2020.2497-2508.pdf

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Substitution of sweet potato flour and corn starch to the growth, survival rate, feed conversion ratio and body chemical composition of juvenile *Litopenaeus vannamei*

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Abstract. For decades, the primary source of carbohydrates for vannamei shrimp (*Litopenaeus vannamei*) relied on the use of the more expensive corn starch. The combination of corn starch and sweet potato flour is expected to reduce the feed cost to a more affordable cost for the fish farmers. This study aimed to identify the most optimum substitution level between corn starch and sweet potato in the vannamei shrimp feed formulation. This study employed a completely randomized design with five formulations and three replications. The observed treatments were treatment A: 100% sweet potato flour and 0% corn starch, treatment B: 75% sweet potato flour and 25% corn starch, treatment C: 50% sweet potato flour and 50% corn starch, treatment D: 25% sweet potato and 75% corn starch, treatment E: 0% sweet potato flour and 100% corn starch. The results indicated that the combination of 50% sweet potato flour with 50% corn starch was the optimum treatment and the best body chemical composition, feed conversion ratio (FCR) and specific growth rate (SGR) were obtained. This study provides a scientific recommendation to the feed industry of vannamei shrimp and therefore, and the aim is to make the cost for the feed less expensive and affordable to the farmers.

Key Words: Aquaculture, chemical composition, feed, growth, substitution.

Introduction. Indonesia is a maritime country with a complex hydrography. Whiteleg shrimp (*Litopenaeus vannamei*) is one of the prominent fishery commodities with high export value (Ayisi et al 2017; Dastidar et al 2013; Haryotejo 2015; Hendradjat & Mangampa 2016; Statistics Indonesia 2016). Whiteleg shrimp is one type of crustacean farmed through aquaculture with higher productivity rate compared to other types of crustaceans including the giant tiger prawn *(Penaeus monodon)* (Liao & Chien 2011). South Sulawesi is one of the provinces in Indonesia that has great potential to be a whiteleg shrimp supplier. Although the export value of the whiteleg shrimp previously declined, in 2018, the total of whiteleg shrimp export value from South Sulawesi reached 4.462 tons, worth USD 42.8 million. There was an indication of decreasing shrimp production in South Sulawesi over the years. Therefore, in 2008, a national program entitled "Gerakan Kebangkitan Udang" (Shrimp Economic Revival Movement) was launched by the provincial government of South Sulawesi.

There were several factors contributing to the decrease of whiteleg shrimp production. One of the primary factors responsible for the failure in whiteleg shrimp farming in Indonesia was the inappropriate aquaculture technology and the inappropriate feeding according to the Water Environmental Carrying Capacity (WECC) (Zainuddin et al 2013; Zainuddin et al 2014). So far, feed cost contributed to 50% (Jatobá 2014), 58% (Son et al 2011) and 60-70% (Cummins et al 2013) of the production cost. Therefore, the government focused on the rearing, feeding, and nutrition development of whiteleg shrimp where feed cost could be potentially reduced (Lemos et al 2009; Tran et al 2013; Varadharajan & Pushparajan 2013).

Few studies have ventured into alternative feed development. The aim of alternative feed production is to minimize the use of animal protein substituted with plant protein (Davis & Arnold 2000; Divakaran et al 2000; Hernandez et al 2004; Tacon et al 2002). However, the use of both protein sources should be managed carefully. The excessive supplementation of protein would cause the expensive cost for feed production and produce waste that may degrade the water quality (Hari et al 2006; Lemos et al 2009; Wang et al 2015). Protein need for whiteleg shrimp's optimum growth, according to more sources, ranges from 30-44% (Cuzon et al 2004; Jatobá 2014; Wang et al 2015), 30-35% (Kumaran et al 2017) and above 30% (Kureshy & Davis 2002).

Several types of shrimp feed in South Sulawesi contained approximately 28-41% protein (Zainuddin et al 2014). Therefore, the use of excessive protein content in feed should be limited and optimized for growth (Bulbul et al 2015; Oujifard et al 2012). Other energy sources such as carbohydrates (protein-sparing effect by carbohydrates) with lower cost could be an alternative (Peres & Oliva-Teles 2002; Xia et al 2015; Zainuddin et al 2019). Many studies have reported the capable use of carbohydrates as an energy source (Cuzon et al 2004; Lee 2003; Zainuddin et al 2014). Carbohydrates have the potential to be an alternative low-cost energy source for shrimp growth (Niu et al 2012; Simon 2009; Wang et al 2014). So far, carbohydrate sources used in feed are microalgae or binders such as cassava flour, bran, wheat flour, or sago (Niu et al 2012; Wang et al 2015). Some studies have reported the use of corn starch in feed production (Guo et al 2006). However, less concern and emphasis were made to seek for the potential sources of carbohydrates used as shrimp feedstuff.

