

## EFFECTS OF DIFFERENT INCLUSION LEVELS OF RICE BRAN IN THE DIETS ON THE PERFORMANCE OF AFRICAN CATFISH (*Clarias gariepinus*) JUVENILES

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### ABSTRACT

A five-week feeding trial was conducted to evaluate the possibility of rice bran meal (RB) substituting dietary soybean meal (SBM) in the diet of African catfish, *Clarias gariepinus* juvenile. Five experimental diets were formulated with RB replacing 0 (RB0), 20 (RB20), 40 (RB40), 60 (RB60) and 80% (RB80) of SBM protein. Another two experimental diets with intermediate (40%) and maximum (80%) inclusion levels of RB were added with feed enzyme (Allzyme SSF) to investigate its effects in improving the feed utilization (RB40SSF and RB80SSF, respectively). The control diet (RB0) was formulated using fish meal and SBM as sources of dietary protein. All diets were isoproteic (30% protein), isolipidic (12% lipid) and isoenergetic (3.83kcal/g). There were no significant differences ( $P>0.05$ ) in terms of weight gain (WG), specific growth rate (SGR), total feed intake (TFI), feed conversion ratio (FCR), survival rate (SR) and protein efficiency among all dietary treatments. Different inclusion levels of RB in diets did not affect the body indices of experimental fish. No definite trend was detected in whole body protein and lipid contents with increased dietary RB. It is concluded that RB can substitute SBM in the diet for *C. gariepinus* at up to 80% without significantly compromising growth and feed utilization. The addition of Allzyme SSF enzyme at 0.02g/100g diet has little contribution in improving growth and feed utilization performances.

**Key words:** African catfish; *Clarias gariepinus*; feed; rice bran meal; soybean meal.

### INTRODUCTION

African catfish, *Clarias gariepinus* is considered as one of the most important aquaculture species in Malaysia and many parts of the world because of its good taste, fast growth and tolerance to high density culture system (Marimuthu *et al.*, 2010). Unfortunately, the intensive farming of catfish is challenged by the increasing cost of the feed as it depends greatly on high protein commercial diets. *C. gariepinus* farming is only economically feasible when it is based on feed compound of locally available agricultural by-product (Degani *et al.*, 1998). Thus, more efforts are made to include the cheaper source of plant protein to partially replace the expensive fish meal. SBM is widely used as an alternative ingredient due to its high protein content (approximately 48%), good amino acid profile (El-Sayed, 1999) and lower cost with steady supply compared to other plant protein sources

(Storebakken *et al.*, 2000). Nevertheless, SBM production is limited to certain geographical areas only. Many other countries have to import this commodity. As such, this raises the cost of feed for the *C. gariepinus*.

Rice bran is a by-product of the rice milling industry which was produced during the conversion of paddy into white rice. Annually, about 63 to 76 million tons of rice bran is produced globally and more than 90% is traded cheaply as animal feed (Kahlon, 2009). It is a locally-available protein source in Malaysia as rice is a staple food of the country. In aquaculture industry, rice bran is commonly added in fish diets to supply energy and reduce feed cost, especially in the developing countries (FAO, 1979). Therefore, investigation on the use of rice bran in *C. gariepinus* diets deserved a better understanding in order to develop the cost-effective diets based on locally available feed ingredients.

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## MATERIALS AND METHODS

### Fish husbandry and feeding

The experiment was carried out at the Borneo Marine Research Institute, Universiti Malaysia Sabah for 5 weeks. Triplicate groups of African catfish, *Clarias gariepinus* juveniles with mean body weight  $9.85 \pm 0.87$  g and mean total length of  $11.23 \pm 0.32$  cm were obtained from the Fish Hatchery of Universiti Malaysia Sabah and were randomly distributed into 21 fiberglass tanks at a stocking density of 15 fish per tank (40 L). Water recirculating system using conditioned tap water was implemented in the present study. Filter media (corals and sponge) was cleaned fortnightly. Fish were fed twice a day at 8 a.m. and 3 p.m. daily close to apparent satiation.

