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Short Communication

Replacement of moist ingredients in the feed training of carnivorous fish

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ABSTRACT - The study evaluated the replacement of bovine heart by gelatin in the feed training of carnivorous fish, using giant trahira (*Hoplias lacerdae*) as an experimental model. A completely randomized design with four treatments and five repetitions was employed. The treatments were composed of wet ingredients beef heart (control), gelatin diluted in water, gelatin diluted in beef heart broth, and gelatin diluted in water mixed with fish meal. The fish (3.22±0.03 cm and 0.57±0.01 g) were conditioned to accept industrialized diets by the technique of gradual feed ingredients transition in the diet. Gains in weight and length, efficiency of feed training, specific growth rate, cannibalism, mortality and survival rates were evaluated. There was significant difference in weight and length gains and specific growth rate, whereby the use of bovine heart gave the best results. Greater efficiency of feed training was observed for fish fed diets containing beef heart and gelatin diluted in water mixed with fish meal. The high survival rates and the absence of significant differences among treatments for rates of cannibalism, mortality and survival indicate the feasibility of using gelatin as a moist ingredient in the feed training of carnivorous fish.

Key Words: bovine heart, dietary transition, gelatin, gradual feed ingredients transition, palatability

Introduction

Freshwater fish production in Brazil increased from 209,812 tons in 2007 to 337,353 tons in 2009. The main species produced are tilapia (*Oreochromis* sp.), carp (*Cyprinus carpio*) and tambaqui (*Colossoma macropomum*) (MPA, 2010). According to data presented by the Ministry of Fisheries and Aquaculture, carnivorous species probably represent only 4% of the national production. The low production of carnivorous fish can be explained by the fact that most of these species need feed training to accept industrialized diets (Moura et al., 2000; Salaro et al., 2003).

Among the various techniques for the feed training of fish, the gradual feed ingredient transition is the one most frequently used. This technique has shown good results for several carnivore species (Kubitza & Losvhisn 1997; Luz et al., 2000; Moura et al., 2000; Salaro et al., 2003; Soares et al., 2007; Salaro et al., 2011) because of the high survival rates, uniformity of size and low rates of cannibalism (Luz et al., 2002; Soares et al., 2007; Feiden et al., 2008; Salaro et al., 2011). This technique requires specialized labor, however, which can make production more expensive. Another important point to note is the difficulty of making pellets for starter diets because of the moistness of the mixture.

In an attempt to minimize production costs and solve the difficulty of making the diet pellets, gelatin has been mixed with industrialized ration during the feed training of carnivorous fish, but only empirically by fish farmers in the production of fingerling surubins (*Pseudoplatystoma* sp.). Studies are therefore needed to evaluate the potential of gelatin in diets for the feed training of carnivorous fish.

The palatability of the diet is considered one of the critical factors in the success of feed training of carnivorous fish (Soares et al., 2007). The feeding behavior of fish in response to the flavors of different substance classes (amino acids, sugars, organic acids, betaine, nucleic acids, nucleotides, etc.) is species-specific (Kasumyan & Deving, 2003), and thus studies are needed to evaluate such substances in order to stimulate feed consumption during the feed training of different carnivore species.

Therefore, the objective of this study was to evaluate the replacement of beef heart by gelatin (pure or in association with fish meal or beef heart broth) in the feed training of

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carnivorous fish, using giant trahira (*Hoplias lacerdae*) as an experimental model.

Material and Methods

This experiment was conducted at the Laboratório de Nutrição de Peixes, Setor de Piscicultura, Departamento de Biologia Animal, UFV, Viçosa, Minas Gerais, Brazil.