A study performed by Zainuddin et al (2017) indicated that in Indonesia, particularly in South Sulawesi, another available potential carbohydrate source is sweet potato flour. According to Zainuddin et al (2017) several carbohydrate sources such as sweet potato, bran, cassava, corn have a great potential to supply the carbohydrate component in shrimp feed since they contained lactose, glucose, and starch. Carbohydrates also contain oligosaccharide extracts that may serve as an aquaculture probiotic (Lesmanawati et al 2013; Theresia et al 2019). Therefore, in this study, the use of corn starch and sweet potato flour was performed to produce various less expensive carbohydrate sources. The use of alternative feed from local feedstuff as a carbohydrate source is expected to optimize potentially the growth of juvenile whiteleg shrimp. This study aimed to identify the optimum substitution level between corn starch and sweet potato in the vannamei shrimp feed formulation.

Material and Method

Experimental diets formulation. The production of formulated feed was initiated by collecting the feedstuff from local farmers in Maros, South Sulawesi. The employed formulated feedstuff contained 25% isoprotein. The sources of feedstuff consisted of fish meal, soy flour, shrimp cephalothorax flour, corn starch, sweet potato flour, vitamin, and mineral mix (Table 1). Formulated pelleted feed was dried to reduce its moisture content to 10%.

The proximate analysis encompassed crude protein content (CP), crude fat content (CF), water content, crude fiber content (CF), nitrogen-free extract (NFE) and ash of the feed following the protocols suggested by the Association of Official Analytical Chemists (2005). The results of the feed proximate analysis are presented in Table 2 and the analysis was carried out in the Chemical Nutrition Laboratory, Faculty of Animal Science, Hasanuddin University.

Feed production was initiated by grinding all the collected feedstuff. All the collected ingredients were weighted according to the percentage and nutritional needs of *Litopenaeus vannamei*. All the ingredients were mixed and stirred to promote the homogeneity of the mixture. Fish oil, vitamin and mineral mix and warm water were mixed to produce a paste. The paste was converted to pellets by the feed pelleting machine.

Table 1

The composition and the percentage of experimental ingredients

Ingredients	Treatment (%)				
	A	*B*	*C*	*D*	*E*
Sweet potato flour	0	10	20	30	40
Corn Starch	40	30	20	10	0
Fish meal	20	20	20	20	20
Cephalothorax flour	10	10	10	10	10
Soy flour	20	20	20	20	20
Fish oil	4	4	4	4	4
Vitamin mix	3	3	3	3	3
Mineral mix	3	3	3	3	3

Table 2

Proximate composition of the feeds used in the treatments

Treatment	*Composition (%)*					
	Water	CP	CF	CFib	NFE	Ash
A	6.37	25.99	11.88	2.44	49.40	10.41
B	7.25	25.89	12.03	2.71	50.86	8.52
C	6.58	25.38	11.98	2.39	52.41	7.81
D	7.13	25.16	11.63	2.83	54.66	5.72
E	7.32	25.46	11.38	2.91	55.54	4.70

Feed trial and experimental design. The experiment was conducted on juvenile whiteleg shrimp with an initial individual weight of 0.77±0.01g. The shrimps were collected from the local shrimp nursery in Pangkep. The whiteleg shrimps were accommodated to the experimental formulated feed. Stocking density of the experimental animals used was 15 shrimps/tank. The tanks used in this study were glass tanks with a size of 60cmx50cmx50 cm and 15 liters volume each. The research took place from April to October 2017 in the mini hatchery of the Faculty of Marine and Fisheries Science, Hasanuddin University. The shrimp were acclimated in the hatchery for 1 week and reared for 8 weeks. We used 15 tanks that were filled with 120 liters of water each. We used sea water diluted with freshwater to obtain a salinity of 20 ppt. The percentage of feed provided to the whiteleg shrimp was 10% of the juvenile body weight per day (Zainuddin et al 2014). The feeding was performed with a frequency of 4 times a day at the following hours: 06:00, 10:00, 14:00, 18:00 (Lestari et al 2019; Zainuddin et al 2014). The experimental formulated feed consisted of treatment A, B, C, D, and E with the following specifications: treatment A consisted of 100% sweet potato flour and 0% corn starch, treatment B consisted of 75% sweet potato flour and 25% corn starch, treatment C consisted of 50% sweet potato flour and 50% corn starch, treatment D consisted of 25% sweet potato and 75% corn starch, treatment E consisted of 0% sweet potato flour and 100% corn starch. Each treatment had 3 replications and was applied randomly to shrimp rearing media.