### Feed Formulation and diets

Seven diets were formulated to be isoproteic (30% dietary protein), isolipidic (12% dietary lipid) and isoenergetic (3.83kcal/g). The control diet was formulated using a fixed amount of Danish fish meal at 21% and SBM at 9%, without RB. In the experimental diets, SBM was replaced with RB at 5 different inclusion levels (0, 20, 40, 60 and 80%). Two diets, each from intermediate and maximum inclusions of RB were added with solid state fermentation enzyme, Allzyme® SSF, Alltech Inc. (40% and 80% rice bran + enzyme Allzyme® SSF). Amount of enzyme added was standardized at 0.02 g per 100 g diet (Table 1).

Other ingredients were fish oil, tapioca starch,  $\alpha$ -cellulose, carboxymethylcellulose (CMC), dicalcium phosphate, vitamin and mineral premixes. Major ingredients and experimental diets were analysed for proximate composition using standard methods described by AOAC (1997). The dry ingredients were mixed prior to the addition of fish oil and water when preparing the experimental diets. The moist dough was extruded through a mincer

and pelleted through a 3 mm die. The pellet then underwent oven drying for six hours at 40°C. Dry pellet was then sieved to eliminate dust. The finish product was kept in the refrigerator.

### Data calculation and Analysis

Experimental fish were individually measured at the beginning and the end of the feeding trial. Every two weeks interval, they were bulk-weighed. Experimental fish were anesthetized using a commercial anaesthetic ( $\alpha$ -methylquinoline) before measurement. Mortality and feed intake were monitored and recorded daily.

At the beginning and the end of the feeding trial, 5 fish per tank were sampled for whole-body proximate analysis. WG, SGR, SR, TFI, FCR, hepatosomatic index (HSI), viscerasomatic index (VSI), protein efficiency ratio (PER) and net protein utilization (NPU) were calculated at the end of the feeding trial using the following formula:

$$\text{Body Weight gain (WG) (\%)} = [\text{Final body weight} - \text{Initial body weight} / \text{Initial body weight}] \times 100$$

$$\text{Specific Growth Rate (SGR) (\%/d)} = [\ln(\text{final wt.}) - \ln(\text{initial wt.}) / \text{days}] \times 100$$

$$\text{Survival Rate (\%)} (\text{SR}) = (\text{final fish number} - \text{initial fish number}) / \text{initial fish number}$$

$$\text{Feed Intake (FI) (g)} = \text{Total feed intake per fish for 5 weeks}$$

$$\text{Feed conversion ratio (FCR)} = \text{dry feed consumed (kg)} / \text{wet weight gain (kg)}$$

$$\text{Protein efficiency ratio (PER)} = \text{wet weight gain (g)} / \text{total protein intake (g)}$$

$$\text{Net protein utilization (NPU)} = 100 \times (\text{final} - \text{initial fish body protein}) / \text{total protein intake}$$

$$\text{Hepatosomatic index (HSI)} = [\text{liver weight (g)} / \text{fish weight (g)}] \times 100$$

$$\text{Viscerasomatic index (VSI)} = [\text{viscera weight (g)} / \text{fish weight (g)}] \times 100$$

**Table 1.** Ingredient composition (g/100g diet) of experimental diets

Ingredients	Diets						
	RB0	RB20	RB40	RB60	RB80	RB40SSF	RB80SSF
Fish meal	32.08	32.08	32.08	32.08	32.08	32.08	32.08
Soybean Meal	19.51	15.61	11.71	7.81	3.90	11.71	3.90
Rice Bran	0.00	15.21	30.41	45.62	60.82	30.41	60.82
Dietary lipid source	9.74	7.34	4.94	2.54	2.56	4.94	2.56
CMC	2.36	2.36	2.36	2.36	2.36	2.36	2.36
Vitamin Premix	3.00	3.00	3.00	3.00	3.00	3.00	3.00
Mineral Premix	2.50	2.50	2.50	2.50	2.50	2.50	2.50
Dicalcium phosphate	1.00	1.00	1.00	1.00	1.00	1.00	1.00
Alpha cellulose	8.94	7.48	6.02	4.55	0.53	6.00	0.51
Tapioca	28.50	21.46	14.43	7.39	0.35	14.43	0.35
Allzyme® SSF	–	–	–	–	–	0.02	0.02

WG, SGR, SR, FCR, PER, NPU, and whole-body proximate composition were analyzed using one-way ANOVA. Differences among treatments were compared using Tukey's HSD test. Values were considered significantly different at  $P < 0.05$ .

