JPPIPA 11(2) (2025)

Jurnal Penelitian Pendidikan IPA

Journal of Research in Science Education

http://jppipa.unram.ac.id/index.php/jppipa/index

Physicochemical Properties of Yoghurt Analog from Peanut and Soy Milk

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Received: December 16, 2024
Revised: January 30, 2025
Accepted: February 25, 2025
Published: February 28, 2025

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DOI: 10.29303/jppipa.v11i2.10058

Abstract: The global demand for plant-based dairy alternatives is driven by health, environmental, and food allergy or intolerance concerns. This movement has promoted plant-based yoghurts, which are nutrient-rich and non-dairy. This study examines the physicochemical and sensory characteristics of yogurts made from peanut milk and soy milk, both with and without the presence of filler, a prebiotic polysaccharide. Peanut milk yogurt exhibited a greater fat content of 11.2% wb, protein levels at 6.2% wb, and increased viscosity when compared to soy yogurt. However, soy yogurt exhibited elevated microbial loads (1.5×10^{11} CFU/mL) and protein content when supplemented with inulin, suggesting improved fermentation activity. Both yogurts displayed pH levels appropriate for consumption (4.1–4.6), with peanut milk yogurt demonstrating a marginally higher acidity compared to soy yogurt. The addition of filler had a notable effect on the sensory profile, enhancing texture, taste, and overall acceptability for both peanut milk yogurt and soy yogurt. It is noteworthy that filler-enriched peanut milk yogurt exhibited diminished microbial loads and decreased viscosity, probably as a result of unutilized carbohydrate content and the presence of inhibitory metabolites from fermentation. Proximate analysis indicated that peanut milk yogurt has the potential to function as an effective plant-based yogurt alternative, exhibiting quality comparable to soy yogurt. Organoleptic testing revealed a clear consumer preference for formulations supplemented with inulin, resulting in ratings ranging from "like slightly" to "like extremely." This thorough examination offers significant perspectives on the possibilities of legume-based yogurts in addressing the increasing consumer interest in healthy, plant-derived dairy substitutes.

Keywords: Inulin; Peanut milk; Plant-based yoghurt; Soy yoghurt

Introduction

The growing demand for plant-based dairy replacements reflects a global shift in dietary preferences driven by health concerns, environmental issues, and food allergies or intolerances. Plant-based yogurts have become a prominent segment within this movement, providing a non-dairy, rich in nutrients option to regular dairy products (Grasso et al., 2020). Legumes, such as soybeans and peanuts, are notable among plant sources for their high protein content, beneficial fats, and bioactive substances. These properties make them promising candidates for yogurt production, with potential nutritional and functional benefits comparable to those found in conventional dairy yogurts (Baskar et al., 2022).

Soy-based yogurt is the most extensively studied plant-based yogurt, with a focus on fermentation profile,

How to Cite:
Kusumahastuti, D. K. A., Lewerissa, K. B., Cahyanti, M. N., Hartini, S., Natalia, R. D., & Nugrahani, Y. F. (2025). Physicochemical Properties of Yoghurt Analog from Peanut and Soy Milk . *Jurnal Penelitian Pendidikan IPA*, *11*(2), 614–620. https://doi.org/10.29303/jppipa.v11i2.10058

sensory characteristics, and functional properties (Dhakal et al., 2023; Otolowo et al., 2022). Numerous studies have shown that fermenting soy milk with lactic acid bacteria produces a product with excellent texture and flavor (Huang et al., 2022). In contrast, research on peanut-based yogurt is limited, often focusing on sensory evaluation rather than a thorough examination of its nutritional or functional properties (Mawunyo Kwasi Fidelis et al., 2014). This discrepancy demonstrates a significant research gap in the comprehensive evaluation of peanut yogurt, especially when compared to the widely reported development of soy-based alternatives.

Another significant yet underexplored component of plant-based yogurt research is the comparison of proximate compositions (Kpodo et al., 2016). Protein, fat, carbohydrate, ash, and moisture content are all key factors in defining yogurt's nutritional quality and customer attraction. While research on soy-based yogurt have yielded good nutritional profiles, the potential for peanut-based yogurt to provide equal or higher benefits is unknown. Furthermore, there are few direct comparisons between these two legume-based yogurts in the literature, leaving numerous questions unanswered about their nutritional advantages.

