Effect of seaweed, *Kappaphycus alvarezii* fermentation by various fermenters combinations as thickener on gel strength, attractiveness and palatability of gel diet in Tilapia, *Oreohromis niloticus*

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**Abstract.** Gel diet is one of the aquaculture diets that use seaweed, *K. alvarezii*, as a thickener made with cooking with water contents of between 50-70%. One effort to increase the nutritional content of seaweed is fermentation. However, fermentation affects the attractiveness, palatability, and gel strength of aquaculture artificial diet. This study aims to compare various fermenter combinations that are most effective in fermenting seaweed, *K. alvarezii* meal in terms of attractiveness, palatability, and gel strength. This research was carried out at the Laboratory of Nutrition and Feed Technology, Faculty of Marine Sciences and Fisheries, Hasanuddin University, Makassar, Indonesia. Experimental fishes used were tilapia size 7, 15, and 28 g. Various combinations of fermenters as treatment were seaweed meal without fermenter (treatment A), + 1.5g *Bacillus* sp. (treatment B), + 1.5g *Bacillus* sp. + 1.5 g of *Rhizopus* sp. (treatment C), + 1.5 g *Bacillus* sp. + 1.5g *Saccharomyces* sp. (treatment D), and + 1.5g *Bacillus* sp. + 1.5g *Aspergillus* sp. (treatment E). Fermentation is carried out on an an-aerobic basis. The parameters measured were attractiveness, palatability, and gel strength. The experimental design used was a completely randomized design with 5 treatments and 5 replications. Data were analyzed by ANOVA and W-Tukey test. The results of this study indicate that the attractiveness and gel strength of the experimental diet were the same in all treatments (p>0.05), while the highest palatability of the experimental diet was obtained in the experimental fish that consumed D diet, namely seaweed meal fermented with 1.5g *Bacillus* sp. + 1.5g *Saccharomyces* sp. Based on the results of this study, it was concluded that tilapia, which consumed the gel diet, contained seaweed fermented with 1.5g *Bacillus* sp. + 1.5g *Saccharomyces* sp. has the best palatability.

1. Introduction

Gel diet is an artificial aquaculture diet of the wet or moist type made with a variety of quality and environmentally friendly raw materials by cooking (without a pellet machine), and using a seaweed, *Kappaphycus alvarezii* as a thickener with a water content of between 50-70% [1,2]. Gel diet began to be developed at Hasanuddin University, Makassar, Indonesia, since 2013. It was further stated that the advantages of gel diet compared to other types of diet are (i) making it very practical and inexpensive and environmentally friendly, (ii) it was believed that all adults could make it, (iii) the tools needed to make it available to every household, using only stoves, pans, and gutters (iv) do not require maintenance tools that require expertise in the machine, (v) the scent spreads quickly to the
water media so that its presence is quickly detected by cultivars or its attractiveness (attractiveness) is high, (vi) its palatability is high, (vii) easy to digest because of its soft texture, and (viii) microorganisms and impurities attached to the raw material can be sterilized through cooking. Seaweed, K. alvarezi is the best thickener compared to several other thickening agents (rice and sago flour) available for the gel diet. Seaweed contains hydrocolloid, which is needed in industry as an emulsion solution, stabilizer, suspension, thickener, and freezing agent or adhesive. However, seaweed has crude fibers that are difficult to digest. For efficient utilization to be more optimal, it is necessary to ferment [3]. Fardiaz (1992) states that fermentation also functions to reduce crude fiber and increase nutrient content [4].

Some types of microorganisms that can act as fermenters in the seaweed fermentation process are Aspergilus sp., Bacillus sp., Rhizopus sp., and Saccharomyces sp. or a combination of two or three fermenters. In addition, due to chemical processes that occur in fermentation cause changes in the attractanity, palatability, and gel strength of aquaculture diet. The scent and taste have a strong influence on attractiveness, palatability, feed efficiency, and growth of cultivation organisms. At the same time, attractiveness and palatability are influenced by fish species and size, temperature, ingredient, diet type, and level of cultivation domestication [5]. According to Halin and Evancho (1992), fermentation can increase the diversity of products with a distinctive taste, scent, and texture [6]. Besides, Pamungkas (2013) added, fermentation can also change complex organic matter into molecules that are simpler and easier to digest so that the attractiveness of diet can be increased. The distinctive scent of stinginf food tends to be preferred by cultivation organisms compared to a diet that does not have a distinctive scent. Furthermore, the level of delicacy or palatability of diet is the ability of cultivation organisms to consume diet to a certain degree of satiety [7]. Saade and Alamsyah (2010) reported that the higher the dose of seaweed, the K. Alvarezi meal, the more the attractiveness and the lower the palatability [8].

