Using of fermented soy pulp as an edible coating material on fish feed pellet in African catfish (*Clarias gariepinus*) production

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Abstract. The quality of fermented soy pulp (FSP) as a coating material on fish feed pellets is significant globally due to the increasing demand for aqua feed industry. Coating technique was employed in this study to assess the physical, biochemical and bacteriologic properties of FSP coated experimental diets additionally to evaluate the loading potency of probiotics on fish feed pellet and growth performance of African catfish *Clarias gariepinus*. FSP was designated as a model edible coating material on fish feed pellet (0%, 25%, 50%, 75% and 100% of FSP) to deliver the probiotic to the fish gut. *Lactobacillus* spp. were employed as a model probiotic and model bacteria for the demonstration of this study to African catfish production. The physical properties of FSP coated diets were evaluated by using different equipment and formulae. FSP coated diets were analyzed for amino acids and bacterial quantification by the High-Performance Liquid chromatography (HPLC) and bacterial plate culture techniques respectively. The growth performance of fish was determined by using various formulae. The physical properties of FSP coated on fish feed pellet diet characteristics in term of feed diameter, expansion rate, bulk density, pellet durability index, floatability and water stability were significantly different ($p < 0.05$) among the experimental diets. The feed colour and odour were more in yellowish black and more strong flavour respectively with increased the different level of FSP coated on fish feed pellet. The distribution of amino acids such as arginine, histidine, leucine, lysine, phenylalanine, glutamic acid, alanine and aspartic acid were the major amino acids within the all five experimental diets. The mean value of lactic acid bacteria (LAB) was significantly ($p < 0.05$) increased with the increased of FSP coating level in the experimental diets. However, the lowest mean value of the total bacteria (TB) and LAB was observed in the control diet (0% FSP). Moreover, the palatability of experimental feed had similar trends among the diets of 0%, 25% and 50% of FSP diet group fish consumed less than 100% of given feed within 5 minutes whereas the palatability of 100% of FSP diet group was observed fish consumed less than 50% of given feed in 5 minutes. The growth parameters of fish in term of final weight, weight gain (%), specific growth rate (SGR) and condition factor were significantly ($p < 0.05$) different among the experimental diets. The highest weight gain and SGR occurred in the 50% FSP diet group fish compared to the other experimental diets. These finding provided a novel insight into plant based FSP coating products which enhanced more efficiently in generating low-cost and healthy aqua feed globally for African catfish and other freshwater fish production.

Key Words: coating, fermentation, soy pulp, aquaculture, African catfish.

Introduction. Aquaculture is rising because of the quickest growing food-producing business within the world due to the increasing demand for fish and seafood. Worldwide, the aquaculture business has vast at a median rate of 8.9% per year since 1970 (FAO 2016; Huang & Nitin 2019). However, aquaculture industries often suffer serious financial losses that threaten their growth and health standing, in the main cause of the outbreaks of assorted diseases (FAO 2016). Different types of chemical and drugs are used to control the diseases in aquaculture industry. Those chemicals and medicines are nearly prohibited by the European Union and other countries because they are harmful for human health and also can lead to environmental contamination. Probiotics are often different rather than medicine and chemicals to boost up useful microbes within the gut of fish for higher growth and health status (Yamamoto et al 2010; Ding et al 2015;

Sharawy et al 2016; Batista et al 2016; Hasan et al 2018; Uczay et al 2019). However, there is a scarcity of techniques for delivering the probiotics to the gut of fish in aquatic environment because the probiotics microbes are killed due to high temperature and pressure during process of fish feed pellet. The methods like as spray and dip techniques have limitation for correct purposeful capability for carrying probiotics on feed pellet to the gut of fish. These issues are connected and could be resolved by the coating techniques of target microbes with edible plant based ingredients like fermented soy pulp (FSP) on the fish feed pellet. Garcia et al (2017) reported that coating mainly improves end-product qualities like flavor, texture, supplement nutrition worth and appearance. Coating formulations, ingredients, and method technologies have developed over time and several other completely different terminologies for coating have get practice to the human and animal feed business (Vonasek et al 2018; Huang & Nitin 2019). However, there is a scarcity of standard concepts, methodology and definitions of coatings on fish feed pellet among the fish feed development business. In this study, the plant based FSP coated on fish feed pellet is incredibility simple and straightforward method for improvement of the nutritional quality, carrying probiotics and value effective in terms of African catfish (*Clarias gariepinus*) growth and health performances.

FSP is currently capturing attention throughout the world because of the value of fish meal high price and inconsistency supply in recent years (Shiu et al 2015). FSP is used as a replacement of fishmeal for protein and energy source for enhanced fish growth and health status. This FSP add further worth to substrates like improvement of organic compound, probiotics inside the gut of fish and improve health performance and additionally to extend economic profits to aquaculture business (Shimeno et al 1993; Sun et al 2007).

Coating technique was employed during this research to assess the physical, biochemical and bacteriologic properties of FSP coated experimental diets additionally to evaluate the loading potency of probiotics on fish feed pellet. The physical quality of FSP coating on fish feed pellet diets is significant for a number of reasons. First of all, transportation and handling in each of the factory and on the farm need fish feed pellets of an explicit integrity while not fines created by attrition stresses (Shi et al 2012; Azarm & Lee 2014; Jiang et al 2018). Different animal species need different physical properties for their respective feeds. For fish feeds, additional pellet characteristics like floating ability, water absorption, palatability, and water stability are necessary compared to other animals feeds (Volpe et al 2012; Shamsuddin et al 2017). Moreover, the hygienic quality of feeds is very important and it is involvement of microbiological management contamination of feeds supported levels of assorted harmful microorganisms. Such quality parameters could also be accustomed value the results of diet formulation, acquisition, expander treatment, pellet binders, die choice (Thomas & Van der Poel 1996).

