies have demonstrated that incorporating pseudocereals such as amaranth into malt can enrich beer with flavonoids and other bioactive compounds [54,55]. These findings suggest that CM can enhance both the nutritional and antioxidant properties of beer.

Table 4Predictive regression models for the physicochemical and technological characteristics of beers according to independent variables.

Characteristics	Dependent Variables	Model ^a	p-value	R ² (predicted)	R ² (adjusted)
Physicochemical	pH	$\begin{array}{l} y = 10.49X_1 + 4.22X_2 + 3.33X_3 \cdot 8.67X_1X_2 \cdot 7.44X_1X_3 + 1.37X_2X_3 \cdot \\ 142.51X_1^2X_2X_3 + 22.73X_1X_2^2X_3 + 86.85X_1X_2X_3^2 \end{array}$	0.0024	98.43	95.28
	Acidity	$y = 0.003X_1 + 0.002X_2 + 0.014X_3$	0.0015	72.67	67.20
	Soluble solids	$y = \text{-}\ 0.166 X_1 \text{-}\ 1.528 X_2 \text{-}\ 0.463 X_3 + 0.031 X_1 X_2 + 0.018 X_1 X_3 \text{-}\ 0.032 X_2 X_3$	0.0321	71.68	51.46
	Absolute density	$\begin{split} y &= 0.0088X_1 + 0.0034X_2 \text{ - } 0.0126X_3 + 0.0002X_1X_2 + 0.0004X_1X_3 \text{ - } \\ 0.00004X_3X_2 \end{split}$	0.0045	87.62	78.78

a Independent variables of the study: Pilsen Malt (X1), Cañihua Malt (X2) and Banana Passionfruit Juice (X3).

3.2. Effects of CM and BPJ on the physicochemical and technological characteristics of craft beers

3.2.1. Physicochemical characteristics

The absolute densities of the 13 experimental treatments ranged from 1.001 g/cm³ (E6, E9) to 1.021 g/cm³ (E8) (Table 3). The mathematical model (Table 4) indicated that both PM and CM had a positive effect on increasing density (coefficients of $0.0088X_1$ and $0.0034X_2$, respectively), whereas BPJ had a negative effect ($-0.0126X_3$), reducing density (Fig. 4). This result is consistent with those of previous studies reporting that fermentable carbohydrate content and alcohol production during fermentation influence the final beer density [56]. In our study, malting increased the carbohydrate content in CM (Table 2), which likely promoted fermentable sugar production, leading to a higher density. Similar trends were observed in beers made with malted quinoa (1.015 g/mL) and unmalted quinoa (1.011 g/mL).

The soluble solids content in the beers varied between 3° Brix (E6, E9) and 5.3° Brix (E8) (Table 3). The mathematical model (Table 4) showed that all three independent variables negatively affected this parameter ($-0.166X_1$, $-1.528X_2$, and $-0.463X_3$), although certain binary interactions between PM and CM ($0.031X_1X_2$) and PM and BPJ ($0.018X_1X_3$) favored an increase in soluble solids (Fig. 4). This suggests that malt content and fruit juice addition influence the total soluble solid content of beer, a pattern also observed in previous studies. For example, Correa et al. [57] reported an increase from 7.5° Brix to 9° Brix with 5 % mango pulp addition, and Sánchez et al. [58] recorded a rise to 6.8° Brix when substituting 15 % purple corn for barley.

The effects of CM and BPJ concentration on pH and titratable acidity are shown in Fig. 4. The pH decreased as the CM concentration increased, likely due to its influence on the fermentation rate. Table 4 indicates that both CM and BPJ significantly affected pH, with a 5 % significance level for both linear and quadratic terms. A similar trend was reported by Kumar et al. [59], indicating a reduction in pH with increasing ingredient concentrations.

