Food Research 7 (1) : 177 - 181 (February 2023)
Journal homepage: https://www.myfoodresearch.com

FOOD RESEARCH

FULL PAPER

Effect of incubation temperature, time and skimmed milk ratio on the quality of peanut kefir

[1,3]Lubna, M.H., [2,*]Rifda, N., [2]Erminawati, M., [2]Tir, Y., [3]Ibrahim, A.I. and [3]Khideer, I.K.

[1]*Post-graduate Student at Faculty of Agriculture, Jenderal Soedirman University, Purwokerto, Indonesia*
[2]*Department of Food Technology, Faculty of Agriculture, Jenderal Soedirman University, Purwokerto, Indonesia*
[3]*Department of Food Science and Technology, Faculty of Agriculture, Omdurman Islamic University, Omdurman, Sudan*

Article history:
Received: 9 October 2021
Received in revised form: 16 November 2021
Accepted: 6 February 2022
Available Online: 10 February 2023

Keywords:
Peanut milk,
Fermentation,
Kefir,
Physicochemical,
Sensory properties

DOI:
https://doi.org/10.26656/fr.2017.7(1).687

Abstract

Peanut milk contains a high percentage of protein, and it is cheap compared to animal milk. This present investigation aimed to study the influence of incubation temperature and skimmed milk ratio on the physicochemical properties, microbiological and sensory characteristics of peanut kefir. The method of the Association of Official Analytical Chemistry International (AOAC) and Indonesian National Standard (SNI) was used to analyze the physicochemical properties of peanut kefir. Analysis of variance (ANOVA) was performed to evaluate the difference between data by SPSS Version 17 and Microsoft Excel 2016. peanut kefir was made by using different incubation temperatures (37 and 45° C), incubation times (18 and 24 hrs) and skimmed milk percentages (3 and 5%). The results showed no significant difference (P≤0.05) between the physicochemical properties of peanut kefir samples in PH, viscosity and colour. There was a significant difference (P≤0.05) between each peanut yoghurt sample in culture cells (TPC) and total solids. General results from this present study established that producing kefir from peanut milk by incubation temperature at 37°C for 18 hrs and skimmed milk ratio of 5%, it is capable of improving the physicochemical and sensory properties of peanut kefir based on reducing the aroma of peanut.

1. Introduction

Globally, fermented beverages are in high demand due to their health and immunity stimulation properties (Puerari *et al*., 2012; Corona *et al*., 2016), also it has health benefits because they contain microorganisms such as Lactic Acid Bacteria (Mizielin *et al*., 2018; Roselló-Soto *et al*., 2018). Kefir is consumed world-widely as a fermented milk beverage, it is very important for promoting-consumers health, also it is a famous drink, especially in the Caucasia areas (Satir and Guzel-Seydim, 2016). The kefir flavour and taste are produced from symbiotic relations between yeasts and some bacteria types such as acetic acid bacteria and lactic acid bacteria (Atalar and Dervisoglu, 2015). Kefir has an important part in human nutrition because of its natural probiotic diversity and nutritive content, it is produced from cow, eve, buffalo and camel milk (Gul *et al*., 2015). On the other hand, it can be produced from soybean and peanut milk. The type of milk, starter culture and production conditions affect the final properties of kefir beverages like microbiological, structural, sensorial, physicochemical and functional properties (Bensmira and Jiang 2015).

Peanut products such as flour and protein concentrates are commonly used to fortify foods due to their high protein content, which has much higher digestibility than wheat and soy flour and compared to animal protein. Peanuts have been used as a source of protein and edible oil and are of high value for human and animal nutrition in many countries. Contains a high value of fat (45-60%), proteins (20-30%), carbohydrates (18-20%), essential vitamins, and minerals such as vitamin A, vitamin E, folic acid, mg, zinc, iron, calcium and dietary fibre (Dogaka, 2015).