Pellets of high physical quality should have properties that give a high nutrition quality for example in terms of higher feed intake and improved biological process worth (Liu et al 2013; De Oliveira et al 2015). For improvement of fish feed product quality in terms of physical, biochemical and biological characterization, data of fundamentals for aggregating particles of a special size, hardness and form is additionally required. From a cost-effective and environmental purpose of read it is vital to make sure that FSP coated on fish feed is well utilized, providing high growth rate, healthiness and at last ensures a prime quality fish feed product. Feed quality is degree inexact term accustomed describe several aspects of feed quality like biological process, physical, hygienic and sensory quality (Thomas & Van der Poel 1996; Hossen et al 2011; Khater et al 2014; Uczay et al 2019). Fish that receive pelleted feeds usually have higher performances in terms of weight gain and lower feed conversion compared with mash feeds as according to various authors (Sun et al 2007; Shiu et al 2015; Kabir et al 2015; Sharawy et al 2016; Hasan et al 2018; Huang & Nitin 2019).

In this research, FSP was designated as a model edible coating material on fish feed pellet to deliver the probiotic to the fish gut. *Lactobacillus* spp. were employed as a model probiotic and model bacteria for the demonstration of this research to African catfish production. Therefore, it is vital to know how the FSP coated on fish feed particles bind and to comprehend insight into binding properties and coating method on fish pellets and their behaviour throughout transportation and storage and performance on fish growth and health status. Finally, this might reveal that data from this study could also be used as a basis for the factors accustomed value plant based FSP coating on fish pellet quality in aqua feed business. This is why the aim of this study was to figure out the physical, biochemical and biological properties of FSP coated on fish feed pellet diets using different level of FSP.

Material and Method

Research location. This study was conducted in the Universiti Malaysia Kelantan Aquaculture Research Pond Complex from the month of July 2018 to March 2019.

Preparation of fermented soy pulp and experimental diets. The raw soy pulp was collected from Prima Mekar Enterprise, Kelantan, Malaysia. The FSP was made through fermentation method by a combination of 0.001% *Lactobacillus* spp., with 10% of molasses in line with the standard methodology Ding et al 2015; Jiang et al 2018) by few modifications. In brief, all ingredients of fermentation were mixed by industrial mixer machine and then kept into a HDPE container for 3 weeks. The FSP was dried within the sun light until less than 10% moisture. The fine powder of FSP was used in coating on the fish feed pellet. Five isonitrogenous (crude protein 32%) diets were formulated according to a standard procedure. The control diet has contained fish feed pellet (0% FSP) while not FSP coating inclusion. The experimental diets were prepared with different levels of FSP 25%, 50%, 75% and 100% of coated on the fish feed pellet (96000 Dindings Super Keli, Malaysia).

Proximate composition and amino acid analysis of the experimental diets. Proximate composition analyses of the experimental diets were performed according to AOAC (1997). The composition of the diets ingredients and proximate composition are given in Table 1. All experimental diets were analysed for amino acids composition using HPLC (High Performance Liquid Chromatography) system (Breeze, Water Corporation, Milford, MA, USA) according to the manufacturer instructions. The amino acid composition of the experimental diets is given in Table 2.

Table 1

Ingredients used and proximate composition of the experimental diets

Ingredients (%)	*Diets (%)*				
	0	*25*	*50*	*75*	*100*
Feed pellet	100	75	50	25	100
Fermented soy pulp	0	25	50	75	100
CMC (binder)	3	3	3	3	3
Proximate composition					
Moisture	5.1	5.62	7.3	8.2	9.1
Protein	32.1	32.21	31.89	31.93	32.2
Lipid	5.4	4.3	4.41	4.64	4.87
Fibre	4.5	4.62	4.76	4.89	5.1
Ash	5.7	5.76	6.1	6.23	6.34
Carbohydrate	45.83	44.6	43.5	41.3	40.32

CMC = carboxymethyl cellulose.

Table 2

Amino acid composition of the experimental (% of total diets amino acids detected)

EAA	*Diets (%)*				
	0	*25*	*50*	*75*	*100*
Arginine	7.58	5.91	4.99	4.20	4.78
Histidine	2.30	6.69	6.45	5.93	4.65
Isoleucine	0.26	0.06	0.32	0.30	0.25
Leucine	1.83	1.80	2.08	2.70	2.22
Lysine	1.66	1.45	1.28	1.35	1.48
Phenylalanine	1.15	3.20	2.40	2.15	2.95
Threonine	0.90	0.29	0.96	0.68	1.06
Valine	0.51	2.09	2.34	2.51	2.54
Methionine	1.02	0.06	0.16	0.75	0.98
NEAA					
Alanine	1.66	0.87	1.62	1.80	2.22
Aspartic acid	2.61	2.62	2.40	2.25	1.97
Glutamic acid	4.74	4.71	3.09	3.80	3.79
Glycine	1.92	0.58	1.22	1.31	1.48
Proline	1.28	0.29	0.72	0.84	0.79
Serine	1.31	0.29	0.96	0.68	0.57
Tyrosine	0.13	0.29	0.16	0.45	0.25
Total	30.86	31.21	31.14	31.67	31.95

EAA = essential amino acid; NEAA = non-essential amino acid.

Measurement of physiological properties of experimental diets

Expansion ratio. About 50 pieces of fish feed pellets were taken for measuring of vertical and horizontal diameter using an electronic calliper (Digimatic Series No. 293, Mitutoyo Co., Tokyo, Japan). For each case, the determination was replicated three times and the mean value was considered. Expansion ratio was calculated by the following equation:

Expansion ratio (%) = (mean value of FSP coated pellet diameter – mean value of original commercial feed diameter) × mean value of original commercial feed diameter - 1) x 100.

