Journal of Indonesia Laboratory Technology of Student (JILTS)
Vol.5, No.2, October 2026, pp. 304~310

Optimization Of Brown Rice Flour (Oryza Sativa L.) As An Alternative Medium For The Growth Of Staphylococcus Aureus Bacteria

Adhela Gladis Adisti[1], Siti Zaetun[2], Thomas Tandi Manu[3], Rohmi[4]
[1-4] Jurusan Teknologi Laboratorium Medis, Poltekkes Kemenkes Mataram, Indonesia
adeliagladis722@gmail.com

ABSTRACT

Background: The high dependence of microbiology laboratories on commercial media like Nutrient Agar (NA), which uses imported components (peptone and meat extract), highlights the need to explore local, cost-effective substrates. Red rice (Oryza sativa L.) is rich in complex carbohydrates, proteins, B-complex vitamins, and micronutrients that could potentially support microbial nutritional needs. **Research Methods:** An in vitro experimental approach with a post-treatment control scheme was employed in this study. The growth media derived from red rice flour was formulated across four gradient levels: 2.5%, 5.0%, 7.5%, and 10.0% (w/v), supplemented with 1.5% agar matrix, then compared with a positive control NA. S. aureus cultures were inoculated and incubated at 37°C for 24–48 hours. Growth parameters were evaluated descriptively based on colony density and thickness. **Research Results**: S. aureus can grow under all treatment variations. A 2.5% concentration results in limited growth, 5% and 7.5% concentrations show moderate growth, while a 10% concentration shows very dense and abundant colonies, comparable on a macroscopic level to standard NA media (fertility score 3). **Conclusion**: Red rice flour has proven to be potentially used as a base material for bacterial inoculum, with a 10% concentration being the most optimal for producing S. aureus growth equivalent to standard media.

Keywords: Alternative Media, Nutrient Agar, Red Rice Flour, Staphylococcus aureus

Article Info

Article history:

Received
August, 2026
Revised
September, 2026
Accepted
October, 2026

Introduction

In microbiology studies and diagnostic laboratory examinations, the provision of responsive and proportionate seed media is an absolute prerequisite for isolating, cultivating, and characterizing microorganisms (Collins et al., 2021). One of the crucial and often evaluated pathogen test isolates is *Staphylococcus aureus.* As a Gram-positive microorganism with a cell structure resembling a grape strand, S. aureus often inhabits skin areas and mucosal membranes and is at risk of triggering mild to systemic pathogenicity (Madigan et al., 2018; Shehade et al., 2025).

Nutrient Agar (NA) is a conventional seed formulation that is non-selective and is standard used in various laboratory facilities. NA formulations generally rely on peptone and *beef extract* as providers of carbon, nitrogen, vitamins, and essential minerals (Cappuccino & Sherman, 2014; Nurhidayanti, 2022). However, the majority of the supply of pepton and meat extract in Indonesia still depends on imported commodities at relatively high prices. This triggers operational deficits in health care facilities with limited facilities and regional educational institutions (Pratomo et al., 2020; Susanti et al., 2022).

The strategy to offset budget constraints and the distribution of imported raw materials is through the substitution of plant-based local organic materials (Marpaung et al., 2022). Research on the exploration of alternative media based on grains and crops such as soybeans, corn, and cassava has shown positive prospects (Suhartati et al., 2018; Martsiningsih et al., 2024; Susilawati et al., 2024). However, the specific use of brown rice flour (Oryza sativa L.) is still very limited.

Brown rice is rich in complex starches (amylose and amylopectin) of 77.6 g per 100 g which acts as a trigger for metabolic energy (Indriyani et al., 2013). In addition, brown rice contains 7.5 g of crude protein, 163 mg of phosphorus, 16 mg of calcium, iron, and thiamine (vitamin B1) which can supply nitrogen and essential enzymatic cofactors for microbes (Almeida et al., 2021; Baptista et al., 2024).

Preliminary studies with a 2% brown rice flour concentration indicated that S. aureus was able to survive. Therefore, this study was designed to diversify and optimize the concentration of brown rice flour (2.5%, 5%, 7.5%, and 10%) as an efficient and economical alternative seed media formula compared to the NA standard media.

Research Methods

A. Research Design and Location

This study uses a pure laboratory experimental model that applies a post-treatment comparison scheme. Formulation, inoculation, and testing activities were carried out at the Microbiology Laboratory, Department of Medical Laboratory Technology, Polytechnic of the Ministry of Health, Mataram in the period from December 2025 to May 2026.

B. Sample Testing and Determination Unit

The study included four levels of variation in material concentration (2.5%; 5%; 7.5%; and 10% w/v) plus one NA comparison group. The number of iterations was based on Federer's ***equation (t - 1)(r - 1) ≥ 15***, so that each treatment group was executed through five iterations per treatment group so that

the total observational sample included 25 treatment cups and 5 control cups (total 30 experimental units).

