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Agro-Processing Utilization of Buni Fruit Juice (Antidesma bunius (L.) Spreng.) in a Pasteurized Milk-Based Beverage: Effects on CIELAB Color, pH, and Titratable Acidity

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Abstract. *Buni fruit is a locally available agricultural commodity with potential application as a natural color-forming ingredient in processed beverages. This study evaluated the effects of different buni fruit juice concentrations on the physicochemical characteristics of a pasteurized milk-based beverage. The experiment followed a completely randomized design with three treatments, consisting of 2.5% (P1), 5.0% (P2), and 7.5% (P3) buni fruit juice, and five independent replicates. Each experimental unit contained 250 mL of fresh milk and was pasteurized at 63 °C for 30 min. The evaluated parameters were pH, titratable acidity expressed as lactic-acid equivalent, and CIELAB color coordinates. Data were analyzed using one-way analysis of variance followed by Tukey's honestly significant difference test at a 5% significance level. Buni fruit juice concentration significantly affected pH, titratable acidity, and all CIELAB color parameters. Increasing the juice concentration reduced the L* value from 85.650 in P1 to 71.986 in P3, indicating a progressive reduction in product lightness. The a* value increased from 0.853 to 3.500, while the b* value decreased from 6.188 to −6.908, demonstrating a color shift toward a darker red-purple appearance. P1 had a significantly higher pH than P2 and P3, whereas P2 and P3 did not differ significantly. Titratable acidity was also higher in P1 than in P2 and P3. Among the evaluated formulations, P3 produced the strongest instrumental color response. However, it could not be defined as the optimum formulation because sensory acceptance, physical stability, microbial quality, antioxidant activity, and refrigerated shelf life were not evaluated. These findings provide preliminary technical evidence supporting the use of buni fruit juice as a local color-forming ingredient in applied beverage processing.*

Keywords: ***buni fruit; agro-processing; natural colorant; pasteurized beverage; CIELAB color; physicochemical properties; titratable acidity.***

INTRODUCTION

The diversification of locally available plant resources into value-added food products is an important component of applied agro-processing. This approach can increase the utilization of underused agricultural commodities, reduce dependence on conventional food ingredients, and create new processing opportunities for rural and small-scale food industries. Fruits with distinctive colors, acidity, and phytochemical characteristics are particularly relevant because they can be processed into juices, concentrates, natural color-forming ingredients, and functional components for formulated foods. Recent developments in plant-based food processing have consequently emphasized the identification, extraction, stabilization, and application of naturally occurring flavonoids and polyphenols in different food matrices (Ciupei et al., 2024; Liga et al., 2023; Ullah et al., 2022).

Buni fruit (*Antidesma bunius* (L.) Spreng.) is a local tropical fruit with potential for further development through postharvest processing. The fruit develops a dark-red to purple-black color during ripening and is traditionally consumed fresh or processed into beverages and other food products. Recent community-based processing activities have demonstrated the feasibility of incorporating buni fruit into wet noodles and gummy confectionery, indicating that the fruit can be developed beyond its conventional uses (Canti & Yanti, 2024). However, the broader utilization of buni fruit remains limited, particularly in standardized food formulations in which ingredient concentration and product quality are evaluated instrumentally. The development of buni fruit juice as a color-forming ingredient may therefore contribute to the valorization of a local agricultural commodity while providing information relevant to small-scale agroindustry.

The technological potential of colored fruits is closely related to the presence of plant pigments and associated phenolic compounds. Flavonoids constitute a major group of plant secondary metabolites that includes flavones, flavonols, flavanones, flavanols, isoflavones, and anthocyanins (Ciupei et al., 2024; Liga et al., 2023). Anthocyanins are water-soluble pigments commonly associated with red, purple, and blue coloration in plant materials. Their extraction and technological performance depend on the plant source, maturity, extraction medium, temperature, pH, oxygen exposure, and processing conditions (Fernandes et al., 2023; Liga et al., 2023). Therefore, the incorporation of a colored fruit juice into a formulated food does not merely add color; it introduces a mixture of acids, soluble solids, pigments, phenolic compounds, and suspended plant material that may modify the physicochemical properties of the product.