The pH of CM- and BPJ-treated beers decreased slightly, whereas total acidity increased at higher BPJ concentrations (Table 3). The acidity of beer is influenced by the type and quantity of juice, as well as the fermentation conditions. The addition of fruit juice tends to lower the pH, reducing the soluble solid content due to sugar conversion into ethanol [60]. Beers with higher BPJ concentrations (10 %, 14 %, and 15 %) exhibited increased volatile acidity (Table 3), consistent with previous studies on fruit beer production, which reported higher levels of volatile compounds [61]. In addition, the low pH resulting from CM addition contributes to the creation of antimicrobial barriers, which is a property of cañihua. pH and acidity are critical parameters in beer production, as they influence the

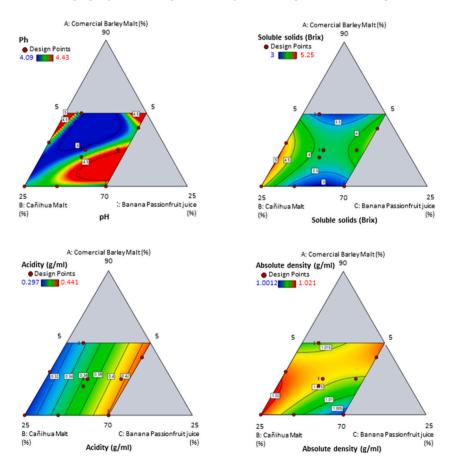


Fig. 4. Response surface 2D plot of the effect of cañihua malt (CM) and banana passionfruit juice (BPJ) on the physicochemical characteristics of craft beers.

chemical stability, microbial control, and sensory acceptance of craft beers [62].

Regarding the alcohol content, ethanol is the main product of alcoholic fermentation, defining beer classification based on alcohol concentration [63]. Our results indicate that beers containing CM and BPJ had higher alcohol content at all stages, although no significant differences were found between samples (Table 3). Rodríguez et al. [64] reported that Ale-type beers typically range between 4 % and 10 % alcohol by volume (v/v), with values between 3.3 % and 6.2 % when adding purple corn extract and passion fruit juice. In our samples, the alcohol content ranged from 4.94 % to 6.19 % (Table 3), which falls within this range.

The reduction in alcohol content with increasing CM concentration may be attributed to its lower carbohydrate content (70.51 g/ 100 g) compared with Pilsen malt (80 g/100 g). Malt plays a key role in alcoholic fermentation. In our study, the control (100 % PM) beer reached 5.07 % alcohol content, whereas beers with pseudocereals typically exceeded this value. For example, Montenegro [7] produced beer with buckwheat and amaranth (4.53 % alcohol), and Soledad & Vilavicencio [65] obtained beer with quinoa and corn (4.21 % alcohol). This trend may be linked to faster glucose degradation in pseudocereals compared to commercial grains.

The increased alcohol content observed in some treatments may be explained by two additional factors. First, *Saccharomyces cerevisiae* SafAle US-05 yeast, which may have undergone autolysis in the high-alcohol environment, releasing polysaccharides that contributed to an increase in the beverage's volume [66]. Second, banana passionfruit juice, which is naturally rich in fermentable sugars such as fructose and glucose, likely enhanced ethanol and carbon dioxide production during fermentation [67].

The addition of fruit juice enhances fermentation. Karabagias et al. [66] found that prickly pear juice addition resulted in an ABV of 5.90 %–9.60 %, while Guimarães [68] reported an ABV of 4.8 % with 15 % camu-camu pulp addition. Kumar et al. [59] recorded 9.25 % ABV when adding 19 % apple juice to a beer made with malt and African millet, demonstrating the impact of raw materials and fermentable sugars on the final beer alcohol content.

3.2.2. Technological characteristics

The EBC values of the experimental treatments ranged from 8.1 to 29.9 (Table 3), indicating light beers (<20 EBC), except for E9 and E11 (>20 EBC), which were slightly darker (Fig. 5). Compared with commercial pale ale beers, which typically range between 17 and 44 EBC [69], the literature suggests that beer color depends on the roasting level, production methods, and malt types [70]. However, no significant differences were observed among the malt formulations.

