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Estimation of Apparent Digestibility Coefficient of Guar, Canola and Meat Meal for *Labeo rohita*

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ABSTRACT

Apparent nutrient (dry matter, crude protein, crude fat & gross energy) digestibility of three feed ingredients guar, canola and meat meal was determined for evaluating the nutrients potential for *Labeo rohita*. For an eight week experiment, a reference diet was mixed with test ingredients in a 70:30 ratio to formulate test diets. Chromic oxide was added as an indigestible marker. The apparent dry matter digestibility was higher (81.13±6.03) for meat meal as compared to canola and guar meal being 70.25±0.95 and 59.91±0.66, respectively. The apparent crude protein digestibility for meat was higher (85.31±4.12) and this was followed by canola (60.67±2.38) and guar meal (50.30±0.94). Apparent crude fat digestibility for meat was higher (92.5±3.07) as compared to canola (87.89±3.00) and guar meal (85.93±7.44). The apparent gross energy digestibility of meat meal was higher (95.54±10.27) than guar (88.32±11.56) and canola meal (66.76±3.49).

Key Words: Nutrient digestibility; *Labeo rohita;* feed ingredients

INTRODUCTION

With the intensification of fish culture operations and constant increases in the cost of many conventional feedstuffs, the need to develop nutritional, economical and efficient feed on the basis of digestibility of respective fish. Assessing the nutritional quality of foodstuffs is best done, by conducting extensive feeding trials but they are time consuming and expensive. The next best method of quality assessment is to measure apparent digestibility coefficients using an *in vivo* procedure. It is said that digestibility is one of the most important aspects in evaluating the efficiency of feedstuffs.

Digestibility describes the fraction of the nutrients of energy in the feedstuff that is not excreted in the faeces. Digestibility is one of the most important aspects in evaluating the efficiency of feedstuffs. The digestibility of nutrients is not precisely defined in many commercial feeds and it is shown that feed performances and digestibility can be increased with the use of enzymes that enhance plant protein use, and by use of extrusion technology (Hasan, 2001).

Determining the digestibility of nutrients in a feedstuffs is important not only to enable formulation of diets that maximize the growth of cultured fish by providing appropriate amounts of available nutrients, but also to limit the wastes produced by the fish. A wide variety of agro based feedstuffs which are rich in protein carbohydrate and energy are available in Pakistan. Out of these feedstuffs, a few have been evaluated for their apparent nutrient and energy digestibility for *Labeo rohita* in Pakistan (Salim *et al.*, 2004) and in India for Indian major carps (Nandeesha *et al.*, 1991; Singh, 1991).

Apparent digestibility coefficient (ADC) vary between fish species and feedstuffs it is important to determine the ADCs of different feedstuffs for optimum inclusion in formulated diets and to allow effective ingredient substitutions for achieving maximum growth. Digestibility estimates also indicate the level of indigestible nutrients voided, accounting for a major portion of aquaculture waste (Cho, 1993).

The method presently used by most fish nutritionist to determine digestibility of feeds, feed ingredients is by using Chromic oxide ($Cr_2 O_3$) as an indigestible marker at 0.5 to 1.0% in test diets (Furukawa & Tsukahara, 1966). By analyzing the feed and faeces for their various components (protein, lipid, carbohydrate & energy etc.) the digestibility of each nutrient can be determined. Technical difficulties associated with digestibility methods for aquatic species include the collection of representative faecal samples, leaching of nutrients from the faeces in contact with water and fracturing of faecal material into small particles with time, a process promotes by aeration and movement of the experimental animals (De Silva & Anderson, 1995).

Presently, there are three systems (Tuf column, Guelph system & St.Poe system) that have been adopted in several laboraties around the world are most likely to produce meaningful estimates of digestibility of nutrient if used correctly (Bureau & Cho, 1999). For the collection of fecal material from water, UA system was developed on settling column principle by utilizing locally available materials and was installed in Fish Nutrition Laboratory, Department of Zoology and Fisheries, University of Agriculture, Faisalabad. Keeping in view the importance of apparent digestibility for the formulation of fish diet, the apparent nutrient digestibility of feedstuffs (guar meal,

canola meal & meat meal) for *Labeo rohita* was determined using chromic oxide as the indigestible marker.