The application of plant-derived pigments in food processing is increasingly relevant because color is one of the first quality attributes perceived by consumers. Nevertheless, natural pigments are generally more sensitive to processing and matrix conditions than conventional synthetic colorants. Their visual expression may change during heating, storage, exposure to light and oxygen, or interaction with other food constituents (Fernandes et al., 2023; Ngo et al., 2022; Rocchetti et al., 2022). Studies on individual citrus flavonoids have similarly shown that extraction conditions and the surrounding chemical environment affect the stability and recovery of plant bioactive compounds (Pyrzynska, 2022; Shilpa et al., 2023). These findings indicate that the performance of fruit-derived ingredients should be evaluated directly in the intended food system rather than predicted solely from the characteristics of the unprocessed fruit.

Instrumental color analysis provides an objective method for evaluating such changes. The Commission Internationale de l'Éclairage L*a*b* system describes color using three coordinates: L* represents lightness from black to white, a* represents the green-to-red axis, and b* represents the blue-to-yellow axis. In a light-colored food matrix, the incorporation of dark-red or purple fruit juice may reduce L*, increase positive a*, and shift b* according to the relative contribution of red, blue, and yellow components. These parameters are therefore useful for determining whether increasing the concentration of buni fruit juice produces a gradual and measurable color response. Instrumental measurements are also preferable to visual descriptions because the apparent color of a formulated beverage may be influenced by pigment concentration, particle dispersion, opacity, and light scattering.

The behavior of plant-derived compounds may become more complex

when they are incorporated into a protein-containing food matrix. Phenolic compounds can interact with proteins, polysaccharides, minerals, and other food constituents through non-covalent or covalent mechanisms (Ngo et al., 2022; Poojary et al., 2023; Rocchetti et al., 2022). Such interactions may influence solubility, turbidity, sedimentation, viscosity, pigment expression, and the accessibility of phenolic compounds. Covalent adducts between phenolic acids and proteins have been identified and quantified in dairy beverages, providing evidence that molecular interactions can occur when plant phenolics are introduced into protein-rich liquid foods (Poojary et al., 2023). In addition, the physicochemical behavior of protein-based beverages is affected by ingredient concentration, heat treatment, acidity, ionic conditions, and the organization of dispersed particles (Le Ba et al., 2025; Sözeri Atik et al., 2024).

A pasteurized milk-based beverage can therefore be used as an application matrix for examining the technological behavior of buni fruit juice. The purpose of using this matrix is not limited to the diversification of an animal-derived product; it also provides an applied model for studying the integration of a local fruit ingredient into a complex processed beverage. Goat milk has previously been processed into yogurt, ice cream, and other diversified products, indicating its suitability for small-scale product innovation (Mutiara Sihombing et al., 2023). Nevertheless, the present study focuses on the processing contribution of buni fruit juice, particularly its effects on measurable color and acidity-related properties.

Acidity is an important factor in the development of fruit-containing beverages because fruit juice can modify the acid–base balance of the product. The addition of organic-acid-containing material may reduce pH, while proteins, phosphates, citrates, and other components of the food matrix can resist changes through their buffering capacity. For this reason, pH and titratable acidity represent different physicochemical properties. pH expresses hydrogen-ion activity, whereas titratable acidity is determined from the volume and concentration of an alkaline solution required to reach a defined endpoint. Valid interpretation of titration results requires complete information regarding sample volume, titrant normality, dilution, endpoint determination, blank correction, and the acid equivalent used in the calculation.

Changes in acidity may also affect the physical behavior of the product. Protein-based food systems are sensitive to pH because protein solubility, aggregation, water retention, and rheological characteristics can change as acidity increases (Le Ba et al., 2025; Sözeri Atik et al., 2024). However, changes in pH alone cannot be used to confirm protein aggregation, product stability, microbial inhibition, or extended shelf life. Those properties require direct measurement. Accordingly, the effects of buni fruit juice should first be described through the parameters that were actually evaluated, without extending the conclusions to unmeasured functional or microbiological properties.

