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Article *in* ANIMAL NUTRITION AND FEED TECHNOLOGY · January 2016
DOI: 10.5958/0974-181X.2016.00005.6

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Animal Nutrition and Feed Technology (2016) 16: 51-60
doi: 10.5958/0974-181X.2016.00005.6

Effect of Shrimp Head Silage Hydrolysate and Distiller's Dried Corn Grain on Digestibility and Growth of Red Tilapia (*Oreochromis mossambicus*)

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(Received July 8, 2014)

ABSTRACT

Valdez-González, F.J, García-Ulloa, M., Hernández-Llamas, A., Rodríguez-Montes de Oca, G.A. and Rodríguez-González, H. 2016. Effect of shrimp head silage hydrolysate and distiller's dried corn grain on digestibility and growth of red tilapia (Oreochromis mossambicus). Animal Nutrition and Feed Technology, 16: 51-60.

We studied the effect of shrimp head silage protein hydrolysate (SHSP) and distillers dried corn grain (DDCG) as ingredients on the digestibility of diets for *Oreochromis mossambicus*. Three diets containing 30% protein, 10% lipid, and 4.9 kcal/g calorific value were prepared: a reference diet and two based on the inclusion of 84.0% of the reference diet, 15% of SHSP or DDCG, and 1% of chromic acid as a marker. Six fish (45.5±2.5g) were introduced in each culture unit (60 L). There were significant differences ($P<0.05$) for the apparent dry matter digestibility among the diets. The SHSP diet obtained the higher dry matter digestibility (86.33±0.8%). While, no differences ($P<0.05$) in the apparent protein digestibility were observed among the diets and ingredients. The mean final weight (58.77±3.8g) was similar for the three experimental treatments. It can be conclude concluded that the red tilapia is able to efficiently consume diets containing these animal and plant-derived ingredients.

Key words: Digestibility, Distillers dried grain, Extrusion, *Oreochromis mossambicus,* Protein hydrolysate

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INTRODUCTION

As feed represents more than 50% of total production costs, rapid expansion of fish farming demands less expensive sources of nutritious fish feeds. Protein is the first limiting nutrient in formulating cost-effective diets because market prices for protein ingredients are high (Guimaraes *et al.*, 2008; Yu *et al.*, 2013). Currently, fishmeal is the predominant protein source in fish feeds (NRC, 1993; Halver and Hardy, 2002) because it contains nearly ideal nutritional quality and low levels of anti-nutritional factors (Zhou *et al.*, 2004). High demand for fishmeal for aquafeeds and other livestock results in rising prices for this product (FAO, 2008). Rapid expansion of the aquaculture industry requires alternative protein sources to sustain further growth. Animal meals, oilseeds, grains, and legumes can provide inexpensive protein sources for fish (Liebert and Portz, 2005; Ghomi *et al.*, 2015). The most important characteristic of any feedstuff is bioavailability, particularly digestible protein, available amino acids, and digestible energy. Understanding of digestibility of ingredients is a basic requirement for formulating diets (Cho and Kaushik, 1990). Coefficients of apparent digestibility provide estimates of nutrient availability in feedstuffs and are used to select ingredients that optimize nutritional value and cost of formulated diets (Fagberno and Jauncey, 1998).

Digestibility of feedstuffs in many fish species, including tilapia, has been reported (Allan *et al.*, 2000; Rawles and Gatlin, 1998; Pereira Da Silva and Pezzatto, 2000; Lee, 2002; Dong *et al.*, 2010). Fish processing waste and distillers dried grains are recommended because they are widely available, cheap, contain nutrients, and may be as partial substitutes for ûsh meal in aquacultural feeds (Glencross *et al.*, 2011). Nutrients and energy digestibility of feed ingredients vary with the process for obtaining them, digestion rate, and feed quality (NRC, 1993). The ingredients like shrimp head silage protein hydrolysate (SHSP) and distillers dried corn grain (DDCG) are easily available and of low cost in the regional market. However, there are no antecedents in the literature of studies of SHSP and DDCG in diets for red tilapia (*Oreochromis mossambicus*). This study assessed whether protein of local and low cost ingredients viz. SHSP and DDCG had acceptable digestibility of dry matter (DM) and crude protein (CP) for red tilapia.

MATERIALS AND METHODS

Diet preparation

The proximate composition of the SHSP and DDCG ingredients are shown in Table 1. Formulation and proximate composition of diets are shown in Table 2. The DDCG was a corn by-product collected from an ethanol factory. The SHSP was obtained from a chitin facility which is extracted according to Cira *et al.* (2002). The fermentation process uses *Lactobacillus* spp. in a reactor. The column reactor consisted of two modules with a mesh between them. The solid (chitin) remained in the upper module and the liquid was gathered in the lower module. After 2 days of fermentation the liquor was taken off from the reactor and used for the feeding trial.

