

Effective utilisation of poultry byproduct meal as an ingredient in the diet of Genetically Improved Farmed Tilapia (GIFT), cultured in reservoir cages in Tamil Nadu, South India

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ABSTRACT

Evaluation of efficiency of poultry byproduct meal (PBM) as alternative protein source by replacing fish meal (FM) in the diets of GIFT strain of tilapia (*Oreochromis niloticus*) was carried out in cages installed in Poondi Reservoir, Tamil Nadu, South India. Six isonitrogenous (30% protein) and isolipidic (7% lipid) diets were formulated using graded levels of PBM protein to replace FM protein. Each diet was fed to two replicate groups of GIFT tilapia with mean initial weight of 20.38±0.0678 g, for 60 days in cages. The best growth performances in terms of mean weight gain (116.72 g), best food conversion ratio (FCR, 1.14) and maximum hepatosomatic index (HIS, 2.47) values were observed in GIFT tilapia fed PBM diet with 40% fish meal replacement. However, no significant (p>0.05) differences were observed in the whole body proximate composition of fish fed control and treatment diets. It was concluded that, poultry byproduct meal can replace 40% fish meal protein in the diets of GIFT tilapia cultured in reservoir cages without compromising growth, FCR and whole body proximate composition. The study suggests that poultry byproduct meal could effectively replace fish meal without affecting growth and feed conversion of the fish. The findings thus may pave a productive way for reducing environmental pressure of disposal of slaughter house waste.

Keywords: Feed ingredient, Fish meal, GIFT tilapia, Poultry byproduct meal, Reservoir cages, Waste utilisation

Introduction

Global poultry production has increased dramatically in the last 20 years with more than 90 million t of chicken meat and 1.1 trillion eggs now produced every year (FAO, 2018; Blake and Tomley, 2014). At present, India stands fifth in the global poultry meat production. Domestic poultry meat production (broiler-carcass weight) is estimated to have increased from less than 1.0 million t in 2000 to 3.4 million t in 2016 (FAO, 2018). The contribution of meat from poultry industry is approximately 36.68%. Slaughtering and processing of poultry birds provide only one third as meat portion while the rest form poultry wastes and byproducts, which need to be effectively processed and utilised. Poultry byproducts include offal, bone, blood, viscera, head, feet and feathers. In India, the total availability of offal/bones, generated from large slaughter houses is estimated to be 21 lakh t per annum (Jayathilakan et al., 2012). Efficient utilisation of byproducts will have direct impact on the economy and environmental pollution of the country. Non-utilisation or underutilisation of byproducts leads to loss of potential revenue as well as increasing cost of disposal of these products. Non-utilisation of animal byproducts in proper manner may create major aesthetic and catastrophic health problems. In many cases poultry wastes have potential for recycling or for conversion into useful products of higher value.

Nile tilapia (*Oreochromis niloticus*) is the most farmed tropical fish species in the world (World Bank, 2013). Global production was 5.5 million t in 2015 (Fitzsimmons, 2016) and is projected to exceed 6.6 million t by 2030 (FAO, 2018). The attributes which make Nile tilapia so suitable for fish farming are its general hardiness, ease of breeding, rapid growth rate, ability to efficiently convert organic and domestic wastes into high quality protein and good taste (Balarin, 1982). Genetically Improved Farmed Tilapia (GIFT) is known to be a fast growing strain of *O. niloticus* which is widely used in a

variety of culture systems in Asia (Dey, 2000). Ng and Hanim (2007) concluded that growth was influenced by the interaction between diet and tilapia genotype and feed conversion ratios were 14 and 33% better in GIFT tilapia compared with red tilapia fed 25 or 35% protein diet respectively. Supporting the culture of GIFT tilapia in enclosed culture systems like cages would help to boost the global aquaculture sector.