The best results were obtained in the variation in the concentration of 10.0% brown rice flour, where all replicas (P1– P5) showed very fertile growth with a score of 3. The morphological appearance of convex, smooth, uniform-sized, and densely crowded colonies covered the scratch area, which was macroscopically identical to the growth in the NA control cup.

C. Media Creation Procedure

The pre-treatment stage begins with washing brown rice followed by a natural drying process. The ingredients are then ground and filtered using a fine sieve until a uniform flour preparation is obtained. The flour preparation was quantified as 2.5 g; 5.0 g; 7.5 g; and 10.0 g, then dispensed into the aqueduct until a final volume of 100 mL was sufficient. The dissolution was assisted by heating on a heating plate with the addition of 1.5 g of a commercial agar compactor. The media formulation was sterilized using an autoclave at a temperature of 121°C and a pressure of 1 atm for 15 minutes. After cooling to a temperature of 45–50°C, the media was poured aseptically into a sterile petri dish and allowed to solidify. The positive control of NA was prepared according to standard laboratory procedures.

D. Inoculation and Growth Assessment

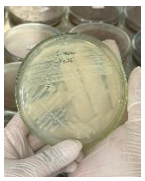
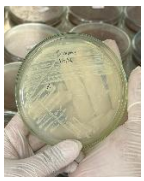
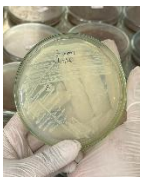
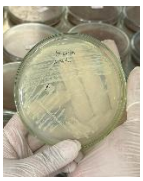
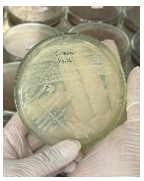
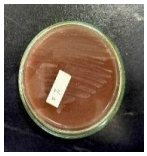
Pure suspension of Staphylococcus aureus was inoculated on the surface of the media using the streak plate method. The entire cup was incubated in an incubator at 37°C for 24–48 hours. Bacterial growth was assessed macroscopically using a standardized score scale:

1. Score 1 (Slightly Fertile): Colonies grow thin, sparse, and spread loosely.
2. Score 2 (Fertile): Colonies compacted moderately along the stroke path.
3. Score 3 (Very Fertile): The colony is very dense, dense, and abundantly crowded.

Research and Discussion Results

The incubation results showed that all variations in the concentration of brown rice flour were able to facilitate the reproduction of Staphylococcus aureus. The density of bacterial colonies at each level of treatment is summarized in Table 1.

Table 1. Density Profile of S. aureus Colonies on Various Test Media Formulations

Treatment	Repetition				
	P1	P2	P3	P4	P5
Control (NA)	 3	 3	 3	 3	 3
Brown Rice Flour 2.5%	 1	 2	 2	 2	 2
Brown Rice Flour 5%	 2	 2	 3	 2	 2
Brown Rice Flour 7.5%	 2	 2	 2	 2	 2
Brown Rice Flour 10%	 3	 3	 3	 3	 3

Description :

Bacterial growth was assessed macroscopically using a standardized score scale:

1. Score 1 (Slightly Fertile): Colonies grow thin, sparse, and spread loosely.
2. Score 2 (Fertile): Colonies compacted moderately along the stroke path.
3. Score 3 (Very Fertile): The colony is very dense, dense, and abundantly crowded.

Based on the exposure of Table 1 data, the standard media of NA produced fertility levels consistent with a score of 3 on all tests. This is natural considering that NA is specially formulated with pure peptone and meat extracts rich in free amino acids and short peptides that are easily absorbed by microbial cells (Atlas, 2010).

In the treatment of brown rice flour the concentration was 2.5%, growth was at a moderate level (dominant score of 2, with 1 cup score of 1). The low density of this colony is due to the limited dissolved nutrients in the substrate, so that cell division and biosynthesis of the cell wall of S. aureus are slow.

An increase in flour content to 5.0% and 7.5% provided growth stability at the fertile level (score 2). The availability of carbohydrate and nitrogen energy was sufficient to support binary cleavage, but had not yet reached maximum efficiency.

The success of this 10% formulation is correlated with the fulfillment of the nutrient adequacy threshold limit. At this concentration, the hydrolyzed starch content provides an abundant supply of carbon, while the brown rice protein fraction provides an intake of glutamic and aspartic acids that support the synthesis of Gram-positive peptidoglycan cells (Baptista et al., 2024). In addition, the natural phosphorus and magnesium content in brown rice acts as important enzymatic cofactors in the microbial glycolysis cycle (Indriyani et al., 2013).