Studies concerning plant polyphenols in food systems have also emphasized that reported biological or preservative potential cannot automatically be transferred to a newly formulated product (Bešlo et al., 2022; Rocchetti et al., 2022; Ullah et al., 2022). Processing may modify the concentration, chemical structure, extractability, and interactions of phenolic compounds. In addition, the stability of plant bioactives may depend on heat treatment, pH, food composition, and storage conditions (Berenshtein et al., 2024; Fernandes et al., 2023; Pyrzynska, 2022). Therefore, although buni fruit is potentially relevant as a source of natural color and plant-

derived compounds, the present study does not claim increased antioxidant activity or functional-food status because those parameters were not measured.

Despite the potential of buni fruit for agro-processing, limited information is available concerning its concentration-dependent effects when applied to a pasteurized milk-based beverage. Existing reports have mainly described general processing applications of buni fruit, whereas quantitative information on its effects on CIELAB color coordinates, pH, and acidity-titration response in a protein-containing beverage remains limited. Furthermore, because the experimental design did not include a formulation without buni fruit juice, the study cannot determine changes relative to an untreated product. The findings must therefore be interpreted as comparisons among the three tested concentrations.

Accordingly, this study aimed to compare the CIELAB color characteristics, pH, and acidity-titration response of pasteurized milk-based beverages containing 2.5%, 5.0%, and 7.5% buni fruit juice. It was hypothesized that increasing the concentration of buni fruit juice would produce a measurable reduction in product lightness, shift the instrumental color toward red-purple coordinates, and alter the acidity-related characteristics of the beverage. The results are expected to provide preliminary technical information for the utilization of buni fruit as a local color-forming ingredient in applied agro-processing.

MATERIALS AND METHODS

Study Location and Period

The experiment was conducted from January to March 2026 at the Laboratory of Campus II, Gowa Agricultural Development Polytechnic, South Sulawesi, Indonesia. The study evaluated the application of buni fruit juice as a local plant-derived ingredient in the production of a pasteurized milk-based beverage.

Materials and Equipment

The principal materials used in the study were 3.75 L of fresh goat milk, fully ripe buni fruit (*Antidesma bunius* (L.) Spreng.), potable water, 0.1 N sodium hydroxide solution, and 1% phenolphthalein indicator. Sugar, salt, synthetic colorants, and additional natural colorants were not included in the beverage formulation.

The processing equipment consisted of a stainless-steel pan, stove, food thermometer, blender, fine-mesh filter or food-grade cheesecloth, stainless-steel stirring spoon, measuring cylinder, beaker, funnel, sterile glass bottles or food-grade plastic containers, and an ice-water bath.

The analytical equipment consisted of a calibrated digital pH meter, a colorimeter or spectrophotometer capable of measuring CIELAB coordinates, an Erlenmeyer flask, burette, burette stand, pipette, and supporting laboratory glassware.

Experimental Design

The experiment was arranged in a completely randomized design with three treatments and five independent replicates, resulting in 15 experimental units. Each replicate was prepared as an independent processing batch.

The treatments were based on the volume of buni fruit juice added relative to the initial milk volume:

P1 = 2.5% buni fruit juice;
P2 = 5.0% buni fruit juice; and
P3 = 7.5% buni fruit juice.

The statistical model was expressed as:

$$Y_{ij} = \mu + \tau_i + \varepsilon_{ij}$$

where:

Y_{ij} = observation from the i-th treatment and j-th replicate;
μ = overall mean;
τ_i = effect of the i-th buni fruit

juice concentration; and
ε_{ij} = random experimental error.

Treatment Formulation

The total milk volume of 3.75 L was divided equally among the 15 experimental units. Each experimental unit therefore contained 250 mL of fresh goat milk.

The volume of buni fruit juice added to each unit was calculated using the following equation:

$$V_j = \left(\frac{C}{100}\right) \times V_m$$

where:
V_j = volume of buni fruit juice added to the beverage;
C = treatment concentration; and
V_m = initial milk volume.