Pale ale is characterized by its reddish hue, bitter hop taste, and medium to high alcohol content, which can contribute to turbidity [71]. The turbidity values ranged from 11 to 96.5 NTU (Table 3). Although no statistically significant differences were found among malt formulations, studies have indicated that malting directly affects grain protein and polyphenol composition. During malting, proteins undergo enzymatic breakdown, releasing peptides and amino acids that form colloidal particles, increasing turbidity. Protein-rich malts, such as cañihua and quinoa [72], enhance proteolytic activity, thereby increasing suspended compounds in craft beers. Additionally, polyphenols in cañihua influence turbidity. During mashing and filtration, incomplete protein coagulation leads to complex formation with tannins (polyphenols) and β -glucans, further increasing turbidity [73]. The high fiber, starch, and polyphenol content in malted cañihua (Table 2) likely contributed to the higher number of suspended particles observed in our samples, aligning with previous studies showing that malted quinoa increases turbidity in beer [74].

Another property influenced by malt protein content is foam capacity, which ranged from 12.5 % (E3) to 101 % (E8) (Table 3). In our study, using 7–21 % canihua malt did not produce significant differences among the treatments. However, in barley and wheat malt beers, higher protein content enhances foam stability, whereas lipids and high ethanol levels negatively affect this property [75].

The bitterness values remained at 16 IBU, except for E1, E3, and E10 (15 IBU, Table 3). No significant differences were observed among experiments. Bitterness primarily arises from iso-alpha acids formed during hop boiling, with pH playing a key role in bitterness perception by affecting hop compound solubility and interaction with taste receptors [76].

3.3. Sensory analysis

The regression analysis of the sensory data indicated that none of the evaluated variables fit the mathematical model ($R^2 < 0.70$). Consequently, Tukey's test was used to differentiate the means. In general, all samples exhibited significant differences in acceptance scores for aroma, color, taste, appearance, and purchase intent, with values ranging between 6 and 7, indicating moderate preference (Table 5). Notably, the experimental beers scored higher in aroma, appearance, and purchase intent than the control beer (100 % PM).

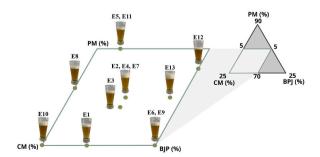


Fig. 5. Distribution of craft beers obtained according to experimental design.

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Table 5Average scores^a of acceptance and purchase intention of the beers.

Experiment	Aroma	Color	Taste	Appearance	Purchase intention
Control	7.24 ^a	7.03°	5.94 ^e	7.41 ^a	3.78 ^c
E1	6.78 ^b	7.21 ^b	6.36 ^c	7.27 ^b	$3.50^{\rm d}$
E2	7.03 ^{ab}	7.16 ^b	6.47 ^c	7.26 ^b	3.59 ^d
E3	7.11 ^a	$7.20^{\rm b}$	7.11 ^a	7.51 ^a	3.63^{d}
E4	7.03^{ab}	7.16 ^b	6.47 ^c	7.26 ^b	3.59 ^d
E5	6.72 ^c	7.23 ^b	5.94 ^e	7.12^{b}	3.48 ^d
E6	6.74 ^c	6.82 ^c	6.22^{d}	7.16 ^b	3.43 ^d
E7	7.03^{ab}	7.16 ^b	6.47 ^c	7.26 ^b	3.59^{d}
E8	6.91 ^b	6.72 ^d	6.05 ^d	6.73°	3.86 ^c
E9	6.47 ^d	6.74 ^d	6.37 ^c	6.84 ^c	3.47 ^d
E10	7.18^{a}	7.41 ^a	6.75 ^b	7.20 ^b	4.06 ^b
E11	$6.82^{\rm b}$	6.92 ^c	5.77 ^e	6.90°	3.30 ^e
E12	6.64 ^c	$7.27^{\rm b}$	6.55 ^c	7.17 ^b	4.23 ^a
E13	6.64 ^c	7.13 ^b	7.02^{a}	7.36 ^a	4.08 ^b

 $^{^{\}rm a}$ Values within a row with different letters are significantly different through Tukey's test (p < 0.05).

