Effects of using puffed feather meal on the growth performance and carcass characteristics of red hybrid tilapia

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Abstract
A study was conducted using juvenile red hybrid tilapia (*Oreochromis* spp.) for a 112-day feeding trial to evaluate the growth performance and carcass characteristics of the fish after been fed with practical diets containing 10 and 15% puffed feather meal. All the experimental feeds were formulated to be isocaloric and isonitrogenous using digestible values of the raw material. For growth parameter of weight gain, feed intake, feed conversion ratio (FCR), specific growth rate (SGR) and protein efficiency ratio (PER), no significant difference was found (p>0.05) indicating that inclusion of up to 15% had no adverse effect on the growth performance of the red hybrid tilapia. Highest average weight gain and SGR were recorded in the control with 197.31±25.43g and 2.19±0.15%, respectively. Lowest FCR was also recorded in the control with a value of 1.55±0.12. Carcass analysis results of fillet percentage and whole body composition did not show significant different (p>0.05) among the test groups. This study showed that inclusion of up to 15% puffed feather meal in the diet of red hybrid tilapia has no detrimental effect on the growth and carcass quality.

Keywords: carcass analysis, puffed feather meal, growth performance, red hybrid tilapia,

Introduction

Searching for new or alternative feed ingredients in any livestock production has been widely studied. In determining the suitability of a source as a feed ingredient, price, nutritional quality, and the availability of the feed source are among the characteristics which need to be taken into account. Efficient absorption and digestion of the feed ingredient by the fish is also important to ensure that the nutrients in the feed ingredient are fully utilized (Koprucu and Ozdemir, 2005). Fishmeal is globally known to be the main source of protein used in many aquaculture diets (Emre *et al.*, 2003), which in turn has several limitations nowadays. Despite being depleting in source and sustainability, the price of fish meal is in fact a bit pricey compared to other protein sources and was predicted to increase in the coming years (Suloma *et al.*, 2014). Due to the increasing demand for aquafeeds (Cashion *et al.*, 2017), exploring new alternatives is now becoming crucial and is one of the important keys in a successful aquaculture industry.

Researchers have made many attempts to partially or totally replace fish meal with less expensive and sustainable protein sources (El-Sayed, 1999; Lim *et al.*, 2005). Plant and animal based sources are the main feed ingredients being explored by many researchers. Besides agricultural waste products and by-products from agro manufacturing are also among the feed sources that can be utilized as feed ingredients in aquaculture feed (Farahiyah *et al.*, 2015). By- products from poultry processing are among the feed ingredients which fall into this category, and are under utilized in some countries (Hasan *et al.*, 1997; Campos *et al.,* 2017). These by products can be divided into two types: poultry offal meal and feather meal. Poultry offal meal generally is discarded internal parts of the poultry that has been processed and transformed into feed meal (Dale *et al.,* 1993), whereas feather meal is the product from the processing of the feathers into palatable meal (Isika *et al.,* 2006). Conventional feather meal is processed using the hydrolyzed technique where the feathers were rendered through high pressure at 207 to 690 kPa and steaming process to break the protein bonding in the keratin structure of the feather, resulting in a 70% digestible crude protein (El Boushy *et al.,* 1990; Isika *et al.,* 2006). This method has been widely practiced in feather meal production worldwide. The nutrient quality of feather meal is also subjected to cooking process of the feathers (Campos *et al.,* 2017). In Canada, recent manufacturing practices have shown improvement in terms of the digestibility of the feather meal (Bureau *et al.,* 2000).

The feather meal in this study was processed using the puffing method which does not involve the commonly used hydrolyzed technique. With this technique, protein digestibility can be enhanced and improved and thus is suitable to be used as feed for fish. The inclusion of feather meal in aquaculture diet which is readily available from many sources can effectively lower the price of aqua-feed and thus help in the utilization of the abundant agricultural by-products (Bishop *et al.,* 1995). Furthermore, feather meal also contains high protein and has a good essential amino acid profile, however, it is deficient in methionine, histidine and lysine (Suloma *et al.,* 2014; Campos *et al.,* 2017).

Materials and Methods

Preparation of feed

Most of the feed ingredients and vitamins used in the study were purchased from a local supplier. Palm kernel expeller was obtained from Sime Darby processing plant in Carey Island, Selangor. The puffed feather meal (PFM) was purchased from Biometrio Fusion Sdn Bhd, Johor. The processing of PFM involved the raw materials been ground and pulverized to very fine particles and mixed thoroughly according to the formulation. The pre mixed ingredients were then extruded though a twin screwed extruder (SIMA Brand) to form extruded pellets. The pellets were then dried at 60˚C.

