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# Effect of fermented seaweed addition on blood glucose level, hepatosomatic index, and gastric evacuation rate of milkfish, *Chanos chanos* larvae

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**Abstract**. Seaweed is a fishery product which can be used as the source of carbohydrate and binder in fish feed composition. However, excessive use of seaweed results in high crude fiber. This disadvantage could be avoided by the fermentation of the seaweeds. The aim of this study was to evaluate a variety of seaweeds fermented using microorganism towards glucose level, hepatosomatic index and gastric evacuation rate to see the impact of seaweed use on milkfish (*Chanos chanos*) larvae. The five types of seaweed used were green strain of *Kappaphycus alvarezii*, brown strain of *K. alvarezii*, *Gracilaria gigas*, *Sargassum* sp. and *Caulerpa* sp. The measurement result of blood glucose level, hepatosomatic index, and gastric evacuation rate consistently showed the highest value on the use of seaweed *G. gigas*.
**Key Words**: fermentation, microorganism, seaweed, feed ingredient, milkfish larvae.

**Introduction**. Milkfish (*Chanos chanos*) is one of the superior brackish water commodities in Indonesia. Many pond fish farmers cultivate this species in traditional system which only relies on live food available in the pond. However, increasing population and increase in demand which are not balanced by the availability of fish farming area makes intensive fish farming system becoming an option. Intensification of milkfish farming business at high stocking density completely depends on artificial food in the form of pellet. This condition causes the feed to be one of production factors that spends the highest cost, reaching approximately 60% of total production cost (De Silva & Hasan 2007).

Several studies (Aslamyah et al 2016, 2017) have been conducted in an effort to suppress the feed cost without reducing the quality of the feed itself. Fish feed quality, in addition to be determined by nutritional content, is also determined by physical quality, particularly the stability in the water. One of the ingredients that determine the quality is the binder. Additional material used as binder highly determines the stability of feed in the water (Meyer & Zein-Eldin 1972).

One of fishery products containing carbohydrate as well as has a function as binder is seaweed. In general, seaweed consists of water (27.8%), protein (5.4%), carbohydrate (33.3%), fat (8.6%), crude fiber (3%) and ash (22.25%). Besides carbohydrate, protein, fat, and fiber, seaweed also contains enzyme, nucleic acid, amino acid, vitamin (A, B, C, D, E, and K), macro minerals such as nitrogen, oxygen, calcium and selenium and also micro minerals like iron, magnesium and sodium (Aslamyah et al 2015). Seaweed has many benefits for humans, environment, and aquaculture so it needs to be studied, especially in South Sulawesi Province (Nursidi et al 2017).

In the previous study, we have reported that the use of artificial feed containing fermented seaweed as binder and carbohydrate source showed increasing growth, survival, digestibility level, and body chemical composition of milkfish juvenile (Aslamyah et al 2016). In this study, results of glycogen level in liver and muscle, blood glucose level, hepatosomatic index, and gastric evacuation rate of milkfish fed artificial feed containing fermented seaweed were reported. This study was aimed specifically to determine the best quality feed ingredient for milkfish by using seaweed as carbohydrate source and binder.

## Material and Method

***Research location***. The research was conducted from April to August 2015 in Fish Breeding Technology Laboratory, Fishery Department, Faculty of Marine and Fishery, Hasanuddin University, Makassar, Indonesia.

***Milkfish juvenile***. Experimental fish used was milkfish juvenile at 5.57±0.22 g in weight. The next procedure was in accordance with the procedure conducted by Aslamyah et al (2016).

***Seaweed***. Seaweeds used in this study were green strain of *Kappaphycus alvarezii*, brown strain of *K. alvarezii*, *Gracilaria gigas, Sargassum* sp. and *Caulerpa* sp. obtained from seaweed farmers in Punaga Village, Takalar Regency. Seaweeds were cleaned and dried, and further crushed into flour.