Development of sustainable aquaculture depends on the establishment of alternative feedstuffs to replace fish meal (FM) (Mankinde and Sonaiya, 2012). Animal proteins are free of anti-nutritional factors, palatable, cheaper and readily available than fish meal thus making them perfect FM replacers for tilapia especially in developing nations (El-Sayed, 1999). Poultry byproduct meal (PBM) is made of ground, rendered, or clean parts of the carcass of slaughtered poultry. PBM is similar to FM in composition except being slightly lower in some amino acids (Galkanda-Arachchige et al., 2019). PBM has been tested with varying levels of success so far in salmon (Yang et al., 2004), seabream (Nengas et al., 1999), channel catfish (Sadiku and Jauncey, 1995), tilapia (El-Sayed, 1998) and common carp (Hasan et al., 1997). FM and PBM are highly digestible in terms of protein (88%) and energy (82%). These digestibility values suggest that PBM could be used in aquafeeds to a level similar to FM (Yang et al., 2004). In this context, the present study was designed to replace fish meal with poultry byproduct meal to develop cost effective feed for GIFT tilapia without affecting the growth performance and feed utilisation.

Materials and methods

Experimental set-up

The experiment was conducted in Poondi Reservoir in Sathyamurthy Sagar Lake, situated in Poondi Village, Tamil Nadu, India. The reservoir receives water from Kosasthalaiyar River and its total capacity is 3231 million cubic feet. GIFT tilapia seeds were procured from State Fisheries Department, Krishnagiri, Tamil Nadu and were acclimated to the experimental conditions for one month before the start of the experiment. A total of 600 juveniles (mean body weight 20.38 \pm 0.0773 g) were uniformly distributed at 50 nos. m³ in 12 square cages of $1 \times 1 \times 1$ m dimension and were fed on test and reference diets to duplicate groups of fishes.

Experimental diets

Proximate composition and limiting amino acid content of feed ingredients used are given in Table 1. Ingredient composition of the experimental diets are given in Table 2. Six isonitrogenous (30% protein) and isolipidic (7% lipid) experimental diets were formulated including one control diet (0PBM) containing fish meal as main protein source. Other diets were formulated using poultry byproduct meal (PBM) to replace fish meal dietary protein at 20, 40, 60, 80 and 100% designated as diet 20PBM, 40PBM, 60PBM, 80PBM and 100PBM respectively. The essential amino acid profiles of all the formulated feeds were estimated and the feeds were supplemented with limiting amino acids, lysine and methionine to meet the required levels for Nile tilapia (NRC, 2011). The ingredients were finely ground, blended as per

Table 1. Proximate composition (g 100 g⁻¹ dry matter), gross energy (KJ g⁻¹) and limiting amino acid content (g 100 g⁻¹ protein) of ingredients

Ingredients	Fish meal	Poultry by product meal	Soybean meal	Maize flour	Wheat flour
Moisture	8.96	6.20	7.63	9.80	11.74
Protein	63.28	56.75	48.70	8.29	10.05
Lipid	6.89	25.76	1.99	4.37	1.56
Ash	20.18	8.39	7.75	1.89	1.69
Fibre	0.37	1.0	7.06	2.78	1.60
Gross energy (kcal kg ⁻¹)	4254	5757	4297	3907	3714
Amino acids (g 100 g ⁻¹ pro	otein)				
Arginine	6.2	6.6	7.4	4.5	4.7
Histidine	2.4	1.8	2.6	2.8	2.3
Isoleucine	4.2	3.9	4.6	3.5	3.4
Leucine	7.2	7.0	7.5	12.0	6.5
Lysine	7.5	4.4	6.1	3.1	2.9
Methionine	2.7	1.4	1.4	2.1	1.6
Phenylalanine	3.9	3.9	5.0	4.8	4.5
Threonine	4.1	3.9	3.9	3.6	2.9
Tryptophan	1.0	0.7	1.3	0.7	1.2
Valine	4.9	5.4	4.8	4.8	4.3

Table 2. Ingredients and proximate composition (% dry matter basis) of experimental diets