Table 1. Formulation of pasteurized milk-based beverages containing buni fruit juice

Treatment	Milk volume	Juice concentration	Buni fruit juice volume	Estimated initial mixture volume
P1	250 mL	2.5% v/v of milk	6.25 mL	256.25 mL
P2	250 mL	5.0% v/v of milk	12.50 mL	262.50 mL
P3	250 mL	7.5% v/v of milk	18.75 mL	268.75 mL

No untreated beverage containing 0% buni fruit juice was included. Therefore, the experiment compared the three tested buni fruit juice concentrations rather than their effects relative to a control beverage.

Preparation of Buni Fruit Juice

Fully ripe buni fruits were selected based on their dark-red to purple-black color. Fruits showing visible fungal growth, decay, fermentation, or severe mechanical damage were discarded.

The selected fruits were thoroughly washed under running potable water and drained. The fruits were blended with potable water at a fruit-to-water ratio of 1:1 (w/v). Blending was conducted at high speed for 3 min until a uniform fruit slurry was obtained.

The slurry was filtered through a fine-mesh filter or clean food-grade cheesecloth to remove seeds and coarse particles. The resulting buni fruit juice was collected in a clean, closed container and used immediately for beverage preparation.

When immediate use was not possible, the juice was stored at 4 °C for no longer than 24 h before processing.

Beverage Preparation and Pasteurization

For each experimental unit, 250 mL of fresh goat milk was transferred into a clean stainless-steel processing vessel. Buni fruit juice was added according to the assigned treatment concentration.

The mixture was stirred using a stainless-steel spoon until visually homogeneous. Each batch was pasteurized using the low-temperature, long-time method at 63 °C for 30 min.

The product temperature was monitored continuously using a food thermometer. Gentle stirring was maintained during heating to promote uniform heat distribution and minimize localized overheating.

Immediately after pasteurization, the beverage was rapidly cooled in an ice-water bath until its temperature reached 4 °C. The cooled beverage was transferred into sterile, tightly closed glass bottles or food-grade plastic containers.

The samples were stored at 4 °C and analyzed within 24 h after processing.

pH Measurement

The pH of each experimental unit was measured using a calibrated digital pH meter. Before analysis, the instrument was calibrated using standard buffer solutions at pH 4.00 and 7.00.

Each beverage sample was gently homogenized, and approximately 20 mL was transferred into a clean beaker. The pH electrode was immersed in the sample without touching the bottom or sides of the container.

The pH value was recorded after the displayed reading had stabilized. The electrode was rinsed with distilled water and gently dried between measurements.

Measurements were conducted at 25 °C. Each experimental unit was measured three times, and the technical readings were averaged to obtain one pH value per experimental unit.

Titratable Acidity Measurement

Titratable acidity was determined using an acid–base titration method. A 10 mL beverage sample was transferred into an Erlenmeyer flask.

Three to five drops of 1% phenolphthalein indicator were added to the sample. The sample was then titrated using 0.1 N sodium hydroxide solution until a faint-pink color persisted for approximately 30 s.

The volume of sodium hydroxide required to reach the endpoint was recorded in milliliters.

Titratable acidity was expressed as percentage lactic-acid equivalent and calculated using the following equation:

$$\text{Titratable acidity (\%)} = \frac{V_{\text{NaOH}} \times N_{\text{NaOH}} \times 90.08 \times DF}{V_s \times 10}$$

where:

V_{NaOH} = volume of sodium hydroxide used during titration, in mL;

N_{NaOH} = normality of the sodium hydroxide solution;

90.08 = molecular mass of lactic acid;

DF = dilution factor; and

V_s = volume of the beverage sample, in mL.

Because a 10 mL sample, 0.1 N sodium hydroxide, and no additional dilution were used, the equation was simplified to:

$$\text{Titratable acidity (\%)} = V_{\text{NaOH}} \times 0.09008$$

The resulting values were expressed as percentage lactic-acid equivalent.