The bulk density. Bulk density was determined by loose running feed into a measuring cylinder with a known volume like as some 1 kg of feed poured into a modified 1000 mL measuring cylinder. The content of the measuring cylinder was weighed on the Mettler Toledo scale and the experiment was repeated three times.

Pellet durability index. Pellet durability index (PDI) was measured employing a Tumbling box tester (Seedburo, Chicago IL, USA) for tumbling. In brief, three samples of approximately 100 g of pellets were weighed into the analytical instrument and for tumbling over a period of 10 min at a speed of 50 rpm. After testing, the feed samples were sieved to remove dust following standard sieve sizes (2 mm). PDI is defined as the weight of pellets remaining on the sieve divided by the total weight of pellets before tumbling and multiplied by 100.

Water stability. Water stability was measured as the ratio of the pellets retained on a wire mesh screen after immersion of 25 g of each replicate feeds in 1000 mL water for 20 min and then oven-drying at 105°C for 24 h to whole pellets at the start. The weight obtained was the left over from the initial weight after immersion due to disintegration for each test period i.e. weight of whole pellets. The water stability was calculated as the proportion of the weight of retained (whole) pellets against the initial total sample dry weight. Water stability (%) = 100* Weight of retained whole pellets/Initial total weight of pellets.

Floatability. Floatability of each replicate feeds was tested using 1000 mL beaker. The characteristics of 50 pellets as floater were dropped into the water of the beakers and observed for 20 minutes. At the end of each observation, the quantity of feed pellet as a floater on the water was recorded consequently. The mean numbers of the floating feed were expressed as percentage of the initial number.

Swelling test. Swelling variations between dry feed pellets and pellets immersed in water for various periods of time were examined in the laboratory. Ten pellets of each type were randomly chosen, the diameter of each fish feed pellets were measured (mm). Pellets were left on the surface of the water still they sank, then they were left submerged for 2, 6, 10 minutes. At the end of the immersion period they were gently retrieved and water in excess was drained by placing pellets on an absorbent paper. Finally, the particles of feeds were re-measured to get dimension after immersion over the period of time. The calculation of swelling was represented in percentage.

Organoleptic and palatability test. The organoleptic test specially the colour, odour of each feeds treatments was determined by the sensory organs through visual observation of the experimental feeds. The water colour change was also reported during the observation. For palatability test, all fish were acclimatized to the laboratory condition for seven days before experiment started. The experimental fish were fed with commercial diets twice daily at the satiation level and the palatability of the feed was recorded. In brief, experimental tanks were equipped with aeration. Comparisons of palatability were created by dropping at the same time of the experimental fish feeds on the water surface as distant from the fish tank. As shortly as the particles of feed hit the water, the fish swam towards the feeds. The reaction of the fish to each experimental feed was observed, associated an estimate product of that was most well-linked preference. Tests were repeated three times.

Bacterial load determination in diets*.* Total viable lactic acid bacteria (LAB) and total bacteria (TB) within the experimental diets were determined according to the modification of the bacterial plate culture techniques. In brief, the experimental diets were weighed and transferred to a sterile tube wherein it was homogenized in sterile saline solution (1 g feed/9 mL saline solution). The homogenate was serially diluted to 10^{-7} with the sterile saline solution. Ten µL of the diluted sample were spread onto de Man, Rogosa, and Sharp agar (MRS, HiMedia, India) or Tryptic Soy Agar (TSA, HiMedia, India) in triplicate plates for determination of total viable LAB (aerobic activity) and total counts of bacteria, respectively. Plates were incubated for 36 hours in an incubator at 37^{o}C. The plates were removed after the colony is visible and viable cell counts were expressed as colony forming units (CFU/g feed).

Fish growth parameters measurement*.* The African catfish juvenile was purchased from a local hatchery and raised within the UMK Aquaculture Research Pond Complex. The fish was acclimatized for some days to increase the size up to fingerling using available catfish commercial feed containing 32% protein and 5% lipid. At the start of the trial, the African catfish were fasted for 24 hours and therefore the total length and weight were measured when being anesthetized with MS-222 at 0.1 g L^{-1} water. The experimental fish was randomly allotted into 15 plastic tanks (capacity: 500 L per tank) covered by plastic black nets during the fish growth trial. The stocking density was 70 fish per tank. Each trial has consisted of three biological replicates. Fish was fed to apparent satiation by hand at two times on 9:30 and 18:30 hours daily for 12 weeks. Water quality conditions were maintained among the subsequent ranges throughout the experimental period: temperature, 29±2°C; pH, 7.5±0.5; dissolved oxygen, 5.25±0.5 mg L^{-1}.

At the end of the feeding trial, fish was fasted for 24 hours before harvested. Total numbers of fish sample in each tank was counted, and total body length and weight of fish were measured. After this, 9 fish from each tank was anesthetized to collect the liver for the biometric analyses. Then the collected samples were analyzed to estimation of the fish growth morphology, body indices and nutrient utilization using by the following formulae:

Survival rate (%) = 100* (number of surviving fish / total number of fish at the start of experiment)

Weight gain (%) = 100* (final weight-initial weight) / initial weight)

Relative growth (%) = 100* (final weight - initial weight / initial weight)

Specific growth rate (%) = 100* (Final weight - initial weight) / day of experiment

Hepatosomatic index (%) = 100* (weight of liver / total body weight)

Feed efficiency = live weight gain / total feed intake

***Statistical analysis**.* All data were analysed using IBM SPSS software (SPSS Inc., version 17.0, Chicago. IL, USA). One-way analysis of variance (ANOVA) was done to identify the mean value of each parameter; significantly differences among the experimental diets were evaluated using Duncan test. A p-value < 0.05 was considered significant. The results are presented as mean±standard deviation.