***Fermentation process***. Fermentation process was done by using 3 species of microorganisms with fermentation composition as follows: a). *Bacillus* sp. bacteria of 2 mL/100 g seaweed powder; b). 1.5% of ragi tape (yeast) as the source of *Rhizopu*s sp.; c). 1.5% baker's yeast as the source of *Saccharomyces* sp.; d) mix of *Bacillus* sp., *Rhizopu*s sp., *Saccharomyces* sp. at ratio of 1 mL:1 g:1 g/100 g seaweed flour, and control treatment resulted from fermentation process without using any bacteria. The best fermentation product was applied as fish feed mix to be used in further treatments.

Fermentation was started by weighing each of 100 g seaweed and further seaweeds were put into plastic with sealing. Every package was added with microorganisms solved in 20 mL sugar cane drop in each treatment by evenly sprayed it using sprayer. The package was tightly closed and incubated for 72 hours. After 72 hours, seaweed flour was steamed in boiled water for 1-2 minutes to inactivate the microorganism activity.

***Blood glucose level***. Observation of blood glucose level on experimental fish was done at the end of study. Milkfish was fasted for 24 hours and further fed to full. Experimental fish was transferred into container of large basin with aeration. Collection of experimental fish blood was started at hour-0 and hour- 1, 2, 4, 5, 6, 8, 10, 12, 14, 16, and 20 post prandial. Sample of experimental fish blood was collected from the gills and caudal vena by using spuit of 1 mL volume. Measurement of blood glucose level was performed through colorimetric analysis using reagent kit of Gluco Dr AGM 2100.

***Hepatosomatic index***. Hepatosomatic index was calculated by comparing the total weight of fish and the weight of liver. At first, fish was weighed for its weight, after that fish was dissected on ice. Dissection was carefully and quickly performed. The liver was weighed. The value of hepatosomatic index was measured based on the equation below:

$$HI = HP/W$$

where: IH = hepatosomatic index;
HP = hepatosomatic weight (g);
W = fish weight (g).

***Gastric evacuation rate***. Observation of gastric evacuation rate (GER) was done at the end of study. Furthermore, the measurement method applied has followed the procedure conducted by Lee et al (2000) which has been modified. Milkfish was fasted for 24 hours, and further fed to full. Fish stomach content was obtained after feeding, and later this process was done at interval of one hour. Stomach content collection was stopped when experimental fish stomach was empty. Feed percentage in digestive track was calculated using the formula:

$$\% \text{ GER} = \frac{\text{Feed in stomach}}{\text{Feed eaten}} \times 100$$

***Data analysis***. All data obtained in this study were analyzed through the analysis of variance and further was continued by response test at significance level of 5% using SPSS 12.0 and descriptively, that was by comparing the results obtained between treatments, and also comparing with supporting literature.

**Result and Discussion**

***Blood glucose level***. Blood glucose level of milkfish juvenile immediately increased after fish consumed feed (Figure 1). The highest blood glucose level reached its peak at hour-3 and experienced a decrease at hour-4 until it reached the lowest level at hour-16. The highest glucose level was shown by feed contained *G. gigas* seaweed of 138 mg $dL^{-1}$, followed by the green strain of *K. alvarezii* which amounted to 126 mg $dL^{-1}$, brown strain of *K. alvarezii* of 122 mg $dL^{-1}$, *Sargassum* sp. of 118 mg $dL^{-1}$, and the lowest was found in *Caulerpa* sp. of 86.5 mg $dL^{-1}$.

