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Chemical Characteristics and Microbial Identification of Fish Fermented Food by Flores Ethnic

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ABSTRACT

Mudu and Mbarase are two local fish-fermented foods that have been known for a long time in the culture of the Flores community. These two fermented foods were made using a high-salt fermentation process. Mudu is made from fish (tuna and *Rastrelliger* family) innards while Mbarase is made from rabbitfish (*Siganus* Sp.). This study aimed to analyze the chemical characteristics and identify microbial content in these two samples. The chemical characteristic here was proximate analysis and the microbial identification was done for lactic acid bacteria (LAB) identification. Proximate analysis of samples was done using the AOAC standard method. LAB identification was done by morphology observation with a clear zone around the colony, gram test, and catalase test. For the proximate analysis, Mudu contains 21.022% carbohydrate, 11.8% protein, 0.42% fat, 6.587% ash, 60.171% water, and 1.94% fiber. Mbarase contains 7.732% carbohydrates, 25.19% protein, 4.558% fat, 5.13% ash, 57.29% water, and 0.16% fiber. A clear zone can be seen in the Mbarase sample and slightly seen in the Mudu sample. Lactic acid bacteria isolated from Mbarase have antibacterial activity to E. coli and A. hydrophilla. Mbarase and Mudu may become a good source of nutrients as they have high protein, low fat, and ash. These fish fermented food also have probiotic potentials as it has antibacterial activity against pathogen bacteria.

Keywords: Fish fermentation, mudu, mbarase, proximate, microbe

Introduction

Fermentation is known as one of the oldest methods of food preservation and the most economical. Fermentation is a process that helps break down large organic molecules into simpler ones via the action of microorganisms (Sharma et al., 2020). Fermentation can increase the shelf life of food, reduce volume, shorten cooking times (Surono, 2016), remove unwanted ingredients from raw materials (Sharma et al., 2020), and change the organoleptic characteristics of foods including flavors, aromas, and textures. The fermentation process also improves digestibility and nutritional quality by enriching food substrates with vitamins, proteins, essential amino acids, and essential fatty acids (Nuraida, 2015) and consists of higher in vitro antioxidant capacity Sharma et al., 2020). Bacteria, yeast, and molds are the most common microorganisms involved in food fermentation.

Lactic Acid Bacteria (LAB) is the dominant microbiota in fermented foods and beverages LAB has been considered the most critical part contributing to beneficial effects in fermentation (Sharma et al., 2020). LAB-fermented foods feature the use of a variety of raw materials, including

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fruits, vegetables, cassava, meat, milk, cereals, legumes, and fish (Nuraida, 2015; Afrianti, 2014; Singh et al., 2014; Amelia et al., 2021; Ngasotter et al., 2020; Marti-Quijal et al., 2020). In some traditional fermentation, LAB was used for spontaneous fermentation at an aerobic condition because it is already present in the raw materials used for fermentation. Studies have demonstrated that LAB is part of the normal intestinal microbiota in fish. Some important fermented fish products that are eaten day by day in Indonesia are Bekasam (freshwater fish), Rusip (anchovy or Bilis fish), Chao (Tembang fish) (Ngasotter et al., 2020), Bakasang from innards of Skipjack tuna (Lawalata et al., 2011), Inasua (Mahulette et al., 2020), Mudu (innards of skipjack, rastrelliger), and Mbarase (rabbit fish).

Mbarase is fish-fermented food from Siganus sp. Its baby fish (1-2 cm) was cleaned and added with salt then stored in a plastic bottle. The bottle was covered (anaerobic fermentation) and fermented for at least 7 days. Mbarase can stored for one year, therefore it can be sold freshly or after being saved for a certain time. Mbarase is an integral part of the food and culture of the Sikka people. It can be developed commercially to support livelihood. Mbarase is sold in the local traditional market in Sikka, Flores Island and the price is Rp 35.000-50.000/bottle.

Mudu was fermented from innards of skipjack, tuna, or rastrelliger fish. The innards were cleaned and added with salt then stored in a covered jar for at least 7 days (anaerobic fermentation). Unlike mbarase, mudu is not made for commercial purposes. Mudu is consumed by adding some condiments (onion, lime, chili, basil) to become 'sambal mudu'. Mudu can also be stored for a year.