Table 1. Proximate composition (%) of the shrimp head silage protein hydrolysate (SHSP), and distillers dried corn grains (DDCG).

Attributes	Ingredients	
	SHSP	DDCG
Moisture	5.38±0.88	8.36±0.12
Crude protein	38.61±0.31	25.53±0.26
Ether extract	0.03±0.01	9.28±0.14
Ash	8.15±0.24	4.52±0.10
Fibre	ND	6.74±0.23
Nitrogen free extract	53.26	53.94

ND= not detected

Table 2. Ingredients composition (g/kg) and proximate analysis (% DM) of the experimental diets for tilapia.

Ingredients	Diet		
	Reference	SHSP	DDCG
Ingredient composition			
SHSP		150	-
DDCG	-	-	150
Fish meal	200	50	50
Soybean meal	150	224	260
Wheat meal	419	331	309
Fish oil	35	42	35
Soy lecithin	35	42	35
Vitamins premix	1	1	1
Minerals premix	10	10	10
Chromic oxide	10	10	10
Starch	100	100	100
Binder (grenetine)	40	40	40
Proximate composition			
Crude protein	29.35±0.14	29.88±0.08	30.21±0.29
Ether extract	10.30±0.2	10.47±0.03	9.75±0.03
Moisture	9.15±0.27	7.81±0.19	8.71±0.11
Ash	7.11±0.02	7.41±0.01	7.60±0.22
Crude fibre	0.71±0.03	1.40±0.03	0.02±0.01
Nitrogen-free extract	55.97	52.51	55.02
Gross energy (kcal/g)	4.94±0.02	4.98±0.03	4.50±0.02

Three diets were prepared: one was a reference diet and the other two were experimental diets. The two experimental diets contained 15% each of the SHHP or DDCG. All the three diets contained 30% protein, 10% lipid, and had an energy value of 4.9 kcal/g, in addition to 1% chromic oxide as a marker. The reference diet was formulated and produced in the Nutrition Laboratory of CIIDIR.

Diets were prepared with a single screw laboratory extruder (C.W. Brabender Instruments, Inc., South Hackensack, NJ, USA) with a 19 mm screw-diameter. Extruded pellets were cooled and dried, using ambient conditions (25°C, 65% humidity), packed in plastic bags, and stored at 4°C. Triplicate samples of each diet were used for chemical analysis.

Feeding trial

A single-factor, completely randomized experimental design with six replicates per treatment was used. Eighteen 60-L plastic tanks supplied with filtered and sterilized freshwater were used as experimental units. Each tank was stocked with 6 fish (45.5±2.5 g) obtained from the CIIDIR hatchery. Six replicates per treatment were used. Tilapia were acclimated for one week and fed with the reference diet. Afterwards, experimental diets were offered to apparent satiation twice a day for 54d. Three hours after feeding, faeces were collected from each tank using a Pasteur pipette with a siphon. Feces were washed with distilled water, lyophilized, and frozen at -70 °C for later analysis. Crude protein was estimated by Kjeldahl method (AOAC, 1990) and chromic oxide in the faeces and diets were assayed, following the methods by Bolin *et al.* (1952). Temperature, dissolved oxygen and pH were kept at 21.5±2.5°C, 4.8±1.2 mg/ L, and pH 7-8 during the trial. Gain in weight of tilapia was measured after 26 and 54 days.

Calculation of digestibility and growth rate

The percentage of apparent DM digestibility (ADMD) and apparent protein digestibility of diets (APD) were calculated using the equations of Maynard *et al.* (1981).

ADMD= 100-100 [(%Cr in diet)/ (%Cr in faeces)]

APD=100-100 [(%Cr in diet)/ (%N in diet)] × [(%N in faeces)/ (%Cr in faeces)]

Where, Cr is chromic acid and N is the content of the selected nutrient (protein).

The apparent DM digestibility of the ingredient (IADMD) and apparent digestibility of the ingredient (IAPD) was calculated according to Cho and Slinger (1979).

IADMD= [(100 × ADMD of TD)-((100-% TI) × ADMD of RD)]/ (% TI)

IAPD= [(100 × APD of TD × % CN in TD)-((100-%TI) × APD of RD × % CP in RD)]/(%TI × % CP in TI)

Where, ADMD of TD is the apparent dry matter digestibility of the tested diet, TI is the tested ingredient, and ADMD of RD is the apparent DM digestibility of the reference diet, APD of TD is the apparent nutrient digestibility of the tested diet, CP in TD is the concentration of protein of the reference diet, TI is the tested ingredient, APD of RD is the apparent protein digestibility of the reference diet, CP is the concentration of protein of the reference diet and CP in TI is the concentration of protein in the tested ingredient.

