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Development and Characterization of Jack Bean (*Canavalia ensiformis*) Plant-Based Yoghurt: Quality Modulation by Different Sucrose Levels

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Abstract

This study evaluated the effect of different sucrose concentrations on the physicochemical and microbiological characteristics of jack bean (*Canavalia ensiformis*)-based plant yoghurt. The treatments consisted of four sucrose concentrations, namely 0%, 3%, 6%, and 10%. The parameters analyzed included viable lactic acid bacteria (LAB) counts, titratable acidity (TA), pH, and total soluble solids (TSS). The results showed that within the tested sucrose concentration range, no significant differences ($p > 0.05$) were observed in viable LAB counts, TA, and pH values among treatments. All treatments produced LAB counts above 10^7 CFU/mL and TA values within the Indonesian National Standard (SNI, 2009) requirements for yoghurt products, indicating that all formulations fulfilled the criteria for plant-based yoghurt. Although the 10% sucrose treatment showed the highest LAB count and lower pH values, these results were not significantly different from the other treatments. In contrast, sucrose concentration significantly affected ($p < 0.05$) the TSS values, with increasing sucrose concentration resulting in higher TSS values. The highest TSS value was obtained in the 10% sucrose treatment. Overall, the addition of sucrose within the studied range did not significantly alter the microbiological quality, acidity, or pH of jack bean yoghurt, but it did significantly increase its total soluble solids content.

Keywords: *Canavalia Ensiformis*; Plant-Based Yoghurt; Lactic Acid Bacteria; Sucrose Concentration; Total Soluble Solids; Titratable Acidity

1. Introduction

The growing interest in plant-based fermented beverages has encouraged the exploration of alternative raw materials beyond conventional dairy substrates. Legume-based beverages are particularly promising because they contain proteins, carbohydrates, minerals, and other soluble components that can support lactic acid fermentation and contribute to product quality. Lentil, soybean, rice, oat, and chickpea-based fermented beverages have been reported as viable matrices for the development of yogurt-like products with acceptable microbiological and physicochemical characteristics (Boeck et al., 2022; Deziderio et al., 2023; Diez-Ozaeta et al., 2024; Mesquita et al., 2020).

Jack bean, locally known in Indonesia as *kacang koro pedang* (*Canavalia ensiformis*), is an underutilized tropical legume with potential application as a raw material for plant-based fermented beverages. This legume contains substantial amounts of carbohydrate and protein, making it technically relevant for further processing into milk-like extracts and fermented products. However, pretreatment is generally needed to improve its processability and to reduce undesirable characteristics associated with raw legumes. Previous studies have shown that soaking in sodium bicarbonate solution, dehulling, wet grinding, filtration, and heating are suitable steps for preparing jack bean milk as a non-dairy beverage base (Virly et al., 2024).

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The potential of jack bean extract as a fermentation medium has also been demonstrated. Fermented jack bean milk inoculated with selected lactic acid bacteria was shown to support cell growth, acid formation, and pH reduction after 24 h of fermentation. This indicates that jack bean extract contains fermentable components that can be utilized by lactic acid bacteria during product development(Sari et al., 2025).

In fermented plant-based beverages, formulation factors may influence both microbial activity and final physicochemical properties. Sucrose is commonly incorporated not only to improve sweetness, but also to provide an additional soluble carbohydrate fraction in the product. Depending on the substrate and starter culture, sucrose may affect microbial metabolism, acidification behavior, and the concentration of soluble solids. In plant-based fermented beverages, sucrose supplementation has been reported not to alter the rate of bacterial growth in some formulations, suggesting that its effect may be more pronounced on physicochemical attributes than on microbial proliferation itself. (Deziderio et al., 2023). Sugar concentration has also been investigated as a factor affecting growth and product stability in fermented chickpea coconut beverages (Mesquita et al., 2020).