Although mbarase and mudu have long been known as local dishes in the Flores community, there are no published studies that evaluate their nutritional and microbial content or moreover the microbial potential of them. Some publications about mbarase are only on its economic potential (trader's income and consumer behavior, while for mudu, there are no publications found yet. Therefore, this study aimed to determine the crude nutritional content of mbarase and mudu. Microbial identification was also carried out to identify LAB in these two fermented foods.

Material and Methods

Samples of Mbarase were purchased from the local market in Sikka, Flores Island. A sample of Mudu was obtained from East Flores Regency (Solor Island). The products were stored in clear plastic bottles as containers. Samples were transported to the Biology Laboratory, University of Muhammadiyah Kupang, East of Nusa Tenggara. Samples were kept at 4°C until analysis. Then, the sample was examined in the laboratory for its chemical qualities, identification of LAB content, and LAB antibacterial potential to pathogen bacteria.

Analysis of chemical characteristics

This analysis was conducted to determine the crude nutritional content of samples, including carbohydrates, protein, fat, water, ash, and fiber. The analysis of the sample was carried out using the standard method of the Association of Official Analytical Chemists (AOAC, 2005), which has been mentioned in a previous study (Ilyanie et al., 2020). This analysis was done in the Nutrition Lab of Agricultural Polytechnic Kupang and BPOM (Agency for Drug and Food Control), Kupang.

Water content

Samples were placed in the crucible and dried in an oven (T 105°C) until constant weight. The difference between the wet weight and dry weight of the sample was calculated to obtain the percentage of water content in the samples.

Ash analysis

Three grams of sample were burnt and ashed in a furnace (T 525°C, 24 h) until ash formed. Ash then cooled in a desiccator. The percentage of total crude ash was calculated using this equation.

Crude ash (%) dry basis = [W1 / W2] × 100

W1 = weight of samples after ashing; W2 = weight of samples before ashing.

Protein analysis

The protein content of samples was measured using nitrogen assay based on the Kjeldahl method. This method includes the digestion and distillation process. The percentage of crude protein was expressed as a total nitrogen percentage and multiplied by a factor of 6.25 (nitrogen-protein conversion factor for fish and seafood products).

Crude protein (%) = Nitrogen (%) in samples × 6.25

Fat analysis

Fat was estimated using Soxhlet extraction with petroleum ether as the solvent. The fat content of each sample was determined after oven-drying the extracted fats overnight. The fat content of samples was calculated using this equation.

Fat (%) = [(W1 – W2) / W0] × 100

W0 = weight of the sample after treatment; W1 = total weight of extraction beaker with boiling stones and extracted fats; and W2 = total weight of extraction beaker and boiling stones.

Carbohydrate analysis

Total carbohydrate was calculated using the difference of the value of each nutrient using this equation.

Carbohydrates (%) = 100 – [Moisture (%) + Ash (%) + Protein (%) + Fat (%)]

Isolation of Lactic Acid Bacteria (LAB)

Samples were put into sterile Erlenmeyer and diluted with sterilized physiologic NaCl with 10-1, 10-2, and 10-3 dilutions. One ml of the previous dilution was inoculated in medium NA and then incubated at 37°C for 24h. The colony which indicated a clear zone shows that the colony is lactic acid bacteria.

Purifying culture bacteria

Purifying was started by choosing colonies around the clear zone. Ose was sterilized and touched to the surface of colony bacteria then inoculated at the surface of medium NA using scratch method to get separated colonies. Last, it was incubated (T 37°C, 24 h).

Characterization of bacterial probiotic

Assessment of gram

Observation of colony morphology was done through the gram staining technique. First, bacteria were placed in an objective glass and fixed. About 2-3 drips of gram A (crystal violet) were dripped in colony bacteria, and stored for 60 seconds. Then, the preparation was cleaned using flow water and dried. About 2-3 drip of gram B (lugol) was dripped in prepared and stored for 60 seconds. The preparation was then cleaned with flow water and dried. After that, the preparation was dripped with 2-3 drips of alcohol-acetone and stored for 60 seconds, cleaned again, and dried. The preparation was dripped with safranin 2-3 drips and stored for 60 seconds, then cleaned and dried. Lastly, this preparation was observed under a microscope.

Catalase test

Isolated bacteria were taken for 1 ose (round ose) from each culture stock and then dipped in H2O2 which had been dropped before on the object glass. A positive result is indicated by the appearance of gas bubbles in ose.