These findings are in line with the principles of developing alternative media based on local raw materials (Siti Juariah, 2021; Nurhidayanti, 2022). With a concentration of 10%, brown rice flour is suitable as a substitute for basic organic matter that is cheap, easy to obtain, and environmentally friendly for *Staphylococcus aureus cultures* in the laboratory.

Conclusion

A. Conclusion

Based on a series of tests that have been carried out, it can be concluded that:

1. Brown rice flour-based seed media (Oryza sativa L.) was proven to be able to support the growth of Staphylococcus aureus bacteria in all test concentration variations.
2. There was a positive correlation between the concentration of brown rice flour and the fertility rate of the colony. The formulation of the concentration of 10.0% provided the most optimal performance and produced an equivalent growth response (score of 3 / very fertile) with the standard control medium *of Nutrient Agar* (NA).

B. Suggestions

Further research is needed by adding quantitative tests (counting the number of colonies / CFU per mL) and comparing the performance of isolation using selective media such as *Blood Agar Plate* (BAP) or *Mannitol Salt Agar* (MSA).

BIBLIOGRAPHY

Almeida, R.L.J., T. dos Santos Pereira, R.D. Almeida, Â.M. Santiago, W.I.M. de Lima Marsiglia, E.H. Nabeshima, L. de Sousa Conrado dan R.P. de Gusmão. 2021. *Rheological and technological characterization of red rice modified starch and jaboticaba peel powder mixtures*. *Scientific Reports*. 11(1): 1–11.

Anggraheni, D.A., Pestariati and S. Arifin. 2023. *Comparison of agar nutrient media with beef, goat meat and chicken as a growth medium for Staphylococcus aureus*. *Science Health Analyst*. 12(1): 1–4.

Atlas, R.M. 2010. *Handbook of Microbiological Media*. 4th Edition. CRC Press. Boca Raton.

Cappuccino, J.G. and N. Sherman. 2014. *Microbiology: A Laboratory Manual*. 10th Edition. Pearson Education. Boston.

Madigan, M.T., K.S. Bender, D.H. Buckley, W.M. Sattley dan D.A. Stahl. 2018. *Brock Biology of Microorganisms*. Edisi ke-15. Pearson. New York.

Baptista, E., A. Liberal, R.V.C. Cardoso, A. Fernandes, M.I. Dias, T.C.S.P. Pires, R.C. Calhelha, P.A. Garcia, I.C.F.R. Ferreira dan J.C.M. Barreira. 2024. *Chemical and bioactive properties of red rice with potential pharmaceutical use*. *Molecules*. 29: 2265.

Daun, A., M. Rhizophora, P.S. Pharmacy, F. Medicine and U. Mataram. 2024. *Factors influencing activity against Staphylococcus aureus bacteria*. 5: 3829–3836.

Halim, N. 2025. *Scientific Papers*. www.smapda-karangmojo.sch.id.

Hu, Z., Z. Zhu, H.K. Ono, S. Hirose, Y. Hara-kudo, S. Li dan D. Hu. 2025. *Effects of nutritional conditions on growth, biofilm formation, and enterotoxin production in Staphylococcus aureus associated with food poisoning*. 1–14.

Indriyani, F., Nurhidajah dan A. Suyanto. 2013. *Physical, chemical and organoleptic characteristics of brown rice flour based on the variation of drying time*. *Jurnal Pangan dan Gizi*. 4(8): 27–34.

Kasmawati, K. 2023. *The relationship between brown rice consumption and blood sugar in patients with type II diabetes mellitus at the Minasa Upa Makassar Health Center*. *Amsir Multidisciplinary Scientific Journal*. 1(2): 250–266.

Lipinwati, B. dan H. Justitia. 2023. *Staphylococcus aureus antibiotics resistance pattern in osteomyelitis cases at hospital in Jambi City*. *Jambi Medical Journal*. 11(4): 435–441.

Marpaung, J.K., S. Purba, D.J. Barus and D. Iqlima. 2022. *Creation of alternative growth media in microorganisms using porang tubers (Amorphophallus oncophyllus Prain) against Staphylococcus aureus, Escherichia coli and Candida albicans*. *Tekesnos Journal*. 4(1): 363–372.

Martsiningsih, M.A., F. Dwi Astuti, M. Kasiyati, U. Rahmawati, M. Rahayu and A. Puspa Aeni. 2024. *Mixture of cassava infusion (Manihot esculenta), soybean (Glycine max (L.) Merrill) and yeast extract as an alternative medium for the growth of Staphylococcus aureus bacteria*. *The Journal of Muhammadiyah Medical Laboratory Technologist*. 7(1): 66–73.

Nurhidayanti, N. 2022. *Comparison of soybean alternative media and agar nutrient media against the growth of Staphylococcus aureus bacteria*. *Indobioscience*. 4(2): 47.