Total lactic acid bacteria, titratable acidity, and pH are commonly used to describe the microbiological and acidification profile of fermented beverages. Total LAB indicates the viable bacterial population developed in the product, while titratable acidity reflects the total amount of organic acids formed during fermentation. The pH value provides complementary information by indicating the degree of acidity in the beverage system. These parameters do not always respond proportionally to changes in sucrose concentration because fermentation behavior is also governed by starter adaptation, substrate complexity, buffering components, and the duration of incubation (Sari et al., 2025; Diez-Ozaeta et al., 2024). Total soluble solids represent another important quality parameter in fermented beverages. The value measured as °Brix reflects the presence of soluble compounds, particularly added sugars, residual sugars, and other dissolved solids remaining after fermentation. For this reason, formulations with higher sucrose addition are expected to show more evident changes in total soluble solids than in microbial population or acidity-related parameters. Refractometric measurement of total soluble solids has been widely applied in fermented beverage studies to monitor changes in dissolved components during and after fermentation (Diez-Ozaeta et al., 2024; Lugo-Zarate et al., 2024).

Although jack bean has begun to receive attention as a raw material for plant-based beverages and fermented products, information concerning the effect of sucrose concentration on the quality characteristics of fermented jack bean beverage remains limited. Existing studies have primarily focused on jack bean milk pretreatment, fermentation performance of selected lactic acid bacteria, and changes in functional properties during fermentation (Virly et al., 2024; Sari et al., 2025). Further evaluation is therefore needed to determine whether sucrose addition mainly alters fermentation-related parameters or instead contributes more directly to the soluble solid fraction of the final beverage. This study aimed to evaluate the effect of different sucrose concentrations on fermented jack bean beverage based on total lactic acid bacteria, titratable acidity, pH, and total soluble solids. The findings are expected to provide a clearer understanding of the role of sucrose in shaping the microbiological and physicochemical characteristics of jack bean-based fermented beverages.

2. Materials and Methods

2.1. Materials

The main raw material used in this study was white jack bean (*C. ensiformis*). Additional ingredients included distilled water, granulated sucrose, sodium bicarbonate, and commercial plain yogurt used as the fermentation starter. The analytical reagents consisted of 0.85% physiological saline solution, de Man–Rogosa–Sharpe Agar (MRSA), phenolphthalein indicator, and 0.1 N sodium hydroxide solution. The equipment used included plastic basins, blender, cooking pan, stove, thermometer, measuring cylinders, digital balance, filter cloth, sterile spoons, glass jars, beakers, pH meter, refractometer, Petri dishes, pipettes, micropipettes, incubator, and colony counter. The general selection of materials and processing tools followed the requirements of jack bean milk and fermented beverage preparation described in previous studies (Virly et al., 2024; Sari et al., 2025).

2.2. Preparation of Jack Bean Extract

Jack bean extract was prepared with modifications from methods reported for jack bean milk processing by Virly et al. (2024) and Sari et al. (2025). Jack beans were weighed at 600 g and soaked in 0.25% sodium bicarbonate solution using a bean-to-solution ratio of 1:5 (w/v) for 24 h at room temperature. Sodium bicarbonate soaking has been applied in jack bean milk preparation to support pretreatment and facilitate subsequent processing. After soaking, the beans were rinsed thoroughly under running water and manually dehulled. The dehulled beans were then blended with water at a bean-to-water ratio of 1:2 (w/v) until a homogeneous slurry was obtained. The slurry was filtered using a filter cloth to separate the liquid extract from the solid residue. Similar wet grinding and filtration steps have been used in previous jack bean milk preparation studies.

The resulting jack bean extract was pasteurized at 85–90°C for 10–15 min using a double-boiling method. Heating treatment is commonly included in plant-based milk preparation to stabilize the liquid extract and reduce microbial contamination before fermentation. The pasteurized extract was then cooled to room temperature prior to inoculation.

2.3. Preparation of Fermented Jack Bean Beverage

The fermentation procedure was adapted with modifications from jack bean milk fermentation described by Sari et al. (2025). Sterile fermentation jars were prepared, and sucrose was added according to the treatment level of 0%, 3%, 6%, or 10%. A volume of 150 mL of pasteurized jack bean extract was transferred into each jar.