Understanding the proximate composition of these products will be critical not only for nutritional optimization, but also for product stability and adherence to food regulations. These proximate values influence features such as texture, water holding capacity, and fermentation efficiency, all of which are important in consumer satisfaction and industrial scaling-up operations (Hartono et al., 2024). This gap must be bridged by rigorous comparative research to assist the development of plant-based yogurt that fulfills high quality, nutritional balance, and commercial viability standards.

In order to address these knowledge gaps, this study compares and evaluates the proximate composition of yogurt prepared from peanuts and soybeans. The results of this study will be very helpful in shedding light on the nutraceutical potential of these legumes as a base for plant-based yogurts. Therefore, it is anticipated that the findings will support in directing future studies and products being developed in this emerging field of plant-based dairy products.

Method

Materials in this research are: peanut and soybean from traditional market in Salatiga.

Preparation of Peanut and Soy Milk (Ojo & Arowolo, 2019).

The initial phase consists of preparing beans milk through the process of washing and soaking peanuts for an extended period, ideally overnight. The beans undergo a peeling process followed by blanching in warm water for a duration of 2 minutes. The beans are combined with water in a 1:2 ratio and blended until a smooth consistency is achieved. The mixture is then strained through a filter and transferred into a pot. The beans milk undergoes pasteurization for 30 minutes at a temperature of 85°C, followed by cooling to 42°C to create the ideal growth conditions for the starter culture.

P*reparation of Beans Milk Yogurt (Ojo & Arowolo, 2019)*

The beans milk is chilled before the starter culture is introduced. Each milk is divided into two bottles, with one bottle containing 1.5% filler. The milk undergoes incubation at a temperature range of 35-37°C for different time intervals of 0, 3, 4.5, 6, and 7.5 hours. Following the incubation period, the yogurt is placed in a refrigerator at 4°C to effectively stop the fermentation process.

Analysis of Physicochemical Properties

pH Measurement (Rasbawati et al., 2019)

pH measurements were conducted with a pH meter. Before calibration with a buffer solution of pH 7, the electrode was initially cleansed with distilled water and then wiped with tissue. Finally, the electrode was immersed in the yogurt sample, and readings were obtained after stabilization. Universal pH indicators were additionally employed for verification purposes.

Total Acid Testing (Fauziah et al., 2023)

1 mL sample was introduced into an Erlenmeyer flask, subsequently diluted with 9 mL of distilled water, and combined with 3 drops of a 1% phenolphthalein indicator. The sample was titrated with 0.1 N NaOH until a pink endpoint was achieved. The calculation of the total acid percentage was performed using Formula 1.

$$\%\ \text{Total acid} = \frac{\text{V NaOH} \times \text{N NaOH} \times 90 \times 100}{\text{Vsampel} \times 1000} \quad (1)$$

Proximate Examination

Ash Content *Determination (Ojo & Arowolo, 2019)*

The samples were ashed in a furnace at 600°C for 6 hours until white or gray ash was formed in porcelain crucibles. The crucibles were weighed after cooling in a desiccator. The ash content was determined using:

$$\%\ \text{Ash content} = \frac{\text{Wc-Wa}}{\text{Wb-Wa}} \times 100\% \quad (2)$$

Where:

Wa represents the weight of an unfilled crucible.

The weight of the crucible with the sample prior to ashing is denoted as Wb.

Wc is the weight of the crucible after ashing with the sample.

Determination of Fat Content (Harmawan, 2022)

Fat content was assessed through the utilization of a separatory funnel. The sample was vigorously stirring and allowed to separate after being mixed with n-hexane. The n-hexane layer went through evaporation via a rotary evaporator, followed by drying the extracted fat at 105 °C, cooled, and weighted. Fat content was determined as follows:

$$\text{Fat content (\%)} = \frac{W2}{W1} \times 100\% \quad (3)$$

Where:
W1 represents the mass prior to the extraction process.
W2 represents the weight following the extraction process.

Determination of Protein Content (Asa et al., 2023)

The Kjeldahl method was employed to figure out the protein content. A 2 g sample had digestion with 7.5 g of Na_2SO_4, 0.4 g of $CuSO_4 \cdot 5H_2O$, and 10 mL of concentrated H_2SO_4 until a clear solution was achieved. The solution underwent dilution, distillation, and titration using 0.1 N HCl. The calculation of protein content was performed as follows:

$$\text{\% Nitrogen} = \frac{0{,}00014 \times \text{V titrant} \times 50}{\text{sample weight}} \times 100\% \quad (4)$$

Determination of Moisture Content (Ojo & Arowolo, 2019)

The samples were desiccated in an oven at 105°C for 3 hours after being weighed. After cooling in a desiccator, the samples were reweighed until a consistent weight was achieved. Moisture content was determined as follows:

$$\text{Moisture Content (\%)} = \frac{(W1\text{-}W2) \times 100\%}{W1\text{-}W} \quad (5)$$

Where:
W1 represents the mass prior to the drying process.
W2 represents the weight following the drying process.
W represents the weight of the empty crucible.