MATERIALS AND METHODS

The present study was carried out for the estimation of apparent digestibility coefficient (ADC) of guar, canola and meat meal for *Labeo rohita*. The experiment was conducted in Fish Nutrition Laboratory, Department of Zoology and Fisheries, University of Agriculture, Faisalabad.

Experimental fish. *Labeo rohita*, fingerlings were purchased from the Government Fish Seed Hatchery, Satiana, Road, Faisalabad. The fingerlings were acclimatized for one week in glass aquaria (37 x 29 x 45 cm). During this period fish were fed once daily to apparent satiation on the reference diet used in subsequent digestibility study (Allan & Rowland, 1992). Before the start of experiment, fish were treated with 5 g/L sodium chloride (NaCl) to ensure fish were free of ectoparasites and to prevent fungal infection (Rowland & Ingram, 1991).

Feed ingredient and diet preparation. Each test diet was composed of 70% reference diet and 30% test ingredient (guar, canola & meat meal) on dry weight basis. Chromic oxide was used an inert marker and incorporated into the reference diet and test diets at 1.0% inclusion level. The percentage of ingredients and calculated chemical composition (Win feed program) of reference and three test diets are shown in Table I. Reference and test ingredients were ground and sieved for incorporation into diets. All dry ingredients were mixed in mixer for 30 minutes, where after, fish oil was gradually added, while mixing constantly. Eighty five (85) ml of water per 100 g of feed was slowly blended into the mixer, resulting in a suitably texture, dough, as for fish food (Lovell, 1989). Drying was carried out in a convection oven at 35^{o}C for 48 h. The dry product was cut into pellets of 2.5 mm diameter. The above procedure was followed to produce a reference and three test diets.

Experimental system. An eight week digestibility experiment was conducted by using UA System in which settling column was used to separate the faecal material of fish from effluent water. Water temperature remained (30-32^{o}C) during the study period. Air pumps were used to maintain the level of dissolved oxygen (5-5.5 mg L^{-1}).

Feeding protocol and faecal collection. After acclimatization, fingerlings were transferred into glass aquaria ((37 x 29 x 45 cm) via random interspersion. For each treatment two replicates were used and in each replicate ten fingerlings were stocked (average weight 16 gm). Fishes were fed at the rate of 2% of live wet weight on their prescribed diet twice daily (morning & afternoon) in the feeding chamber. After a feeding session of 2-3 h, fingerlings were shifted in UA System for faecal collection. Faecal collection continued for 60 days when it was judged that a sufficient sample had been collected for chemical analysis.

Table I. Ingredients percentage and chemical composition of reference and test diets

Ingredients	Reference diet	Test diet 1 (guar meal)	Test diet II (canola meal)	Test diet III (meat meal)
Fish meal	59.03	34.59	36.98	28.95
Rice broken	7.03	7.16	5.56	8.78
Rice polish	13.34	10.29	9.43	11.05
Wheat bran	13.78	11.23	8.68	10.9
Fish oil	4.83	4.73	7.34	8.32
Vitamin premix	1	1	1	1
Chromic oxide	1	1	1	1
Test Ingredient-I (Guar)		30	----	----
Test Ingredient-II (Canola)	---	---	30	----
Test Ingredient-III (Meat meal)	----	---	----	30
Total	100.01	100	100	100
Chemical composition				
Dry matter (%)	90.5	91.24	90.59	91.52
Crude protein (%)	30	30	30	30
Crude fat (%)	10.65	10	11.9	13.55
Crude fiber (%)	4.42	6.64	4.89	4.49
Gross energy (kcal/kg)	2700	2736.03	2700	2700
Ash (%)	16.66	11.71	13.78	11.01

Analytical procedure. A representative sample of feed or oven dried faeces was homogenized using a motor and pestle and analyzed essentially by AOAC (1990) procedures: dry matter (DM) by oven drying at 105ºC for 16 h; crude protein (CP) by micro-kjeldahl analysis and gross-energy by oxygen bomb calorimeter. Crude fat was determined following petroleum ether extraction method (Bligh & Dyer, 1959) through 10454 soxtec system HTz and chromic oxide estimation by using acid digestion method, (Divakaran *et al*., 2002), through UV-VIS 2001 spectrophotometer.