Digestibility study

The experimental diet consisted of 70% of the reference diet (basal diet with fish meal as the sole protein source) and 30% of the test ingredient (puffed feather meal). Digestibility of the test ingredient was performed by total collection method. Ten juvenile red hybrid tilapia fish, *Oreochromis* sp., with mean weight of ±20 g were placed in a 100-L aquarium glass tank. The experiment consisted of two treatments (reference diet and experimental diet) with 3 replicates for each treatment.

The fish were fed once a day till satiation in the morning at 0900. All uneaten feed were collected an h after feeding and subsequently, after 4-5 h of feeding and fresh feaces were collected by siphoning. Feaces were also collected every morning before feeding time. All feacal samples were collected and pooled until sufficient for chemical analysis. The feacal samples were dried in the oven at temperature of 60˚C until dry, ground using a laboratory grinder and kept in a freezer until further analysis. Analyses of dry matter, protein, amino acids and energy content of the feed and feaces were determined and used in calculating digestibility value according to the equation of Belal (2005) and Lim *et al.,*(2005). All chemical analyes were performed following the AOAC methods (2012).

Feeding trial

A total of 540 all male red hybrid tilapia (monosexs), *Oreochromis* spp., juveniles (mean initial weight of 18±1g) were used in this study. The fish were purchased from a local hatchery in Rawang, Selangor. The fish were allocated in a completely randomized design to three treatments with six replications per treatment and were adapted to the diets for two wks prior to the feeding trial. Fish were stocked in an outdoor culture system with water capacity of 800 litres using 1000-litre polyethylene tank and a normal photoperiod. Water exchange (50-70%) was performed twice a week and the frequency was later adjusted according to the water quality. Water quality parameters were observed and recorded every alternate week to ensure that it was maintained and in the range for cultured freshwater fish (Mjoun *et al.,*2010).

Treatments used in this study consisted of three practical diets: T1- control; T2- inclusion of 10% PFM and T3 – inclusion of 15% PFM. The experimental feeds were formulated to be approximately isonitrogenous (32% CP) and isocaloric (13MJ/kg), calculated using digestible values of protein and energy of the feed ingredient (Table 1). The formulation was based on least cost feed formulation. All diets contained minimum nutrients needed for the growth of tilapia. The fish were fed twice a day at 4% of total body weight for the first 12 wks which was later reduced to 3% total body weight for the next 4 wks. The duration of this feeding trial was 16 wks (112 d). Fish were weighed every fortnight to collect the weight gain data and adjust the feed portion. At the beginning of the experiment, 15 fishes were culled to determine the initial whole body composition of the fish. By end of the experiment, 18 fishes were sampled from each treatment to determine the final whole body composition. Growth parameters: feed intake, body weight, weight gain, survival rate, feed conversion ratio (FCR), specific growth rate (SGR) and protein efficiency ratio (PER) were recorded and calculated by the end of the experiment. Results were analysed using general linear model and treatment means were compared by Tukey's test using SAS Version 9.3.

Table 1. Formulation of experimental diets (g $100g^{-1}$ dry diet), proximate composition of the diet based on analysis, and the calculated values of digestible energy and digestible protein

Ingredient	T1	T2	T3
Cassava	12.00	12.00	12.00
Soybean meal	20.50	28.80	19.30
Fish meal	27.80	7.00	7.00
Corn	10.00	10.00	13.00
Rice bran	6.00	6.00	7.00
Wheat pollard	6.00	6.00	6.50
Feather meal	0.00	10.00	15.00
Palm kernel expeller	10.00	12.50	12.50
Crude palm oil	4.00	4.00	4.00
Limestone	0.50	0.50	0.50
DCP	0.20	0.20	0.20

Ingredient	T1	T2	T3
Methionine	0.50	0.50	0.50
Lysine	0.50	0.50	0.50
Premix	2.00	2.00	2.00
Total	100.00	100.00	100.00
Nutrients level (as is basis)			
Dry matter, %	92.45	90.25	88.50
Digestible protein, %	32.00	32.00	32.00
Crude fiber, %	3.95	4.55	4.75
Ether extract, %	7.35	4.55	5.05
Ash, %	7.25	5.20	4.75
Digestible energy (MJ/kg)	13.00	13.00	13.00

*All ingredients used in the formulation was based from the digestible values of tilapia

Results and Discussion

Digestibility values of feather meal

The digestibility values of dry matter, crude protein, digestible energy and amino acid are shown in Table 2. The digestibility coefficent of crude protein was 0.99 with digestible energy of 0.95. The amino acid digestibility values were above 90% except for lysine which was 89%. This is in contrast to Guimares *et al.* (2008) where only 78.5% protein was found to be digestible in Nile tilapia. The nutritional values of the feather meal varied according to batches. It is well known that feathers contain mainly keratin protein which has a high percentage of amino acid cysteine (Hasan *et al.,* 1997). In the puffed feather meal, however, the value of cysteine was undetected, indicating that the process of puffing might have destroyed the amino acid cysteine. Hasan *et al.* 1997 mentioned that autoclaving the feed material might reduce cysteine percentage from 10 to 3.5% and thus made the feathers more soluble. The high digestibility values of PFM indicated that it could be a good protein source for tilapia.