Ingredients	0PBM (Control)	20PBM	40PBM	60PBM	80PBM	100PBM
Fish meal (Anchovy) ¹	33	26.5	20	13.2	6.63	0
Soy bean meal ¹	9	9.2	9.3	9.6	10	10.5
Poultry byproduct meal ²	0	7.4	14.8	22.2	29.6	37
Maize flour ¹	28.49	27.90	32.82	36	37.73	38.74
Wheat flour ¹	23.11	22.86	17.20	13.46	10.76	8.50
Palm oil ¹	2.81	2.30	1.78	1.17	0.55	0.27
Vitamin premix ³	1	1	1	1	1	1
Mineral premix ⁴	1	1	1	1	1	1
L-Lysine ⁵	0.44	0.60	0.80	0.98	1.26	1.34
DL-Methionine ⁶	1.15	1.24	1.30	1.39	1.47	1.65
Proximate composition (%	dry matter basis)					
Moisture	10.77	8.69	9.88	10.06	9.70	10.02
Protein	31.40	31.91	31.69	31.64	30.06	30.49
Lipid	7.03	6.98	7.12	7.34	6.94	7.48
Ash	9.46	8.80	7.91	7.10	6.23	5.44
Fibre	1.32	1.26	1.43	1.34	1.36	1.05
Gross energy (kJ g ⁻¹)	17.31	17.94	18.03	18.20	18.47	18.70

¹ Hakita Feeds Pvt Ltd., Kodambakkam, Chennai, India

formulation and passed through a single screw extruder (SFT 65, UNITECH Ltd., New Delhi) to obtain 2 mm floating pellets. The prepared diets were stored in zip-lock polythene packs in a cool dry place until used.

Feeding trial and fish sampling

Each diet was fed to two replicate groups of GIFT tilapia juveniles according to the feeding chart given by NFDB (2016), for a period of for 60 days. The feed was given in three split doses daily at 08:00, 10:00 and at 16.00 hrs. The fishes were weighed every fortnight and the amount of feed adjusted accordingly. Fishes were harvested on termination of 60 days of feeding trial and length and weight of the fish were recorded. Ten fish from each cage were sampled for whole body proximate analysis.

Proximate analysis of experimental diets and experimental fishes

The moisture, crude protein, lipid, ash, fiber, nitrogen free extract (NFE) and gross energy in the poultry byproduct meal incorporated diets and whole body of the fishes were analysed according to AOAC (1995). Moisture was determined by oven drying at 105-110°C for 6 h and protein by Micro Kjeldhal method after acid digestion. Lipid was determined by Soxhlet's method by

extracting in ether which is continuously volatilised at 60-80°C. Crude fibre was estimated by estimating dried fat free residues after digestion with dilute acid (0.255N) and alkali (0.313N). Ash was determined by ignition at 600°C for 6 h in a muffle furnace. The gross energy (GE) was estimated using digital bomb calorimeter (Model No. RSB, Rajdhani Scientific Inst. Co., New Delhi, India). The limiting amino acids were analysed after acid hydrolysis with 6 M HCl using HPLC (Waters Binary Pump 1525). Gross energy was calculated based on conversion factors for carbohydrate, protein and lipid as 17.2; 23.6 and 39.5 kJ g⁻¹ respectively.

Survival rate

Survival rate was calculated at the end of the experiment by counting the number of fishes in each cage and is estimated as follows:

Survival (%) = Number of fishes harvested/Number of fishes stocked x 100

Growth parameters

Growth parameters of the experimental fishes were assessed by taking their body weight at the end of the feeding trial. After weighing, GIFT tilapia juveniles were dissected to remove the liver for determination of

² Pragathi Broilers and Farms, Thiruvallur, Tamil Nadu, India

³Composition of vitamin premix (quantity kg⁻¹): Vit. A - 1,00,00,000 IU, Vit. B1-5,000 mg, Vit. B2 - 5,000 mg, Vit. B3- 6,000 mg, Vit. B5 -6,000 mg, Vit. B6 - 6,000 mg, Vit. C - 60,000 mg, Vit. D3 - 20,00,000 IU, Vit. E - 10,000 EU, Vit. H - 200 mg.