## RESULTS

### Proximate composition of ingredients and experimental diets

All values of the proximate compositions of the ingredients and the seven experimental diets are presented in Table 2. RB used in the experimental diets has relatively lower crude protein level (13.16%) compared to SBM (48.05%). In addition, it also have higher crude lipid content (17.73%) compared to the almost negligible crude lipid in the defatted SBM (0.65%). RB also contained higher moisture and ash as compared to SBM. The analysed crude protein and lipid contents of experimental diets correspond to the calculated values.

### Performance analysis

Performance analysis were indicated as a growth performance, TFI, FCR, Survival rate, PER and NPU. At the end of the feeding trial, there were no significant differences ( $P > 0.05$ ) in terms of WG and SGR of fish in all treatments. WG ranged from 250.87 to 329.54% at the end of the feeding trial. SGR was the highest in RB0 and RB40SSF (1.80%/d) and the lowest in RB80SSF (1.55%/d). Even though the growth performances were not statistically difference from each other, there was a trend of reduced growth with increasing levels of rice bran in the diet. Similarly, there was no significant difference detected in the feed intake ( $P$

$> 0.05$ ) of fish in all treatments. The results showed that the experimental fish favoured RB60 the most with the highest TFI value ( $30.34 \pm 3.06$ g) while RB80SSF was the least favoured diet ( $23.54 \pm 0.96$ g). Generally, it can be seen that as inclusion of rice bran increased, the FCR performance became poorer. FCR of diets added with enzyme Allzyme<sup>®</sup> SSF were poorer compared to diets without the enzyme. SR of fish with different diet treatments was not significantly different ( $P > 0.05$ ) from each other with values ranged from 93.33 to 100.00%. PER values of all experimental diets decreased with the increase of RB in the diet. PER of RB0 was the highest among all diets and was significantly different ( $P < 0.05$ ) from RB80 and RB60. Both diets containing enzyme had higher PER than those without. Fish fed with RB80SSF showed the highest net protein utilization (NPU) (70.50%) while fish fed with RB40SSF showed the lowest NPU (54.79%). The summary of these results are as shown in Table 3.

### Hepatosomatic index (HSI) and Viscerosomatic Index (VSI)

The HSI values of the experimental fish were not significantly different ( $P > 0.05$ ) in all dietary treatments and were in close proximity to each other ranging from  $1.91 \pm 0.17$  to  $1.14 \pm 0.08$ . In addition, the VSI values of the experimental fish were also not significantly influenced by the inclusion of rice bran in the diets with the highest value observed in RB20 ( $6.09 \pm 1.41$ ). The summary of the HSI and VSI values of the experimental fish are shown in Table 4.

### Carcass Composition of *C. gariepinus* Juvenile

The proximate carcass compositions of the experimental fish are presented in Table 5. Moisture content of experimental fish carcass was not

