E-ISSN: 2347-5129
P-ISSN: 2394-0506
(ICV-Poland) Impact Value: 76.37
(GIF) Impact Factor: 0.549
IJFAS 2023; 11(4): 37-40

www.fisheriesjournal.com
Received: 26-04-2023
Accepted: 28-05-2023

Adistia Rahmaniar
Student, Faculty of Fisheries and Marine Sciences, Padjajaran University, Indonesia

Iskandar
Lecturer, Faculty of Fisheries and Marine Sciences, Padjajaran University, Indonesia

Fittrie M Pratiwy
Lecturer, Faculty of Fisheries and Marine Sciences, Padjajaran University, Indonesia

Roffi Grandiosa Herman
Lecturer, Faculty of Fisheries and Marine Sciences, Padjajaran University, Indonesia

Corresponding Author:
Adistia Rahmaniar
Student, Faculty of Fisheries and Marine Sciences, Padjajaran University, Indonesia

Comparison of fermentation procedures *Lemna* sp. On artificial feed

Adistia Rahmaniar, Iskandar, Fittrie M Pratiwy and Roffi Grandiosa Herman

DOI: https://doi.org/10.22271/fish.2023.v11.i4a.2823

Abstract
Feed is one of the important factors in aquaculture with a high enough content value for fish. So the need to formulate high-quality fish feed that suits the needs of fish increases. *Lemna* sp. is a water weed that can be used as an alternative ingredient for fish feed. This is because *Lemna* sp. has a high content of nutrients. A high crude fibre content in vegetable ingredients causes one of the obstacles to use as fish feed ingredients. However, this can be overcome by processing feed ingredients using the fermentation method. With the help of fermenter solutions, the fermentation process can produce simple combinations. This makes it easier to decompose than before. The fermentation procedure can affect the nutrient content of nutrients in *Lemna* sp. with flour and molasses and probiotics during the three-day incubation period is the best fermentation procedure. Fermentation is influenced by several factors, including the duration of incubation, number of inoculums, dose of probiotics, particle size, and pH.

Keywords: Influence, *Lemna* sp., fermentation procedure, fermentation *Lemna* sp.

1. Introduction
Feed is one of the important aspects that must be considered in cultivation activities. This is because feed is a source of growth energy for fish [1]. The feed should be given to fish effectively and efficiently. This is so that the fish body can use feed for growth. With the development of aquaculture, there is an increasing need to formulate feed that suits the needs of fish in a quality manner [2]. *Lemna* sp. is an alternative fish feed ingredient that can be found in shallow waters, swamps, and lakes [3]. *Lemna* sp. is a water weed that has a high nutrient content but is difficult to control. Based on [5], *Lemna* sp. contains high protein and minerals. *Lemna* sp. consists of a protein content of 10-45%, fiber 7-14%, carbohydrates 35%, fat 3-7%, and mineral and vitamin content is quite high [6].

With *Lemna* sp. which contains high crude fiber and there are anti-nutritional substances and amino acid composition, causing one of the obstacles to using *Lemna* sp. as vegetable raw materials in the fish feed that are different from animal protein raw materials [7]. Based on [7], *Lemna* sp. contains crude fiber of 20.08%. But this can be minimized with fermentation technology. This is because the fermentation process can improve the properties of raw materials. In addition, the fermentation process can increase the absorption of nutrients and produce a taste and aroma that fish like. This causes the fermentation process to produce compounds simpler and easier to digest than before [8]. Based on [9], fermentation quality is influenced by temperature, pH, nature, and composition of the medium, dissolved O2 and CO2, and incubation period. In addition, the smaller the particle size in the fermentation process, the more microbes come into contact with the material in starch degradation. And more and more amount of bound water is released so that the texture of the material becomes soft and porous [10].

2. Fermented *Lemna* sp
Fermentation is a process to simplify organic compounds and involves microorganisms [8]. The principle of fermentation is a biological process using microorganisms, either naturally or by adding yeast/fungal inoculum [11].