Statistical analysis. The observed parameters in this study encompassed the specific growth rate (SGR), survival rate (SR), feed conversion ratio (FCR) and body chemical composition of shrimp. By the end of the study, the bodyweight of the shrimps in each tank was calculated to measure their specific growth rate (SGR) and survival rate (SR) based on the method performed by Braga et al (2016), Niu et al (2012) and Zhang et al (2012). The amount of consumed feed during the rearing was calculated to identify its feed conversion ratio (FCR) based on the method performed by Hulefeld et al (2018). The formula for each calculation of the parameters is presented as follows:

- Specific growth rate: $SGR = 100 \times (LnWt - LnW_0) / t$

SGR = specific growth rate (%)
Wt = average weight of shrimp at time t (g)
Wo = average weight of shrimp in the initial research (g)
t = total days of culture period of research (day)

- Survival rate: SR = Nt / No x 100

SR = survival rate (%)
Nt = total survived shrimp
No = total shrimp stocked

- Feed conversion ratio (FCR): FCR = TFB / TB

TFB = total amount of feed broadcasted (g)
TB = total biomass of shrimp (g)

The body chemical composition was analyzed by proximate analysis at the end of the experiment by employing the AOAC method (AOAC 2005; Braga et al 2016; Dararat et al 2012). The chemical composition of the body shows crude protein (CP), crude fat (CF), crude fiber (CFib), nitrogen-free extract (NFE), ash, and glycogen (Glc).

Research data was statistically analyzed using the analysis of variance (ANOVA) using the SPSS software version 24. All results are expressed as average ± standard deviation (SD). The influence of the different treatments was followed by the Tukey test at 95% confidence level. This test determines the difference of influence between treatments.

Results. Average weight gain of individual shrimp during the study is presented in figure 1. The pattern of all treatments had a linear tendency. It can be observed that the substitution of 50% sweet potato flour and 50% corn starch (treatment C) indicated the most significant treatment compared to all other treatments.

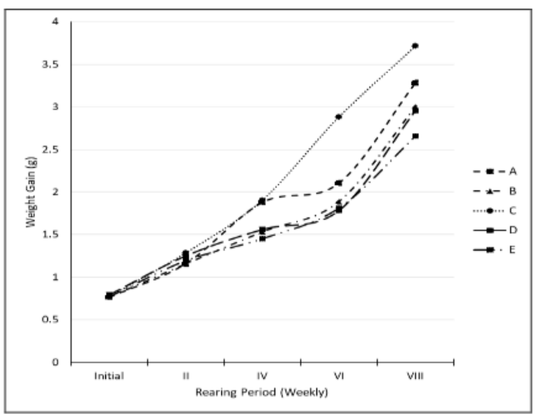

Figure 1. Body weight gain of juvenile whiteleg shrimp during the experiment.

The average rate of specific growth (SGR), survival rate (SR) and feed conversion ratio (FCR). The results of the analysis of variance showed that the treatment of substitution level of sweet potato flour and corn starch in the whiteleg shrimp feed could significantly affect ($p<0.05$) the specific growth rate and survival rate of juvenile whiteleg shrimp (Table 3). Means in the same column with different superscript letters ($^{a, b, c}$) are significantly different ($p<0.05$).