Inhibition test of bacterial pathogens

To study the probiotic potential of this isolated bacteria it is necessary to test the bacteria inhibition to bacterial pathogens. These two pathogen bacteria were *Aeromonas hydrophila* and *Staphylococcus aureus*. The method here was the spread plate method. Bacterial pathogens and bacteria with probiotic potential were suspended until they had the same turbidity visually as Mc Farland suspension, i.e., 106 CFU/ml. Bacterial pathogens *(A. hydrophila* and *S. aureus*) were isolated to petri that contain NA media by using the spread plate method. A hole (well) was made in the center of the media and 0.5 mL of LAB suspension was put in the well, then it was incubated at 35°C for 24h. The observation was done after incubation by observing the inhibition indication through the appearance of the clear zone in media that indicated the inhibition potential to pathogen bacteria. This clear zone was measured using a ruler (mm). The diameter of the inhibition zone was classified as weak (5 mm), moderate (5-10 mm), strong (10-20 mm), and very strong (20-30 mm) (Saham et al., 2022).

Results and Discussion

The chemical characteristics of Mbarase and Mudu which include carbohydrate, protein, fat, water, ash, and fiber are shown in Table 1. Different kinds of fish part material for fermentation cause different chemical characteristics. Mbarase has a higher percentage of protein and fat than Mudu, as it is made from Siganus sp baby fish. Mudu has a higher content of carbohydrates, water, ash, and fibre, than in Mbarase, as it is made from fish innards.

Table 1. Chemical characteristics of *Mbarase* and *Mudu*

Sample	Chemical Characteristic					
	Carbohydrate (%)	Protein (%)	Fat (%)	Water (%)	Ash (%)	Fiber (%)
Mbarase	7.732	25.19	4.558	57.290	5.130	0.16
Mudu	21.022	11.8	0.420	60.171	6.587	1.94

Mudu has higher carbohydrates than mbarase. This is caused by different parts of fish that are used in fermentation. Mudu in this study was made from skipjack innards which is a big fish, while mbarase was obtained from small fish (rabbitfish) with size 1-2 cm fermentation. In comparison with rusip which is also made from small fish (anchovy), mbarase also has smaller carbohydrates. Rusip has 14,117% of carbohydrate. The higher content of carbohydrates is due to the addition of any carbon source (palm sugar) (Putri et al., 2014). Nevertheless, in comparison to fresh material, carbohydrate content will be reduced in fish fermented food, as a consequence of the use of biomolecules by microorganisms in the fermentation process (Ilyanie et al., 2020). A previous study reported the carbohydrate content in fresh and fermented anchovy (rusip) was approximately between 5.1127% and 14.1171% (Putri et al., 2014).

Protein content in mbarase was higher than mudu, which that caused by different parts of fish that are used for fermentation. This high content of protein in mbarase is caused by 2 factors; the source of raw material which is rabbitfish with high protein and the presence of lactic acid bacteria, a source of single-cell protein, that grows during fermentation (Rinto, 2017). Fresh rabbitfish has 15-24% protein (Wahyuningtyas et al., 2017). The protein value in mudu is similar to bakasang which has 12-17% of protein 9 Hursepuny et al., 2021). Fat in mbarase was higher than mudu, as contributed by fresh rabbitfish that consist of 0.1-22% of fat (Wahyuningtyas et al., 2017). This fat value in mbarase is also higher than in rusip, 0.952%, and decreases during fermentation in comparison with the fat value in fresh anchovy, 2.6086% (Putri et al., 2014). Although there is no data for fresh rabbitfish before fermentation in this study, some study shows the decreasing of fat content during fermentation, that caused by fat degradation by microbial

activity and enzymes in those fish (Putri et al., 2014; Rinto, 2017; Wahyuningtyas et al., 2017; Hursepuny et al., 2021; Neti, 2007).