Nile tilapia is one of the fish that is in high demand of the international community because of its distinctive, delicious meat taste, high nutritional content, easily obtained and easily produced. This fish is classified as an omnivore, so it can consume food, in the form of animals and plants. Nile tilapia detect diet with the nose and eyes. The mechanism of the response of fish to an artificial diet starts from chemicals that are diffused from the diet into the water will stimulate fish chemosensory cells. Eating habits of fish are strongly influenced by the nature of the mixture of chemicals present in the diet so that the cells in the fish must be stimulated to stimulate response to feed. Eating behavior in fish shows that olfactory (sense of smell) and gustatory (sense of taste) are sensitive to ingredients similar to a fish diet. Olfactory is a long-distance sense, while gustatory is a short distance sense. Olfactory plays a role in giving signals to approach diet, while gustatory plays an important role in the decision to accept or reject diet [9,10]. However, information about various combinations of fermenters in seaweed fermentation as a thickener of the gel diet in tilapia is still rare.

2. Research methodology
2.1. Experimental diet
The experimental diet used was the gel diet. The Diet formulation and nutrients content of the experimental diet are shown in Table 1.
Table 1. Diet formulation and nutrient content of the experimental diet.

| Ingredients (%) | Fermenters combinations |
|-----------------|-------------------------|
|                 | A   | B   | C   | D   |
| Local fish meal| 45  | 45  | 45  | 45  |
| Shrimp head meal| 5   | 5   | 5   | 5   |
| Soybean meal    | 10  | 10  | 10  | 10  |
| Fine bran       | 5   | 5   | 5   | 5   |
| Seaweed meal without fermenter| 20 | -   | -   | -   |
| Seaweed meal + Bacillus sp. | -   | 20  | -   | -   |
| Seaweed meal + Bacillus sp. + Rhizopus sp. | -   | -   | 20  | -   |
| Seaweed meal + Bacillus sp. + Saccharomyces sp. | -   | -   | -   | 20  |
| Seaweed meal + Bacillus sp. + Aspergillus sp. | -   | -   | -   | -   |
| Cassava meal    | 5   | 5   | 5   | 5   |
| Fish oil        | 6   | 6   | 6   | 6   |
| Micro mix. 1    | 4   | 4   | 4   | 4   |
| Total           | 100 | 100 | 100 | 100 |

Nutrients content

| Nutrients content | A     | B     | C     | D     |
|-------------------|-------|-------|-------|-------|
| Crude protein (%) | 42.13 | 47.93 | 45.07 | 60.57 |
| Crude lipid (%)   | 5.01  | 8.42  | 5.54  | 4.75  |
| Nitrogen free extract (%) | 25.42 | 15.90 | 22.03 | 9.50  |

1) Vitamin A 900 IU, B1 2,400 mg, B2 4,350 mg, B6 900 mg, C 192,400 mg, D3 300,000 IU, E 2,250 mg, K3 360 mg, Capanthotenate 1,350 mg, inositol 33,750 mg, lysin 36,000 mg, methionine 16,500 mg, folic acid 450 mg, biotin 300 mg, nicotinamide 6,000 mg, choline chloride 4,500 mg, Co, Cu, I, Mn, Se, Zn, enzymes of protease, amylase dan cellulose + 1 kg lactose.

2) Gross energy: 1 g protein: 5.6 kcal/g, 1 g lipid: 9.4 kcal/g, dan 1 g carbohydrate: 4.1 kcal/g.