Table 3

The average rate of specific growth rate (SGR), survival rate (SR) and feed conversion ratio (FCR) of juvenile whiteleg shrimp during the experiment

Treatment	*SGR (%/day)*	*Survival rate (%)*	*FCR*
A	3.61 ± 0.25^{b}	$67.00\pm0,00^{b}$	$1.90\pm0,10^{b}$
B	3.57 ± 0.40^{b}	$73.00\pm11,55^{ab}$	$1.87\pm0,15^{b}$
C	5.91 ± 0.54^{a}	$84.00\pm3,84^{a}$	$1.40\pm0,10^{a}$
D	2.87 ± 0.26^{bc}	$67.00\pm0,00^{b}$	$1.93\pm0,06^{bc}$
E	2.51 ± 0.31^{c}	$67.00\pm0,00^{b}$	$1.93\pm0,12^{c}$

Based on table 3, the substitution of 50% sweet potato flour and 50% corn starch was significantly different ($p<0.05$) from other treatments, while the substitution of 100% sweet potato flour and 0% corn starch, 75% sweet potato flour and 25% corn starch, and 25% sweet potato and 75% corn starch were not significantly different ($p>0.05$) in relation to the specific growth rate of juvenile whiteleg shrimp. The substitution of 25% sweet potato flour and 75% corn starch and the substitution of 0% sweet potato flour and 100% corn starch were also not significantly different ($p>0.05$) in the specific growth rate of juvenile whiteleg shrimp. Furthermore, the substitution of 50% sweet potato flour and 50% corn starch significantly different ($p<0.05$) from the other treatments except with a substitution of 75% sweet potato flour and 25% corn starch where the treatment was not significantly different ($p>0.05$) in the survival rate of juvenile whiteleg shrimp. The substitution formulation of 100% sweet potato flour and 0% corn starch, 75% sweet potato flour and 25% corn starch, 0% sweet potato flour 0% and 100% corn starch were not significantly different ($p>0.05$) in the survival rate of juvenile whiteleg shrimp.

Table 3 showed that the substitution of sweet potato flour and corn starch significantly affected ($p<0.05$) the feed conversion ratio. Table 3 indicated that the substitution of 50% sweet potato flour and 50% corn starch was significantly different ($p<0.05$) from the other treatments. The substitution of sweet potato flour 100% and corn starch 0% and the substitution of 75% sweet potato flour and 25% corn starch were not significantly different ($p>0.05$) from the substitution of 25% sweet potato flour and 75% corn starch. The substitution of 25% sweet potato flour and 75% corn starch was not significantly different ($p>0.05$) from the substitution of 0% sweet potato flour and 100% corn starch.

Body chemical composition. The results of body chemical composition analysis by the end of the experiment are presented in Table 4. Means in the same column with different superscript letters ($^{a,\ b,\ c}$) are significantly different ($p<0.05$) and the following abbreviations were used: CP for crude protein (%), CF for crude fat (%), CFib for crude fiber (%), NFE for nitrogen-free extract (%), Glc for glycogen (g/100g sample).

Table 4

The body chemical composition of the experimental shrimp in final dry weight

Treatment	*CP*	*CF*	*CFib*	*NFE*	*Ash*	*Glc*
A	76.22 ± 0.76^{a}	0.42 ± 0.05^{ab}	1.36 ± 0.31	10.60 ± 1.53^{b}	11.41 ± 1.14^{a}	16.59 ± 0.39^{b}
B	76.23 ± 0.37^{a}	0.42 ± 0.04^{a}	1.31 ± 0.04	6.05 ± 0.12^{a}	15.99 ± 0.48^{b}	17.55 ± 0.08^{cd}
C	78.33 ± 0.14^{b}	0.40 ± 0.00^{ab}	1.06 ± 0.02	9.67 ± 0.82^{ab}	10.54 ± 0.97^{a}	18.21 ± 0.12^{d}
D	77.76 ± 0.67^{b}	0.49 ± 0.02^{b}	1.19 ± 0.14	9.40 ± 0.90^{ab}	11.16 ± 0.38^{a}	15.15 ± 0.17^{a}
E	71.82 ± 0.20^{c}	0.74 ± 0.04^{c}	1.12 ± 0.07	12.74 ± 2.96^{b}	13.58 ± 2.72^{ab}	17.38 ± 0.37^{c}

Based on table 4, the substitution of sweet potato flour and corn starch significantly ($p<0.05$) affected the body chemical composition of the experimental shrimp encompassing the crude protein, crude fat, nitrogen-free extract, ash, and glycogen content. In contrast, the substitution of sweet potato and corn starch did not significantly affect ($p>0.05$) the crude fiber. The protein content of whiteleg shrimp was higher with the substitution of 50% sweet potato flour and 50% corn starch compared to other treatments. The fat and ash content of experimental shrimp were significantly lower ($p<0.05$) in the substitution of 50% sweet potato and 50% corn starch compared to the other treatments. The body glycogen content resulted from the substitution of 50% sweet potato flour and 50% corn starch was significantly higher ($p<0.05$) compared to other treatments.