The valuable contents of peanuts have encouraged some suggestions to increase their consumption (outside of cookies) such as imitation milk, which is produced from vegetables, such as almonds, peanuts and soybeans (Yadav, 2016). Some reports promote the idea of

*Corresponding author.
Email: *rnaufalin@yahoo.co.id*

eISSN: 2550-2166 / © 2023 The Authors.
Published by Rynnye Lyan Resources

reducing dairy products and replacing milk with other resources like vegetables, fruits or cereals (Murevanhema, 2013). Groundnut consumption except cookies can be increased by consuming vegetable milk, fermented products or a combination of cow milk due to the nutritional benefits of vegetable proteins, essential fatty acids and rich in minerals (Aidoo *et al*., 2010).

It is, therefore, necessary to adequately research the possibility of peanut processing to be more useful, edible milk or fermented peanut milk products. In the current study, peanut milk was produced and used as the main raw material for kefir production, which claims to have health benefits and exploit the potential market for ground-nut milk and fermented products. However, problems arise such as what is the appropriate temperature for fermentation and what is the appropriate time as well as what are the sensory flavour, aroma and physicochemical characteristics such as viscosity, total solid, Ph and colour of the product. Therefore, this study aimed to study the effect of time, the temperature of incubation and adding of skim milk on physicochemical as total solids, viscosity, colour and sensory properties as flavour and aroma of fermented peanut milk by kefir.

2. Materials and methods

2.1 Location of the study

The research was conducted from February 2021 to April 2021 at the Agricultural Technique Laboratory of Universitas Jenderal Soedirman -Purwokerto -Indonesia.

2.2 Collection of samples

Peanut samples were collected from Nganjuk City, and kefir culture was obtained from the Faculty of Animal Science, Universitas Jenderal Soedirman, Indonesia.

2.3 Manufacturing peanut kefir

Peanut milk was produced by heating at 80°C for 5 mins (steam sterilizer), then cooled to 40°C. Then, skimmed milk was mixed at the levels of 3% and 5% and kefir culture was added at the level of 5% and incubated for 18 and 24 hrs at 37°C and 45°C. The samples were coded as follow: T9 = 37°C / 24 hrs / skimmed milk 5%, T10 = 37°C /24 hrs / skimmed milk 3%, T11 = 37°C /18 hrs / skimmed milk5%, T12 = 37°C /18 hrs / skimmed milk 3%, T13 = 45°C /24 hrs / skimmed milk 5%, T14 = 45°C /24 hrs / skimmed milk 3%, T15 = 45°C /18 hrs / skimmed milk 5%, T16 = 45°C /18 hrs / skimmed milk 3%.

2.4 Physical, chemical and microbiological analysis

2.4.1 Total solids

According to the method of Almeida *et al*. (2010), the total solids of peanut kefir were determined by using a refract meter (PZO-RL1, Warsaw, Poland).

2.4.2 pH determination

According to the method recommended by the AOAC (2005), the pH value of yoghurt was determined by using a digital pH meter. The pH meter was calibrated using pH 7, pH 4, and pH 10. The pH meter was dipped into a 5 mL beaker.

2.4.3 Viscosity determination

According to the procedures method recommended by AOAC (2005), described by Ibrahim *et al*. (2016), the viscosity of peanut kefir was measured by using a digital viscometer and was expressed as a percentage.

2.4.4 Microbiological analysis

The calculation of the total plate count was determined by using the method of Mohammad and El-Zubeir (2011), Igbabul *et al*. (2014) and Ibrahim *et al*. (2020). The sample volume of 1 mL of peanut kefir was put into 9 mL of sterile distilled water solution with a dilution of 10^{-1}, 10^{-2}, 10^{-3} then vortexed. The dilution of 10^{-3} (0.1 mL) was plated out into Plate Count Agar media in petri dish. Incubation was conducted at 37°C for 24 hrs.

2.4.5 Colour

The colour of peanut kefir samples was measured by using a spectrophotometer (Minolta camera Co., Osaka, Japan), according to the method of (Aguirre *et al*., 2009; Chugh *et al*., 2014). Lightness to darkness (L*) (100 to 0), redness (+) to greenness (-) (a*) and yellowness (+) to blueness (-) (b*).