**Table 2.** Proximate composition of protein sources and experimental diets (% dry matter basis; mean $\pm$ SD)

	Fishmeal	Soybean meal	Rice bran				
Moisture	7.50	4.02	10.05				
Ash	12.95	7.28	7.89				
Lipid	7.21	0.65	17.73				
Protein	70.78	48.05	13.16				
Fiber	0.00	3.08	7.89				
	Diets						
	RB0	RB20	RB40	RB60	RB80	RB40SSF	RB80SSF
Moisture	10.69 $\pm$ 0.22 <sup>a</sup>	11.96 $\pm$ 0.36 <sup>ab</sup>	12.08 $\pm$ 0.57 <sup>ab</sup>	11.94 $\pm$ 0.48 <sup>ab</sup>	12.72 $\pm$ 0.35 <sup>b</sup>	12.42 $\pm$ 0.60 <sup>b</sup>	12.31 $\pm$ 0.37 <sup>b</sup>
Ash	9.62 $\pm$ 0.24 <sup>a</sup>	10.42 $\pm$ 0.04 <sup>b</sup>	11.61 $\pm$ 0.03 <sup>c</sup>	12.48 $\pm$ 0.06 <sup>d</sup>	13.41 $\pm$ 0.08 <sup>e</sup>	11.38 $\pm$ 0.15 <sup>c</sup>	13.29 $\pm$ 0.24 <sup>e</sup>
Lipid	9.78 $\pm$ 0.34 <sup>a</sup>	9.83 $\pm$ 0.60 <sup>a</sup>	10.16 $\pm$ 0.30 <sup>a</sup>	10.23 $\pm$ 0.64 <sup>a</sup>	12.05 $\pm$ 0.46 <sup>bc</sup>	10.88 $\pm$ 0.18 <sup>ab</sup>	12.45 $\pm$ 0.28 <sup>c</sup>
Protein	32.2 $\pm$ 0.02 <sup>a</sup>	32.28 $\pm$ 0.05 <sup>a</sup>	32.67 $\pm$ 0.09 <sup>b</sup>	33.24 $\pm$ 0.23 <sup>c</sup>	33.51 $\pm$ 0.03 <sup>c</sup>	32.67 $\pm$ 0.10 <sup>b</sup>	33.53 $\pm$ 0.07 <sup>c</sup>
Fiber	4.02 $\pm$ 0.05 <sup>a</sup>	3.92 $\pm$ 0.04 <sup>a</sup>	3.81 $\pm$ 0.03 <sup>a</sup>	3.81 $\pm$ 0.23 <sup>a</sup>	3.97 $\pm$ 0.06 <sup>a</sup>	3.95 $\pm$ 0.06 <sup>a</sup>	2.93 $\pm$ 0.11 <sup>b</sup>

Figures in the same row with same superscripts are not significantly different ( $P > 0.05$ )

**Table 3.** Growth performance, feed utilization and survival rate of the experimental fish (mean±SD)

Parameter	Diets						
	RB0	RB20	RB40	RB60	RB80	RB40SSF	RB80SSF
Weight gain (%)	329.54±49.51 <sup>a</sup>	309.00±21.07 <sup>a</sup>	319.03±6.51 <sup>a</sup>	279.34±11.46 <sup>a</sup>	277.64±9.91 <sup>a</sup>	327.00±26.77 <sup>a</sup>	250.87±32.63 <sup>a</sup>
Specific growth rate (%/d)	1.80±0.14 <sup>a</sup>	1.75±0.06 <sup>a</sup>	1.78±0.02 <sup>a</sup>	1.65±0.04 <sup>a</sup>	1.65±0.03 <sup>a</sup>	1.80±0.08 <sup>a</sup>	1.55±0.12 <sup>a</sup>
Total Feed Intake (g)	26.32±0.97 <sup>a</sup>	24.96±1.56 <sup>a</sup>	26.67±2.62 <sup>a</sup>	30.34±3.06 <sup>a</sup>	24.16±2.58 <sup>a</sup>	29.95±2.53 <sup>a</sup>	23.54±0.96 <sup>a</sup>
Feed Conversion Ratio	1.16±0.09 <sup>a</sup>	1.16±0.08 <sup>a</sup>	1.19±0.08 <sup>a</sup>	1.53±0.21 <sup>a</sup>	1.27±0.05 <sup>a</sup>	1.39±0.19 <sup>a</sup>	1.36±0.12 <sup>a</sup>
Survival Rate (%)	95.55±3.14 <sup>a</sup>	95.56±3.14 <sup>a</sup>	95.56±3.14 <sup>a</sup>	93.33±5.44 <sup>a</sup>	97.78±3.14 <sup>a</sup>	100.00±0.00 <sup>a</sup>	95.56±3.14 <sup>a</sup>
Protein Efficiency Ratio, PER	4.35±0.28 <sup>a</sup>	3.80±0.21 <sup>ab</sup>	3.03±0.55 <sup>ab</sup>	2.97±0.15 <sup>b</sup>	2.49±0.06 <sup>b</sup>	3.39±0.21 <sup>ab</sup>	3.34±0.26 <sup>ab</sup>
Net Protein Utilisation, NPU	62.64	61.81	63.91	63.98	68.27	54.79	70.5

Figures in the same row with same superscripts are not significantly different ( $P > 0.05$ )