Results

***Experimental diet proximate composition and amino acid profile**.* Ingredients and proximate composition of the experimental diets are shown in Table 1. The proximate composition such as protein, lipid and carbohydrates was numerically almost similar among the experimental FSP coated diets in this study. The protein content of all diets was 32%. Furthermore, the amino acids profile of the experimental diets is shown in Table 2. The distribution of amino acids such as arginine, histidine, leucine, lysine, phenylalanine, glutamic acid, alanine and aspartic acid were the major amino acids within the all five FSP coated experimental diets. The value of histidine was highest in all FSP coated diets compared to control diets.

***Measurement of physiological properties of experimental diets**.* The parameters of the physical properties of FSP coated on the fish feed pellets of experimental diets of African catfish are summarized in Table 3. The physical properties of FSP coated on fish feed pellet diets characteristics in term of feed diameter, expansion rate, bulk density, pellet durability index, floatability and water stability were significantly different ($p < 0.05$) among the experimental diets in this study. The actual diameter, the expansion rate and bulk density of experimental diets have followed the similar trend. The mean value of these parameters was significantly ($p < 0.05$) increased with increased of different level of FSP coated on feed pellet. However, the PDI and floatability were significantly ($p < 0.05$) higher in control diets compared to other experimental diets but there were no significant ($p > 0.05$) differences of PDI between the diet 25% and 50% of FSP and also there was no significant ($p > 0.05$) difference of floatability among the diets of 0%, 25% and 50% of FSP. Although the lowest mean value of PDI (85.66%) and floatability (84.66%) was in 100% of FSP diet compared to the others experimental diets. Furthermore, the water stability was significantly ($p < 0.05$) higher in 100% of FSP diet compared to others experimental diets whereas there is no significant ($p > 0.05$) difference of water stability between 50% and 75% of FSP fish feed diets. However, the lowest mean value (74.66%) of water stability was observed in the diet of control at 0% of FSP. The swelling of the experimental diets showed associate considerable diameter changes once three different periods of immersion that showed in Figure 1. The swelling of control diet was significantly ($p < 0.05$) higher compared to other experimental diets. The examined particles reach a maximum of 168% of diameter increase in control diet after 10 minutes of immersion in water.

Table 3
Physical properties of experimental diets. Data expressed as mean±standard deviation (SD)

Parameters	*Diets (%)*				
	0	*25*	*50*	*75*	*100*
Feed diameter (mm)	3.16±0.05^{e}	4.03±0.05^{d}	5.06±0.05^{c}	6.10±0.10^{b}	7.16±0.05^{a}
ER (%)	3.16±0.05^{e}	26.93±5.19^{d}	53.00±3.40^{c}	77.93±5.19^{b}	104.00±3.40^{a}
Bulk density (kg m^{-3})	582.00±4.58^{e}	610.00±3.60^{d}	633.33±2.08^{c}	658.33±2.51^{b}	686.66±2.08^{a}
PDI (%)	97.50±0.50^{a}	92.50±0.86^{b}	91.50±0.50^{b}	89.33±1.25^{c}	85.66±1.04^{d}
Floatability (%)	99.33±1.15^{a}	98.00±2.00^{ab}	96.00±2.00^{ab}	94.00±2.00^{b}	84.66±3.05^{c}
Water stability (%)	74.66±2.30^{d}	80.00±4.00^{b}	85.33±2.30^{c}	85.33±2.30^{c}	90.66±2.30^{a}

ER = expansion rate; PDI = pellet durability index. Different superscripts in each row indicate significant difference ($p < 0.05$).