Discussion. The substitution of 50% sweet potato flour and 50% corn starch contributed to the most significant effect ($p<0.05$) because these two components contributed positively to the specific growth rate and survival rate of whiteleg shrimp. Both types of carbohydrate sources supplied a similar contribution to the specific growth rate and survival rate of whiteleg shrimp. According to studies performed by Krogdahl et al (2005), Niu et al (2012) and Simon (2009), different protein source could affect the shrimp and fish growth. The specific growth rate or survival rate of juvenile whiteleg shrimp would be reduced if the sweet potato flour percentage is increased to 75% and the corn starch is increased to 75% or 100%. The results can be described based on carbohydrate roles. The researchers assumed that the glucose and fructose content of sweet potato flour were higher compared to other carbohydrate sources (Rosas et al 2001; Theresia et al 2019). This is like starch content in corn starch compared to others. Consequently, the combination of 50%:50% of both produced better results. In relation to that, a few studies also mentioned that the alternative feed treatment with carbohydrate source could promote a significant effect on the juvenile whiteleg shrimp's growth (Amaya et al 2007; Davis et al 2002; Hasan 2001; Rosas et al 2001).

One determining factor in improving carbohydrate digestibility of the feed is the frequency of feeding (Zainuddin et al 2014). The results Zainuddin et al (2014) indicated that the feeding frequency of four times per day had a positive effect to the juvenile whiteleg shrimp growth and survival rate. The increase in growth was also affected by the feed factor and consumption behavior (Oujifard et al 2012). Growth can only occur when the consumed feed is higher than the needs of body maintenance. This is in line with the studies performed by Cuzon et al (2004) and Hasan (2001), indicating that the growth rate and survival rate are affected by the feed availability, age, water quality and water salinity (Diaz et al 2001; Hurtado et al 2006; Jannathulla et al 2019; Saoud et al 2003). The salinity level categorized as appropriate for whiteleg shrimp (Zainuddin et al 2019) ranged from 5-50 ppt (Hurtado et al 2006), 25-26.7 ppt (Diaz et al 2001) and 30-35 ppt (Saoud et al 2003). Braga et al (2016), Ghufron et al (2018), Soemardjati and Suriawan (2006) added that in addition to the feed quality, quantity, and the frequency, the supplied feed also affected the shrimp growth. The supplied feed should be appropriate and accurate to prevent feed deficiency (Hari et al 2006). Zainuddin (2014) stated that frequent feeding could upgrade the shrimp's capability of utilizing carbohydrate.

The highest feed efficiency was found in the substitution of 50% sweet potato and corn starch substitution. The lowest FCR was observed in the treatment of 50% sweet potato and corn starch substitution. FCR showed the conversion value of feed utilized by a reared animal to produce the body weight gain. The lowest FCR was found in the treatment where sweet potato flour and corn starch ratio was 50%:50%. The lowest generated FCR was 1.40, indicating that to produce 1 kg of shrimp, 1.40 kg of feed would be required. In comparison with other studies, the obtained FCR in this study was lower compared to the results (De Carvalho et al 2016; Qiu et al 2017; Olmos et al 2011;

Sookying & Davis 2011; Suárez et al 2009). This indicated that the supplied feed substitution contributed to the low conversion and was categorized as effective to improve reared shrimp biomass (Araneda et al 2008; Oujifard et al 2012).

The results of this study confirmed the argument that juvenile whiteleg shrimp could utilize higher amounts of carbohydrates. Although all the experimental feed from the treatments contained protein below 30%, the shrimp were able to utilize the feed (Tahe & Suwoyo 2011). A similar study was also performed by Hari et al (2006) with a protein content of 25% and 40%. The results indicated that the protein content of 25% with high carbohydrate content significantly promoted *Penaeus monodon*'s higher growth compared to the treatment of experimental feed with the protein content of 40%. In a study performed by Zainuddin et al (2014), it was identified that the use of feed with 30-50% carbohydrate content provided an equal contribution of glycogen and body chemical composition in juvenile whiteleg shrimp.