Growth rate (g/d) was estimated as the slope of a simple linear model fitted to growth data for each treatment. The net gain (g) was obtained with the difference between the final and initial weights for each diet.

Statistical analysis

The values of digestibility of diets and ingredients and weight were tested for normality and homogeneity of variance using Statistica 7.0 (StatSoft, Tulsa, OK) software. One-way parametric ANOVA and Tukey's multiple-range test were used to compare mean values of digestibility. Differences in growth rate among treatments were tested by comparing slope estimates using procedures available in GraphPad Prism 5 (GraphPad, San Diego, CAL). Significance was set at $P<0.05$.

RESULTS

Calculations of ADMD, APD, IADMD and IAPD for the diets are presented in Table 3. Mean values of ADMD varied from 80.50 to 86.33%. There were significant differences ($P<0.05$) between the reference and SHSP diet, but no differences were detected between these diets and the DDCG diet. No significant differences were detected in values of APD among the diets, which averaged 95.54%. There were no significant differences ($P>0.05$) in IADMD and IAPD among ingredients, averaging 80.56 and 89.40%, respectively.

The mean initial weight, final weight and net gain (45.5 ± 2.5; 58.77 ± 3.82 and 13.21 ± 3.6g, respectively) were similar ($P>0.05$) among the three diets (Table 4).

Table 3. Apparent digestibility (%) of dry matter and crude protein of ingredients and experimental diets for pre-adult red tilapia.

Dietary groups[†]	Diet		Ingredient	
	ADMD	APD	ADMD	APD
Reference	$80.5^{b}\pm5.9$	$86.4^{a}\pm5.6$	-	-
SHSP	$86.3^{a}\pm0.8$	$87.9^{a}\pm2.3$	$80.2^{a}\pm5.2$	$81.7^{a}\pm4.4$
DDCG	$82.6^{ab}\pm3.8$	$89.4^{a}\pm4.1$	$87.6^{a}\pm5.6$	$91.3^{a}\pm4.0$

[†]Dietary groups include a reference diet or that containing 15% each of shrimp head silage protein hydrolysate (SHSP) and Distillers dried corn grain (DDCG) as ingredients

*Values in a column with different superscripts are significantly different ($P<0.05$).

Table 4. Initial weight, final weight and net gain of red tilapia (*Oreochromis mossambicus*) fed with the experimental diets.

	Reference	SHSP	DDCG
Initial weight	$45.7^{a}\pm2.5$	$45.4^{a}\pm2.58$	$45.5^{a}\pm2.51$
Final weight	$60.8^{a}\pm4.5$	$56.6^{a}\pm3.4$	$58.9^{a}\pm3.5$
Net gain	$15.1^{a}\pm5.6$	$11.2^{a}\pm1.1$	$13.3^{a}\pm4.2$

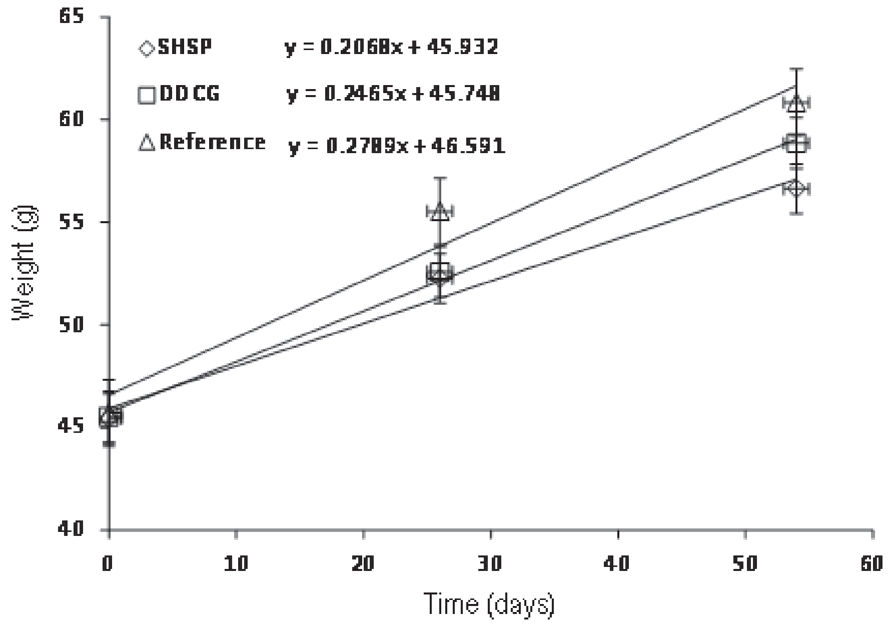

Fig. 1. Growth response of tilapia to diets using shrimp head silage protein hydrolysate (SHSP) and distillers dried corn grain as ingredients (DDCG), and a reference diet. Vertical bars indicate mean standard error.