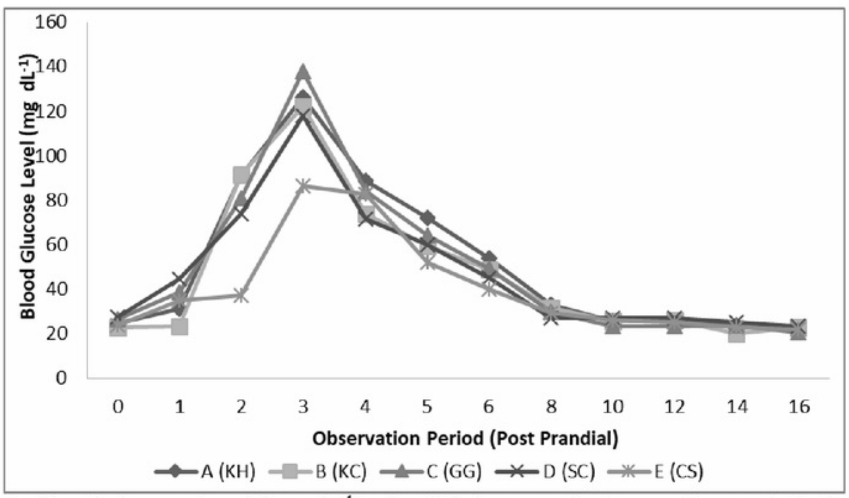


Figure 1. Blood glucose level (mg $dL^{-1}$) of milkfish on each observation period of various fermented seaweed (KH = green strain of *K. alvarezii*, KC = brown strain of *K. alvarezii*, GG = *G. gigas,* SC = *Sargassum* sp., and CS = *Caulerpa* sp.).

***Hepatosomatic index***. The results of the measurement of the hepatosomatic index of experimental fish treated with various fermented seaweeds for 60 days of maintenance, at the beginning and the end of study, are presented in Table 1.

Table 1

Hepatosomatic index of milkfish towards various fermented seaweeds

| *Treatment* | *Hepatosomatic index* |
|---|---|
| Initial value | 1.19±1.19 |
| A (KH) | 2.03±0.29 |
| B (KC) | 2.02±0.12 |
| C (GG) | 2.07±0.15 |
| D (SC) | 2.04±0.21 |
| E (CS) | 2.05±0.04 |

KH = green strain of *K. alvarezii*, KC = brown strain of *K. alvarezii*, GG = *G. gigas,* SC = *Sargassum* sp., and CS = *Caulerpa* sp.

The measurement result indicated that the highest hepatosomatic index was shown by the feed contained *G. gigas* even though the result of variance analysis revealed that the finding was not significantly different ($p > 0.05$) towards milkfish hepatosomatic at the end of maintenance.

***Gastric evacuation rate***. Gastric evacuation rate of experimental fish showed different time periods on each treatment. The fastest gastric emptying was found milkfish given feed contained *Caulerpa* sp. that was at hour-5 post prandial, while gastric evacuation rate in milkfish treated feed contained green strain of *K. alvarezii* strain, brown strain of *K. alvarezii*, *G. gigas,* and *Sargassum* sp. occurred at hour-6 (Table 2).

Table 2

Gastric evacuation rate (%) of milkfish on each observation period of various fermented seaweeds

| *Hour post prandial (hour)* | *Gastric evacuation rate (%)* | | | | |
|---|---|---|---|---|---|
| | *A (KH)* | *B (KC)* | *C (GG)* | *D (DP)* | *E (CP)* |
| 0 | 0 | 0 | 0 | 0 | 0 |
| 1 | 94.38±0.78 | 93.94±1.41 | 93.17±2.41 | 96.28±1.64 | 92.05±2.72 |
| 2 | 72.57±8.35 | 74.36±1.33 | 79.41±1.92 | 78.74±0.75 | 66.44±3.17 |
| 3 | 51.86±1.73 | 57.57±9.88 | 55.74±6.39 | 61.38±7.47 | 46.94±1.78 |
| 4 | 31.3±7.62 | 28.54±4.56 | 38.10±1.3 | 28.8±2.79 | 36.50±9.72 |
| 5 | 19.97±3.18 | 20.63±4.11 | 27.49±0.8 | 14.43±3.16 | 16.44±2.3 |
| 6 | 13.74±1.52 | 10.81±6.34 | 14.31±3.91 | 10.65±1.26 | 0 |
| 7 | 0 | 0 | 0 | 0 | 0 |
| 8 | 0 | 0 | 0 | 0 | 0 |
| 9 | 0 | 0 | 0 | 0 | 0 |

KH = green strain of *K. alvarezii*, KC = brown strain of *K. alvarezii*, GG = *G. gigas,* SC = *Sargassum* sp., and CS = *Caulerpa* sp.