2.4.5 Sensory evaluation

The sensory assessment tests of peanut kefir were carried out in triplicate by twenty people of trained panellists according to the method of Arioui *et al*. (2016) and Ibrahim, Rifda, Erminawati and Hidayah (2020). The tests were conducted to examine the attributes of colour, smell, taste, and preference for peanut kefir with a range of 1-7. Each sample was given a code of two-digit numbers randomly.

2.5 Statistical analysis

The study was designed with a randomized complete block design (RBD) with 3 replications. The data were analyzed by analysis of variance two-way (ANOVA).

https://doi.org/10.26656/fr.2017.7(1).687

Then, to differentiate between the means was used Duncan Multiple Range Test. Then, if the effect was significant, the treatment differences were tested by (LSD), according to Ibrahim *et al*. (2021).

3. Results and discussion

3.1 Effect of incubation temperature, time and skimmed milk ratio on physicochemical properties of peanut kefir

The effects of incubation temperature, time, and skimmed milk ratio on the physicochemical properties of peanut kefir are presented in Table 1. The pH result values were 3.69, 4.39, 3.63, 4.69, 3.97, 4.62, 3.78 and 4.58. These results showed that there was no significant difference (P≤0.05) between the pH values of peanut kefir samples. One of these results achieved is that adding skimmed milk has a significant effect on decreasing pH. This decrease in pH value may be due to growth and an increase in the number of culture cells (Atalar and Dervisoglu, 2015). The total solids result of peanut kefir were 7.40, 5.00, 6.20, 5.20, 4.13, 3.80, 4.10 and 4.00. These results showed there were significant differences (P≤0.05) between peanut kefir samples. These differences may be due to incubation temperature and time, and also the addition of skimmed milk. The use of a high-temperature 45˚C led to a decrease in viscosity and total solids of peanut kefir. Julianto *et al*. (2016) suggested that the fermentation temperature of 45ºC and cultures concentration of 5% have significant effects on decreasing the viscosity, total solids and pH of fermented milk. Ibrahim *et al*. (2019) revealed that the higher the concentration culture and incubation temperature of 40-45ºC increased the number of bacteria cells and decreased pH in the final product.

The viscosity values of peanut kefir showed in Table 1. The results obtained for peanut kefir were 25.94, 24.24, 34.20, 34.62, 10.98, 12.70, 16.22 and 14.12. The result showed there were no significant differences (P≥0.05) between the samples. These differences in viscosity values of peanut kefir may be due to skimmed milk ratio (3-5%) and using different temperatures in incubation (37-45˚C). The incubation temperature of 45˚C for a long time led to decreased viscosity of peanut kefir. Julianto *et al*. (2016) suggested that the fermentation temperature of 45ºC and cultures concentration of 5% have significant effects on decreased viscosity and pH of fermented milk. Ibrahim *et al*. (2019) revealed that the higher the concentration culture and incubation temperature of 40-45ºC increased the number of bacteria cells in the final product.

Table 1. Effect of incubation temperature, time and skimmed milk ratio on physicochemical properties of peanut kefir

Treatments	pH	Total solids	Viscosity
T9	3.69±0.21[a]	7.40±0.55[d]	25.94±3.31[a]
T10	4.39±0.43[a]	5.00±0.00[b]	24.24±2.35[a]
T11	3.63±0.14[a]	6.20±0.27[c]	34.20±2.60[a]
T12	4.69±0.20[a]	5.20±0.27[b]	34.62±15.32[a]
T13	3.97±0.42[a]	4.13±0.25[a]	10.98±2.16[a]
T14	4.62±0.05[a]	3.80±0.45[a]	12.70±7.51[a]
T15	3.78±0.04[a]	4.10±0.00[a]	16.22±0.49[a]
T16	4.58±0.15[a]	4.00±0.00[a]	14.12±4.51[a]

Values are presented as the mean±SD. Values with different superscript within the same column are significantly different.