A completely randomized design with four treatments and five repetitions was employed. The treatments consisted of the following wet ingredients: beef heart (control), gelatin diluted in water, gelatin diluted in beef heart broth and gelatin diluted in water mixed with fish meal. Unflavored commercial gelatin was used for the three last-mentioned treatments. In the treatment of gelatin diluted in water, the gelatin was dissolved in water at 50 °C and mixed with ration powder. The diet of gelatin diluted in beef heart broth used 200 g of ground beef heart, which was boiled in 500 mL of water for 30 minutes. The broth was sieved (0.5 mm) and mixed with gelatin and ration powder. The diet of gelatin diluted in water mixed with fish meal consisted of sieved fish meal (0.5 mm) mixed with ration powder and gelatin dissolved in water at 50 °C. All diets were stored in a freezer (-18 °C) and thawed daily to make the pellets (1 mm) just before the feeding of the fish.

The fish were conditioned to accept industrialized diets by the technique of gradual transition of ingredients in accordance with Luz et al. (2002), with modifications to the transition time of the diets, over 22 days (Table 1). The ingredients used in the manufacture of the test diets were analyzed for dry matter, ether extract, ash, according to AOAC (1995), and crude protein, according Detmann et al. (2012). With the data obtained from the analysis of the ingredients, chemical compositions of the different test diets were calculated (Table 2).

Table 2 - Calculated chemical composition (natural matter) of diets used in the feed training of giant trahira juvenile (*Hoplias lacerdae*)

Treatments	Chemical composition (g/kg)				
	Diets	Moist	Crude protein	Ether extract	Ash
Bovine heart	Diet 1	641.12	400.60	30.00	24.00
	Diet 2	520.34	388.20	40.00	38.00
	Diet 3	399.56	375.80	50.00	52.00
	Diet 4	278.78	363.40	60.00	66.00
	Diet 5	158.00	351.00	70.00	80.00
Gelatin dissolved in water	Diet 1	508.00	317.00	23.33	28.89
	Diet 2	454.79	321.26	30.57	36.80
	Diet 3	383.86	328.46	39.98	47.11
	Diet 4	289.24	338.25	52.50	60.83
	Diet 5	158.00	351.00	70.00	80.00
Gelatin dissolved in broth of bovine heart	Diet 1	501.68	339.22	25.56	37.78
	Diet 2	449.45	340.09	32.45	44.33
	Diet 3	379.79	342.79	41.41	52.84
	Diet 4	286.88	346.58	53.33	64.17
	Diet 5	158.00	351.00	70.00	80.00
Gelatin dissolved in water mixed with fish meal	Diet 1	500.79	332.73	18.00	57.42
	Diet 2	447.74	336.67	25.35	64.74
	Diet 3	374.62	348.65	33.13	83.73
	Diet 4	285.19	347.10	49.50	76.88
	Diet 5	158.00	351.00	70.00	80.00

Juvenile giant trahira of 3.22±0.03 cm standard length and 0.57±0.01 g in weight were stocked (7.2 animals L^{-1}) in 20 aquariums (35 × 30 × 14 cm) with five liters of water, constant aeration and temperature controlled (27 °C) by a heater and thermostat. Saline water was used throughout the experimental period at the concentration of 4 g of common salt L^{-1} (Cisne®, Brazilian Industry - 390 mg common sodium/g salt, 25 mg of iodine/g of salt) (Salaro et al., 2011). The aquariums for all treatments were covered by a screen of 2 mm in diameter to prevent the escape of fish.

Before the first feeding of the day, the water temperature (27±1 °C) was measured by an alcohol thermometer. Every

Table 1 - Quantity of ingredients and number of days of the diets used in the feed training of giant trahira (*Hoplias lacerdae*)