The fermentation process is carried out using a fermenter solution [12]. According to [13], probiotics used in the fermentation process contain microorganisms that can increase the nutrient content in fish feed. In line with research [14], the fermentation process will produce better nutritional value and improve the basic properties of raw materials such as increasing nutrient absorption, removing toxic substances, and causing a taste and aroma that fish like. Factors that affect the fermentation process are the incubation period, number of *starters*, fermentation media, temperature, oxygen, and pH [15].

Based on [7], *Lemna* fermentation can be done by weighing the harvested *Lemna* and then cleaning it to determine the dose of probiotics to be given. The dried *Lemna* that was weighed and then put in a solution of fermentor derived from probiotics and molasses with a probiotic dose of 5% with a temperature of 34 °C was stored in a ziplock that had been perforated to obtain aerobic conditions with an incubation period of 7 days. The success of the fermentation process can be said to be successful by showing the characteristic sour smell of fermentation [14]. Based on [7], the nutritional content of *Lemna* sp. before fermentation is 94.12% moisture content, 15.92% ash content, 19.17% protein, 18.37% crude fiber, 2.70% fat, and 43.84% carbohydrates. While the nutritional content of *Lemna* sp. after fermentation is moisture content 7.45%, ash content 20.76%, protein 23.47%, crude fiber 13.57%, fat 2.29%, and carbohydrates 39.91%.

Research [16], the addition of fermented *Lemna* flour by 10% mixed with soybean flour in carp feed gives the best results so that it can be used in cultivation. Based on research [4], the utilization of fermented *Lemna* by 10% in carp feed provides more optimal results when compared to feeding using soybean flour. In addition, research [17] shows the use of fermented *Lemna* can replace soybean flour as feed for Siamese catfish.

3. *Lemna* Fermentation Procedure

Fermentation yields *Lemna* SP flour is obtained on the basis of a proximate test. The proximate test consists of the following procedures

1. Fermentation of *Lemna* sp. by mixing *Lemna* sp. 100 grams with fermenter solution using a ratio of molasses and BIOM-S probiotics, which is 1: 1 then given 100 ml of water (homogenization) and incubated for 7 days. (Dry *Lemna* sp.) [18].
2. Fermentation of *Lemna* sp. using *Lemna* sp. as much as 100 grams mixed with BIOM-S probiotics as much as 7 ml and incubated for 7 days. (Dry *Lemna* sp.) [19].
3. Fermentation of *Lemna* sp. was done by mixing 2 kg of *Lemna* sp. who were given a solution of 3% molasses fermenter and 10 ml of water given BIOM-S probiotics as much as 3ml / litre then incubated for 7 days (dry *Lemna* sp.) [20].

Fermented flour *Lemna* sp. by mixing *Lemna* sp. flour as much as 100 grams with fermenter solution using a ratio of molasses and BIOM-S probiotics, which is 1: 1 then given 100 ml of water (homogenization) and incubated for 3 days. (*Lemna* sp. flour) [18]. With such procedures have proximate test results shown in Table 1.

Table 1: Proximate Test Results of *Lemna* sp. Flour Fermentation Procedure

Procedure	Incubation Period (days)	Womb							
		Water (%)	Ash (%)	Protein (%)	Crude Fiber (%)	Fat (%)	BETN (%)	TDN (%)	Metabolic Energy (Kcal/kg)
Molasses: BIOM-S	7	11, 49	9, 68	22, 50	18, 22	1, 73	47, 87	65, 53	3264
BIOM-S	7	8, 39	10, 32	22, 62	20, 73	1, 61	44, 72	62, 16	3386
Molasses and Probiotics	7	8, 59	10, 21	22, 57	16, 73	1, 73	48, 76	66, 76	3359
Molasses and Probiotics	3	9, 89	9, 36	23, 93	17, 73	1, 37	47, 61	66, 50	3345

***Source*:** Results of Ruminant Animal Nutrition and Animal Food Chemistry Laboratory Analysis of Faculty of Animal Husbandry, Padjadjaran University 2023

All procedures undergo incubation treatment at room temperature 29 °C±1. Based on the data in Table 1, the procedure using molasses and probiotics with an incubation period of three days (in the form of flour) is the best procedure. It is based on the best CP (*crude protein*). *Lemna sp.* flour that has been fermented using BIOM-S probiotics containing Lactobacillus sp., *Saccharomyces* sp., *and* Bacillus *sp*. [20].