After the temperature of the extract decreased to room temperature, commercial plain yogurt was added at 10% of the total volume as the starter culture and mixed until homogeneous. The use of lactic acid bacterial inoculum in plant-based legume extracts follows the general principle of fermented beverage preparation previously applied in jack bean milk and other plant-based beverages.

The inoculated samples were incubated for 24 h at room temperature under dark conditions. After fermentation, the process was terminated by immediately transferring the products into refrigerated storage prior to analysis. A 24 h fermentation period has been used in jack bean milk fermentation studies to observe bacterial growth, acid production, and pH changes in the fermented substrate.

2.4. Determination of Total Lactic Acid Bacteria

Enumeration of total lactic acid bacteria was performed using the pour plate method on MRSA medium, adapted from Sari et al. (2025). One milliliter of fermented beverage sample was aseptically transferred into 9 mL of sterile 0.85% physiological saline solution to obtain a 10^{-1} dilution. Serial decimal dilutions were then prepared up to 10^{-6}. Aliquots of 1 mL from selected dilutions, particularly 10^{-4}, 10^{-5}, and 10^{-6}, were transferred into sterile Petri dishes. Molten MRSA medium was poured into each plate at approximately 15–20 mL and mixed carefully. The plates were allowed to solidify and incubated in an inverted position at 37°C for 48 h. Similar incubation conditions for viable LAB enumeration in fermented jack bean milk have been reported previously. After incubation, colonies were counted using a colony counter.

2.5. Determination of Titratable Acidity

Titratable acidity was determined by titration with sodium hydroxide solution using phenolphthalein as an indicator, adapted from the analytical approach reported by Sari et al. (2025) and Diez-Ozaeta et al. (2024). A 10 mL aliquot of fermented jack bean beverage was transferred into a 100 mL volumetric flask and diluted with distilled water to the calibration mark. The mixture was homogenized and filtered before titration. A 10 mL portion of the filtrate was transferred into an Erlenmeyer flask, followed by the addition of two to three drops of 0.1% phenolphthalein indicator. The solution was titrated with 0.1 N NaOH until a stable pale pink color appeared. Titration with NaOH is commonly employed to estimate acid accumulation in fermented beverage systems and is generally expressed as lactic acid equivalent. (Sari et al., 2025; Diez-Ozaeta et al., 2024).

2.6. Measurement of pH

The pH measurement was conducted using a calibrated pH meter, following procedures applied in fermented jack bean milk and plant-based fermented beverage studies. (Diez-Ozaeta et al., 2024). Prior to analysis, the pH meter was calibrated using standard buffer solutions at pH 4.00, 7.00, and 10.00. A 25 mL sample was transferred into a clean beaker. The pH electrode was rinsed with distilled water, gently dried, and immersed in the sample. The pH value was recorded after the reading stabilized. The electrode was rinsed again between samples to avoid carry-over contamination. pH monitoring is widely used to evaluate acidification in fermented plant-based beverage systems (Diez-Ozaeta et al., 2024).

2.7. Determination of Total Soluble Solids

Total soluble solids were measured using a refractometer and expressed as °Brix. The method was adapted from refractometric procedures used in fermented beverage characterization by Lugo-Zarate et al. (2024). Before measurement, the refractometer was calibrated using distilled water. A drop of the sample was placed on the prism surface, and the reading displayed by the instrument was recorded as the total soluble solids value. The prism was cleaned carefully between measurements. Refractometric determination of °Brix has been applied to describe the soluble fraction of fermented beverages, particularly in relation to residual sugars and soluble metabolic products.

2.8. Statistical Analysis

The data were analyzed using analysis of variance (ANOVA) to determine the effect of sucrose concentration on total lactic acid bacteria, titratable acidity, pH, and total soluble solids. The experiment consisted of four independent replications for each treatment, while each laboratory determination was conducted in duplicate. When the ANOVA results indicated a significant difference at $p < 0.05$, further mean comparison was performed using Duncan's Multiple Range Test (DMRT). The use of ANOVA followed by post hoc multiple comparison is commonly applied in food fermentation studies to identify treatment effects among formulations.