Table 2 shows the colour values of peanut kefir. The peanut kefir samples' colour ranged from white to yellowish-white. According to Table 3, the total plate count bacteria values (10 CFU/mL) were 9.00, 36.00, 16.67, 9.67, 11.00, 8.67, 23.67 and 17.00. These results indicate that the number of microbes in all samples of peanut kefir is still in the range of Indonesian standards, and does not have a negative effect on the quality of the final product.

3.2 Sensory evaluation of peanut kefir

Table 4 shows the sensory evaluation of peanut kefir. Peanut kefir produced by kefir grains does not affect the colour of the peanut kefir produced. It shows that the process of kefir production does not produce materials that change the colour of the peanut kefir produced.

Table 2. Influence of incubation temperature, time and skimmed milk ratio on peanut kefir colour

Treatments	Colour		
	A (Redness (+) to greenness (-) (a*)	B (Yellowness (+) to blueness (-) (b*)	L (Lightness to darkness (L*) (100 to 0)
T9	15.23±1.76[a]	23.77±1.16[a]	71.83±1.26[a]
T10	14.73±1.48[a]	22.77±4.09[a]	73.50±1.13[a]
T11	18.40±3.56[a]	24.73±3.51[a]	74.77±1.07[a]
T12	18.33±6.73[a]	21.07±4.82[a]	74.20±6.26[a]
T13	17.87±2.52[a]	23.70±0.66[a]	71.47±6.50[a]
T14	15.70±1.61[a]	21.10±0.90[a]	68.63±3.93[a]
T15	18.10±2.36[a]	22.87±0.75[a]	66.07±9.78[a]
T16	16.50±2.17[a]	26.70±5.80[a]	73.03±2.70[a]

Values are presented as the mean±SD. Values with different superscript within the same column are significantly different.

https://doi.org/10.26656/fr.2017.7(1).687

Table 3. Microbiology analysis (Total Plate Count)

Treatments	TPC (CFU/mL)
T9	9.00±1.00[a]
T10	36.00±7.21[d]
T11	16.67±0.58[b]
T12	9.67±1.53[a]
T13	11.00±3.46[ab]
T14	8.67±0.58[a]
T15	23.67±2.08[c]
T16	17.00±2.00[b]

Values are presented as the mean±SD. Values with different superscript within the same column are significantly different.

However, the level of skimmed milk and kefir culture added to the peanut milk significantly influences the taste and smell, and also has implications for the level of preference for the peanut kefir produced. The results suggested that the higher incubation temperature for a long time gives the more acidic flavour of peanut kefir, but the panellists are still tolerating a more acidic flavour at the level of 5% kefir grain supplementation. The taste of peanut kefir produced will have an impact on preference, which shows that the higher level of incubation temperature for a long time affects the lower the level of preference. It indicates that the average panellist favour kefir as it does not have a highly acidic taste. The highest preference level was given to peanut kefir produced with the supplementation of kefir grain at a level of 5%, skimmed milk at 5%, and incubation temperature of 37°C for 18 hrs. Ibrahim *et al*. (2019) revealed that the higher concentration of culture with increased incubation temperature for a long time increased the smell of fermented milk. Also, the reduction of pH value enhanced the sour and unique flavour of fermented milk products.

4. Conclusion

The result indicated that a product like kefir can be produced from peanut milk which has excellent physicochemical properties. These results show that incubation temperature, time and skimmed milk addition ratio have no significant effect (P>0.05) on some physical and chemical properties of peanut kefir, such as colour, pH, and viscosity, whilst they have a significant effect (P>0.05) on total solids. General results from this present study established that the best peanut milk kefir can be produced by the addition of 5% skimmed milk, and 5% kefir grain and incubated for 18 hrs at 37°C based on physicochemical, microbiological, and sensory characteristics.

Acknowledgements

The researchers are grateful to the Ministry of Research Technology and Higher Education and the Research and Community Service Institute at Sudirman State University for funding and support. Also, a lot of thanks go to the staff of the Agricultural Technology Laboratory for support and guidance.

References

AOAC (Association of Official Analytical Chemists) (2005). Official methods of analysis. 16th ed., Maryland, USA: AOAC.