RESULTS

The proximate nutrient analysis of feed, faeces and estimation of chromic oxide are shown in Table II. Apparent nutrient digestibility (%) of dry matter, crude protein, crude fat and gross energy of individual feed ingredients (Mean ± SE, n=2) are shown in Table III. Apparent nutrient digestibility coefficient of dry matter was highest for meat meal, (81.13±6.03), followed by canola and guar meal (70.25±0.95) and (59.91±0.66), respectively. The analysis of variance of dry matter digestibility (%) of all the three test ingredients were significant ($P<0.05$). The comparison of means of test ingredients for dry matter (Table IV) revealed that there was non-significant difference between test ingredients-I (guar) and II (canola) but there was significant difference between test ingredient-I (guar)and test ingredient-III (meat meal). The test ingredient-II (canola) showed non-significant difference from test ingredient-III (meat meal).

Table II. Proximate nutrient analysis of feed, faeces and estimation of chromic oxide (Cr_2O_3)

Component	Reference Diet	Test Diet-I (Guarmeal)	Test Diet-II (Canolmeal)	Test Diet-III (Meat Meal)
		Feed		
Dry matter (%)	91.59±0.29	98.56±0.33	98.92±0.06	98,87±0.90
Crude protein (%)	30.00±0.00	28.33±0.28	28.92±0.24	29.63±0.32
Crude fat (%)	4.73±0.24	6.97±0.02	6.47±0.02	4.47±0.04
Gross energy kcal/g	1.63±0.01	3.15±0.07	2.89±0.03	2.83±0.00
Chromic oxide (%)	0.87±0.01	0.98±0.00	0.96±0.00	0.87±0.05
		Faeces		
Dry matter (%)	95.22±0.55	74.58±0.37	71.08±0.06	73.08±0.07
Crude protein (%)	11.56±0.23	11.48±0.23	10.52±0.43	9.13±0.12
Crude fat (%)	3.36±0.43	3.38±0.19	2.93±0.01	2.28±0.01
Gross energy kcal/g	1.05±0.01	1.61±0.01	1.48±0.03	1.53±0.02
Chromic oxide (%)	1.13±0.04	1.08±0.02	1.05±0.01	1.04±0.03

Table III. Apparent nutrient digestibility coefficient (%) of test ingredients (Mean ± SE, n =2) using chromic oxide as marker

Test ingredients	Dry matter	Crude protein	Crude fat	Gross energy
Test ingredient-I (guar)	59.91±0.66	50.30±0.94	85.93±7.44	88.32±11.56
Test ingredient-II (canola)	70.25±0.95	60.67±2.38	87.89±3.00	66.76±3.49
Test ingredient-III (meat meal)	81.13±6.03	85.31±4.12	92 .50±3.07	95.54±10.27

Table IV. Comparison of means of test ingredients for dry matter and crude Protein

Comparison of means of test ingredients for dry matter		Comparison of means of test ingredients for crude protein	
Ingredients	**Mean**	**Ingredients**	**Mean**
Guar	59.91B	Guar	50.30B
Canola	70.25AB	Canola	60.67B
Meat meal	81.13A	Meat meal	85.31A
The ingredients followed by the same letters are non significantly different at 5% level of significance using Tukey's Test		The test ingredients followed by different letters are significantly different at 5% level of significance using Tukey's Test	

The apparent crude protein digestibility for meat meal was higher (85.31±4.12) and this was followed by canola (60.67±2.38) and guar meal (50.30±0.94).The analysis of variance of crude protein affirmed that the apparent crude protein digestibility(%) of all the three test ingredients were highly-significant (P<0.05). The comparison of means of test ingredients for crude protein (Table IV) showed that the digestibility percentage of crude protein for test ingredient-I (guar) was non-significantly different from test ingredient-II canola). Whereas test ingredient-I (guar) was significantly different from that of test ingredient-III (meat meal) and similarly, test ingredient-II (canola) was also significantly different from test ingredient-III (meat meal).