3. Results and discussion

3.1. Viable cells

Total lactic acid bacteria (LAB) is one of the important parameters used to determine the quality of both dairy and plant-based yoghurt (Guntiyastutik et al., 2020). According to the Indonesian National Standard (SNI, 2009), the total LAB count in yoghurt should be at least 10^7 CFU/mL. The results of the total LAB analysis of jack bean (*Canavalia ensiformis*)-based plant yoghurt beverages showed that the average LAB counts of all treatments met the required criteria for plant-based yoghurt products.

Based on the results, no significant differences ($p > 0.05$) were observed among treatments with different sucrose concentrations. The addition of sucrose may promote the growth of LAB because sucrose serves as an additional substrate that can be utilized as a nutrient source for microbial metabolism (Nurhasanah et al., 2022). The total LAB analysis also indicated that jack bean yoghurt containing the highest sucrose concentration (10%) produced the highest bacterial count. This result suggests that increasing sucrose concentration promoted greater microbial growth without inhibiting LAB proliferation due to excessive substrate levels. In addition, the protein content of jack bean may also contribute to the total LAB count, as proteins can serve as important substrates that support optimal microbial growth, thereby increasing LAB populations (Rosida et al., 2020). Moreover, a previous study reported that the addition of 8% and 12% sucrose significantly increased the total lactic acid bacteria (LAB) population of butterfly pea (*Clitoria ternatea* L.) yoghurt compared to the control yoghurt, whereas treatments with 0% and 4% sucrose showed no significant differences. In butterfly pea yogurt, LAB counts increased significantly with sucrose levels, peaking at 12% sucrose (2.86×10^7 CFU/mL) (Suharman et al., 2021).

3.2. Titratable acidity

Titratable acidity (TA) analysis is used to determine the amount of acid produced by lactic acid bacteria (LAB) during the fermentation process (Octaviani et al., 2024). The results showed that there were no significant differences ($p > 0.05$) in TA values among all treatments. The TA value of yoghurt products should range from 0.5% to 2.0% (SNI, 2009). The results obtained in this study met the required standard criteria, indicating that the products can be classified as plant-based yoghurt.

Higher TA values are associated with the increased conversion of substrates, sugars, and other nutrients into lactic acid by LAB during fermentation (Djali et al., 2017). The results of the TA analysis of jack bean (*Canavalia ensiformis*)-based plant yoghurt showed that treatments with sucrose addition produced higher total acid values compared to the treatment without sucrose addition. In a previous study, higher sucrose concentrations (8% and 12%) significantly increased the lactic acid content of butterfly pea (*Clitoria ternatea* L.) yoghurt compared to the control, while the addition of 0% and 4% sucrose showed no significant differences. The butterfly pea yogurt with 12% sucrose exhibited the highest lactic acid concentration (2.0%) (Suharman et al., 2021). This finding reported that sucrose, as an additional substrate, enriched the nutrient content available for LAB metabolism and was subsequently converted into lactic acid. Therefore, higher lactic acid production resulted in higher total acidity values in the yoghurt products.

3.3. pH

In this study, there were no significant differences ($p > 0.05$) for all treatments. The decrease in pH value in yoghurt products is generally caused by lactic acid bacteria (LAB) converting sugars into organic acids, mainly lactic acid. These organic acids release H^+ ions during fermentation, resulting in increased acidity due to lactic acid production by LAB from the carbohydrate content of jack bean-based products (Anggraini et al., 2021). The total LAB count is generally closely associated with pH values in yoghurt products, where higher LAB populations lead to lower pH values (Rosida et al., 2020).