Beers containing up to 10 % BPJ and between 15 % and 16 % CM (E3, E4, E2, and E7) received the highest acceptance scores for aroma and appearance. However, no clear color or taste patterns were identified. The highest overall evaluation scores were achieved by samples E3 (16 % CM: 10 % BPJ) and E10 (25 % CM: 5 % BPJ), whereas the highest purchase intent scores were recorded for sample E12 (7 % CM: 15 % BPJ).

These results contrast with Deželak et al. [77], reporting lower sensory acceptance when malt was partially substituted with pseudocereals like buckwheat and quinoa, primarily due to increased astringency and bitterness. According to Cela et al. [72], these differences may be attributable to variations in consumer preferences across regions. Recent studies by Salvador-Reyes et al. [78,79] have shown that Andean grain-based products generally receive higher acceptance than those made with conventional cereals, which may explain the favorable perception of beers formulated with CM. Additionally, the incorporation of banana passionfruit juice may have mitigated the dominant flavors and aromas typically associated with pseudocereal-based malt, as previously reported by Paiva et al. [80] showed that the addition of goji berry enhances the acceptance of amber-type beer.

The CATA analysis allowed for the identification and quantification of specific sensory attributes that influenced beer acceptance. According to Cochran's Q test (Supplementary Material 2, Table S1), 34 out of the 37 evaluated attributes were significant for differentiating the samples ($p \le 0.10$). Among these, the descriptors "glossy," "golden color," and "toasted cereal aroma" were the most frequently selected, with an incidence above 50 %, highlighting their relevance in consumer perception.

Correspondence analysis (Fig. 6a) showed that components F1 and F2 accounted for 71.30 % of the variability in the data. The first component (F1 = 51.53 %) primarily differentiated samples based on color, taste, and mouthfeel, whereas the second component (F2 = 19.77 %) reflected a lesser influence of attributes related to aroma and appearance. Samples E2, E7, E4, and E3, which contained 10 % BPJ and 15-16 % CM, clustered near the control (C) and were associated with positive descriptors such as "foamy appearance," "intense caramel color," "intense sweet taste," "refreshing," "effervescent," and "delicate mouthfeel" (Fig. 6b). This suggests that these ingredient combinations preserved the sensory characteristics similar to those of the control, which may have contributed to their higher acceptance.

Conversely, beers formulated with higher CM concentrations (E1, E8, and E10) or higher BPJ levels (E12 and E13) were described as "viscous," with a "foamy appearance" and a "toasted cereal" aroma and taste. The viscosity and toasted cereal flavor can be attributed to the physicochemical properties of canihua, which has a high protein and fiber content that influences the texture and flavor profile of the final product [81,82]. On the other hand, samples E6 and E9, which contained equal proportions of CM and BPJ (15 %), were associated with attributes such as "citrus aroma," "fruity taste," and "golden color" but also with "opaque appearance." Samples E5 and E11, which contained the lowest concentrations of both ingredients, were described with less favorable terms such as "acidic taste," "citrus," and "watery mouthfeel."

The penalty-lift analysis (Fig. 7a and b) revealed that attributes such as "golden color," "glossy appearance," and "foamy texture" were the main drivers of acceptance, increasing sensory scores by up to +0.40 points. These attributes were predominantly associated with beers containing higher concentrations of CM or BPJ (E1, E8, E10, E12, and E13), where the combined presence of these ingredients ranged from 25 % to 30 %. In contrast, descriptors such as "opaque appearance" and "watery mouthfeel" significantly penalized the samples, decreasing acceptance scores by up to -0.60 points. Opacity, which is often perceived negatively in craft beers, may have contributed to the lower acceptance of these samples, as consumers tend to associate reduced transparency with lower quality [83]. Similarly, a watery mouthfeel may indicate a lack of body and density, which also diminishes consumer preference [84]. These terms were most commonly associated with samples E5 and E11, which contained the lowest concentrations of CM and BPJ. Therefore, the results suggest that the inclusion of CM and BPJ improves the sensory acceptability of craft beers, provided they are used at concentrations above 10 % to positively impact key sensory attributes.