The stages of making a gel diet are first to mix the raw materials until dough are homogeneous, adding 30mL/100g of water to the raw material mixture and cooking until bubbles appear from the bottom of the pan during cooking stirring. This cooking is done with the strength of a small fire. After cooking, pour into the chamfer and stored at room temperature or left to cool. Then the cold diet is stored in the freezer for at least 1 hour and then cut into small pieces according to the size of the mouth opening of the cultivation organism and then stored again until use. An-aerobic fermented seaweed and incubated for 72 hours. The manufacture of an experimental diet was carried out at the Laboratory of Nutrition and Feed Technology, and the measurements of the attractancy, palatability, and gel strength of the experimental diet carried out at the Hatchery of the Faculty of Marine and Fisheries Sciences, Hasanuddin University, Makassar, Indonesia.

2.2. Experimental fish
The experimental fish used in this study were male GIFT tilapia weighing 7, 15, and 28g, obtained from Bantimurung Hatchery Center (BHC), Maros Regency, South Sulawesi Province, Indonesia. Experimental fish acclimatized in an aquarium measuring 60 x 30 x 30 cm was 10 fish/aquarium. During acclimatization, the experimental fishes were given an experimental diet of unfermented seaweed meal three times a day, i.e., morning, noon, and afternoon by satiation.
2.3. Measured parameters

2.3.1. Attractiveness. The attractiveness of the experimental diet is calculated based on the length of time it takes for the fish to eat the first time the experimental diet is expressed in units of cm/sec. Attractiveness testing has modified the method of [11]. First, the experimental fish was put into the "Aquarium Attractinity Test" (AAT) measuring 60 x 30 x 30 cm. ATT is equipped with a set of aeration. On the side of the aquarium covered with black plastic, and the top is covered with a net. This is so that the experimental fish will be protected from outside disturbances.

Stages of attractiveness measurement are experimental fish weighing 7 g and 28 g inserted and maintained in ATT, and given experimental diet by satiation for a week or until the fish are benign. On the following morning (day eight), (i) the volume of water is reduced to 1 cm above the body of the experimental fish. (ii) Install the separator (A) on one side of the AAT with a distance of 20 cm from the AAT wall, while simultaneously directing the experimental fish to gather on that side (D). (iii) Placing a sufficient experimental diet on the opposite side to the position of experimental fish (C). (iv) Lifting the separator and activating the stopwatch simultaneously, and then the experimental fish moves towards the experimental diet. (v) The stopwatch is deactivated when the experimental fish consumes the first time the experimental diet. (vi) Record the time taken for the experimental fish to eat the first experimental diet in units of cm/sec. (vii) The volume of water in the aquarium is increased up to 20 cm. Furthermore, (viii) feeding in the afternoon and evening. On the following morning, points ii to viii are repeated until the fifth day. For details, see Figure 1.

2.3.2. Palatability. Measuring the experimental diet patability is done by modifying the method [11]. Stages of palatability measurement of the experimental diet were experimental fish weighing 7 and 28 g inserted and maintained in an aquarium measuring 37 x 40 x 34 cm and given experimental diet for one week or experimental fish to be benign with the intention that the experimental fish are accustomed to the experimental diet and research environment. On the following morning (8th day), experimental diet (first time) slowly by satiation until the experimental fish dodge or reject the experimental diet, and rest for 15 minutes, (ii) then given the experimental diet (the second time) until the experimental diet rejects the given experimental diet. (ii) Weigh or add an experimental diet (points I + ii) in the morning in units of g/fish. On the following morning (9th day), points i-iii are repeated until the fifth day. The requirements for the attractiveness and palatability tests are that the experimental fish are in benign conditions, the water temperature is always the same during the measurement, and the experimental fish have not received feed in the morning [5].

2.3.3. Gel strength. The strength of the experimental diet was analyzed by the method of Balange and Benjakul (2003) [12] using Textur Analyzer XT Plus. The stages of gel strength analysis were to turn on the power "ON," install the probe in accordance with the parameters to be measured, the
sample is placed in the middle position, turn on the computer then open the texture analyzer program, open the application guide window, select food then the parameters to be measured. After the parameters and the tool settings have been determined, click load project, then open the Textur Analyzer window, open a new sheet by clicking on a new paper box, clicking "TA" then a TA window will appear, type in a name, graphic and file name then press OK, if the file has finished stored, then pressing the power "Off" as a sign of the analysis is complete.