Determination of Carbohydrate Content (Ojo & Arowolo, 2019)

The carbohydrate content was calculated by deducting the percentages of moisture, protein, fat, and ash from a total of 100%.

Count of Lactic Acid Bacteria (Nugroho et al., 2023)

The Standard Plate Count method was employed to ascertain the count on de Man Rogosa and Sharpe Agar (MRSA). There were serial dilutions of the yogurt sample (1:9) prepared up to 10^{-8}. Samples that were diluted to concentrations of 10^{-7} and 10^{-8} were plated on MRSA and incubated at a temperature of 37°C for a duration of 48 hours. Bacterial colonies were quantified by employing the following method:

$$\text{CFU/mL} = \frac{\text{colony number per plate}}{2} \times \frac{1}{\text{dilution factor}} \quad (6)$$

Measurement of Viscosity (Ojo & Arowolo, 2019)

The measurement of viscosity was conducted utilizing a viscometer. The spindle readings were taken after the yogurt sample was transferred into a beaker.

Sensory Analysis (Ojo & Arowolo, 2019)

Ten panelists assessed sensory parameters, such as aroma, appearance, color, flavor, texture, consistency, and overall acceptance, in accordance with a hedonic scale that ranged from "like extremely" to "dislike extremely." Each sample was presented in a randomized cup, with a capacity of 25 mL, following overnight storage at 4–5°C.

Result and Discussion

Figures 1 a and b showed the peanut yogurt and soy yogurt, respectively in this study.

a b

Figure 1. Peanut yogurt (a) and Soy yogurt (b)

Physicochemical properties of peanut milk yoghurt

Proximate analysis, viscosity, pH, acidity, and total bacteria of peanut milk yoghurt with and without inulin addition are shown in Table 1. It is clear that peanut milk is quite suitable for lactic acid bacteria to propagate. The number of bacteria reach 11 log 10 CFU/ml for control and the one with inulin addition. Many studies presented the positive effect of inulin for the growth of lactic acid bacteria, as it can serve as prebiotic (Teferra, 2021). However, data in this study show lower microbial load in yoghurt with inulin compared to control. One possible mechanism for lower microbial load is the accumulation of some inhibitory metabolites produced during fermentation (Ojo & Arowolo, 2019). However, more investigations are needed to clearly explain this phenomenon. The finding also relates to the carbohydrate content in yoghurt with inulin. Its carbohydrate content is much higher than that of control, since inulin is a polysaccharide and a member of carbohydrate. High value carbohydrate content can also indicate that the prebiotic has not yet effectively consumed by the bacteria. This situation leads to lower total bacterial count.

Table 1. Proximate Analysis, Viscosity, pH, Acidity, and Total Bacteria of Peanut Yoghurt Control and With Filler Addition

Parameter	Control	With Filler
Water (%wb)	78.4± 0.64	77.5 ± 2.1
Ash (%wb)	0.6 ± 0.0	0.6 ± 0.0
Fat (%wb)	11.2 ± 1.2	10.5± 0.5
Protein (%wb)	6.2± 0.5	4.7 ± 0.7
Carbohydrate (%wb)	3.6 ± 0.6	6.6 ± 2.5
Viscocity (mPa.s)	1072.97 ± 25.42	897.27 ± 138.99
pH	4.5 ± 0.1	4.4 ± 0.0
Acidity (%)	0.97 ± 0.07	0.88 ± 0.47
Total bacteria (CFU/mL)	1.6×10^{11}	1.3×10^{11}

Higher microbial load in control causes higher titratable acidity. This is reasonable since higher microbial load leads to higher organic acid productivity. The pH values of control and the one with inulin addition do not much differ (pH 4,5 vs pH 4.4). The suitable pH value for consumption in peanut milk yoghurt was in the range of 4.1-4.6 (Arslan, 2018). Data shows that the pH values for both treatments are between those ranges. These pH value causes protein aggregation, which eventually affect the final texture of the product (Dhakal et al., 2023).