Instrumental Color Measurement

Instrumental color was evaluated using the CIELAB color system. The measured parameters were L^*, a^*, and b^*. L^* represents lightness on a scale ranging from 0 for black to 100 for white. The a^* coordinate represents the green-to-red axis, with negative values indicating green and positive values indicating red. The b^* coordinate represents the blue-to-yellow axis, with negative values indicating blue and positive values indicating yellow.

Before measurement, each beverage sample was gently homogenized and transferred into a clean transparent sample container. A consistent sample volume and depth were maintained for all measurements.

The instrument was calibrated using the reference standard supplied by the manufacturer. Measurements were performed using a D65 illuminant, a 10° standard observer angle, and an 8 mm measurement aperture.

Each experimental unit was measured three times. The technical readings were averaged to obtain one L^*, a^*, and b^* value for each experimental unit.

Statistical Analysis

The data were analyzed using Python version 3.13.5 with SciPy version 1.17.0 and statsmodels version 0.14.6. Results were expressed as mean ± standard deviation from five independent replicates.

The effects of buni fruit juice concentration on pH, titratable acidity, L^*, a^*, and b^* were initially evaluated using one-way analysis of variance.

The normality of the model residuals was evaluated using the Shapiro–Wilk test, while homogeneity of variance was assessed using Levene's test.

When the treatment effect was significant, pairwise comparisons among treatment means were performed using Tukey's honestly significant difference test at a significance level of 5%.

The significance level was set at:

$$\alpha = 0.05$$

RESULTS AND DUSCUSSION

Overall Effects of Buni Fruit Juice Concentration

The concentration of buni fruit juice significantly affected the physicochemical characteristics of the pasteurized milk-based beverage. The mean pH, titratable acidity, and CIELAB color coordinates are presented in Table 2.

Table 2. Physicochemical characteristics of pasteurized milk-based beverages containing different concentrations of buni fruit juice

Parameter	P1: 2.5%	P2: 5.0%	P3: 7.5%	F-value	ANOVA p-value
pH	4.958 ± 0.758[a]	4.076 ± 0.032[b]	4.048 ± 0.013[b]	6.982	0.0097
Titratable acidity (% lactic acid)	0.232 ± 0.027[a]	0.081 ± 0.061[b]	0.065 ± 0.008[b]	28.24	<0.001
L*	85.650 ± 0.966[a]	77.404 ± 0.808[b]	71.986 ± 1.079[c]	258.077	<0.001
a*	0.853 ± 0.432[b]	1.874 ± 0.733[b]	3.500 ± 0.777[a]	20.138	<0.001
b*	6.188 ± 1.285[a]	2.721 ± 0.853[b]	−6.908 ± 1.645[c]	135.759	<0.001

Values are expressed as mean ± standard deviation from five independent replicates. Within the same row, values followed by different superscript letters differ significantly according to Tukey's honestly significant difference test at $p < 0.05$.

The residuals of the L*, a*, and b* data satisfied the normality assumption, and the variances were homogeneous. The color results could therefore be interpreted directly using conventional one-way analysis of variance and Tukey's test.

The residuals of the pH and titratable-acidity data did not satisfy the normality assumption. Consequently, the conventional analysis was accompanied by robust sensitivity analyses. For pH, conventional analysis of variance indicated a significant treatment effect at $p = 0.0097$, while the Kruskal–Wallis test also detected a difference among treatments at $p = 0.020$. However, Welch's analysis of variance produced $p = 0.0586$. The pH result should therefore be interpreted cautiously because the conclusion was sensitive to the statistical method used.

For titratable acidity, the treatment effect remained significant using conventional analysis of variance, Welch's analysis of variance, and the Kruskal–Wallis test. The effect of treatment on titratable acidity was therefore more statistically robust, although the unusual direction of the response requires careful interpretation.

Instrumental Color Characteristics

Buni fruit juice concentration had a highly significant effect on all CIELAB color coordinates. Increasing the juice concentration progressively reduced L*, increased a*, and shifted b* from positive to negative.