2.4. Treatments and experiment design
This study uses a completely randomized design with 5 treatments and 5 replications. The treatments in this study are:

a) Seaweed flour without fermenters (Treatment A)
b) Seaweed flour with fermented Bacillus sp. (Treatment B)
c) Seaweed flour with fermented Bacillus sp. + Rhizopus sp. (Treatment C)
d) Seaweed flour with fermented Bacillus sp. + Saccharomyces sp. (Treatment D)
e) Seaweed flour with fermented Bacillus sp. + Aspergilus sp. (Treatment E)

2.5. Data analysis
Research data were analyzed by ANOVA. The treatments that affect the parameters were tested further by the W-Tuckey test.

3. Research results
3.1. Attractiveness
The average attractiveness value of a gel diet containing seaweed meal fermented by various combinations of fermenters as a thickening agent in male nile tilapia, O. niloticus small (weight 7 g), and large (weight 28 g) can be seen in Table 2.

| Combination fermenters            | Average attractiveness (cm/sec.) |
|-----------------------------------|----------------------------------|
|                                   | Small fish                       | Large fish                       |
| Seaweed meal without fermenter (A)| 2.76 ± 0.40a                     | 4.03 ± 1.65a                     |
| Seaweed meal + Bacillus sp. (B)   | 6.90 ± 2.26a                     | 9.30 ± 3.17a                     |
| Seaweed meal + Bacillus sp. + Rhizopus sp. (C) | 4.43 ± 2.57a | 6.03 ± 2.62a                     |
| Seaweed meal + Bacillus sp. + Saccharomyces sp. (D) | 3.00 ± 1.11a | 6.67 ± 2.08a                     |
| Seaweed meal + Bacillus sp. + Aspergilus sp. (E) | 6.70 ± 4.98a | 5.10 ± 2.00a                     |

The same superscript letters in the same column show the difference of not significant between treatments (p>0.05).
The average attractiveness of experimental diet containing seaweed, *K. alvarezi* meal was fermented by various combinations of fermenters in small fish (weight 7g) between 2.76 ± 0.40 to 6.90 ± 2.26 cm/sec., and in fish large (weight 28g) between 4.03 ± 1.65 with 9.30 ± 3.17 cm/sec. Based on the analysis of variance (ANOVA), it was shown that the experimental fish that consumed the experimental diet contained seaweed, *K. alvarezi* meal fermented by various fermenters as a thickener had no significant effect (p > 0.05) on attractiveness both in small and large fishes.

### 3.2. Palatability

The average palatability value of an experimental diet containing seaweed, *K. Alvarezii* meal is fermented by various combinations of fermenters as a thickening agent in nile tilapia, *O. niloticus* small size (weight 7 g) and large size (weight 15 g) at morning and daytime can be seen in Table 3.

The highest average palatability of experimental diet containing seaweed, *K. Alvarezii* meal fermented by various fermenter combinations in small experimental fish (weight 7g) in the morning was obtained between 1.38 ± 0.18 with 1.80 ± 0.05 g/fish while average palatability during the daytime was obtained between 1.26 ± 0.28 with 1.97 ± 0.06 g/fish.

Based on the results of the analysis of variance (ANOVA), it was shown that the experimental fish that consumed the experimental diet contained seaweed, *K. alvarezi* meal fermented by various fermenter combinations as thickener had a significant effect (p<0.05) on the palatability of the experimental diet on small fish both at mealtime morning and daytime. W-Tukey test results showed that the average palatability in the morning, treatment E was significantly different from treatments A and D, whereas treatments B and C were the same, while the results of the W-Tukey test on average palatability during the daytime showed that treatment E was significantly different from treatment A, whereas, between treatments B, C and D were the same.