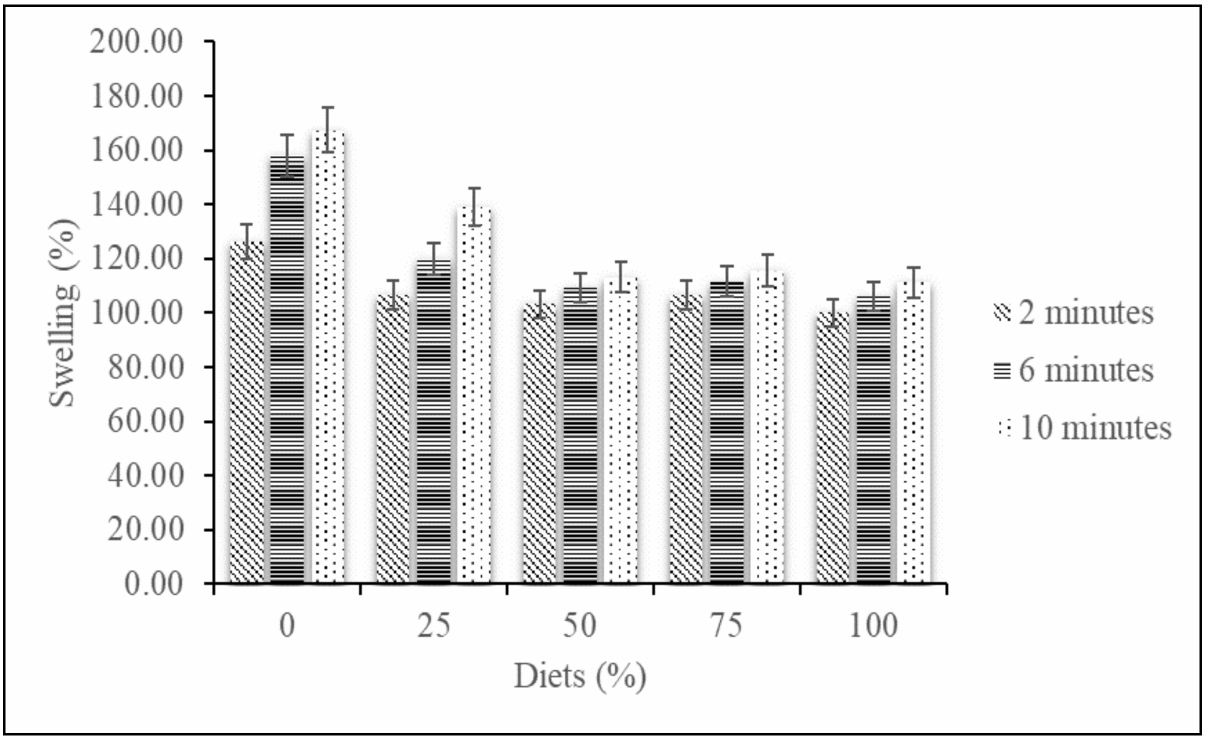

Figure 1. Swelling trends over the time period (2, 6 and 10 minutes) of experimental diets at 0, 25, 50, 75 and 100% of fermented soy pulp coated on fish feed pellet.

Organoleptic and palatability test. Table 4 presented the organoleptic and palatability observation treated with test diets. From the results it is depicted that there was considerable variation of feed colour and odour within feeding treatments in this study. The feed colour and odour were more in yellowish black and more strong flavour respectively with increased the different level of FSP coated on fish feed pellet. Moreover, the palatability of experiment all feed was similar trends among the diets of 0%, 25% and 50% of FSP group fish consumed less than 100% of given feed within 5 minutes whereas the palatability of 100% of FSP diet group was observed fish consumed less than 50% of given feed in 5 minutes.

Table 4

Organoleptic and palatability characteristics of experimental diets

Parameters	Diets (%)				
	0	25	50	75	100
Feed colour	Ash	Brown yellowish black	Turbid yellowish black	Deep yellowish black	Deeper yellowish black
Feed odour	Fishy odour	Characteristic flavour	Powerful flavour	Strong flavour	More strong flavour
Water colour	Turbid	Brownish turbid	Yellowish turbid	More yellowish turbid	Strong yellowish turbid
Palatability	++++	++++	++++	+++	++

+: fish consumed less than 25% of given feed in 5 min; ++: fish consumed less than 50% of given feed in 5 min; +++: fish consumed less than 75% of given feed in 5 min; ++++: fish consumed less than 100% of given feed in 5 min.

Bacterial load determination in experimental diets. TB and LAB of FSP coated on the fish feed pellets of the experimental diets of African catfish are represented in Table 5. The mean value of TB was significantly ($p < 0.05$) higher in 100% of FSP diet compared to other experimental diets whereas there was no significant ($p > 0.05$) difference of TB quantity between 0% and 25% of FSP fish feed diets. On the other hand, the mean value of LAB was significantly ($p < 0.05$) increased with the increased of FSP inclusion level in the experimental diets. However, the lowest mean value of TB and LAB was observed in the diet of 0% of FSP diet.

Table 5

Total bacteria (TB) and lactic acid bacteria (LAB) quantity in the experiment diets. Data expressed as mean±standard deviation (SD)

Parameters	Diets (%)				
	0	25	50	75	100
TB count (CFU g^{-1} of feed) x 10^9	1.80 ± 0.26^d	3.36 ± 0.30^d	5.86 ± 0.25^c	13.03 ± 1.10^b	21.16 ± 1.75^a
LAB count (CFU g^{-1} of feed) x 10^7	1.00 ± 0.10^e	5.53 ± 0.60^d	6.86 ± 0.35^c	8.43 ± 0.50^b	12.40 ± 0.95^a

Different superscripts in each row indicate significant difference ($p < 0.05$).

Fish growth performance. Table 6 presented the growth performance and feed utilization of African catfish fed with different levels of FSP coated on experimental diets. The growth parameters in term of final weight, weight gain (%), specific growth rate (%) and condition factor were significantly ($p < 0.05$) different among the experimental diets. The highest weight gain and SGR (1.73%) occurred in the 50% of FSP diet group fish compared to other experimental diets. Similarly, African catfish in the 50% of FSP diet group had the lowest significantly ($p < 0.05$) food conservation rate compared to other experimental diets. On the other hand, the overall survival rate was more that 90% of the African catfish and isplayed no significant ($p > 0.05$) differences in the mean values among the different diets tested at the end of the experiment.

Table 6

Growth performance of African catfish fed with experimental diets for 12 weeks. Data expressed as mean±standard deviation (SD)

Parameters	Diets (%)				
	0	25	50	75	100
Initial weight (g)	23.83 ±2.21	23.70 ±2.06	24.10 ±1.05	24.00 ±2.08	23.70 ±1.73
Final weight (g)	242.33 $\pm 2.08^{d}$	306.33 $\pm 5.50^{b}$	397.66 $\pm 3.51^{a}$	277.66 $\pm 3.78^{c}$	177.66 $\pm 1.52^{e}$
Weight gain (%)	992.98 $\pm 101.09^{c}$	1200.27 $\pm 133.01^{b}$	1552.41 $\pm 81.67^{a}$	1064.03 $\pm 122.59^{bc}$	652.58 $\pm 60.53^{d}$
Specific growth rate (%)	1.44 $\pm 0.06^{c}$	1.59 $\pm 0.06^{b}$	1.73 $\pm 0.03^{a}$	1.52 $\pm 0.06b^{c}$	1.25 $\pm 0.05^{d}$
Condition factor	0.76 $\pm 0.09^{ab}$	0.84 $\pm 0.07^{a}$	0.86 $\pm 0.06^{a}$	0.82 $\pm 0.05^{a}$	0.69 $\pm 0.04^{b}$
Survival rate (%)	90.95 ±2.97	95.23 ±3.59	96.57 ±1.43	92.38 ±2.97	91.90 ±2.18
Feed conversion rate	1.62 $\pm 0.06^{b}$	1.22 $\pm 0.03^{d}$	1.01 $\pm 0.01^{e}$	1.39 $\pm 0.03^{c}$	2.19 $\pm 0.05^{a}$

Different superscripts in each row indicate significant difference ($p < 0.05$).