Apparent crude fat digestibility for meat meal was higher (92.5±3.07) as compared to canola (87.89±3.00) and guar meal (85.93±7.44). The analysis of variance of crude fat digestibility showed that the apparent crude fat digestibility (%) of test ingredients were non-significant (P>0.05).

The apparent gross energy digestibility of meat meal was higher (95.54±10.27) than guar (88.32±11.56) and canola meal (66.76±3.49). The analysis of variance concluded that the apparent gross energy digestibility (%) of the three test ingredients were non-significant (P>0.05).

DISCUSSION

The apparent digestibility of nutrients in the test ingredients was higher in animal ingredient (meat meal) than plant ingredients (canola & guar meal). The apparent digestibility of dry matter was comparatively higher for meat meal (81.13±6.03) than guar meal (59.91±0.66) and canola meal (70.25±0.95). The low digestibility of dry matter for plant ingredients in the present study may be due to higher carbohydrate contents. Several other studies reported low dry matter digestibility coefficients in plant protein with high carbohydrate contents (Allan *et al*., 2000; Laining *et al*., 2003). However, Sugiura *et al.* (1998) suggested that fish cannot utilize non-protein component from plant material effectively because of the presence of starch and fibers.

The apparent crude protein digestibility (APD) was also higher in animal ingredient as compared to plant ingredient. The apparent digestibility for crude protein was higher in meat meal (85.31±4.12) as compared to guar meal (50.30±0.94) and canola meal (60.67±2.38). The higher apparent digestibility for meat meal might be the contribution of amino acid profile which, are well balanced in meat meal than in guar meal and canola meal. The low APD in plant ingredients might be due to higher contents of carbohydrates.

The current study showed that the crude fat in meat meal (92.50%±3.07) and canola meal (87.89%±3.00) was well digested by *Labeo rohita.* Whereas the digestibility for guar meal (85.93%±7.44) was lower. By comparison, the digestibility of crude fat in meat meal was nearly same to the value (92%) reported by Mark *et al.* (2005). However, fat digestibility of present study was higher than the value (59%) as reported by Gaylord and Gatlin (1996). They concluded that some of the difference in lipid digestibility values for red drum compared to other species might be attributed to differences in techniques used to extract lipid.

Similarly the apparent energy digestibility (AED) of meat meal was also higher (95.54±10.27) than guar meal (88.32±11.56) and canola meal (66.76±3.49). The AED of guar meal and canola meal in current study was comparatively lower than meat meal. The lower AED of

plant ingredients could be attributed to their higher carbohydrate contents and poor digestibility by carnivorous fish (Lupatsch *et al*., 1997). Similar confirmation was reported by Storebakken *et al*. (1998). They concluded that increased dietary carbohydrate (10-20%) reduced dry matter, energy and fat digestibility but had little effect on protein digestibility for rainbow trout. According to Rawles *et al*. (2000) meat meal appeared to be the best ingredient for sunshine bass diets in terms of over all nutrient profile and digestibility of nutrients.

CONCLUSION

In conclusion, *Labeo rohita* was able to digest energy and nutrients (protein, fat & dry matter) of the animal ingredients (meat meal) more efficiently than plant ingredients (canola meal & guar meal), though the percentage of nutrient digestibility of guar meal and canola meal was comparatively less but all the digestibility values were somewhat near to standard digestible values of carps (NRC, 1993). The data established in this study will provide the basis for inclusion of meat meal as well as guar meal for the formulation of diet for *Labeo rohita*.

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