The average palatabilities of experimental diets containing seaweed, *K. alvarezi* meal were fermented by various combinations of fermenters in large fish in the morning between 0.14 ± 0.03 and 0.26 ± 0.03 g/fish, and when daytime between 0.12 ± 0.01 and 0.23 ± 0.03 g/fish. Based on the results of the analysis of variance (ANOVA) showed that large size experimental fish (15g) which consumed an experimental diet containing seaweed, *K. alvarezi* meal fermented by various fermenter combinations as thickener had a significant effect (p<0.05) on the palatability of the experimental diet at the morning and daytime mealtime. W-Tukey test results showed that the average palatability in the morning, E and C treatments were significantly different from other treatments, and treatments A, B, and D were not significantly different from each other, whereas W-Tukey test results on average palatability in the daytime showed that treatment D was significantly different (p < 0.05) from treatment A, and treatments B, C, and E were the same.

Table 3. Average palatabilities in nile tilapia, *O. niloticus* small and large sizes that consume an experimental diet in the morning and daytime using seaweed, *K. Alvarezii* meal, which is fermented with various combinations of fermenters as a thickener.

| Combinations fermenters | Small fish | Large fish |
|-------------------------|------------|------------|
|                         | Morning    | Daytime    | Morning    | Daytime    |
| Seaweed meal without ferments (A) | 1.38 ± 0.18<sup>b</sup> | 1.26 ± 0.28<sup>b</sup> | 0.14 ± 0.03<sup>b</sup> | 0.12 ± 0.01<sup>b</sup> |
| Seaweed meal + Bacillus sp. (B) | 1.63 ± 0.22<sup>ab</sup> | 1.62 ± 0.22<sup>ab</sup> | 0.16 ± 0.01<sup>b</sup> | 0.18 ± 0.06<sup>ab</sup> |
| Seaweed meal + Bacillus sp. + Rhizopus sp. (C) | 1.45 ± 0.11<sup>ab</sup> | 1.68 ± 0.02<sup>ab</sup> | 0.26 ± 0.02<sup>a</sup> | 0.22 ± 0.07<sup>ab</sup> |
| Seaweed meal + Bacillus sp. + Saccharomyces sp. (D) | 1.39 ± 0.10<sup>b</sup> | 1.70 ± 0.22<sup>a</sup> | 0.16 ± 0.04<sup>b</sup> | 0.25 ± 0.02<sup>a</sup> |
The different superscript letters in the same column show the significant difference between treatments ($p<0.05$).

### 3.3. Gel strength

The average gel strength of the gel diet containing seaweed meal fermented by various combinations of fermenters as a thickening agent can be seen in Table 4.

**Table 4.** The average gel strength of the experimental diet containing seaweed, *K. Alvarezii* meal, which was fermented with various combinations of fermenters as a thickener.

| Combinations fermenters                      | Average gel strength (g/cm$^2$) |
|-----------------------------------------------|---------------------------------|
| Seaweed meal without fermenter (A)            | 100.97±14.24a                   |
| Seaweed meal + *Bacillus* sp. (B)              | 81.89±2.40a                     |
| Seaweed meal + *Bacillus* sp. + *Rhizopus* sp.(C) | 72.99±11.95a                   |
| Seaweed meal +*Bacillus* sp. +*Saccharomyces* sp.(D) | 83.30±0.92a                   |
| Seaweed meal + *Bacillus* sp. + *Aspergillus* sp. (E) | 76.70±5.71a                   |

The same superscript letters in the same column show the difference of not significant between treatments ($p>0.05$).

An experimental diet containing a seaweed meal fermented by various fermenter combinations had an average gel strength between 76.70 ± 5.71 with 100.97 ± 14.24 g/cm$^2$. Variance analysis results showed that seaweed fermentation by various fermenter combinations did not significantly affect the gel strength of the experimental diet ($p>0.05$). This means that both unfermented seaweed and those fermented by various fermenter combinations have the same average gel strength.

### 4. Discussion

#### 4.1. Attractancy

The attractiveness of a gel diet containing seaweed fermented by various combinations of fermenters as thickening agents is the same both without fermenter (seaweed meal without fermentation) and those fermented by various combinations of fermenters (*Bacillus* sp., *Aspergillus* sp., *Saccharomyces* sp. and *Rhizopus* sp.). This indicates that the combination of fermenters in seaweed fermentation did not significantly change the scent of the test diet, and the experimental fish was not affected by the scent of the experimental diet in detecting the presence of feed in its medium. This is also related to the level of perfection of the role of olfactory (sense of smell) and gustatory (sense of taste) of the experimental fish.