Lightness

The L* value decreased significantly from 85.650 in P1 to 77.404 in P2 and 71.986 in P3. All treatments differed significantly from one another. The reduction in L* indicates that the beverage became progressively darker as the concentration of buni fruit juice increased.

Milk is naturally light and opaque because its dispersed fat globules and protein particles scatter incident light. Incorporation of the dark-red to purple buni fruit juice increased the proportion of colored plant material in the beverage and reduced its apparent lightness. The progressive response across the three formulations suggests that the instrumental color was concentration-dependent rather than resulting from random variation.

The decrease in L* may be associated with the increasing concentration of natural pigments and suspended fruit-derived particles. Flavonoids include anthocyanins, which are associated with red, purple, and blue colors in plant materials (Ciupei et al., 2024; Liga et al., 2023). However, the present study did not determine total anthocyanin concentration or identify individual pigments. The reduction in lightness should therefore be interpreted as a measured color response to buni fruit juice addition, not as direct evidence of a specific anthocyanin concentration.

The high F-value obtained for L* and the relatively small standard deviations indicate that lightness was the most consistent color response among the evaluated parameters. From an agro-processing perspective, this result demonstrates that the visual intensity of the beverage can be adjusted by modifying the concentration of buni fruit juice.

Red–Green Coordinate

The a* value increased from 0.853 in P1 to 1.874 in P2 and 3.500 in P3. Positive a* values indicate movement toward the red axis. P3 differed significantly from P1 and P2, whereas the difference between P1 and P2 was not significant according to Tukey's test.

These results indicate that the red component of the beverage became clearly distinguishable at the highest tested concentration. Although the mean a* value increased numerically between P1 and P2, the variation among replicates was sufficiently large that the two treatments could not be separated statistically. The response therefore appeared to become more pronounced when the buni fruit juice concentration reached 7.5%.

The increased positive a* value is consistent with the incorporation of red-purple pigments from ripe buni fruit. The expression of fruit-derived pigments in a processed food depends on pigment structure, concentration, extraction conditions, pH, temperature, and interactions with other food constituents (Fernandes et al., 2023; Liga et al., 2023; Ngo et al., 2022). Because the buni fruit was blended with water at a ratio of 1:1 and filtered before use, the measured response represents the behavior of the resulting juice in the beverage matrix rather than the pigment concentration of the intact fruit.

Blue–Yellow Coordinate

The b* value declined significantly from 6.188 in P1 to 2.721 in P2 and −6.908 in P3. All three treatments differed significantly. Positive b* values in P1 and P2 indicate a yellow component, whereas the negative value in P3 indicates a shift toward the blue axis.

The change from a positive to a negative b* value was the clearest

indication that the color profile of the beverage changed qualitatively at the highest juice concentration. The combination of a positive a* value and a negative b* value in P3 is consistent with a red-purple or purple-blue appearance.

At lower buni fruit juice concentrations, the natural light-yellow appearance of the milk matrix may still have contributed substantially to the measured color. At 7.5%, the contribution of the fruit-derived pigments was sufficiently strong to shift the b* coordinate below zero.

Pigment behavior in protein-containing foods can also be influenced by interactions with macromolecules. Phenolic compounds can associate with proteins and other food constituents through non-covalent and, under some conditions, covalent interactions (Ngo et al., 2022; Rocchetti et al., 2022). Phenolic acid–protein adducts have been identified in dairy beverages, demonstrating that molecular interactions can occur in this type of matrix (Poojary et al., 2023). Nevertheless, the present experiment did not directly evaluate pigment–protein binding, and such interactions should be regarded only as a possible contributing mechanism.

Overall, P3 produced the darkest, reddest, and most blue-purple instrumental color profile. This does not necessarily mean that P3 had the best or most acceptable color because no sensory evaluation was conducted. The term "strongest color response" is therefore more appropriate than "best color."

pH

The mean pH decreased from 4.958 in P1 to 4.076 in P2 and 4.048 in P3. According to conventional analysis of variance and Tukey's test, P1 differed significantly from P2 and P3, whereas P2 and P3 did not differ.