The use of carbohydrates in feed is particularly important due to some reasons: (a) carbohydrates are a cheaper energy source compared to the protein. Carbohydrate use could reduce feed cost that in turn, would decrease the total production cost (Simon 2009; Suárez et al 2009; Yang et al 2009). (b) At a certain level, carbohydrates could replace the energy from protein (*protein-sparing effect*) and due to its efficiency, the utilization of protein feed for growth can be improved (Wang et al 2015). (c) As a binder, carbohydrates (especially from certain feedstuff) could improve the physical quality of feed and reduce the percentage of feed dust (Son et al 2011). (d) As a nitrogen-free extract component, the supplementation of carbohydrates in feed could reduce the nitrogen waste to minimize the negative effect of feed on the environment (Dararat et al 2012; Olmos et al 2011).

Some results indicated similarities with a study performed by Hari et al (2006), stating that the carbohydrate source played an important role in reducing the ammonia content of nitrogen and in reducing the use of protein as an energy source (Braga et al 2016; Cuzon et al 2004; Samocha 2004). The quality of the carbohydrate relies on its sources. Sweet potato flour contains a high concentration of oligosaccharide (Viet et al 2018), glucose and fructose. Each component accounts for 4.49% and 4.23% respectively and has better content than other feedstuff sources such as cassava, sago, wheat, and corn (Zainuddin et al 2017).

The findings from the previous studies showed that the carbohydrate component in feed played an important role in boosting the metabolism, growth, and survival rate of shrimp or fish. Hari et al (2006) stated that the increase of growth was also affected by the feed factors, especially the inorganic nitrogen and carbohydrate content. Growth can only occur when the consumed feed is higher than the needs of body maintenance.

The results of this study indicated that the protein content of the experimental shrimp was identified in the treatment of 50% sweet potato and 50% corn starch substitution. This indicated the response of the protein-sparring effect by the carbohydrates (Rosas et al 2001). The results also showed that the treatment of 50% sweet potato flour and 50% corn starch substitution can lower the fat content of shrimps. The results indicated that the treatment of 50% sweet potato flour and corn starch substitution could deposit the glycogen significantly better compared to other treatments. The increase of carbohydrates and fat ration of feed was not linear with the increase of glycogen content in shrimps. The carbohydrates and fat ratio accounted for 4:5 in the substitution of 50% sweet potato flour and 50% corn starch were proven to deposit more glycogen compared to the carbohydrate and fat ratio accounted for 5:1 in the substitution of 0% sweet potato flour and 100% corn starch. The result of the study is different from the study performed by Li et al (2013) and Dong et al (2018), reporting that the glycogen content of juvenile blunt snout bream (*Megalobrama amblycephala*) increased as the ratio of the feeds carbohydrates and fat increased. The result of this study was like the result obtained by Ali & Jauncey (2004) stating that the glycogen content of African catfish was not significantly affected ($p>0.05$) by the increase of feed carbohydrate and lipid. The carbohydrate content in sweet potato flour and corn starch also affected the chemical glycogen content in shrimp's body. In the muscles, glycogen is a primary energy storage component that can amount to almost 2% of the shrimp total mass (Nur 2011).

Conclusion. Based on the results of the study, it can be concluded that the substitution of 50% sweet potato flour and 50 % corn starch was categorized as the most effective treatment regarding the specific growth rate, survival rate, feed conversion ratio and chemical body composition of juvenile whiteleg shrimp. Further studies on the combinations of carbohydrates and proteins in whiteleg shrimp rearing is recommendable.

Acknowledgements. The authors would like to express their gratitude to the Ministry of Research, Technology and Higher Education that funded this research through the Higher Education Advanced Applied Research Program in 2017-2019. The authors would also like to express their gratitude to the laboratory staff that helped to analyze the research results and other colleagues that contributed to the completion of this research.

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Received: 30 March 2020. Accepted: 23 April 2020. Published online: 11 September 2020.
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How to cite this article:
Zainuddin, Aslamyah S., Nur K., Hadijah, 2020 Substitution of sweet potato flour and corn starch to the growth, survival rate, feed conversion ratio and body chemical composition of juvenile *Litopenaeus vannamei*. AACL Bioflux 13(5):2497-2508.

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