⁴Composition of mineral premix (quantity kg⁻¹): Magnesium - 2,800 mg, Iodine - 7.4 mg, Iron - 7,400 mg, Copper - 1,200 mg, Manganese - 11,600 mg, Zinc - 9,800 mg, Cobalt chloride - 4 mg, Potassium - 100 mg, Selenium - 4 mg, Calcium carbonate - 27.25%, Phosphorous - 7.45 mg, Sulphur - 0.7 mg, Sodium - 6 mg, Calpan - 200 mg, Aluminium - 1,500 mg and Choline chloride - 10,000 mg

⁵Ajinomoto Heartland, Inc., Chicago (L-lysine HCL - 98.5%)

⁶Evonik AG, Germany (DL-methionine: MetAMINO® - 99%)

hepatosomatic index (HSI). Moisture was removed with the help of blotting paper and then the liver was weighed (g). Growth was determined by evaluating the following growth and nutrient utilisation indices:

Mean weight gain Final body weight - Initial (MWG)(g)body weight [(Final body weight - Initial body Percentage weight gain (PWG) (%) weight) /Initial body weight] x100 Food conversion Total feed consumed (g)/ ratio (FCR) Wet weight gain (g) [(Ln final mean weight - Ln initial Specific growth rate (SGR) (%) mean weight)/number of days] x 100 Protein efficiency Wet wet gain (g)/Protein ratio (PER) ingested (g) [Weight of liver/Weight of the fish] Hepatosomatic Index (HSI)

Cage monitoring and water quality parameters

The cages were cleaned regularly to avoid net clogging and to ensure proper water exchange. Fish were monitored regularly to assess the feed intake and health status. Dead fish and leftover feed were removed promptly from the cages. Water quality parameters such as temperature, pH, dissolved oxygen, transparency and conductivity were monitored every 15 days following standard methods (APHA, 2005).

Statistical analysis

All the data were subjected to one way analysis of variance (ANOVA), which was carried out to find out whether there is any significant difference among the growth related parameters, whereas the Tukey's multiple range test was used to compare treatment means. Statistical analysis was performed using the software SPSS 20.0 for windows (SPSS Inc., Chicago, USA).

Results

The range of water quality parameters recorded during the feeding trial were, water temperature: 25.5-27°C, dissolved oxygen: 5.5-6.5 mg l⁻¹, pH: 7.5-8.0, Hardness: 120-130 ppm, alkalinity:140-170 ppm, ammonia-N: 0.2-0.4 ppm, nitrite-N: 0.1-0.2 ppm, nitrate-N: 0.06-0.08 ppm.

Survival and growth parameters

At the end of the 60 days feeding trial, the survival rate (%) was not significantly different between different treatments (Table 3). Percentage weight gain (PWG) was highest (569.73 g) in GIFT tilapia fed on 40PBM diet, which was significantly different (p<0.05) from the percentage weight gain in fish fed on 0PBM, 20PBM, 60PBM and 80PBM diets. Lowest percentage weight gain was recorded in the fishes fed with 100PBM, though it was not significantly different (p>0.05) from fishes fed with 60PBM diet.

The highest specific growth rate (SGR) of 3.16 was observed in fish fed on 40PBM diet and lowest SGR (2.18) was recorded in group fed with 100PBM diet. SGR values of 60PBM, 80PBM and 100PBM diets were lower than control diet.

Maximum protein efficiency ratio (PER) was recorded in 40PBM (2.91), followed by 0PBM (2.66) and 20PBM (2.79) diet groups. Lowest PER was 100PBM (2.22) followed by 80PBM (2.36) diet groups. GIFT tilapia fed with 40PBM diet showed good food conversion ratio (FCR) of 1.14, followed by 0PBM (1.24) and 20PBM (1.19). Poor FCR was observed in 100PBM and 80PBM diet groups. Maximum hepatosomatic Index (HSI) value of 2.47 was observed in fish fed 20PBM diet compared to other experimental diets.

Initial and final whole body proximate composition

Initial moisture content, crude protein, ether extract and ash content of GIFT tilapia were 71.53±0.8, 16.63±0.6, 6.28±0.3 and 4.23±0.2 respectively (Table 4). No significant differences (p>0.05) were observed in the proximate composition of GIFT tilapia fed control and treatment diets.