Fish feeding habits are strongly influenced by the nature of the mixture of chemicals found in the diet; the cells of the komosensori in fish must be stimulated to cause a response to feed. Feeding behavior in fish shows that olfactory (sense of smell) and gustatory (sense of taste) are sensitive to food ingredients similar to fish food. Olfactory is a long-distance sense, while gustatory is a short distance sense. Olfactory plays a role in giving signals to approach food, while gustatory plays an important role in the decision to accept or reject food [9,10]. However, there is a tendency that large-sized Nile tilapia (28g) is more quickly attracted to the experimental diet than small-sized fish (7g), although the attractiveness of diet E is slower to be responded to by large-sized test fish (28g) than small-size (7g). It is suspected that olfactory and gustatory large fish have a perfect role than small
size fish. The average attractiveness of dried artificial diet (pellets) containing seaweed as a binder was 8.04 ± 0.36 to 12.80 ± 0.30 cm/sec [5] and 7.70 cm/sec. In nile tilapia [13], who consumed a gel diet. This is influenced by the high lipid and protein content in the diet and makes the distinctive scent/odor or attractiveness value of the diet increased and more favored by nile tilapia. The more pungent the scent of diet, the higher the attractiveness of the feed [5]. It was further stated that the attractancy was influenced by the scent or odor of the diet.

4.2. Palatability

In general, the high palatability of a gel diet containing seaweed fermented by various combinations of fermenters consumed by small (7g) and large (15g) nile tilapia both in the morning and daytime is caused by the work of fermenters that can increase the palatability of the gel diet. This stimulates nile tilapia to increase their consumption. Forbes (1986) states that the more palatable feed given, the higher the consumption of feed or the palatability of the feed [14]. Furthermore, Saade (2014) reported that the higher the dose of seaweed meal in koi fish that consumed the gel diet, the higher the palatability [1].

Khasani (2013) suggests that fermentation can change low palatability into high palatability or from disliking to being liked [15]. This occurs in feed B, C, D, and E in small and large size nile tilapia both in the morning and daytime that have higher palatability than feed A. Fermentation also plays a role in reducing crude fiber, changing complex organic materials such as protein and carbohydrates, becoming molecules that are simpler and easier to digest. In accordance with the opinion of Saade (2011), it also states a similar statement, namely palatability related to taste, feeding habits (the type of diet, time and method of feeding), quality of diet (nutrient content, freshness, scent, and others) [5]. There is a tendency for palatability is higher during the daytime than in the morning, especially in small size nile tilapia. Presumably, this is because the temperature increases so that the metabolic process increases, and the rate of consumption also increases, temperature changes will affect the process of taking food, metabolic processes and enzymatic processes [16].

Fermentation technology is mostly carried out to increase the nutritional value of local feed ingredients or the origin of the waste. The palatability test results in this study indicate that treatment E is a seaweed meal with fermenter Bacillus sp. + Aspergillus sp. has a tendency to palatability higher than other fermenter combinations. This can be seen in Table 3, which is small size fish in the morning and daytime, and large size fish in the morning, but in the daytime, there is a tendency for fermented seaweed meal to be better than unfermented.

The results of this study are in accordance with nile tilapia, which consume a diet containing fermented moringa leaf meal using Aspergillus sp. as a fermenter, able to increase its palatability [17]. Pamungkas (2013) suggests that high palatability of diet can increase the growth of catfish larvae [7]. Furthermore, [18] also reported that the use of palm kernel cake fermented with Rhizopus sp. as much as 18% in the carp diet significantly increased the feed consumption, feed efficiency, and weight gain.

Basically, the attractiveness of aquaculture feed is produced from its protein or amino acid content [19]. The higher the protein value, the more the attractiveness of the diet increases. Increasing protein and amino acids in feed ingredients can be carried out through the fermentation process. Wandsansari state that fermentation using molds makes it possible to overhaul the components of material that are difficult to digest, become more digestible, increase protein content, and reduce crude fiber. Increased protein can be a good attractant and cause stimulation in fish by the senses of taste, odor, and texture of the diet [20]. The scent of feed gives a signal to the cultivation organism to recognize its diet as its food source. It was further stated that the use of Aspergillus sp. in the palm kernel cake fermentation can increase protein content and reduce crude fiber. Fermentation technology with Aspergillus sp. can increase the protein content in sago [21].