4. Results and Discussion

Based on the results of proximate analysis, the best procedure for *Lemna* fermentation is in the form of flour using molasses and BIOM-S probiotics in a ratio of 1: 1 diluted with 100 ml of water with an incubation period of 3 days. After fermentation, *Lemna* flour undergoes an increase in protein levels caused by the microbial work process in fermentation. The increase in protein comes from an increase in fungal biomass derived from single-cell protein (SCP) [7]. *Lactobacillus* sp. in probiotics also increases the secretion of proteolytic enzymes (digestibility of feed) by breaking down proteins into amino acids that can be absorbed faster by the intestine. With the fermentation process, the energy required will be less so that excess energy will be used for growth [21]. According to [1], the fermentation process in feed results in higher feed absorption by fish. Furthermore, the fermentation process is able to convert long-chain proteins into short-chain peptide bonds so that they are easily absorbed by fish for growth.

Decrease in crude fibre content in fermented *Lemna* flour due to the presence of cellulase enzymes. Cellulase is a group of fibrinolytic enzymes capable of hydrolyzing fibres in plant cell walls into glucose. The decrease in crude fiber due to fermentation time goes hand in hand with the growth of mycelium, at the same time cellulose and hemicellulose will be degraded [22]. This statement is not in line with research that has been conducted by [23] if *Lemna* fermented using bacillus probiotics with an incubation period of 48 hours can benefit poultry, this can be seen from the growth curve of B. KATMIRA1933 *subtilis* reached 9.48 Log CFU/g after 24 hours, and 11.31 Log CFU/g after 48 hours, while *B. amyloliquefaciens* B-1895 reached 8.47 Log CFU/g after 24 hours, and 10.17 Log CFU/g after 48 hours. The number of probiotics given is related to the size of the microbial population which has the opportunity to determine the speed of development which will further facilitate the substrate so that it can affect the final product [24].

The large number of inoculum used, the time used to increase the crude protein content is faster. The length of fermentation time and the right amount of inoculum are factors that determine the crude fiber content that will be produced from the fermentation process [25]. Based on [15], the process of fermentation of *Lemna* with probiotics as much as 5% produces good nutritional value when compared to the percentage of probiotics 1% and 3%. Modification of crude fiber in *Lemna* that has gone through the fermentation process occurs due to the activity of bacteria contained in the probiotics used.

pH is one of the important factors in the fermentation process because it can affect microbial growth conditions [26]. According to [27], pH is related to the acidity level of the medium that will determine the growth of microorganisms. pH that is too low or high can trigger microbial cell death [27]. The high rate of microbial death can affect the speed of fermentation because the number of microbes to break down glucose is reduced. In addition, [28] optimal pH values also affect microbial growth, ranging from 3-6.

Particle size has an important role in the fermentation process of solid media related to the surface area to volume ratio of the substrate. The ratio of surface area to volume increases with the decrease in particle size. The optimal particle size of the substrate affects the ability to penetrate the mycelium of microorganisms into the substrate [29]. Microorganisms that have low penetrating ability require a higher surface area to volume ratio, in other words, require smaller particle sizes [30]. Fermented sweet potatoes with a thickness of 1 mm can increase viscosity values, reduce moisture and starch content, and are not easily retrograded [10].