3.4. Physicochemical and technological optimization

Optimization was conducted using the desirability function to determine the ideal formulation of PM, CM, and BPJ. The significant

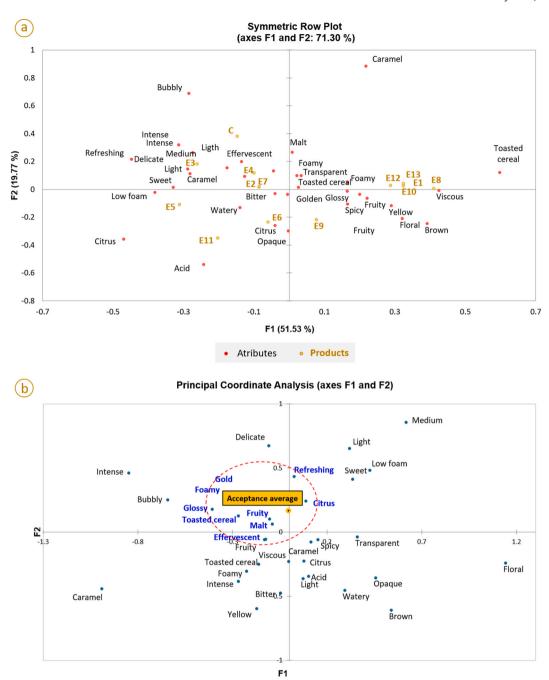


Fig. 6. a) Correspondence analysis using Hellinger distance, b) representation of first and second dimensions of the Principal Coordinate Analysis (PCoA), showing the CATA terms and overall liking.

variables identified in previous analyses (p < 0.05, $R^2 > 0.70$) were weighted based on their importance, ranging from 1 (low) to 5 (high), in accordance with the study objectives. A minimization criterion was applied for pH and acidity to comply with standard guidelines, whereas the soluble solid content was maximized to enhance the contribution of BPJ. The absolute density was maintained within the acceptable range of 1.00-1.04 g/cm³, as established by the Beer Judge Certification Program [85].

The evaluated criteria and the optimal formulation (OPM1) identified through the mathematical model are presented in Table 6. The OPM1 formulation consisted of 73.09 % PM, 21.91 % CM, and 5 % BPJ, achieving a desirability level of 94.8 %. This formulation demonstrated an optimal balance between physicochemical and technological parameters, particularly foam stability and acidity regulation, suggesting that this combination maximizes yield in craft beer production.

Based on the sensory analysis results, the OPM1 formulation is expected to achieve high consumer acceptance. Samples with similar

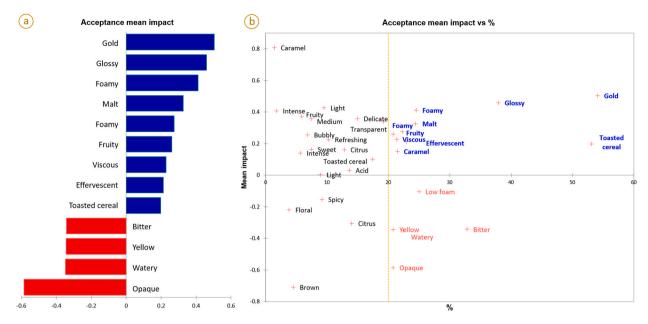


Fig. 7. Penalty-lift analysis: a) terms with highest positive (blue) or negative (red) mean impact, used by more than 20 % of consumers, b) mean impact vs. answers (%). Attributes located in the lower part of the graphic are negative.