Discussion. This is the first report on the FSP as a coating material on fish feed pellet for delivery probiotics to the fish gut along with used as a protein supplement of African catfish production. The quantity of probiotics on fish feed is an important issue in aquaculture and is one of the limiting factors for enhancing the health status and growth performance of fish. In this study, authors observed the physical, biochemical and bacteriological properties of FSP coated experimental diet to understand the feed utilization and growth performance of African catfish. The present results offer the first data of the important role of edible FSP level in African catfish production.

Experimental diet proximate composition and amino acid profile. The properties of ingredients used in the formulation of experimental diets were based on the proximate analysis of the dry ingredients with a ready to stay up the protein, lipid and carbohydrate contents in all type of experimental fish feed pellets at different levels of FSP with 0.001% probiotics like as *Lactobacillus* spp. In this study it is observed that FSP has high quality protein, especially essential amino acids quantity. Among plant protein sources, FSP is one of the foremost appropriate sources for coating on fish feed to African catfish production because of its protein levels as well as amino acid profile that match the fish requirements. It may be clear that FSP coated diets have abundant higher concentrations of free amino acids profile that increases the flavour and promote the changes of biochemical properties of the feed or diets (Dajanta et al 2011; Li et al 2013).

Physiological properties of experimental diets. The worth of physical characteristics of fish pellet like feed diameter, expansion rate and bulk density are essential for better utilization of place. These results indicated that the bulk density of the fish feed pellets increases with increasing the feed diameter and therefore the expansion rate. These findings are in agreement with the numerous researchers (Thomas & Van der Poel 1996; Saalah et al 2010; Khater et al 2014; Syamsu et al 2015). Furthermore, the feed floating significantly decreased as the composition of percentages of FSP coated on fish feed pellet increases. Fish feed pellet requires good quality physical structure and prolonged feed flotation time. In this study the data indicated that the mean floatability of the fish feed pellets decrease with the increasing of the fish pellet sizes. There are many factors that have an effect on the PDI, floating and water stability being influenced by the various kind of natural, modified or artificial substances used as binder for fish feed, the formula variation of feed, size of the pellet (Shamsuddin et al 2017) and also the composition of diets, feed manufacturing process (Ruscoe et al 2005; Solomon et al

2011). Obaldo et al (2002) reported that larger diameter pellets provided a higher degree of pellet stability in water.

The swelling of all diets exaggerated over the periods but the pattern of swelling decreased with the increasing of FSP inclusion level as a coating within the experimental diets. Feed swelling is a vital quality parameter within the producing of aqua feeds. Highest pellet swelling refers to the pellet physical integrity with minimal disintegration and nutrient leach (Fagbenro & Jauncey 1995; Obaldo et al 2002; Volpe et al 2012).

Organoleptic and palatability test. From the results it is depicted that there was considerable variation of feed colour and odour within feeding treatments in this study. Hossen et al (2011) described that ordinarily the colours of fish feeds of brown; deep brown colour indicates the microbial contamination. Odour may immediately indicate any distinction within the traditional smell. In this study the odour of feed felt somewhat mixed smell of dry FSP and molasses cake. The similar trends were also observed within the water colour change which increased from yellowish to more deep yellowish brown with the increasing of the level of FSP coated fish diets once diets were exposed to water. Shamsuddin et al (2017) observed that the water in all tanks turned turbid like pale yellowish green and the colour of water in the diets with palm kernel based feeds for shrimp tanks showed a turbid yellow colour. In order to improve palatability of African catfish feed, attractants or stimulants have drawn serious attention particularly once the plant protein source are included at high level. Tantikitti (2014) reported that palatability is influenced by the nutrient and the chemical content of the diets, the nutritional requirement of the fish, and the fish past feeding behaviour and other low molecular weight organic compounds such as organic acids, nucleotides and nucleosides (Hossen et al 2011; Li et al 2013; De Oliveira et al 2015). Fish use all senses like smell, taste and sight to discriminate among the feeds that given pleasant or unpleasant feelings related to ingestion. It is also shown that the turbidity of water increased with the decreased the fish palatability. The more turbidity could be due to nutrient leaching, especially the molasses within the coated FSP that is 100%. However, the size of the pellet in regard to the mouth gap size of the fish and perhaps the texture of the feed may be considerably important to maximise feed consumption and to minimize feed wastages (Tantikitti 2014).

Bacterial load determination in experimental diets. Lactic acid bacteria strains with FSP as an aquaculture feed additives have been shown to promote fish growth reported by numerous researchers (Li et al 2013; Liu et al 2013; De Oliveira et al 2015). Recently, the utilization of by-products of soy pulp from the soy industry has become widespread. In this study, the results indicated that TB and LAB might have ability to multiply throughout soy pulp fermentation process and FSP as a coating material may be the best carrier of probiotic like *Lactobacillus* spp. on the fish feed pellet deliver to the fish gut. Numerous researchers reported that besides plant protein nutritional value as a source of energy and amino acids, plant macromolecule is additionally used to modulate the physicochemical, bacteriological and sensory properties of diets (Rashad et al 2011; Liu et al 2013; De Oliveira et al 2015; Batista et al 2016; Hasan et al 2018; Huang & Nitin 2019). In this study FSP may be a good edible material that has reuse value and biotransformation prospects, and it has been described as a possible supply of supplement protein as a growth booster in useful diet for African catfish aquaculture development.