5. Conclusion

The high crude fiber in *Lemna* sp is an obstacle to being used as feed. The solution is to reduce crude fiber by fermentation. The use of correct fermentation procedures can increase the content contained in *Lemna* sp. as an artificial feed. The best procedure for *Lemna* fermentation is in the form of flour using molasses and BIOM-S probiotics with a ratio of 1: 1 diluted with 100 ml of water with an incubation period of 3 days where the results of the proximate analysis that has been carried out produce that it has a water content of 9.89%, ash 9.36%, crude protein 23.93%, crude fiber 17.73%, crude fat 1.37%, BETN 47.61%, TDN 66.5%, and energy 3345 kcal/kg. It is based on the best CP (crude protein). It is influenced by time, the number of inoculums, particle size and pH.

6. Reference

1. Hadijah I, Mustahal AN, Son. Effect of prebiotics in commercial feed on catfish growth (*Pangasius sp.*). Journal of Fisheries and Marine. 2015;5(1):33-40.
2. Kallau M. Increased bioavailability of vegetable fish feed with fermenting agents; c2013. p. 463-475.
3. Prasetyowati L. The Effect of Variations in the Addition of Duckweed (*Lemna* sp) in Feed and Its Application as Catfish Feed (*Clarias sp*). Agroteknose (Journal of Agricultural Technology and Engineering). 2016;7(2):21-31.
4. Winarti W, Subandiyono S, Sudaryono A. Utilization of Fermentation of *Lemna* Sp. Flour in Artificial Feed on the Growth of Goldfish (*Cyprinus carpio*). Journal of Aquaculture Science Technology. 2017;1(2):88-94.
5. Nopriani U, PDMH K, Prihantoro I. Productivity of Duckweed (*Lemna minor*) as an alternative forage fodder with different light intensity. JITV. 2014;19(4):272-286.
6. Iqbal S. Duckweed cultivation. Potentials, Possibilities and Limitations for Combined Wastewater Treatment and Animal Feed Production in Developing Countries. SANDEC Report No. 6/99. 1999;6(99):1-89.
7. Zidni, Iskandar, Andriani Y. Fermentation of *Lemna* sp. as Fish Feed Ingredients to Increase the Provision of Animal Protein Sources for the Community. National Seminar on Building Food Security through Local Commodity Empowerment; c2016 Nov. p. 1-6.
8. Ulungkas W. Fermentation technology is an alternative solution in an effort to utilize local feed ingredients. Aquaculture Media. 2011;6(1):43. https://doi.org/10.15578/ma.6.1.2011.43-48
9. Suryaningrum LH. Microbial application in an effort to improve the quality of fish feed raw materials through fermentation. Proceedings of Biology Achieving Sustainable Development Goals with Biodiversity in the Face of Climate Change; c2021 Nov. p. 204-210.
10. Pusparani T, Yuwono SS. The Effect of Natural Fermentation on Sweet Potato Chips (*Ipomoea batatas*) on the Physical Properties of Sweet Potato Flour Fermentation. Food and Agroindustry. 2014;2(2):59-69.
11. Prabowo A. The use of feed fermentation technology in the integration system of cattle-corn crops. Triton Journal: Agriculture. 2016;7(2):99-106.
12. Aslamsyah S, MY Karim, Badraeni. Dosing effects of mixed microorganisms. In fermented feed, raw materials contain *Sargassum* sp. Against Growth Performance, Body Chemical Composition and Hepatosomatic Index of Whitefish, (*Chanos chanos* Forsskal). Torani: Journal of Fisheries and Marine Sciences. 2018;1(2):59-70.
13. Linayati TA, Prasetyo, TY Mardiana. Growth Rate Performance of Milkfish (*Chanos chanos*) Fed with Enrichment Probiotics. Journal of R&D Pekalongan City. 2021;19(1):64-71.
14. Iskandar I, Andriani Y, Rostika R, Zidni I, Riyanti NA. The Effect of Using *Lemna* Sp. Fermentation on Fish Feed on the Growth Rate of Nilem Goldfish (*Osteochilus hasselti*). World News of Natural Sciences. 2019 Jul;26:157-166.
15. Sarungu YT, Ngatin A, Sihombing RP. Fermentation of hay as an additional feed for ruminants. Liquids. 2020;13(1):24-29.
16. Sukran SH, Suharman I, Adelina A. Effect of Fermentation of *Lemna* Leaf Meal (*Lemna minor*) on Feed Formulation on the Growth of Gurame Fish (*Osphronemus goramy*). Aquatic Science. 2021;9(2):86.
17. Syarif MI, Adelina, Suharman I. The effect of the use of *Lemna* flour (*Lemna minor*) fermented using kombucha on the growth of Siamese catfish fry (*Pangasianodon hypophthalmus*). 2022;10(2):120-128.
18. Zahidah, Gunawan W, Subhan U. Population growth of Daphnia spp. fed with floating net caramba cultivation waste fertilizer (KJA) in Cirata Reservoir which has been fermented EM4. Aquatic Journal. 2012;3(1):84-94.
19. Dwi Cahya M. Evaluation of Fermented Food Waste Feed Biomaterials and Their Application to the Performance of Sangkuriang Catfish (*Clarias gariepinus*). Thesis. Fisheries Study Program, Faculty of Fisheries and Marine Sciences. Padjadjaran University; c2022.