Table 2. Nutrient digestibility of PFM in tilapia fingerlings (average weight ±20 g)

Nutrient	Digestible coefficient	Total PFM	Digestible PFM
Dry matter (%)	0.97	94.45	91.79
Protein (%)	0.99	93.28	92.51
Energy (MJ/kg)	0.95	23.02	21.87
Amino acids (%)			
Arg	0.97	5.83	5.63
Thr	0.95	3.98	3.80
Cys	na	na	na
Val	0.97	5.58	5.39
Met	0.94	0.56	0.53
Ile	0.97	3.77	3.66
Leu	0.96	6.30	6.06

Nutrient	Digestible coefficient	Total PFM	Digestible PFM
Tyr	0.90	0.97	0.88
Phe	0.96	4.27	4.08
His	0.92	0.58	0.54
Trp	na	na	na
Lys	0.89	0.97	0.87

na =not available

Growth performance of tilapia

Table 3 shows the growth performance of tilapia fed PFM over a 112-d period of culturing. All treatments did not show any significant difference in weight gain, feed intake, FCR, SGR and PER except for the survival rate where T2 (10% PFM inclusion) and T3 (15% PFM inclusion) showed ~~a~~ significant difference ($p<0.05$). The highest average weight gain was from the control, followed by 15% PFM inclusion while the 10% PFM inclusion had the lowest weight gain. Feed intake however was lowest in 15% PFM inclusion, showing that the higher the inclusion of the PFM in the diet, the lesser was the intake. This could be due to the lesser amount of fishmeal used in the feed, which reduced the palatability of the feed. Squid oil has been widely and normally used by feed manufacturers to increase the palatability of the feed in which it serves as an attractant. In this study however, no squid oil was used in the practical diets, which could be the reason of the low feed intake as more feather meal was included.

Fish in the control group showed the best FCR with 1.55, followed by the 15% PFM inclusion (1.63) and 10% PFM inclusion (1.70). Yong *et al.* (2018) reported that feather meal could be included up to 15% in tilapia diet, replacing 100% of fishmeal in the formulation and was better when compared to the control without feather meal inclusion. Higher inclusion of feather meal was suspected to be possible without compromising the growth performance (Yong *et al.*, 2018).

Most studies conducted often measured growth performance and nutrient utilization to determine the efficacy of the feedstuff as feed for fish. Other than growth parameters, changes in immune parameters were also said to be the criteria to be looked at (Campos *et al.*, 2017). Hybrid tilapia were reported by Zhang *et al.* *(*2014) to show less stress in the immune parameters when fed with 12% of feather meal.

Specific growth rate was also found to be highest in the control group. In term of the protein efficiency ratio, fish in the control group were more efficient in utilizing the protein, but not significantly different from other treatments. Treatment with 15% inclusion of PFM had better PER than the treatment with 10% PFM inclusion and the best survival rate with less than 1% mortality. In a study conducted using Nile tilapia fries, inclusion of up to 9.9% of feather meal in the total diet reduced the performance of the fish (Bishop *et al.*, 1995), whereas Tacon *et al.* (1983) found that replacement of up to 14.3% of feather meal in the diet of Nile tilapia fries did not have any significant difference in terms of growth , feed utilization and also body composition compared to the control. Campos *et al.* (2017) included 12.5% of hydrolyzed feather meal in the diet of European seabass, and indicated no negative impact on the immune system, growth and composition of fatty acids.

In this study, a balanced composition of nutrients based on calculation, using digestible values of nutrients of the feed ingredients in the formulation was used. Furthermore, feather meal used in this study was prepared using a puffing method which improved the digestibility of the product in fish. From the growth study, inclusion of up to 15% of PFM did not have any adverse effect on the growth performance of tilapia. FAO (1988) had recommended for omnivorous fish, the maximum inclusion of hydrolyzed feather meal was 10% in the diet.

However, in this study we found out that inclusion of up to 15% of PFM did not give any detrimental effects on the growth performance of the red hybrid tilapia, suggesting that it was possible to include feather meal at that percentage in tilapia diet. Bureau *et al.,* (2000) incorporated up to 15% of feather meal in rainbow trout diets and concluded that no detrimental effect affecting the growth performance of the fish could be cautioned. For Indian major carp, it could be used up to 20% inclusion which served 50% of the dietary protein in the feed without compromising the growth performance (Hasan *et al.*, 1997).