Aguirre, B.D., Mawson, R., Versteeg, K. and Cánovas, B.V.G. (2009). Composition Properties, Physicochemical Characteristics and Shelf Life of whole Milk After Thermal and Thermo-Sonication Treatments. *Journal of Food Quality*, 3(32), 283-302. https://doi.org/10.1111/j.1745-4557.2009.00250.x

Aidoo, H., Sakyi-Dawson, E., Tano-Debrah, K. and Alia, F.K. (2010). Development and characterization of dehydrated peanut–cowpea milk powder for use as a dairy milk substitute in chocolate manufacture. *Food Research International*, 43(1), 79-85. https://doi.org/10.1016/j.foodres.2009.08.018

Almeida, L.F.D., Gisely, M.F., Cavalcante, M.T., Castro, R.D. and Cavalcanti, A.L. (2010). Cariogenic and erosive potential of industrialized fruit juices available in Brazil. *Brazil Journal Science*, 9(3), 351

Table 4. Sensory evaluation of peanut kefir.

Treatments	Colour	Taste	Peanut Aroma	Texture	Flavour	Overall acceptability
T9	3.45±0.89[a]	3.64±1.07[a]	2.64±1.23[a]	4.55±0.66[d]	2.36±1.07[a]	2.27±0.90[a]
T10	3.55±0.78[a]	3.18±1.27[a]	2.73±1.21[a]	4.18±1.03[cd]	2.27±0.86[a]	2.09±0.67[a]
T11	3.55±0.66[a]	2.36±0.88[a]	3.73±0.75[a]	4.09±0.90[cd]	2.64±0.77[a]	2.55±0.78[a]
T12	4.09±0.79[a]	3.09±1.44[a]	3.18±1.27[a]	3.82±0.83[bcd]	1.82±1.03[a]	1.64±0.88[a]
T13	3.91±0.67[a]	3.09±1.31[a]	2.91±0.90[a]	3.64±0.48[ab]	2.64±0.77[a]	2.36±0.88[a]
T14	3.64±0.64[a]	2.82±0.72[a]	3.36±0.88[a]	3.18±0.83[ab]	2.45±0.78[a]	2.18±0.72[a]
T15	3.55±0.89[a]	2.64±1.30[a]	3.27±1.35[a]	3.36±0.98[ab]	1.82±0.57[a]	2.00±0.85[a]
T16	3.64±1.07[a]	2.82±1.59[a]	3.09±1.38[a]	2.82±0.94[a]	1.64±0.88[a]	1.64±0.88[a]

Values are presented as the mean±SD. Values with different superscript within the same column are significantly different.

-357.

Arioui, F., Ait, S.D. and Cheriguene, A. (2015). Physicochemical and sensory quality of yogurt incorporated with pectin from peed of *Citrus sinensis*. *Food science and Nutrition*, 5(2), 358-365. https://doi.org/10.1002/fsn3.400

Atalar, I. and Dervisoglu, M. (2015). Optimization of spray drying process parameters for kefir powder using response surface methodology. *Food Science and Technology*, 60(2), 751-757. https://doi.org/10.1016/j.lwt.2014.10.023

Bensmira, M. and Jiang, B. (2014). Total phenolic compounds and antioxidant activity of a novel peanut-based kefir. *Food Science and Biotechnology*, 24(3), 1055-1060. https://doi.org/10.1007/s10068-015-0135-7

Chugh, A., Khanal, D., Walkling-Ribeiro, M., Duizer, L. and Griffiths, M.W. (2014). Change in color and volatile composition of skim milk processed with pulsed electric field and microfiltration or heat pasteurization. *Journal of Food*, 3(2), 250-368. https://doi.org/10.3390/foods3020250

Corona, O., Randazzo, W., Miceli, A., Guarcello, R., Francesca, N., Erten, H., Moschetti, G. and Settanni, L. (2016). Characterization of kefir-like beverages produced from vegetable juices. *Food Science and Technology*, 66, 572-581. https://doi.org/10.1016/j.lwt.2015.11.014