Fitriyah, D., D.P. Putri Ayu, S.D. Dewi Puspita, R.C. Kartika and M. Ubaidillah. 2022. *Nutritional content and antimicrobial activity of brown rice extract*. *Journal of Food and Nutrition*. 12(2): 30–36.

Pratomo, M.D., D.W. Wardani, N.A. Revonagara, D. Ersyah, D. Setijawati, H.S. Yufidasari and A.A. Jaziri. 2020. *Characteristics of peptone from kurisi fish waste (Nemipterus sp.) as a halal-guaranteed bacterial growth medium*. *Journal of Aquaculture and Fish Health*. 9(2): 104–113.

Sahgal, A. 2024. *Identification of Staphylococcus aureus bacteria in cleaners at the Banjardowo landfill, Jombang District, Marpaung Regency, J.K., S. Purba, D.J. Barus and D. Iqlima.* 2022. *Creation of alternative growth media in microorganisms using porang tubers (Amorphophallus oncophyllus Prain) against Staphylococcus aureus, Escherichia coli and Candida albicans*. *Tekesnos Journal*. 4(1): 363–372.

Martsiningsih, M.A., F. Dwi Astuti, M. Kasiyati, U. Rahmawati, M. Rahayu and A. Puspa Aeni. 2024. *Mixture of cassava infusion (Manihot esculenta), soybean (Glycine max (L.) Merrill) and yeast extract as an*

alternative medium for the growth of Staphylococcus aureus bacteria. *The Journal of Muhammadiyah Medical Laboratory Technologist*. 7(1): 66–73.

Nurhidayanti, N. 2022. *Comparison of soybean alternative media and agar nutrient media against the growth of Staphylococcus aureus bacteria*. *Indobioscience*. 4(2): 47.

Fitriyah, D., D.P. Putri Ayu, S.D. Dewi Puspita, R.C. Kartika and M. Ubaidillah. 2022. *Nutritional content and antimicrobial activity of brown rice extract*. *Journal of Food and Nutrition*. 12(2): 30–36.

Pratomo, M.D., D.W. Wardani, N.A. Revonagara, D. Ersyah, D. Setijawati, H.S. Yufidasari and A.A. Jaziri. 2020. *Characteristics of peptone from kurisi fish waste (Nemipterus sp.) as a halal-guaranteed bacterial growth medium*. *Journal of Aquaculture and Fish Health*. 9(2): 104–113.

Sahgal, A. 2024. *Identification of Staphylococcus aureus bacteria in cleaners at the Banjardowo landfill, Jombang District, Jombang Regency*. 4(1): 9–15.

Shehade, S.M., A.S.M. Juma, M.H. Ubeid dan S.M. Al-Awqati. 2025. *Staphylococcus aureus nasal carriage among medical students at Damascus University, Syria*. *Medical Journal of Babylon*. 22(Suppl 1): S128–S133.

Siti Juariah, R. 2021. *Alternative growth media of Staphylococcus aureus from durian seeds (Durio zibethinus murr). Meditory: The Journal of Medical Laboratory*. 9(1): 19–25.

S. S. 4(1): 9–15.

Shehade, S.M., A.S.M. Juma, M.H. Ubeid dan S.M. Al-Awqati. 2025. *Staphylococcus aureus nasal carriage among medical students at Damascus University, Syria*. *Medical Journal of Babylon*. 22(Suppl 1): S128–S133.

Siti Juariah, R. 2021. *Alternative growth media of Staphylococcus aureus from durian seeds (Durio zibethinus murr). Meditory: The Journal of Medical Laboratory*. 9(1): 19–25.

Susanti, M., S. Khalimatusa'diah and A. Rasyid. 2022. *Utilization of a variety of carbohydrate sources from palawija as an alternative synthetic medium for bacterial growth*. *The Journal of Science and Biology Education*. 7(2): 61–67.

Susilawati, N.M., A. Astuti dan A. Peni. 2024. *Utilizing maize cob media as a replacement for nutrient agar media in the growth of Escherichia coli and Staphylococcus aureus bacteria*. *International Journal of Educational Research*. 1(1): 28–35.

Talapan, D., A.M. Sandu dan A. Rafila. 2023. *Antimicrobial resistance of Staphylococcus aureus isolated between 2017 and 2022 from infections at a tertiary care hospital in Romania*. *Antibiotics*. 12(6).

Wijerathna, R., N. Asanthi, W. Ratnasooriya, R. Pathirana dan N. Nelumdeniya. 2018. *Evaluation of in vitro antibacterial activity and phytochemical profile of aqueous leaf extract of Asystasia variabilis*. *Journal of Pharmacognosy and Phytochemistry*. 7(4): 639–642.