Diets		Diet 1	Diet 2	Diet 3	Diet 4	Diet 5
Time (days)	Ingredients (g/kg)	6	4	4	4	4
Treatments						
Bovine heart	Commercial ration	200.00	400.00	600.00	800.00	1000.00
	Bovine heart	800.00	600.00	400.00	200.00	0.00
Gelatin dissolved in water	Commercial ration	333.33	436.66	571.11	750.00	1000.00
	Gelatin	222.22	186.66	142.22	83.33	0.00
	Water	444.44	376.66	286.66	166.66	0.00
Gelatin dissolved in broth of bovine heart	Commercial ration	333.33	436.66	571.11	750.00	1000.00
	Gelatin	222.22	186.66	142.22	83.33	0.00
	Broth of bovine heart	444.44	376.66	286.66	166.66	0.00
Gelatin dissolved in water mixed with fish meal	Commercial ration	66.66	175.55	228.88	600.00	1000.00
	Gelatin	222.22	186.66	142.22	83.33	0.00
	Water	444.44	376.66	286.66	166.66	0.00
	Fish meal	266.66	261.11	342.22	150.00	0.00

week, the pH was measured with the aid of a portable potentiometer, oxygen dissolved with a portable oximeter and ammonia measured through a commercial kit, maintained at 6.8±0.3, 6.0±0.5 mg L^{-1} and 0.2±0.0 mg L^{-1}, respectively.

Fish were fed three times a day (06h00, 12h00 and 18h00) to satiety, until the time was not observed over the attack of pellets by the fish. During the feeding of the fish, the aquariums were checked for cannibalism, according to Salaro et al. (2011), and dead fish. Every day, at 18h30, 100% of the water volume was exchanged to maintain water quality. During this procedure, which lasted about 30 seconds, the juveniles were caught by hand net and immediately transferred to another clean aquarium in the same experimental conditions (Salaro et al., 2011).

At the end of the experiment, all fish were counted, weighed on a precision scale (model MB45 Toledo® 0.01 g) and their standard length was measured with a digital caliper (model series 500 Absolute Coolant Proof® of 0.0001 m) for the calculation of weight (WG) and length (LG) gains, feed training efficiency (FTE), specific growth rate (SGR) and cannibalism, mortality and survival rates. The cannibalism rate (CR) was calculated by the formula: CR = 100 - (SR + MR) in which SR = survival rate and MR = mortality rate. The feed training efficiency was calculated as follows: NCF/NLF * 100, in which NCF = number of conditioned fish (fish with body weight greater than the initial average weight) and NLF = number of live fish.

The effect of the wet ingredients during the feed training of juvenile giant trahira on the parameters of feed training efficiency, weight and length gains and specific growth rate was evaluated by analysis of variance. In the event of a significant F test, the Tukey test was performed at 5% significance level. The effect of the wet ingredients during feed training of juvenile giant trahira in terms of cannibalism, mortality and survival rates was assessed by the non-parametric Kruskal-Wallis test at 5% significance level.

Results and Discussion

The commercial gelatin associated with fish meal provided higher feed training efficiency, with a value equivalent to that observed for fish in the control group (beef heart). The efficiency of feed training of the fish in treatments gelatin diluted in water and mixed with ration powder and gelatin diluted in beef heart broth was lower ($P<0.01$) than in the other treatments, although the values were satisfactory for carnivorous fish (Table 3). These results indicate the feasibility of using gelatin as a moist ingredient in the feed training of carnivorous fish. The diet containing beef heart provided better performance ($P<0.01$) than diets containing gelatin as moist ingredient. Fish meal added to the diet results in better growth performance than gelatin diluted in water and diluted beef heart broth. There were no significant differences among the wet ingredients with regard to cannibalism, mortality and survival rates (Table 3).

The better productive performance of animals conditioned by the technique of gradual transition using wet ingredients like beef heart (control) can be explained by the better palatability of the diet, which led to rapid acceptance of feed by fish and therefore to its faster intake. The addition of fish meal probably also provided improved flavor. In both treatments, immediate acceptance of the diets was observed on the first day of training, when the animals caught and ate the pellets as soon as they were offered, indicating the attractiveness and palatability of beef heart and fish meal. The immediate acceptance of feed by fingerling and juvenile giant trahira during feed training with beef heart as a moist ingredient was also observed by Luz et al. (2002) and Salaro et al. (2011).