Protein content in control is much higher than that of the one with inulin addition. Taking into consideration, that the microbial load in control is much higher than that of inulin addition, it is predicted that proteolysis activity from the starter in control causes higher protein content (Ojo & Arowolo, 2019). It is commonly known that protein contents are much affected by the proteolytic activity.

The results show that the viscosity of peanut milk yoghurt with inulin addition has lower viscosity than that of control. Similar result was found by (Bulca & Büyükgümüş, 2024). In that study, exopolysaccharides (EPS) were added instead of insulin, and addition of EPS caused decrease in viscosity. Both inulin and exopolysaccharides are polysaccharides but differ in type. However, there are conflicting result regarding the addition of polysaccharides to yoghurt product (Ramchandran & Shah, 2009) . For example, studies by (Rezaei et al., 2014) found that adding inulin resulted an increase in viscosity. It is believed that fiber addition improved the texture and viscosity. The discrepancy of the result on the viscosity of peanut milk yoghurt can be caused by different strains and strain's concentration added to yoghurt. In addition, some researcher found that type of protein, and protein content also affect the viscosity of yoghurt (Bulca & Büyükgümüş, 2024). The higher the protein content, the higher the viscosity. Based on Table 1, it can be seen that protein content of control is much higher than that of yoghurt with inulin. Lower viscosity designated the incompatibility of insulin to form gel in yoghurt (Zhou et al., 2024). Other authors also indicated negative effect of inulin addition to yoghurt when added excessively. It can cause viscosity decreased and increase syneresis (Zhou et al., 2024).

Other parameter observed in this study, i.e. water, ash, and fat content of both yoghurt products do not vary. Water contents are in the range of 77-78%wb, ash content is 0.6%wb, and fat content around 10-11%wb. The fat content of yoghurt is much higher compared to other macronutrients content. Peanut is a legume rich in essential fatty acids, such as oleic and linoleic acids (Bulca & Büyükgümüş, 2024) and other authors stated that the fat content of peanut is around 45-60%(Gamli & Atasoy, 2018).

Physicochemical properties of soy milk yoghurt

Table 2 shows the proximate analysis, viscosity, pH, acidity, and total microbial load of soy yoghurt with and without inulin addition. The water content of soy yoghurt is in the range of 87.3-87.9%. The content is lower than that reported by other authors, i.e. in the of 91,9% (Farinde et al., 2009), 94-95% (Mehaya et al., 2023). The discrepancy may be due to different starter used in those studies.

Table 2. Proximate Analysis, Viscosity, pH, Acidity, and Total Bacteria of Soy Yoghurt Control and With Inulin Addition

Parameter	Control	With Inulin
Water (%wb)	87.9 ± 0.48	87.3 ± 1.7
Ash (%wb)	0.7 ± 0.0	0.6 ± 0.0
Fat (%wb)	3.6 ± 0.2	4.3 ± 0.3
Protein (%wb)	5.5 ± 0.3	5.9 ± 0.6
Carbohydrate (%wb)	2.2 ± 0.2	1.9 ± 0.7
Viscocity (mPa.s)	1284.0 ± 0.6	1151.0 ± 13.9
pH	4.6 ± 0.2	4.6 ± 0.2
Acidity (%)	0.82 ± 0.03	0.65 ± 0.02
Total bacteria (CFU/mL)	1.3×10^{11}	1.5×10^{11}

The microbial loads of soymilk yoghurt were higher in inulin added soy milk. Inulin is known as prebiotic and therefore can increase the viability of lactic acid bacteria. Nevertheless, some authors indicated that bacterial growth varies with strains (Donkor et al., 2007).

Protein content of soy yoghurt was higher in inulin added yoghurt. There is a linear correlation between high microbial loads and protein content. Higher viable bacteria mean higher proteolysis activity, which leads to higher protein content (Shahandasht et al., 2024). In case of carbohydrate content, it seems that carbohydrate as a carbon source is effectively taken by bacteria in yoghurt with inulin addition, so they propagate more than that of control. Proteolytic activity of the higher microbial load in soy yoghurt with inulin can be a cause for lower viscosity in the product (Fang et al., 2024). However, viscosity of the product was affected by the type of strain (Lim, 2016). Study by (Lim, 2016) revealed that total viable of *S. thermophilus* (1.8 x10^{10} CFU/mL) cause higher viscosity than that with *L. bulgaricus* (2.6 x 10^8 CFU/mL) in soy yoghurt.

In this study, soy yoghurt with and without inulin addition have the same pH, but different acidity. This phenomenon (similar pH but difference acidity) also occurs in peanut milk with inulin addition (see Table 1). It is predicted that inulin addition somehow affects the buffer system in the product.