**Discussion**. The use of fermented seaweed as feed ingredient has been widely reported (Zaki et al 1994; Norambuena et al 2015; Ilias et al 2015; Aslamyah et al 2017). The addition of fermented seaweed in feed for milkfish juveniles as the source of carbohydrate showed an increase in fish blood glucose level which reached its peak at hour-3 postprandial for all types of seaweeds. Nutrient intake is a factor that affects the glucose response (Martinez-Porchas et al 2009). Feed containing different fat and protein will result in different blood glucose levels as well (Cheng et al 2006). The highest peak was seen at the use of *G. gigas* seaweed of 138 mg $dL^{-1}$ and the lowest was found in *Caulerpa* sp. of 86.5 mg $dL^{-1}$. These values indicated that the use of *G. gigas* seaweed was able to supply adequate glucose which has a main role in animal bioenergy that will be converted into chemical energy (ATP) released in the form of mechanical energy (Lucas 1996).

The role of seaweed in improving nutrient absorption (Natify et al 2015) and metabolism process (Mustafa et al 1995) was also seen in the increasing hepatosomatic index of milkfish. Hepatosomatic index becomes an indicator of energy status in liver (Hismayasari et al 2015) and is important in estimating nutritional value in feed (Ighwela et al 2014). The highest value of hepatosomatic index was found in the use of *G. gigas*, while the lowest was obtained by the green strain of *K. alvarezii* despite overall difference in the value of hepatosomatic index in all types of seaweed did not show a significant difference ($p > 0.05$). It reflected the role of seaweed in increasing hepatosomatic index in milkfish larvae. Result obtained was in line with the finding of study conducted by Mustafa et al (1995) which found an increase in hepatosomatic index through the use of *Porphyra* sp. in red sea bream (*Pagrus major*).

Gastric evacuation rate showed stomach emptying at hour-7 in all types of seaweeds, except for in *Caulerpa* sp. namely at hour-6. Feed contained *G. gigas* indicated the most feed consumed at hour-6 which amounted to 14.31±3.91, while the least was found in *Caulerpa* sp. This finding showed that feed containing *G. gigas* was longer digested in stomach. It was due to the reason that *G. gigas* feed contained more carbohydrate and other nutrient component compared to other types of seaweed by considering the value of blood glucose level and hepatosomatic index which placed *G. gigas* to reach the highest value. Hepatosomatic index also increased along with the increasing intake of feed contained carbohydrate in *Oreochromis niloticus* (Ighwela et al 2014) and *Cyprinus carpio communis* (Ahmad et al 2012).

**Conclusions**. Seaweed *G. gigas* can be used as a source of carbohydrates through fermentation. Based on blood glucose level, hepatosomatic index, and gastric evacuation rate, this seaweed showed the highest values compared to the other species. Results of this study showed that the use of *G. gigas* seaweed fermented using mix of microorganisms was the best feed ingredient which was able to fulfill nutrient requirement for the growth of milkfish larvae.

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**References**

Ahmad M., Qureshi T. A., Singh A. B., 2012 Effect of dietary protein, lipid and carbohydrate contents on the viscera composition and organ indices of *Cyprinus carpio communis* fingerlings. African Journal of Biotechnology 11(33):8361-8366.

Aslamyah S., Karim M. Y., Badraeni, 2015 [Final Report: Diversification of fermented seaweed as a source of carbohydrates and binders in cheaper artificial feed and environmentally friendly to support the intensification of milkfish aquaculture in South Sulawesi]. PENPRINAS MP3EI 2011-2025 – Hasanuddin University, Makassar. [in Indonesian]

Aslamyah S., Karim Y., Badraeni, Tahya A. M., 2016 Seaweed as a source of carbohydrates in the feed of milk fish (*Chanos chanos* Forsskal). International Journal of PharmTech Research 9(11):64-67.