20. Iskandar, Pitaloka FA, Lewaru MW, Haetami K. Utilization of Fermented *Lemna* as a Source of Feed for the Growth of Sangkuriang Catfish (*Clarias sp*.). Asian Journal of Fisheries and Waters Research. 2022;20(6):86-94.
21. Mardhiana A, ID Buwono, Y Andriani, Iskandar. Commercial Probiotic Supplementation in Artificial Feed for Growth Induction of Sangkuriang Catfish (*Clarias gariepinus*). Journal of Fisheries and Marine. 2017;8(2):133-139.
22. Khairudin, Adelina, Suharman I. The Effect of *Lemna* Leaf Fermentation (*Lemna minor*) on Feed on the Growth of Gurame Fish (*Osphronemus goramy*). 2021;9(2):108-115.
23. Mahoney R, Sunday R, Huang Q, Dai W, Cao Y, Liu G, *et al.* Fermented Duckweed as a Potential Feed Additive with Poultry Beneficial Basil Probiotics. Probiotics and Antimicrobial Proteins. 2021;13(5):1425-1432.
24. Kumajas NJ, JSIT Onibala. Effect of Inoculum Dose and Incubation Time of *Phanerochaete chrysosporium* and Trichoderma reesei Fermentation Mixture on Hyacinth Nutrient Content. Zootech. 2022;42(1):97-104.
25. Montesqrit, Mirzah, Pratiwi S. The effect of fermentation duration and dose of *Bacillus amyloliquefaciens* inoculum on the nutritional content of paitan leaves (*Tithonia diversifolia*). Pastura. 2022;11(2):91.
26. Hendrawan Y, SH Sumarlan, CP Rani. Effect of pH and Fermentation Temperature on Ethanol Production from Rice Straw Hydrolysis. Journal of Tropical Agricultural Engineering and Biosystems. 2017;5(1):1-8.
27. Taslim M, M Mailoa, M Rijal. Effect of pH, and Duration of Fermentation on Ethanol Production of *Sargassum crassifolium*. Journal of Biology, Science & Education. 2017;6(1):13-25.
28. Mayangsari V, A Abtokhi. Analysis of the Effect of Temperature Variation and Time of Hydrolysis Process on Glucose Levels in the Utilization of *Lemna minor* as Bioethanol. Journal of Neutrinos. 2014;7(1):16-22.
29. Khasanah LU, BK Ananditho, AE Nugraheni. Effect of preliminary treatment of solid fermentation and liquid fermentation on yield and quality characteristics of cinnamon leaf essential oil. Agritech. 2014;34(1):36-42.
30. Indriani DO, LNI Syamsudin, FH Sriherfyna, AK Wardani. Reversal of *Aspergillus Niger* by Solid State Fermentation Method and Its Application in Industry: A Review of the Literature. Journal of Food and Agroindustry. 2015;3(4):1405-1411.