Fat content in soy yoghurt with inulin addition was less than that of control. Further study needs to be done to investigate this condition.

Proximate analysis comparison of peanut milk yoghurt and soy yoghurt

Based on Table 1 and 2, it clearly seen that the water content of soy yoghurt approximately 12% higher than those of peanut yoghurt. This is understandable since the water content of soy milk is 88-91%(Aydar et al., 2020) and peanut milk is 75.8% (Singh et al., 2018). In case of ash contents, the content of soy yoghurt and peanut milk yoghurt is quite similar. This also occurs in pH value for both yoghurts. Both types of yoghurt have pH values that are in the range of suitable pH for plant-based yoghurt (Arslan, 2018).

The microbial loads in soy yoghurt are much higher than that in peanut milk yoghurt. This indicated that soy milk is more compatible for lactic acid bacteria to grow. Many researches have been done to investigate lactic acid bacteria fermentation in soy milk (Harper et al., 2022). Many authors had reported that proteolytic activity in soy milk was in line with the bacterial growth (Donkor et al., 2007).

The fat content of peanut milk yoghurt is much higher than that of soy yoghurt. Peanut milk, like other peanut products are rich in fat (49.24/100 g) (Paul et al., 2020). The viscosity of soy yoghurt is higher than the viscosity of peanut milk yoghurt. This finding was correlated well with other study, which stated that the viscosity and firmness of peanut milk yoghurt were low compared to soy yoghurt (Bansal et al., 2015).

Organoleptic test on peanut milk yogurt and soy yoghurt

Table 3 shows the data of organoleptic test for both type of yoghurt. It is clearly seen that for most parameters the one with inulin addition has higher score than those of control, except for color in soy yoghurt, and texture in peanut milk yoghurt. The hedonic test for soy yoghurt has higher score than peanut milk yoghurt. Soy yoghurt is more available in the market than peanut milk yoghurt, therefore panelists are more familiar with the taste, aroma, color, and texture of soy yoghurt. In addition, peanut has peanut-beany flavor. It also has light brown color and peanut smell that make the product is less acceptable (Sakthi et al., 2020). Interesting to see, the inulin addition can increase the panelists' preference for both type of plant-based yoghurt. The best formulations for soy yoghurt and peanut milk yoghurt are those with inulin addition, and their score for all parameter tested are in the range of 4-5.0 (like slightly- like extremely).

Table 3. Organoleptic test on peanut milk yoghurt and soy yoghurt (control and with inulin addition)

	Soy yoghurt		Peanut milk yoghurt	
	Control	With inulin	Control	With inulin
Aroma	4.4	4.6	3.8	4.0
Color	5.1	5.0	4.6	4.9
Taste	3.9	4.6	3.6	4.0
Texture	3.6	4.5	4.5	4.1

Conclusion

Peanut milk and soy yoghurt were successfully made with and without inulin addition. Data showed that peanut yoghurt without inulin addition (control) has higher protein, fat, acidity content, and microbial load compared to the one with inulin addition. Proximate analysis showed that peanut milk yoghurt could be an option for plant-based yoghurt analog. In regard to soy yoghurt, it can be concluded that the microbial loads are higher in soy yoghurt with inulin addition, followed by higher protein content due to proteolytic activities of microbial enzyme. In case of carbohydrate content, it seems that carbohydrate as a carbon source is effectively taken by bacteria in yoghurt with inulin. Both types of yoghurt have pH values that are in the range of suitable pH for plant-based yoghurt. For sensory test, the best formulation of plant-based yoghurt is the one with inulin addition. Both have sensory assessment in the rage of like slightly to like extremely in terms of aroma, color, taste, and texture.

Acknowledgments

The authors are grateful for the financial support from Satya Wacana Christian University with Contract Number: 075/SPK-PF/RIK/08/2024.

Author Contributions

Dewi Kurnianingsih Arum Kusumahastuti, Margareta Novian Cahyanti, Sri Hartini and Karina B. Lewerissa conducted the research methods, formal analysis, wrote and revised the manuscript. Rifka Dwi Natalia and Yulia Frida Nugrahani performed the experiments. All authors agreed with the final version of the manuscript.

Funding
This research were funded by Direktorat Riset dan Pengabdian Masyarakat Satya Wacana Christian University, Contract Number: 075/SPK-PF/RIK/08/2024.

Conflicts of Interest
The authors declare no conflict of interest. The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript; or in the decision to publish the results.

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