The water and ash content of mbarase and mudu were slightly similar. The high content of water correlates to the source of fish and process cleaning and washing before fermentation. During fermentation, water content will increase. Carbohydrate degradation in fish will produce water, whereas glucose will turn into lactic acid and water. Protein in fish also will turn into dipeptides, peptides, and amino acids that release water (Putri et al., 2014; Rinto, 2017; Wahyuningtyas et al., 2017; Hursepuny et al., 2021; Neti, 2007; Chan et al., 2023). The ash content in food refers to the presence of minerals and trace elements left or not destroyed after the sample has been heated by high temperatures (Kari et al., 2022). Ash content in mbarase and mudu is lower than rusip (50,070%). Ash content correlates to the salt content that is added for fermentation. Mineral in fresh fish only comes from the fish, and adding salt for fermentation, will increase mineral content (Putri et al., 2014). However, these percentages of ash may be different due to the source of fish, feeding behavior, environment, and ecosystem (Kari et al., 2022). The crude fiber content of mbarase and mudu in this study was low. This low content of crude fiber is because fish are not vegetable resources, but its animal resource, so it doesn't have much fiber (Putri et al., 2014).

Mbarase and Mudu fermentations are spontaneous fermentation at anaerobic conditions by utilizing microbe (lactic acid bacteria, LAB) which is already present in raw fish. This fermentation environment was controlled by adding high salt. The roles of salt are 1) to promote the growth of LAB over spoilage bacteria, and 2) to inhibit pectinolytic and proteolytic enzymes that can cause softening and putrefaction (Nuraida, 2015). In this study, LAB identification was done by morphology observation with a clear zone around the colony, gram test, and catalase test. A clear zone can be seen clearly in Mbarase sample and slightly seen in Mudu sample. From three repetitions of LAB identification in mudu, the clear zone was not too clear in two of them, only one was slightly seen. Bubble gas appearing in the catalase test means that the catalase test was positive (Nadia et al., 2020), indicating that lactic acid bacteria has not yet been identified. There was no clear zone not due to the absence of LAB, but allegedly due to the fermentation time. Fermentation time of 2-4 weeks the bacteria were in the lag phase (adaptation to the environment) in which the microbes have not yet divided, bacteria acclimatizing to environmental conditions (pH, temperature, nutrients) (Hursepuny et al., 2021). Mudu in this research was freshly fermented for 2 weeks.

Mbarase shows different results in microbial identification. In terms of colony morphology, Mbarase has a milky white color, opaque, entire margin, and there is a clear zone around the colony. Purple results from gram staining indicate gram-positive bacteria. In the catalase test no bubble gas appeared means negative catalase and indicated that LAB was identified. LAB in the catalase test is negative because it does not produce the enzyme catalase. This enzyme can convert hydrogen peroxide into water and oxygen, which is related to the ability of LAB which only needs a little oxygen to be able to live (Nadia et al., 2020).

LAB ferments carbohydrates to almost entirely lactic acid (homo-fermentation) or a mixture of lactic acid, carbon dioxide, acetic acid, and/or ethanol (hetero-fermentation). Other compounds that are also produced are diacetyl, acetaldehyde, and hydrogen peroxide. These compounds impacted the flavor and texture of fermented food. They also contribute to the inhibition of undesirable microbes (Nuraida, 2015). LAB that was isolated from Mbarase explored its potential as a probiotic against two pathogen bacteria, i.e., *Staphylococcus aureus* and *Aeromonas hydropila*. The average of each inhibition zone diameter of Mbarase against these two bacteria is shown in Table 2. The inhibitory zone produced for these two pathogen bacteria can be categorized as strong.

Table 2. The inhibition zone of *Mbarase* to *S. aureus* and *A. hydropila*

Bacteria	Inhibition zone (cm)			Average
	1	**2**	**3**	
S. aureus	10	13.5	10.5	11.3
A.hydropila	14	13	8	11.7

The antibacterial potential of LAB isolated from fermented food to pathogen bacteria has also been reported before. LAB-isolated from Bakasang sample had inhibitory activity against pathogenic bacteria and spoilage bacteria (*E. coli, S. aureus,* and *P. fluorescens*). The diameter of the inhibition zones was varied and ranged between 3 – 15 mm. The activity of LAB culture as inhibitory to bacterial pathogens and spoilage bacteria is supported by the acid and the components of metabolites produced. The acid produced by LAB has the effect of antimicrobial against enteric pathogens. In addition to producing acid, LAB also produces other compounds inhibiting such as diacetyl, and hydrogen peroxide, and some strains produce bacteriocins (Lawalata et al., 2011). LAB-isolated from Plaa-som, a Thai fermented food, showed an ability to suppress and eliminate pathogenic bacteria such as *E. coli* and *Salmonella spp*. within 24h. The inhibitory activities are due to the production of organic acids by LAB (Saithong et al., 2010). LAB isolated from Bekasam had antimicrobial activity to *E. coli, S. typhimurium, B. cereus*, and *S. aureus* (Choesri et al., 2015).