Table 3. Growth performance of tilapia (mean±SD) fed with PFM diet for 112 days

Treatment	T1	T2	T3
Initial weight (g)	18.78±3.01[a]	17.67±3.13[a]	18.89±3.69[a]
Final weight (g)	216.82±27.39[a]	189.33±8.59[a]	187.87±18.62[a]
Weight gain (g)	197.31±25.43[a]	169.88±7.52[a]	170.34±21.95[a]
Feed intake (g)	303.76±38.74[a]	289.45±28.01[a]	275.82±30.34[a]
FCR	1.55±0.12[a]	1.70±0.14[a]	1.63±0.23[a]
SGR (%.day^{-1})	2.19±0.15[a]	2.13±0.15[a]	2.06±0.10[a]
PER	2.04±0.15[a]	1.85±0.15[a]	1.94±0.29[a]
Survival (%)	98.33±3.65[ab]	90.67±5.06[b]	99.44±1.36[a]

[ab] Means with different superscript differs significantly ($p<0.05$) within the same row
FCR- Feed conversion ratio; SGR-Specific growth rate; PER-Protein efficiency ratio

Carcass analysis

Fish samples were taken after the 112-d feeding trial for carcass analysis. Fish were culled and filleted to collect the carcass data. The fish were dressed by removing the internal parts and weighed to calculate dressing percentage. The fish were then filleted laterally from the upper head of the fish right until the end to the caudal peduncle, and the flesh was cut up until the bone. The fillet was then weighed to record its weight, and to calculate the fillet to bone ratio. No significant difference was found among the treatments in term of the quality of the fillet (Table 4). Highest fillet percentage was found in the control group, followed by 10% PFM inclusion which was slightly higher than 15% PFM inclusion.

Table 4. Fillet analysis of tilapia (mean±SD) at 112 days of culture

Treatment	T1	T2	T3
Live weight (g)	484.00±91.68	409.83±48.66	456.83±55.56
Dressing weight (g)	399.83±82.10	340.33±38.23	381.50±54.18
Fillet weight (g)	192.67±40.64	163.00±22.09	182.83±31.87
Fillet percentage (%)	48.17±1.31	47.78±1.45	47.76±2.12

*No significant difference for all treatments within the same row (p>0.05)

The fillet of the fish was also subjected to whole proximate composition to see its nutrient content at the end of the 112-d feeding trial. The fillet proximate composition of the tilapia is shown in Table 5.

Table 5. The proximate composition of tilapia fillet (mean±SD)

Nutrient	Initial	T1	T2	T3
Dry matter (%)	91.50^{b}	95.72±0.15^{a}	95.07±0.67^{a}	95.30±0.57^{a}
Crude protein (%)	74.60^{a}	78.12±3.03^{a}	74.68±3.26^{a}	75.68±2.36^{a}
Lipid (%)	3.60^{b}	10.80±1.04^{a}	11.53±2.75^{a}	13.40±2.09^{a}
Crude fiber (%)	2.00^{b}	4.08±0.80^{a}	3.05±0.49^{ab}	3.02±0.69^{ab}
Ash (%)	8.80^{a}	5.64±0.39^{b}	5.70±0.50^{b}	5.58±0.13^{b}
Gross energy (MJ/kg)	14.66^{b}	17.51±0.23^{a}	17.44±0.61^{a}	17.89±0.48^{a}

^{ab} Means with different superscript differs significantly (p<0.05) within the same row

At the beginning of the experiment, 15 fish were culled and filleted prior to the start of the experiment to determine fillet proximate composition and the fillet percentage. The crude protein, lipid, ash and energy content in the initial fish flesh was 74.60%, 3.60%, 2.00% and 14.77 MJ/kg, respectively, and were lower when compared to the treatments. Among the treatments, protein composition was found to be lowest in treatment with 10% PFM (74.68%), while the highest protein percentage was in the control (78.12%). All nutrients in the fillet were not significantly different among the treatments (p>0.05). Ash was highest in the initial fillet with 8.80% and was significantly different (p<0.05) compared to the treatments. Lipid content was highest in the 15% PFM inclusion treatment, and as the inclusion of the PFM increased in the diet, the content of lipid also increased. Crude fiber on the other hand was highest in the control group (4.08%) and the percentage decreased when the inclusion of PFM was increased. Gross energy was highest in 15% PFM fillet (17.89 MJ/kg) which could be associated with the high lipid content in the treatment.

Conclusion

Our findings indicate that inclusion up to 15% of puffed feather meal in the diet of red hybrid tilapia resulted in no detrimental effects on growth characteristics and nutrient utilization. Quality of the nutrients in the fish fillet was similar with the initial fillet except for lipid concentration which increased with the increase of PFM in the diets.

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