Table 6Parameters selected for the desirability optimization method, optimal beer formulation.

Name	Criteria	Lower Limit	Upper Limit	Importance	OPM 1
X ₁ : Pilsen malt (PM)	is in range	70	80	3	73.09
X ₂ : Cañihua malt (CM)	is in range	7	25	3	21.91
X ₃ : Banana Passionfruit Juice (BPJ)	is in range	5	15	3	5
pH	minimize	4.09	4.43	3	4.06
Soluble solids (Brix)	maximize	3	5.25	3	5.05
Acidity (g/ml)	minimize	0.30	0.44	5	0.31
Turbidity (NTU)	none	11	96.50	3	14.79
Absolute density (g/ml)	is in range	1.00	1.04	3	1.02
Viscosity	none	1.32	1.47	3	1.45
Color (EBC)	none	8.11	33.39	3	11.81
Alcohol Content (%)	none	4.94	6.19	3	5.38
Foaming Capacity	none	12.5	110	3	80.24
Bitterness (IBU)	none	15	16	3	15.61
Desirability					0.948

CM and BPJ concentrations, such as E10, received higher scores for aroma and appearance, with descriptors including "refreshing," "foamy appearance," and "intense caramel color." These sensory characteristics, combined with the physicochemical findings, suggest that the optimized formulation provides a well-balanced and appealing sensory experience for consumers.

The controlled inclusion of BPJ ensured that the fruity profile remained subtle, preventing it from overpowering the malt notes. Meanwhile, the higher proportion of CM contributed to improved body, texture, and foam stability, attributes that are key to enhancing the overall acceptability of the final product.

4. Conclusion

This study optimized the formulation of an Ale-type craft beer enriched with cañihua malt (CM) and banana passionfruit juice (BPJ) using a D-optimal experimental design. The optimal formulation, consisting of 21.91 % CM and 5 % BPJ, provided a well-balanced combination of physicochemical and technological parameters, including foam stability, density, and acidity. The malting process significantly enhanced the nutritional profile of cañihua, increasing its dietary fiber content, phenolic compounds, and antioxidant capacity. These improvements contributed to greater product stability, enhanced body, and improved foaming capacity. Additionally, the incorporation of BPJ introduced fresh and fruity notes, which were positively perceived in the sensory evaluations.

The craft beers formulated in this study achieved high acceptance scores, particularly in terms of aroma, appearance, and body, along with a strong purchase intent from consumers. These findings suggest that the combination of CM and BPJ represents a

promising alternative for developing differentiated craft beers, enhancing both sensory appeal and nutritional value while promoting the utilization of native Andean ingredients in the brewing industry.

CRediT authorship contribution statement

Luz Maria Paucar-Menacho: Writing – review & editing, Writing – original draft, Visualization, Validation, Supervision, Software, Resources, Project administration, Methodology, Investigation, Funding acquisition, Formal analysis, Data curation, Conceptualization. Rebeca Salvador-Reyes: Writing – review & editing, Writing – original draft, Visualization, Methodology, Investigation, Data curation. Williams Esteward Castillo-Martinez: Writing – original draft, Validation, Resources, Project administration, Methodology, Formal analysis, Data curation. Alicia Lavado-Cruz: Writing – original draft, Validation, Methodology, Investigation, Formal analysis. Anggie Verona-Ruiz: Writing – original draft, Validation, Methodology, Formal analysis. Jordy Campos-Rodriguez: Writing – original draft, Visualization, Methodology, Formal analysis. Wilson Daniel Simpalo-Lopez: Writing – original draft, Validation, Methodology, Investigation, Formal analysis, Data curation. William López-Rodriguez: Methodology, Formal analysis. Soledad Quezada-Berrú: Visualization.

Data and code availablity statement

Data will be made available on request. For requesting data, please write to the corresponding author (luzpaucar@uns.edu.pe).

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.heliyon.2025.e42610.

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