In this research, we didn't identify specific LAB species isolated from Mbarase. LAB-isolated from fish-fermented food show different species of LAB, such as *L. plantarum* and *L. reuteri* in Plaa-som (Hursepuny et al., 2021), LAB genus Leuconostoc, Lactococcus, Pediococcus, Enterococcus and Lactobacillus in Loah Ko-Dalla (Yonle & Pal, 2014), *Pediococcus acidilactici in* Bakasang (Lawalata et al., 2011), *Lb. plantaru, Lb. curvatus, Lb. murinus,* and *Strep. thermophilus* was isolated from Ikan Peda (Surono, 2016). Therefore, future study is needed to identify LAB species in Mbarase. Nevertheless, the presence of antimicrobial activity against pathogen bacteria and nutritional content in this fermented food indicated that Mbarase as a local probiotic food with a simple fermentation process, has cheap, and economic benefits.

Conclusion

Mbarase and Mudu have different proximate composition values due to their different raw fish used in the making of each product. Mbarase has higher protein, and fat, and is also lower in carbohydrates than Mudu. Lactic acid bacteria that are isolated from Mbarase have antibacterial potential to pathogen bacteria. As these two fish-fermented foods have high protein content, low in fat and ash, Mbarase and Mudu may become a source of nutrients. They also have potential as probiotic foods.

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References

Afrianti, L. H. (2014). *Teknologi pengawetan pangan*. Bandung: Alfabeta

Amelia, R., Philip, K., Pratama, Y. E., & Purwati, E. (2021). Characterization and probiotic potential of lactic acid bacteria isolated from dadiah sampled in West Sumatra, *Food Sci. Technol,* Campinas, *41*(2), 746-752. https://doi.org/10.1590/fst.30020

Chan, S. X. Y., Fitri, N., Mio Asni, N. S., et al. (2023). A comprehensive review with future insights on the processing and safety of fermented fish and the associated changes. Foods, 12(3), 558. https://doi.org/10.3390/foods12030558

Choesri, D., Rusmana, I., Suwanto, A., & Mubarik, N. R. (2015). Characterization of lactic acid bacteria isolated from an Indonesian fermented fish (bekasam) and their antimicrobial activity against pathogenic bacteria. Emirates Journal of Food and Agriculture 25(6), 489-494. https://doi.org/10.9755/ejfa.

Hursepuny, J. J., Moniharapon, T., & Mailoa, M. M. (2021). Chemical and microbial characteristics bakasang of innards yellow fin tuna (*Thunnus albacares*). *Inasua-Jurnal Teknologi Hasil Perikanan, 1*(2), 1-5. https://doi.org/10.30598/inasua.2021.1.2.86

Ilyanie, H. Y., Huda-Faujan, N., & Ida Muryany, M. Y. (2020). Comparative proximate composition of Malaysian Fermented Shrimp Products. *Malays. Appl. Biol*., *49*(3), 139–144. https://doi.org/10.55230/mabjournal.v49i3.1557

Kari N. M., Ahmad, F., & Ayun, M. N. A. (2022). Proximate composition, amino acid composition and food product of anchovy: a review. *Food research, 6*(4), 16-29. https://doi.org/10.26656/fr.2017.6(4).419

Lawalata, H. J., Sembiring, L., & Rahayu, E. S. (2011). Molecular identification of lactic acid bacteria producing antimicrobial agents from Bakasang, An Indonesian Traditional Fermented Fish Product. *Indonesian Journal of Biotechnology, 16*(2), 93-99. https://doi.org/10.22146/ijbiotech.16368

Mahulette, F., Mubarik, N. R., Suwanto, A., & Widanarni. (2020). Microbiological and physicochemical characteristics of inasua traditional fish fermented from Maluku Island. *Biosaintifika Journal of Biology & Biology Education, 10*(2), 298-305. http://doi.org.10.15294/biosaintifika.v10i2.13537