**Table 4.** Hepatosomatic and viscerosomatic indices of the experimental fish (mean±SD)

	Diets						
	RB0	RB20	RB40	RB60	RB80	RB40SSF	RB80SSF
HSI	1.76±0.09	1.62±0.43	1.91±0.17	1.49±0.48	1.39±0.23	1.14±0.08	1.20±0.15
VSI	5.86±0.80	6.09±1.41	5.04±0.36	5.18±0.70	5.59±0.74	5.09±0.33	5.17±0.58

Initial HSI = 1.21±0.37, initial VSI = 4.98±0.21

**Table 5.** Whole-body proximate composition of the experimental fish (% of body wet weight; mean±SD)

	Diets						
	RB0	RB20	RB40	RB60	RB80	RB40SSF	RB80SSF
Moisture	71.90±1.03 <sup>a</sup>	71.75±0.40 <sup>a</sup>	75.15±5.20 <sup>a</sup>	73.34±1.33 <sup>a</sup>	72.32±0.27 <sup>a</sup>	72.91±1.02 <sup>a</sup>	71.96±1.18 <sup>a</sup>
Ash	3.37±0.03 <sup>ab</sup>	3.44±0.03 <sup>b</sup>	3.05±0.08 <sup>a</sup>	3.48±0.10 <sup>b</sup>	3.33±0.11 <sup>ab</sup>	3.50±0.07 <sup>b</sup>	3.53±0.18 <sup>b</sup>
Protein	15.31±0.30 <sup>c</sup>	14.92±0.48 <sup>bc</sup>	13.80±0.27 <sup>a</sup>	14.09±0.20 <sup>ab</sup>	14.27±0.38 <sup>abc</sup>	14.38±0.13 <sup>abc</sup>	14.76±0.33 <sup>abc</sup>
Lipid	7.58±0.03 <sup>bcd</sup>	7.63±0.13 <sup>bcd</sup>	6.85±0.09 <sup>ab</sup>	6.22±0.45 <sup>a</sup>	8.16±0.26 <sup>d</sup>	7.15±0.26 <sup>bc</sup>	7.66±0.06 <sup>cd</sup>

Whole body proximate for initial shrimp was 70.31±0.34% moisture, 12.08±0.62% protein, 2.35±0.38% lipid, and 3.57±1.13% ash. Figures in the same row with same superscripts are not significantly different ( $P > 0.05$ )

significantly different ( $P > 0.05$ ) in all treatments. Ash content of experimental fish carcass did not show any a clear trend as a result of rice bran inclusion in the diets. Meanwhile, the crude protein content of the experimental fish carcass composition

was the highest in RB0 (15.31±0.30%) and lowest in RB40 (13.80±0.27%). Carcass lipid was significantly affected by the dietary treatments with content ranged from 6.22±0.4% (RB60) to 8.16±0.26% (RB80).

**Table 6.** Estimated feed cost for production of African catfish

	Diets						
	RB0	RB20	RB40	RB60	RB80	RB40SSF	RB80SSF
Estimated feed cost per kg (RM)	2.54	2.33	2.09	1.89	1.60	2.09	1.60
FCR	1.16	1.16	1.19	1.53	1.27	1.39	1.36
Feed cost per kg fish (RM)	2.95	2.70	2.49	2.89	2.03	2.91	2.18
% Survival	95.55	95.56	95.56	93.33	97.78	100.00	95.56
Final feeding cost/ kg fish production (RM)	3.08	2.83	2.60	3.10	2.08	2.91	2.28

### Cost Evaluation

The estimated cost of the diets is presented in Table 6. Diet cost decreased 8.27–15.34% when soybean meal replacement with RB meal increased every 20% in the diets. The addition of the Allzyme<sup>®</sup> SSF enzyme increased the price of feed by 16–6.88%. Taking into account the economic conversion ratio (ECR) and survival rate, RB80 is the most economical feed.