Similar results from stimulating consumption of dry diets were observed when freeze-dried krill meal was used in diets for fingerlings of largemouth bass (*Micropterus salmoides*) during feed training (Kubitza & Lovshin, 1997). Other ingredients such as beef heart, eviscerated sardines, and tilapia fillets as moist ingredients in diets for feed training of black bass (*Micropterus salmoides*) also resulted in greater acceptance of the diets by the fish (Feiden et al., 2008).

The good results for feed training efficiency, survival, cannibalism and mortality rates (Tables 3 and 4) for fish fed gelatin as moist ingredient support the hypothesis that beef heart can be replaced by gelatin during the feed training of carnivorous fish. The high efficiency of feed training of fish fed gelatin diluted in water with fish meal indicates that fish meal has good palatability, improving the diet acceptance by giant trahira. Therefore, the use of flavors improves the acceptance of gelatin as moist ingredient during the feed training process of fish. Further studies are needed, however, to evaluate the addition of various flavors to commercial gelatin during feed training of other carnivorous fish, since the palatability of the diet is species-specific (Kasumyan & Deving, 2003).

Feiden et al. (2008) obtained survival rates of 80 to 90% during the feed training of fish, which indicates the success of the technique, since the animals become able to accept industrialized diets. Survival rate around 96.66% was found by Luz et al. (2002), during the feed training of fingerlings of giant trahira using the technique of gradual

Table 3 - Mean values of the feed training efficiency and growth performance of juvenile giant trahira (*Hoplias lacerdae*) subjected to feed training

Variables	Treatments				P value	CV (%)
	Bovine heart	Gelatin dissolved in water	Gelatin dissolved in beef heart broth	Gelatin dissolved in water mixed with fish meal		
Efficiency of feed training (%)	97.75A	69.96B	68.49B	92.76A	0.0001	15.26
Weight gain (g)	0.46A	0.06C	0.04C	0.17B	0.0001	29.32
Length gain (cm)	0.60A	0.21C	0.19C	0.37B	0.0001	25.23
Specific growth rate (%/day)	2.65A	0.43C	0.33C	1.18B	0.0001	29.17

Means in the same row followed by different letters differ by the Tukey test (P <0.05).
CV - coefficient of variation.

Table 4 - Average rates of cannibalism, mortality and survival of juvenile giant trahira (*Hoplias lacerdae*) subjected to feed training

Variables	Treatments				P value	CV (%)
	Beef heart	Gelatin dissolved in water	Gelatin dissolved in beef heart broth	Gelatin dissolved in water mixed with fish meal		
Canibalism rate (%)ns	0.00	2.22	1.11	0.00	0.0729	166.74
Mortality rate (%)ns	5.01	5.01	2.78	1.67	0.3971	106.59
Survival rate (%)ns	94.99	92.77	96.11	98.33	0.3963	5.16

Means in the same row followed by different letters differ by the Tukey test (P<0.05).
ns - not significant; CV - coefficient of variation.

transition of the ingredient, with beef heart as moist ingredient. Good survival rates were also observed during feed training for other carnivorous fish species such as black bass (*Micropterus salmoides*) (Feiden et al., 2008), peacock bass (*Cichla* sp.) (Soares et al., 2007), pintado (*Pseudoplatystoma corruscans*) (Campos, 2010) and arapaima (*Arapaima gigas*) (Pereira-Filho & Roubach, 2010).

The high humidity (Table 1 and 2) and viscosity of beef heart hinder the production of pellets for the diets for feed training of carnivorous fish. Therefore, the possibility of using gelatin as moist ingredient during this phase may facilitate the production of pellets, reducing labor during the production phase. The low growth rates of fish fed gelatin, however, indicate the necessity to evaluate new product flavors for addition to the gelatin.

Conclusions

The replacement of beef heart with gelatin as a moist ingredient is viable for the feed training of carnivorous fish. The addition of fish meal to the gelatin improves the feed training efficiency and growth of juvenile giant trahira.

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