The decrease in pH is consistent with the increasing incorporation of an acidic fruit-derived ingredient. Buni fruit juice may introduce organic acids and hydrogen ions into the beverage, thereby changing its acid–base balance. The absence of a significant difference between P2 and P3 suggests that increasing the juice concentration from 5.0% to 7.5% did not produce an additional measurable reduction in pH under the conditions of this experiment.

The milk matrix contains proteins, phosphates, citrates, and other components that contribute to its buffering capacity. Consequently, the relationship between the quantity of fruit juice added and product pH is not necessarily linear. Interactions between phenolic compounds and milk proteins may also modify the physicochemical behavior of protein-containing beverages (Ngo et al., 2022; Poojary et al., 2023; Rocchetti et al., 2022).

The mean pH values of P2 and P3 were close to 4.0. At this level of acidity, the physical stability of a milk-protein system may be affected because protein solubility and aggregation are influenced by pH. Reviews of dairy gel and beverage systems have shown that acidity and processing conditions can alter protein organization and rheological behavior (Le Ba et al., 2025; Sözeri Atik et al., 2024). However, the present study did not measure viscosity, sedimentation, coagulation, whey separation, or particle size. It cannot therefore confirm whether the lower pH caused physical destabilization.

The pH data for P1 showed substantial variation. Three replicates had values between 5.49 and 5.54, whereas the remaining two had values of 4.20 and 4.06. This produced a standard deviation of 0.758, which was considerably larger than the standard deviations of P2 and P3.

The broad distribution of P1 affected the statistical assumptions and explains why the results of conventional analysis of variance, Welch's analysis, and the Kruskal–Wallis test were not completely consistent. Possible causes

include genuine variation among independently prepared batches, differences in fruit-juice acidity, incomplete homogenization, or measurement variation. Because no documented measurement error was identified, all observations were retained.

For this reason, the pH result should be stated conservatively: the available data indicate that P1 generally had a higher pH than P2 and P3, but the magnitude of the difference should be confirmed in a future experiment with tighter process control and an untreated formulation.

Titratable Acidity

The mean titratable acidity was 0.232% lactic-acid equivalent in P1, 0.081% in P2, and 0.065% in P3. P1 differed significantly from P2 and P3, whereas P2 and P3 did not differ.

The acidity values were calculated from the recorded sodium hydroxide volumes using a conversion factor of 0.09008:

Titratable acidity (%) = VNaOH × 0.09008

The mean titrant volumes of 2.58, 0.90, and 0.72 mL therefore corresponded to approximately 0.232%, 0.081%, and 0.065% lactic-acid equivalent, respectively.

The titratable-acidity response did not follow the pattern normally expected from the pH results. As the buni fruit juice concentration increased, pH declined, but the measured titratable acidity also declined. Although pH and titratable acidity are different properties and do not necessarily change in parallel, the magnitude and direction of this result require cautious interpretation.

pH reflects hydrogen-ion activity at the time of measurement, whereas titratable acidity estimates the quantity of alkali needed to reach a specified endpoint. Titratable acidity is influenced by the buffering effects of proteins, phosphates, organic acids, and other ionizable components. Increasing the amount of a juice prepared with a 1:1 fruit-to-water ratio may also have diluted some of the buffering constituents contributed by milk. However, dilution alone may not fully explain the large difference between P1 and the other treatments.

Visual endpoint detection may have affected the acidity measurement. As the beverage became darker and more purple, the faint-pink phenolphthalein endpoint may have been increasingly difficult to identify. This may have caused the endpoint to be recorded prematurely in the more intensely colored P2 and P3 samples, resulting in a lower apparent sodium hydroxide volume.

The P2 acidity data also showed considerable dispersion. One replicate required 2.1 mL of sodium hydroxide, whereas the remaining four required only 0.4–0.7 mL. This produced a standard deviation of 0.061% lactic acid, equivalent to approximately 76% of the P2 mean. Despite this variation, conventional analysis of variance, Welch's analysis of variance, and the Kruskal–Wallis test all detected a treatment effect.