Table 3. Growth performances and feed utilisation of GIFT tilapia fed PBM supplemented diets

Treatment	Mean initial	Mean final	Mean weight	PWG	SGR	PER	FCR	HSI	Survival (%)
	weight (g)	weight (g)	gain (g)						
0PBM	20.43±0.85	116.59±24.4 d	96.16±23.54 ^d	470.64±27.4d	2.90±5.57°	2.66±5.32d	1.24±0.62b	2.34 ±0.04b	98±2.11
20PBM	20.41±0.91	123.31±28.6 e	102.90±27.68e	504.18±30.25e	2.99±5.73bc	2.79 ± 4.16^{e}	1.19 ± 0.79^{b}	$2.47\ \pm0.07^{\rm d}$	98±3.1
40PBM	20.48 ± 0.88	$137.21\pm22.62^{\mathrm{f}}$	116.72±21.73 ^f	569.73±24.43f	3.16 ± 5.39^{c}	2.91 ± 3.47^{c}	1.14 ± 0.95^{a}	$2.36\!\pm\!0.05^{b}$	98±3.01
60PBM	20.35±0.86	106.31±30.73°	85.96±29.86°	422.31±34.43°	2.75 ± 5.94^{bc}	2.43 ± 3.74^{c}	1.36 ± 0.89^{c}	2.27 ± 0.11^a	100
80PBM	20.32±0.86	86.27±15.84 ^b	65.95 ± 14.98^{b}	324.58±17.27 ^b	2.40±4.84ab	2.36 ± 3.02^{b}	$1.40{\pm}1.1^d$	2.31 ± 0.04^{b}	100
100PBM	20.31 ± 0.84	$75.41{\pm}10.84^a$	55.10±10.01a	$271.31{\pm}11.89^a$	$2.18{\pm}4.26^{a}$	2.22 ± 2.17^{a}	1.49 ± 1.53	12.40±0.03°	98±3.01
p Value	0.065	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.452

Table 4. Whole body proximate composition (%) of GIFT tilapia juveniles fed experimental diets

Parameter	0PBM	20PBM	40PBM	60PBM	80PBM	100PBM	p value
Moisture	68.43 ± 0.28	68.34±0.33	68.38 ± 0.27	68.51±0.19	68.43 ± 0.37	68.34 ± 0.23	0.136
Crude protein	18.54 ± 0.17	18.43 ± 0.15	18.63 ± 0.09	18.48 ± 0.21	18.44 ± 0.14	18.52 ± 0.16	0.075
Crude lipid	6.23 ± 0.07	6.34 ± 0.11	6.53 ± 0.13	6.45 ± 0.05	6.58 ± 0.17	6.36 ± 0.14	0.064
Ash	4.38 ± 0.06	4.49 ± 0.03	4.28 ± 0.07	4.37 ± 0.03	4.44±0.01	4.23 ± 0.05	0.182

Mean value of initial whole body composition: 71.53 % moisture, 16.63% crude protein, 6.28% crude lipid and 4.23% ash Values are expressed as means±SD of two replicates per treatment (n=2)

Discussion

Various researchers have studied the effect of substitution of fish meal with different protein sources on the growth, feed utilisation and whole body composition of fishes (El-Saidy and Gaber, 2002; Abdelghany et al., 2003; Borgeson et al, 2006; Gaber, 2006; Hernandez et al., 2010; Figueiredo-Silva et al., 2015). Poultry byproduct meal (PBM) is a rendered product obtained from the waste of poultry production and processing plants. It is usually made from inedible portions of poultry, excluding feathers. It has been evaluated as a fish meal (FM) replacement in the diets of gibel carp Carassius auratus gibelio (Yang et al., 2006), Nile tilapia Oreochromis niloticus (El-Sayed, 1998), African catfish Clarias gariepinus (Abdel-Warith et al., 2001), chinook salmon Oncorhynchus tshawytscha (Fowler, 1991), rainbow trout Oncorhynchus mykiss (Alexis et al., 1985), coho salmon Oncorhynchus kisutch (Higgs et al., 1979), European eel Anguilla anguilla (Gallagher and Degani, 1988), gilthead seabream *Sparus aurata* (Alexis 1997), red seabream Pagrus major (Takagi et al., 2000), sun shine bass Morone chrysops x M. saxatilis (Webster et al., 1999), red drum Sciaenops ocellatus (Kureshy et al., 2000) and Pacific white shrimp *Penaeus vannamei* (Davis and Arnold, 2000).