Fish growth performance. Numerous researchers (Shimeno et al 1993; Hossen et al 2011; De Oliveira et al 2015; Kabir et al 2015; Sharawy et al 2016; Uczay et al 2019) reported that lactic acid fermentation improved the nutritional value of raw soy pulp by partly eliminating feed allergens and anti-nutritional factor in soy that enhanced the fish growth performance, nutrient digestibility and physiological conditions such as bile status and intestinal microbiota in fish. The highest weight gain and SGR occurred in the 50% of FSP diet group fish compared to other experimental diets. This result indicates that using FSP as a coating material on fish feed pellet improves the growth performance of African catfish. Similar results have been reported by numerous researchers (Shimeno et al 1993; Sun et al 2007; Kabir et l 2015; Hasan et al 2018; Huang & Nitin 2019; Pfeuti et al 2019; Uczay et al 2019). On the other hand, the overall fish survival rate was more that 90% at the end of the experiment. This result indicates that fish have the ability to learn and to perceive tested diet condition in the culture environment. Moreover, in this study, 50% of FSP diet group fish shows a better feed utilization that indicates that this level of FSP coated on fish feed is suitable for African catfish growth and health status. The provision of diet up to 50% of FSP coated improved the growth of African catfish without impairing food efficiency. This plant based development of coating material as well as supplement of protein source for African catfish booster growth and production performance were not explored by other researchers. However, further experiments on protein digestibility, gut morphology and blood parameters, amino acid profile and the gene expression by FSP coated material are needed for African catfish growth and health status.

Conclusions. In summary, this is the first study that evaluated the physical, biochemical and bacteriological properties of FSP coated diets for African catfish production. Results showed that using 50% of FSP inclusion levels as an edible coating material along with as a protein supplement on fish feed pellet for delivering probiotics to the fish gut could have been used in the aqua feed industry for better growth and health status of African catfish as well as other freshwater fish production. These findings provided a novel insight into plant based FSP coating products which enhanced more efficiently in generating low-cost and healthy aqua feed. This study will help the researchers to uncover the critical areas of probiotic delivery techniques and viability of probiotics bacteria like *Lactobacillus* spp. in aqua fish feed pellet that many researchers were not able to explore. Thus, a new theory on using FSP as a plant based coating material as well as a protein supplement for fish booster growth and health status may be arrived at 50% of FSP coated diet.

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References

AOAC, Association of Official Analytical Chemists, 1997 Animal feeds. Chapter 4. In: Official Methods of AOAC International, Vol. 1 (16th edn), Cunniff P. A. (ed), pp. 1-3.

Azarm H. M., Lee S. M., 2014 Effects of partial substitution of dietary fish meal by fermented soybean meal on growth performance, amino acid and biochemical parameters of juvenile black sea bream, *Acanthopagrus schlegeli*. Aquaculture Research 45(6):994-1003.

Batista S., Ozorio R. O. A., Kollias S., Dhanasiri A. K., Lokesh J., Kiron V., Valente L. M. P., Fernandes J. M. O., 2016 Changes in intestinal microbiota, immune- and stress-related transcript levels in Senegalese sole (*Solea senegalensis*) fed plant ingredient diets intercropped with probiotics or immunostimulants. Aquaculture 458:149-157.

Dajanta K., Apichartsrangkoon A., Chukeatirote E., Frazier R. A., 2011 Free amino acid profiles of thua nao, a Thai fermented soybean. Food Chemistry 125(2):342-347.

De Oliveira C. F., Corrêa A. P., Coletto D., Daroit D. J., Cladera-Olivera F., Brandelli A., 2015 Soy protein hydrolysis with microbial protease to improve antioxidant and functional properties. Journal of Food Science Technology 52(2):2668–2678.

Ding Z., Zhang Y., Ye J., Du Z., Kong Y., 2015 An evaluation of replacing fish meal with fermented soybean meal in the diet of *Macrobrachium nipponense*: growth, nonspecific immunity, and resistance to *Aeromonas hydrophila.* Fish and Shellfish Immunology 44(1):295-301.

Fagbenro O. A., Jauncey K., 1995 Water stability, nutrient leaching and nutritional properties of moist fermented fish silage diets. Aquaculture Engineering 14(2):143-153.

FAO, 2016 The state of world fisheries and aquaculture. In: Contributing to food security and nutrition for all. Rome, 200 pp.

Garcia R. B. R., Silva F. S., Kawachi E. Y., 2017 Evaluation of dip and spray coating techniques in corrosion inhibition of AA2024 alloy using a silicon/zirconium sol-gel film as coating. AIP Conference Proceedings 1809(1):020017.

Hasan M. T., Jang W. J., Kim H., Lee B. J., Kim K. W., Hur S. W., Lim S. G., Bai S. C., Kong I. S., 2018 Synergistic effects of dietary *Bacillus* sp. SJ-10 plus β-glucooligosaccharides as a synbiotic on growth performance, innate immunity and streptococcosis resistance in olive flounder (*Paralichthys olivaceus*). Fish and Shellfish Immunology 82:544-553.

Hossen M. N., Das M., Sumi K. R., Hasan M. T., 2011 Effect of storage time on fish feed stored at room temperature and low temperature. Progressive Agriculture 22(1-2):115-122.

Huang K., Nitin N., 2019 Edible bacteriophage based antimicrobial coating on fish feed for enhanced treatment of bacterial infections in aquaculture industry. Aquaculture 502:18-25.

Jiang Y., Zhao P. F., Lin S. M., Tang R. J., Chen Y. J., Luo L., 2018 Partial substitution of soybean meal with fermented soybean residue in diets for juvenile largemouth bass, *Micropterus salmoides*. Aquaculture Nutrition 24(4):1213-1222.