Aslamyah S., Karim M. Y., Badraeni, 2017 Fermentation of seaweed flour with various fermentors to improve the quality of fish feed ingredients. Jurnal Akuakultur Indonesia 16(1):8-14.

Cheng A. C., Chen C. Y., Liou C. H., Chang C. F., 2006 Effects of dietary protein and lipids on blood parameters and superoxide anion production in the grouper, *Epinephelus coioides* (Serranidae: Epinephelinae). Zoological Studies 45:492-502.

De Silva S. S., Hasan M. R., 2007 Feeds and fertilizers: the key to long term sustainability of Asian aquaculture. In: Study and analysis of feeds and fertilizers for sustainable aquaculture development. Hasan M. R., Hecht T., De Silva S. S., Tacon A. G. J. (eds), FAO Fisheries Technical Paper No. 497, FAO, Rome, pp. 19-47.

Hismayasari I. B., Marhendra A. P. W., Rahayu S., Saidin, Supriyadi D. S., 2015 Gonadosomatic index (GSI), hepatosomatic index (HIS) and proportion of oocytes stadia as an indicator of rainbow fish *Melanotaenia boesemani* spawning season. International Journal of Fisheries and Aquatic Studies 2(5):359-362.

Ighwela K. A., Ahmad A. B., Abol-Munafi A. B., 2014 The selection of visceresomatic and hepatosomatic indices for the measurement and analysis of *Oreochromis niloticus* condition fed with varying dietary maltose level. International Journal of Fauna and Biological Studies 1(3):18-20.

Ilias N. N., Jamal P., Jaswir I., Sulaiman S., Zainuddin Z., Azmi A. S., 2015 Potentiality of selected seaweed for the production of nutritious fish feed using solid state fermentation. Journal of Engineering Science and Technology. Special issue on SOMCHE 2014 & RSCE 2014 Conference, January (2015), pp. 30-40.

Lee S. M., Hwang U. G., Cho S. H., 2000 Effects of feeding frequency and dietary moisture content on growth, body composition and gastric evacuation of juvenile Korean rockfish (*Sebastes schlegeli*). Aquaculture 187:399-409.

Lucas A., 1996 Bioenergetics of aquatic animals. Taylor & Francis, 169 pp.

Martinez-Porchas M., Martinez-Cordova L. R., Ramos-Enriquez R., 2009 Cortisol and glucose: reliable indicators of fish stress? Pan-American Journal of Aquatic Sciences 4(2):158-178.

Meyer S. P., Zein-Eldin Z. P., 1972 Binders and pellet stability in development of crustacean diets. Journal of the World Aquaculture Society 3:351–364.

Mustafa G., Wakamatsu S., Takeda T., Umino T., Nakagawa H., 1995 Effect of algae meal as feed additive on growth, feed efficiency, and body composition in read sea bream. Fisheries Science 61(1):25-28.

Natify W., Droussi M., Berday N., Araba A., Benabid M., 2015 Effect of the sea weed *Ulva lactuca* as a feed additive on growth performance, feed utilization and body compotition of Nile tilapia (*Oreochromis niloticus* L.). International Journal of Agronomy and Agricultural Research 7(3):85-92.

Norambuena F., Hermon K., Skrzypczyk V., Emery J. A., Sharon Y., Beard A., Turchini G. M., 2015 Algae in fish feed: performances and fatty acid metabolism in juvenile Atlantic salmon. PLoS ONE 10(4):e0124042.

Nursidi, Ali S. A., Anshary H., Tahya A. M., 2017 Environmental parameters and specific growth of *Kappaphycus alvarezii* in Saugi Island, South Sulawesi Province, Indonesia. AACL Bioflux 10(4):698-702.

Zaki M. A., Nour A. M., Omar E., El-Din A. E. T., 1994 The use of seaweed meal in feeding common crap (*Cyprinus carpio* L.). AJAS 7(2):183-189.

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