Earlier studies have shown that PBM with 60% crude protein and 16-22% ash, if used alone, could generally replace not more than 50% of FM protein, or growth was compromised (Steffens, 1994). However, an improvement in the nutritional quality of PBM has been achieved in recent years. When supplemented with amino acids (lysine, methionine or tryptophan) or combined with other proteins, PBM showed a more pronounced nutritional potential (Webster *et al.*, 1999). High quality PBM now contains about 70% crude protein and relatively low ash content (Davis and Arnold 2000) and can even be used without supplementation, replacing 75% or even 100% of the FM without a significant depression in fish performance (Alexis 1997; Nengas *et al.*, 1999; Takagi *et al.*, 2000).

In comparison with fish meal, the poultry byproduct meal used in this study had almost equally high protein content (56-60%), lower ash content, but higher lipid

content. Results of the present study indicated that the diets with PBM at 20 and 40% of the dietary fish meal protein replacements showed higher growth performances compared to control diet (0PBM), which contained only fish meal. Takagi et al. (2000) found that yearling red seabream fed diets with upto 100% FM replaced by PBM showed a growth performance and feed utilisation similar to or better than fish fed FM-based control diets. Davis and Arnold (2000) reported that replacement of 80% FM protein in practical diets for P. vannamei resulted in a significant increase in weight gain. These varying results may relate to the species tested, but are more likely to be a consequence of the different quality of PBM production, which varies among producers (Dong et al., 1993; Bureau et al., 1999). Dong et al. (1993) found that there were significant differences in proximate composition and protein digestibility in PBM samples obtained from six different manufacturers.

In the present study, with supplementation of required crystal lysine and methionine, growth of GIFT tilapia fed with 20PBM and 40 PBM diets showed similar kind of growth performance as control diet (0PBM) in terms of mean weight gain, PER, FCR, average daily growth and mean feed intake. Fowler (1991) was successful in rearing chinook salmon (*Oncorhynchus tschawytscha*) with a diet containing 20% PBM meal without additional amino acid supplementation. Alexis *et al.* (1985), obtained very good results by feeding rainbow trout, *Salmo gairdneri* with 20% PBM meal, with methionine supplementation.

The best FCR values observed with 40PBM diet suggests that replacement of fish meal protein by PBM at 40% could improve feed utilisation. FCR and PER values were consistent with the values reported in other studies (Fasakin *et al.*, 2005; Prabu *et al.*, 2018). In addition, the results are in agreement with the findings by Hasan *et al.* (1997), who found that FCR and PER were better at lower substitution levels of FM by hydrolysed feather meal (HFM) in Indian major carp diet. Likewise, FCR and PER decreased significantly with diets containing high levels of HFM in Nile tilapia (Tacon *et al.*, 2008; Davies *et al.*, 1989).

GIFT tilapia fed with PBM incorporated diets showed no significant difference in moisture, protein, lipid and ash

content in final fish whole body proximate composition among all the treatments. Similar kind of results were observed by Hernandez et al. (2010) where, addition of PBM in Nile tilapia diets did not significantly affect (p > 0.05) carcass crude protein and moisture, however it is contrary to the carcass crude lipid and ash content, which was significantly different among treatments. According to El-Sayed (1998), PBM incorporated fish meal diet caused significant difference in lipid and ash content of Nile tilapia carcass composition. The present study indicated that PBM in the diet of GIFT tilapia could be used as a main protein source to replace fish meal upto 40% of dietary protein without negative effect on the growth performances and feed utilisation. Optimal replacement of fish meal with PBM in diet for GIFT tilapia requires further investigation with the consideration of essential amino acid requirements.

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