Kabir M. A., Ghaedi A., Talpur A. D., Hashim R., 2015 Effect of dietary protein levels on reproductive development and distribution of amino acids in the body tissues of female *Pangasianodon hypophthalmus* (Sauvage, 1878) broodstock in captivity. Aquaculture Research 46:1736-1747.

Khater El-S. G., Bahnasawy A. H., Samir A., 2014 Physical and mechanical properties of fish feed pellets. Journal of Food Processing Technology 5(10):6 pp.

Li S., Zhu D., Li K., Yang Y., Lei Z., Zhang Z., 2013 Soybean curd residue: composition, utilization, and related limiting factors. Hindawi Publishing Corporation, ISRN Industrial Engineering 2013:423590.

Liu W., Ren P., He S., Xu L., Yang Y., Gu Z., Zhou Z., 2013 Comparison of adhesive gut bacteria composition, immunity, and disease resistance in juvenile hybrid tilapia fed two different *Lactobacillus* strains. Fish and Shellfish Immunology 35(1):54-62.

Obaldo L. G., Divakaran S., Tacon A. G., 2002 Method for determining the physical stability of shrimp feeds in water. Aquaculture Research 33:369-377.

Pfeuti G., Cant J. P., Shoveller A. K., Bureau D. P., 2019 A novel enzymatic pre-treatment improves amino acid utilization in feather meal fed to rainbow trout (*Oncorhynchus mykiss*). Aquaculture Research 50(5):1459-1474.

Rashad M. M., Mahmoud A. E., Abou H. M, Nooman M. U., 2011 Improvement of nutritional quality and antioxidant activities of yeast fermented soybean curd residue. African Journal of Biotechnology 10(30):5750-5759.

Ruscoe I. M., Jones C. M., Jones P. L., Caley P., 2005 The effects of various binders and moisture content on pellet stability of research diets for freshwater crayfish. Aquaculture Nutrition 11(2):87-93.

Saalah S., Shapawi R., Othman N. A., Bono A., 2010 Effect of formula variation in the properties of fish feed pellet. Journal of Applied Sciences 10(21):2537-2543.

Shamsuddin N. S., Daim N., Mamat N. Z., 2017 Water stability of palm kernel-based feeds for shrimp. Symposium on Innovation and Creativity (iMIT-SIC) 2:300-305.

Sharawy Z., Goda A. M. A. S., Hassaan M. S., 2016 Partial or total replacement of fish meal by solid state fermented soybean meal with *Saccharomyces cerevisiae* in diets for Indian prawn shrimp, *Fenneropenaeus indicus*, postlarvae. Animal Feed Science and Technology 212:90-99.

Shi M., Yang Y. N., Guan D., Zhang Y., Zhang Z., 2012 Bioactivity of the crude polysaccharides from fermented soybean curd residue by *Flammulina velutipes*. Carbohyrate Polymer 89(4):1268-1276.

Shimeno S., Mima T., Yamamoto O., Ando Y., 1993 Effects of fermented defatted soybean meal in diet on the growth, feed conversion, and body composition of juvenile yellowtail. Nippon Suisan Gakkaishi 59(11):1883-1888.
Shiu Y. L., Wong S. L., Guei W. C., Shin Y. C., Li C. H., 2015 Increase in the plant protein ratio in the diet of white shrimp, *Litopenaeus vannamei* (Boone), using *Bacillus subtilis* E20-fermented soybean meal as a replacement. Aquaculture Research 46(2):382-394.
Solomon S. G., Ataguba G. A., Abeje A., 2011 Water stability and floatation test of fish pellets using local starch sources and yeast (*Saccahromyces cerevisiae*). International Journal of Latest Trends in Agriculture and Food Sciences 1(1):1-5.
Sun M., Kim Y., Okorie O. E., Devnath S., Yoo G., Lee S., Jo Y. K., Bai S. C., 2007 Use of fermented fisheries by-products and soybean curd residues mixture as a fish meal replacer in diets of juvenile olive flounder, *Paralichthys olivaceus*. Journal of World Aquaculture Society 38(4):543-549.
Syamsu J. A., Yusuf M., Abdullah A., 2015 Evaluation of physical properties of feedstuffs in supporting the development of feed mill at farmers group scale. Journal of Advanced Agricultural Technologies 2(2):147-150.
Tantikitti C., 2014 Feed palatability and the alternative protein sources in shrimp feed. Songklanakarin Journal of Science and Technology 36(1):51-55.
Thomas M., Van der Poel A. F. B., 1996 Physical quality of pelleted animal feed 1. Criteria for pellet quality. Animal Feed Science and Technology 61(1-4):89-112.
Uczay J., Battisti E. K., Lazzari R., Pessatti M. L., Staudt Schneider T. L., Hermes L. B., Peixoto N. C., Fabregat T. E. H. P., 2019 Fish meal replaced by hydrolysed soybean meal in diets increases growth and improves the antioxidant defense system of silver catfish (*Rhamdia quelen*). Aquaculture Research 50(5):1438-1447.
Volpe M. G., Varricchio E., Coccia E., Santagata G., Di Stasio M., Malinconino M., Paolucci M., 2012 Manufacturing pellets with different binders: effect on water stability and feeding response in juvenile *Cherax albidus*. Aquaculture 324-325:104-110.
Vonasek E. L., Choi A. H., Sancez Jr. J., Nitin N., 2018 Incorporating phage therapy into WPI dip coatings for applications on fresh whole and cut fruit and vegetable surfaces. Journal of Food Science 83(7):1871-1879.
Yamamoto T., Iwashita Y., Matsunari H., Sugita T., Furuita H., Akimoto A., Okamatsu K., Suzuki N., 2010 Influence of fermentation conditions for soybean meal in a non-fish meal diet on the growth performance and physiological condition of rainbow trout *Oncorhynchus mykiss.* Aquaculture 309(1-4):173-180.

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