Growth response of tilapia observed during the feeding trial is shown in Fig. 1. Weight significantly increased during the trial, but no significant differences ($P > 0.05$) were detected in the slopes (i.e. growth rates) estimated for the treatments, ranging from 0.20 g/d to 0.28 g/d.

DISCUSSION

Red tilapia had the capability to effectively digest protein from the SHSP and DDCG ingredients in the diet. These findings agree with other reports for the Mozambique tilapia (Davies *et al.*, 1990; Llanes *et al.*, 2010; Olvera-Novoa *et al.*, 2002; Dechavez and Serrano, 2012). On the other hand, no evidence was detected of negative effects on growth of tilapia when SHSP and DDCG were used.

The ADMD significantly increased when SHSP was included. Plascencia-Jatomea *et al.* (2002), concluded that SHSP is an promising alternative protein source for feeding *O. niloticus*, improving the growth ratio at inclusion levels as high as 15%. In our study, no evidence was detected of negative effects on growth of *O. mossambicus* when SHSP and DDCG were used. This is expected because SHSP contains an adequate concentration of essential amino acids for normal tilapia growth (Guimaraes *et al.*, 2008). While chitin and chitosan have an adverse effect on BW and FCR in *O. niloticus* and *O. aureus* (Shiau and Yu, 1999), this effect is prevented with silage, where chitin is separated out. Additional benefits of using shrimp silage proteins in fish diets are larger final weight of fish and a lower FCR (Fox *et al.*, 1994). There are no studies that compare DDCG with solubles for fish, except the study with rainbow trout by Cheng and Hardy (2003), who reported that the apparent digestibility of essential amino acid in these diets were high and ranged from 87.9 to over 90%.

Protein digestibility has been used as an important evaluation tool in protein use in aquaculture. Akiyama *et al.* (1989) observed that apparent digestibility of protein and DM in shrimp meal *in vivo* and *in vitro* is relatively low, as does Rivas-Vega *et al.* (2010) who reported 72.6 and 73.4% digestibility of DM and protein in *O. mossambicus* × *O. niloticus*. These results differ from our findings. The SHSP diet had high digestibility (86.37%). This can be attributed to the lack of chitin in the ingredient (Pacheco *et al.*, 2011).

Protein digestibility decreases with increases in plant content of diets. However, we did not observe a difference in APD among the diets indicating that digestibility did not decline. Digestibility of 89.37% is the first such report of DDCG for tilapia. The APD, IADMD, and IAPD results for animal-based ingredients were similar to plant-based ingredients. Tilapia is apparently able to assimilate and digest a wide variety of feedstuffs, compared with other freshwater tropical species (Davies *et al.*, 2011). The ADMD that we measured for the reference diet (80.50%) was similar to results in Köprücü *et al.* (2004), and Köprücü and Ozdemir (2005) for several tilapia species.

The effect of extrusion on digestibility of feedstuffs is known (Guimaraes *et al.*, 2008). Extrusion is important because cooking of starches provides stiffness, reduces powder in the feed, and increases pellet stability in water (Botting, 1991). Conditioning temperature (70-90-140°C), screw speed (150 rpm) and feed speed (40 rpm) that we used for preparing diets were adequate to obtain the benefits of the extrusion process. The values of overall digestibility of the SHSP and DDCG diets, together with the fact that growth was not negatively affected, suggest that these ingredients can replace at least a portion of the fishmeal in diets for red tilapia. This should contribute to more precisely formulated high-quality diets that reduce production costs.

It may be concluded that tilapia is able to efficiently consume diets containing SHSP, and DDCG as ingredients. The values of protein digestibility of the experimental diets indicate a positive influence of the fermentation process on these ingredients during the extraction process.

ACKNOWLEDGEMENTS

We thank Arturo Polanco Torres for assistance during the trials and Sonia Rocha and Dolores Rondero for biochemical analyses of the samples. Ira Fogel of CIBNOR provided editing services. This study was funded by Secretaria de Investigación y Posgrado of Instituto Politecnico Nacional (SIP-IPN grant 20080126) and Consejo Estatal de Ciencia y Tecnología de Sinaloa (CECyT grant 2008-2009). H.R.G. is a fellow of EDI and COFAA-IPN. F.J.V.G. is a recipient of a student fellowship (CONACyT grand 232894).

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