Higher sucrose concentration resulted in lower pH values. This condition may be attributed to the higher availability of sucrose as a substrate, which promoted LAB growth and consequently increased lactic acid production. During fermentation, the added sucrose was metabolized into secondary metabolites, particularly lactic acid, thereby increasing the acidity of the product. As the lactic acid content increased, the pH value of the plant-based yoghurt decreased along with the reduction of sugar content in the product (Nurhasanah et al., 2022).

3.4. Total soluble solids

The ANOVA analysis revealed that different sucrose concentrations significantly affected ($p < 0.05$) the total soluble solids (TSS) of jack bean (*Canavalia ensiformis*)-based plant yoghurt. The sucrose concentration contributes to the overall solids content in yogurt. The observed TSS values were likely associated with the presence of residual sucrose, lactic acid, and other soluble compounds derived from the raw materials. During fermentation, both added sucrose and naturally occurring sugars in the jack bean were metabolized by lactic acid bacteria (LAB) as energy sources. Consequently, the remaining sugars, together with the organic acids produced during fermentation, contributed to the dissolved solids content of the yoghurt product (Sulityo et al., 2024).

Yoghurt with sucrose addition had higher TSS values than the control treatment. This result was likely due to the higher amounts of residual sugars and lactic acid produced during fermentation as the sucrose concentration increased (Duhe et al., 2025). Among all samples, treatments with 10% sucrose produced the highest TSS value, suggesting that increasing sucrose concentration increased the dissolved solids content of jack bean-based plant yoghurt. According to the Indonesian National Standard (SNI, 2009), yoghurt products should have a minimum TSS value of 8.2%; therefore, treatments with sucrose 6% and 10% fulfilled the required standard.

Table 1 Viable lactic acid bacteria (LAB) counts of jack bean yoghurt with different concentrations of glucose

Sucrose concentration	Viable cells (log CFU/mL)
0%	7.97 ± 0.45^a
3%	7.75 ± 0.47^a
6%	7.86 ± 0.45^a
10%	8.06 ± 0.43^a

Values are presented as mean ± standard deviation. No significant differences among treatments were observed according to ANOVA at $p > 0.05$

Table 2 Titrable acidity of jack bean yoghurt with different concentrations of glucose

Sucrose concentration	Titrable acidity (%)
0%	0.97 ± 0.28^a
3%	1.27 ± 0.51^a
6%	0.99 ± 0.47^a
10%	1.04 ± 0.56^a

Values are presented as mean ± standard deviation. No significant differences among treatments were observed according to ANOVA at $p > 0.05$.

Table 3 pH value of jack bean yoghurt with different concentrations of glucose

Sucrose concentration	pH
0%	4.09 ± 0.10^a
3%	4.01 ± 0.23^a
6%	4.17 ± 0.36^a
10%	4.01 ± 0.20^a

Values are presented as mean ± standard deviation. No significant differences among treatments were observed according to ANOVA at $p > 0.05$.

Table 4 Total soluble solids of jack bean yoghurt with different concentrations of glucose

Sucrose concentration	Total soluble solids (°Brix)
0%	3.42±1.23^{a}
3%	7.04±1.76^{b}
6%	9.24±0.76^{b}
10%	12.58±2.77^{c}

Values are presented as mean ± standard deviation. Different superscript letters within the same column indicate significant differences among treatments according to Duncan's Multiple Range Test (DMRT) at $p < 0.05$

4. Conclusion

Different sucrose concentrations (0–10%) influenced the characteristics of jack bean (*Canavalia ensiformis*)-based plant yoghurt. The addition of sucrose did not significantly affect viable LAB counts, titratable acidity, or pH values, but significantly increased the total soluble solids (TSS) content of the yoghurt products. All treatments met requirements for yoghurt, with LAB counts above 10^7 CFU/mL and titratable acidity values within the acceptable range. Among all treatments, the yoghurt supplemented with 10% sucrose showed the most favorable overall characteristics, including the highest viable LAB count and TSS value, suggesting that this formulation may be the most suitable for jack bean-based plant yoghurt.

Compliance with ethical standards

Disclosure of conflict of interest

No conflict of interest to be disclosed.

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