Gul, O., Mortas, M., Atalar, I., Dervisoglu, M. and Kahyaoglu, T. (2015). Manufacture and characterization of kefir made from cow and buffalo milk, using kefir grain and starter culture. *Journal of Dairy Science,* 98(3), 1517-1525. https://doi.org/10.3168/jds.2014-8755

Ibrahim, A.I., Rifda, N., Erminawati, M., Hidayah, D. and Shima, E.H. (2020). Influence of temperature and time on microbial, physicochemical and functional quality of goat milk. *African Journal of Food Science,* 14(4), 86-91. https://doi.org/10.5897/AJFS2020.1912

Ibrahim, A.I., Rifda, N., Erminawati, M. and Hidayah, D. (2020). Optimization dehydration conditions of cow's and goat milk yogurt powder. *International Journal of Innovative Technology and Exploring Engineering*, 9(3), 68-71. https://doi.org/10.35940/ijitee.C1015.0193S20

Ibrahim, A.I., Rifda, N., Erminawati, M. and Hidayah, D. (2019). Effect of fermentation temperature and culture concentration on microbial and physicochemical properties of cow and goat milk yogurt. *IOP Conf. Series: Earth and Environmental Science,* 406(1), 012009. https://doi.org/10.1088/1755-1315/406/1/012009

Ibrahim, A.I., Nurjanah, S. and Kramadibrat, A.M. (2016). Impact of different extraction methods on physical chemical properties of palm kernel oil (*ElaeisGuineensis*). *Jurnal Teknotan*, 10(1), 11-15. https://doi.org/10.24198/jt.vol10n1.2

Ibrahim, A.I., Rifda, N., Erminawati, M. and Hidayah, D. (2021). Comparative study between cow and goat milk yogurt based on composition and sensory evaluation. *IOP Conference Series: Earth and Environmental Science*, 746, 012001. https://doi.org/10.1088/1755-1315/746/1/012001

Igbabul, B., Shember, J. and Amove, J. (2014). Physicochemical, Microbiological and Sensory Evaluation of Yoghurt Sold in Makurdi Metropolis. *African Journal of Food Science and Technology*, 5 (6), 129-35.

Julianto, B., Rossi, E. and Yusmarini. (2016). Chemical and Microbiology Characteristics of Kefir from Cow Milk with Soy Milk Addition. *Jom Faperta*, 3(1), 1-11.

Mizielin, M. and Topusiewicz, T. (2018). Encapsulation and evaluation of probiotic bacteria survival in simulated gastrointestinal conditions. *Romanian Biotechnological Letters*, 23 (3), 13690–13696.

Mohammad, E.E.B. and El-Zubeir, I.E.M. (2011). Chemical Composition and Microbial Load of Set Yogurt from Fresh and Recombined Milk Powder in Khartoum State, Sudan. *International Journal of Dairy Science*, 6(3), 172-180. https://doi.org/10.3923/ijds.2011.172.180

Puerari, C., Magalhães, K.T. and Schwan, R.F. (2012). New cocoa pulp-based kefir beverages: Microbiological, chemical composition and sensory analysis. *Food Research International*, 48(2), 634–640. https://doi.org/10.1016/j.foodres.2012.06.005

Roselló-Soto, E., Garcia, C., Fessard, A., Barba, F., Munekata, P., Lorenzo, J. and Remize, F. (2018). Nutritional and Microbiological Quality of Tiger Nut Tubers (*Cyperus esculentus*), Derived Plant-Based and Lactic Fermented Beverages. *Fermentation*, 5 (1), 3. https://doi.org/10.3390/fermentation5010003

Satir, G. and Guzel-Seydim, Z.B. (2015). Influence of Kefir fermentation on the bioactive substances of different breed goat milks. Food Science and Technology, 63(2), 852-858. https://doi.org/10.1016/j.lwt.2015.04.057

Yadav, P.B. (2016). Studies on the Preparation of peanut milk and milk powder. Raipur, India: Indira Gandhi Krishi Vishwavidyalaya, MTech. Thesis.