Decreased palatability causes a decrease in the amount of feed consumed, as seen in feed A (without fermentation). Feed without fermentation is not able to remind the scent of feed. This is thought to have caused differences in the level of feed consumption, growth, and feed efficiency.
Cultivation organism response, which tends to be slow to feed, is also thought to be influenced by the nutrient content of the diet. A slow response causes the diet to stay in water for longer, resulting in the dissolution of diet nutrients [22].

4.3. Gel strength
Nutrients that most contribute to the strength of the gel are raw materials for carbohydrate sources. The more/complex carbohydrate molecules, the higher the gel strength of an ingredient, conversely the simpler the carbohydrate molecule, the lower the gel strength, or in other words, the polysaccharide gel strength is higher than monosaccharides. In the process of seaweed fermentation, the polysaccharide content is reformed into monosaccharides so that the opportunity has a lower gel strength value. This is in accordance with the statement of Fardiaz (1992), carbohydrate is the main component that is broken down in the fermentation process, namely polysaccharides or starch broken down into glucose and other monosaccharides [4].

The combination of fermenters in the seaweed fermentation process, which acts as a thickening material in the experimental diet, does not significantly affect the average gel strength. The microbes used in this study are commercial fermenters. The use of microbes that are not goodwill affect its quality as a fermenter in the fermentation process. The fermenters used should be pure culture microbes in the laboratory [23]. It was further stated that other factors affecting fermentation were pH (acidity), time, oxygen, and temperature. Fermentative organisms are able to reduce carbohydrates from macromolecules to macromolecules because fermenters produce invertase and simase enzymes. The role of invertase enzymes is to break down polysaccharides (starch) that have not yet been hydrolyzed and converted into monosaccharides (glucose). While the enzyme zimase acts to convert monosaccharides into ethanol.

The strength of the gel obtained in this study was between 76.70 ± 5.71 with 100.97 ± 14.24 g/cm². The strength of the gel is lower than that obtained by Erjanan et al. (2017), who reported that the strength of the *K. alvarezii* seaweed gel was between 114.57-188.53 g/cm². This is due to the strength of the gel in this study is a mixture of some feed raw materials, not just the strength of seaweed gel [24]. High gel strength has high water stability so that both finned fish that are actively moving in search and passive crustaceans looking for food may have a long time to look for food, and are able to consume the feed they provide.

5. Conclusions and suggestion
5.1. Conclusion
Based on the results of this study, it was concluded that:
1. The combination of fermenters in seaweed, *K. alvarezii* meal fermentation as a thickener does not change not only gel strength but also the attractiveness of the gel diet by nile Tilapia, *O.niloticus* both in small and large fish.
2. The palatability value of gel diet containing seaweed fermented by various combinations of fermenters as thickening agent consumed by small and large tilapia both in the morning and daytime is better than without fermentation.
3. The average palatability value in small size is higher than the large size nile tilapia, which consumes a gel diet containing seaweed fermented by various fermenters as a thickening agent.

5.2. Suggestion
Based on the results of this study, it is recommended to improve the palatability of the gel diet should use fermented seaweed by various fermenter combinations.

References
[1] Saade E and Trijuno D D 2014 Growth response of koi fish fed on the diet containing Euchema cottoni *J. Akuakultur Indones.* 13 140–5
[2] Saade E, Bohari R, Hidayani A A and Haryati 2019 Utilization of gel diet in cultivating of
rabbit fish, Siganus guttatus in floating net cage in the dry and rainy season in the Makassar Strait.