### DISCUSSION

The present study shows that rice bran can replace soybean meal at high replacement levels of up to 80% in the diet of *C. gariepinus* juvenile without adverse effects on growth and feed efficiency. Based on the current findings, growth performance of fish fed different inclusion levels (20%, 40%, 60%, 80%) of RB and that of the control diet (0% rice bran) were not significantly different. SGR and percentage of weight gain of *C. gariepinus* in the present study were higher than the growth obtained in a study on *C. gariepinus* of similar size fed diets containing various proportions of fish meal and plant products by Oso *et al* (2013), indicating the experimental diets were well-utilized by the fish. Dedicated research on the utilization of RB in fish feed is not well-reported compared to other plant protein sources such as SBM, wheat meal, canola meal and maize meal. Nevertheless, a few studies on the digestibility of RB showed that the digestibility coefficients are comparable to other plant protein sources. For example, apparent digestibility coefficients for RB in tropical freshwater catfish were 81% for protein, 85.81% for dry matter and 67.93% for energy (Khan, 2008).

Good performance of *C. gariepinus* fed all experimental diets in the present study might be contributed by the good palatability of rice bran meal-based diets as indicated in the trend of total feed intake of the experimental fish. Studies on the nutritional value of RB showed that RB is superior in term of amino acid composition than other cereal grains (Warren & Farrel 1990; Warren & Farrel 1991). RB is also widely used as an ingredient

in feeds for poultry, cattle, horse, camel and pig. In the present study, all FCR values were below 1.60 indicating efficient utilization of the diets. Nevertheless, there was a trend of poorer FCR with the increasing amount of rice bran in the diets. This is in agreement with Martin *et al* (1998) which reported the depressed growth rate and FCR with increasing amount of rice bran. Reduced digestibility of the diets could also be one of the major limiting factors on the use of many other plant-protein based diets. Similar to growth trend, PER decreased as the RB inclusion increased in the diet. These trends were probably due to the reduced growth rate of the fish and protein intake during feeding trial. NPU however increased slightly with the increase of RB inclusion. Diet RB80SSF showed the highest NPU among all the diets. This means that more protein is absorbed by the fish in extreme inclusion of RB with the help of enzyme in the diet.

Survival rate in all treatments was relatively high, ranged from 93.33-100%. The HSI and VSI of fish fed different diets were not significantly different among groups. In general, the HSI and VSI of the experimental fish decreased with the increasing level of rice bran in the diet. Overall it can be seen that the inclusion of the Allzyme<sup>®</sup> SSF enzyme at 0.02g/100g had little effect in improving growth and feed utilization of diet containing 80% rice bran. It improved the growth performance of fish fed 40% rice bran but did not help in improving the FCR. This may also suggest that higher inclusion of enzyme is needed as inclusion of RB increases.

Except the moisture content, whole-body proximate composition of fish fed experimental diets was influenced by the inclusion of RB in the diets with no definite trend. The lipid content of *C. gariepinus* in the present study was much higher than those reported by Stephen *et al* (2007) which investigated the effects of palm oil-based diets on body composition of African catfish. This shows that body lipid is strongly influenced by the dietary lipid level in the diet.

The price of rice bran meal used in this experiment is cheaper by more than half of the price of soy bean meal used (61.11% of the price of soybean meal). The cost to produce 1 kg of diet

decreased as the amount of the cheaper and locally available rice bran increased in the diet. Increasing rice bran meal for every 20% in the diet reduced the cost of diet by 15.34%. High inclusion of rice bran meal in the diet reduced the production cost of the diet without compromising weight gain, specific growth rate, protein efficiency ratio, survival rate and net protein utilisation. Replacement of soybean meal with rice bran meal at up to 80% was indeed the most economical inclusion. Since there was no significant difference ( $P>0.05$ ) in the performance between diets added with Allzyme® SSF enzyme and those without the enzyme, addition of this enzyme into the feed is not necessary to further reduce the production cost.

The results of this study indicated that rice bran meal can substitute up to 80% of soybean meal protein without significantly affecting the growth, feed utilization, survival rate and body indices of *C. gariepinus*. The findings are considered important because rice bran is locally available in Malaysia and regarded as a by-product of the rice industry. Therefore, the inclusion of rice bran in the diet of *C. gariepinus* will be able to reduce the production cost as shown in our estimated feed cost. Further study on higher inclusion of Allzyme® SSF enzyme is warranted considering the lack of growth-promoting effect as a result of addition of enzyme in the diets.

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