Marti-Quijal, F. J., Remize, F., Meca, G., Ferrer, E., Ruiz, M-J., & Barba, F. J. (2020). Fermentation in fish and by product processing: an overview of current research and future prospect. *Current Opinion in Food Science, 31*, 9-16. http://doi.org./10.1016/j.cofs.2019.08001

Nadia, A. B., Jannah, S. N., & Purwantisari, S. (2020). Isolation and characterization of lactic acid bacteria from *Apis mellifera* stomach and their potential as antibacterial using in vitro test against growth of *Staphylococcus aureus* and *Salmonella typhimurium. NICHE Journal of Tropical Biology, 3*(1), 35-44. https://doi.org/10.14710/niche.3.1.35-44

Neti, Y. (2007). Proses fermentasi rusip yang terbuat dari ikan teri (*Stolephorus* sp). Agritech, 27(1), 1-5. https://doi.org/10.22146/agritech.9488

Ngasotter, S., Waikhom, D., Mukherjee, S., Devi, M. S., & Singh, A. S. (2020). Diversity of Lactic Acid Bacteria (LAB) in fermented fish products: A review. *Int.J.Curr.Microbiol.App.Sci 9*(5), 2238-2249. https://doi.org/10.20546/ijcmas.2020.905.255

Nuraida, L. (2015). A review: Health promoting lactic acid bacteria in traditional Indonesian fermented foods. *Food Science and Human Wellness, 4,* 47-55. http://dx.doi.org/10.1016/j.fshw.2015.06.001

Putri, D. M., Budiharjo, A., & Kusdiyantini, E. (2014). Isolasi, karakterisasi bakteri asam laktat, dan analisis proksimat dari pangan fermentasi rusip ikan teri (*Stolephorus* Sp.). *Jurnal Biologi, 3*(2), 11-19.

Rinto, H. S. (2017). Kajian keamanan dan kualitas rusip bangka. *Prosiding Seminar Nasional dalam Rangka Dies Natalis ke-54 Fakultas Pertanian Universitas Sriwijaya*, Palembang 9 November 2017. https://repository.unsri.ac.id/24114/1/PI_680-685_Rinto-HAfif.pdf

Saithong, P., Panthavee, W., Boonyaratanakornkit, M., & Sikkhamondhol, C. (2010). Use of a starter culture of lactic acid bacteria in Plaa-som, a Thai Fermented Fish. *Journal of Bioscience and Bioengineering, 110*(5), 553–557. https://doi.org/10.1016/j.jbiosc.2010.06.004

Sanam, M. U. E., Detha, A. I. R., & Rohi, N. K. (2022). Detection of antibacterial activity of lactic acid bacteria, isolated from sumba mare's milk, against *Bacillus cereus*, *Staphylococcus aureus*, and *Escherichia coli*. Journal of Advanced Veterinary and Animal Research, 9(1), 53-58. https://doi.org/10.5455/javar.2022.i568

Sharma, R., Gang, P., Kumar, P., Bhatia, S. K., & Kulshrestha, S. (2020). Microbial Fermentation and Its Role in Quality Improvement of Fermented Foods. Fermentation, 6(4), 106. https://doi.org/10.3390/fermentation6040106

Singh, A., Kumar, M., Ghosh, M., & Ganguli, A. (2014). *Traditional foods and beverages as delivery vehicles for probiotics*. In book: Traditional foods and beverages as delivery vehicles for probiotics (pp.foth coming) Publisher: Recent Developments in Biotechnology, 413-439.

Surono, I. S. (2016). *Ethnic fermented foods and beverages of Indonesia*. Ethnic Fermented Foods and Alcoholic Beverages of Asia, 341-382. https://doi.org/10.1007/978-81-322-2800-4_14

Wahyuningtyas, L. A., Nurilmala, M., Sondita, M. F. A., Taurusman, A. A., & Sudrajat, A. O. (2017). Nutritional profile of Rabbitfish (*Siganus* spp.) from the kepulauan seribu (Thousand Islands), Jakarta, Indonesia. *International Food Research Journal 24*(2), 685-690.

Yonle, R., & Pal, J. (2014) Microbial Diversity and Proximate Composition of Loah Ko Dalla, A Smoke-Dried Fish Product of Darjeeling Hills*. Asian Journal of Microbiology, Biotechnology and Environmental Science, 8,* 9–14.