[3] Merdekawati W and Susanto A B 2009 Kandungan dan komposisi pigmen rumput laut serta potensinya untuk kesehatan Squalen Bull. Mar. Fish. Postharvest Biotechnol. 4 41–7

[4] Fardiaz S 1992 Mikrobiologi Pengolahan Pangan Pusat Antar Universitas Pangan dan Gizi (Bogor)

[5] Saade E 2011 Kandungan nutrisi, atraktanitas dan palatabilitas pakan ikan nila GIFT, Oreochromis niloticus yang menggunakan berbagai sumber tepung rumput laut, Euchema cottoni sebagai binder Aquac. Indones. 12 33–41

[6] Halin J H and Evancho G M 1992 The Beneficial Role of Microorganisms in the Safety and Stability of Refrigerated Food (New York: Ellis Horwood)

[7] Pamungkas W 2013 Uji palatabilitas tepung bungkil kelapa sawit yang dihidrolisis dengan enzim rumen dan efek terhadap respon pertumbuhan benih ikan patin siam (Pangasius hypophthalmus Sauvage) Ber. Biol. 12 359–66

[8] Saade E, Aslamyah S and Salam N I 2011 Kualitas pakan buatan udang windu yang menggunakan berbagai dosis tepung rumput laut (Gracilaria gigas) sebagai bahan perekat J. Akuakultur Indones. 10 59–66

[9] Houlihan D F, Boujard T and Jobling M 2006 Food Intake in Fish (London: Blackwell Science)

[10] Michael W C 2006 2006 Chemoreceptions in Evans, D.H. The physiology of fish (CRC Press Boca Raton)

[11] Suresh A V and Nates S 2011 Attractability and palatability of protein ingredients of aquatic and terrestrial animal origin, and their practical value for blue shrimp, Litopenaeus stylirostris fed diets formulated with high levels of poultry byproduct meal Aquaculture 319 132–40

[12] Balange A and Benjakul S 2009 Enhancement of gel strength of bigeye snapper (Priacanthus tayenus) surimi using oxidised phenolic compounds Food Chem. 113 61–70

[13] Pribadi R, Saade E and Tandipayuk H 2016 Pengaruh Metode Pengerasan Terhadap Kualitas Fisik dan Kimiawi Pakan Gel Ikan Koi Cyprinus carpio haematopterus Menggunakan Tepung Rumput Laut Kappaphycus alvarezii sebagai Pengental J. Rumput Laut Indones. 1 108–16

[14] Forbes J M 1986 The voluntary food intake of farm animals Department of Animal Physiology and Nutrition (London: University of Leeds. Butterworths)

[15] Khasani I 2013 Atraktan pada Pakan Ikan: Jenis, Fungsi, dan Respons Ikan Media Akuakultur 8 127–34

[16] Chapman D 1992 Water quality assessments - a guide to use of biota, sediments and water in environmental monitoring (UNESCO/WHO/UNEP)

[17] Ilkwanuddin M, Putra A N and Mustahal M 2018 Utilization of Rice Bran Fermentation with Aspergillus niger on Feed Raw Material of Tilapia (Oreochromis niloticus) J. Perikan. dan Kelaut. 8 79–87

[18] Amri K and Khairuman 2003 Membuat Pakan Ikan Konsumsi (Tangerang: Agromedia Pustaka)

[19] Giri S S 2017 Farm-made Aquafeed, opportunities challenges and policy intervention South Asian Association for Regional Cooperation – SAARC Agriculture Centre (SAC) (Dhaka)

[20] Wandansari B D 2013 Fermentasi rumput laut Eucheuma cottonii oleh Lactobacillus plantarum Chem Info J. 1 64–9

[21] Kompiang, I P, Purwadaria T, Hartati T and Supriyati 1997 Bioconversion of sago Metroxylon sp. Waste Current status of Agricultural Biotechnology in Indonesia AARD Indones. 523–6

[22] Saade E and Aslamyah S 2009 Uji fisik dan kimiawi pakan buatan untuk udang windu penaeus Monodon fab. yang menggunakan berbagai jenis rumput laut sebagai bahan perekat Torani (Jurnal Ilmu Kelaut. dan Perikanan) 19 107–15

[23] Judomidjoyo M, Darwis A A and Saad F G 1990 Teknologi fermentasi Pusat Antar Universitas Bioteknologi
[24] Erjanan S, Dotulong V and Montolalu R I 2017 Mutu Karaginan dan Kekuatan Gel dari Rumput Laut Merah Kappaphycus alvarezii *Media Teknol. Has. Perikan.* 5 36–9