The statistical significance therefore appears robust, but the analytical meaning remains uncertain. Future studies should use a potentiometric endpoint, such as titration to a predetermined pH, rather than relying exclusively on a visual phenolphthalein endpoint in a dark-colored beverage. Automatic or pH-meter-assisted titration would reduce possible interference from the natural color of buni fruit juice.

Implications for Agro-Processing

The color results provide the strongest evidence for the technological application of buni fruit juice. Increasing the concentration produced consistent and measurable changes in L*, a*, and b*, demonstrating that buni fruit juice can function as a natural color-forming ingredient in a pasteurized milk-based beverage.

The 7.5% formulation produced the most pronounced instrumental color response. Nevertheless, the optimum concentration cannot be determined from color measurements alone. A commercially acceptable formulation must also be evaluated for taste, aroma, mouthfeel, physical stability, microbial safety, and consumer acceptance.

The pH values observed in P2 and P3 indicate that higher juice concentrations substantially acidified the beverage. This may influence product stability and sensory characteristics, but these effects were not directly measured. Additional analyses of viscosity, sedimentation, phase separation, and protein aggregation would be necessary to determine whether the formulations remained physically stable during refrigerated storage.

The acidity results also demonstrate the importance of selecting an analytical method suitable for strongly colored products. Instrument-assisted endpoint determination is recommended in subsequent work to minimize subjective interpretation of the phenolphthalein color change.

Study Limitations

The experiment did not include a formulation containing 0% buni fruit juice. Consequently, the study cannot determine the magnitude of change relative to an untreated pasteurized beverage.

The study also did not measure total anthocyanin content, total phenolic content, antioxidant activity, viscosity, phase separation, microbial quality, storage stability, aroma, taste, or consumer acceptance. The results therefore demonstrate concentration-dependent changes in instrumental color, pH, and titratable-acidity response but do not establish functional-food properties, improved shelf life, or an optimum commercial formulation.

The relatively high variability in the P1 pH data and P2 acidity data should also be recognized. Further research should include an untreated control, a larger number of independent batches, characterization of the buni fruit juice before formulation, potentiometric acidity measurement, and evaluation of physical and sensory stability during refrigerated storage.

CONCLUSION

Buni fruit juice concentration significantly influenced the instrumental color, pH, and titratable-acidity response of the pasteurized milk-based beverage. Increasing the concentration from 2.5% to 7.5% progressively reduced the L* value, increased the a* value, and shifted the b* value from positive to negative. These changes indicate that higher buni fruit juice concentrations produced a darker beverage with a stronger red-purple color.

The 2.5% treatment had a higher mean pH than the 5.0% and 7.5% treatments, whereas no significant difference was observed between the two higher concentrations. The measured titratable acidity was also higher in the 2.5% treatment than in the 5.0% and 7.5% treatments. However, the acidity results should be interpreted cautiously because the dark color of the beverage may have interfered with the visual determination of the phenolphthalein endpoint.

Among the tested formulations, the 7.5% treatment produced the strongest instrumental color response and therefore showed the greatest potential for the development of a naturally colored pasteurized beverage. Nevertheless, it cannot yet be identified as the optimum formulation because the study did not include an untreated control or evaluate sensory acceptance, physical stability, microbial quality, antioxidant activity, or refrigerated shelf life.

Overall, the findings provide preliminary technical evidence that buni fruit juice can be utilized as a local color-

forming ingredient in applied beverage processing. For further development, future studies are recommended to include a 0% buni fruit juice control, a larger number of independent processing batches, and physicochemical characterization of the fruit juice before formulation. Potentiometric titration should also be used to minimize color interference during acidity measurement. In addition, further research should assess total anthocyanin content, antioxidant activity, viscosity, phase separation, microbial quality, refrigerated shelf life, and consumer acceptance to determine the most suitable formulation for practical agro-processing applications.

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