academicJournals

Vol. 5(7), pp. 172-176, July, 2013
DOI: 10.5897/IJFA11.055
ISSN 1991-637X ©2013 Academic Journals
http://www.academicjournals.org/IJFA

International Journal of Fisheries and Aquaculture

Full Length Research Paper

Inclusion effect of graded levels of molases in the diet of *Clarias gariepinus* juvenile

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Accepted 22 March, 2013

The effect of molasses as dietary energy replacement for indomie waste in the diet of African catfish was tested on 120 juveniles for 56 days. The fish were evenly distributed over twelve experimental tanks (52.5 × 33.5 × 21 cm); feeding was done by hand to satiation and the water changed every other day to maintain good water quality. Four experimental diets were formulated; the control, without the test ingredient and the other three diets (diets 2, 3 and 4) contained 1.0, 1.5 and 2.0% molasses inclusion respectively. The mean weight gain (MWG) significantly increased (P<0.05) from 1 to 1.5% before declining at 2% inclusion of test ingredient. Diet 3 recorded the highest MWG (117.47±6.17 g), followed by the control diet (106.75±6.49 g). Similar pattern was recorded for the specific growth ratio. There was no significant difference (P>0.05) in the feed conversion ratio. The packed cell volume and the hemoglobin significantly increased (P<0.05) from 1 to 2% inclusion, while the carcass proximate composition for dry matter, crude protein and ether extract did not record any significant difference (P>0.05). The inclusion of molasses in African catfish diet up to the level tested would not have any negative effect on growth and health of fish.

Key words: Molasses, African catfish, molasses replacement, catfish diet.

INTRODUCTION

Global consumption of fish and fishery products have consistently increased over the years and this trend is expected to continue with most of this increase expected from the developing countries (De Silva, 2001). Such growth in the aquaculture industry is likely to have great effect on the demand for feed and raw materials. The gap between local supply and demand for these traditional feed ingredients is expected to widen over the coming decades, providing a compelling reason for exploring the usefulness of locally available, alternative feedstuffs in feed formulation. As a result of this, there has been keen interest in evaluating alternative feed resources like molasses in the present study.

Molasses is a term applied to a variety of by-product feeds derived from sugar-rich crop. Molasses can be produced from citrus, wood sugar, sugar beet and sugar cane (Perez, 1983). The use of molasses in livestock and poultry feeds dates back into the nineteenth century and has been a major subject of several reviewed articles (Scott, 1953; Cleasby, 1963; Van Niekers, 1980; Waldroup, 1981). The extent to which molasses has been used in animal feeds varies from a small amount used to eliminate dust and feed wastage, to serving as the major source of dietary energy. Curtin (1983) also reported that it could be used to increase palatability of many types of rations and source of energy in the form of simple sugars.

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Table 1. Proximate composition of Molasses and Noodle waste.

Parameter	Molasses	Noodle waste
Dry matter	74	92.3
Crude protein	4.2	12.94
Nitrogen free extract	87.2	65.00
Ether extract	0.0	18.05
Crude fibre	0.0	1.25
Ash	7.8	6.5
Calcium	0.55	1.05
Phosphorous	0.10	4.11

Sugarcane molasses contains nutritionally significant levels of calories, the essential minerals, potassium, calcium, magnesium, sulfur, chloride, and iron, and the B vitamin biotin (Leo, 1983). The low levels of protein and very low levels of phosphorous, fiber, fat soluble vitamins, and most trace minerals gives it an "environmentally friendly" feed ingredients characteristic. The major contributors of digestible energy in most feedstuffs are digestible carbohydrate (including sugars), while the major contributors of digestible energy in molasses are the sugars (glucose, fructose and sucrose). These necessitate the trial inclusion of molasses as dietary energy source in the diet of juvenile African catfish in the present study. Attention will be focused on the effects of molasses on growth, blood parameters, carcass composition and economic indices.

MATERIALS AND METHODS

Experimental procedure

The experiment was carried out at the Nutrition Unit of the Department of Marine Sciences, University of Lagos, Akoka, Nigeria and it lasted for 56 days.

A total of 120 African catfish (*Clarias gariepinus*) juveniles were used for the experiment, spread into ten fish per experimental tank (52.5 × 33.5 × 21 cm) and were allowed to acclimatize for two weeks during which the fish were fed with Coppens (2 to 3 mm, a commercial feed).

Throughout the experimental period, water was changed with borehole every other day to maintain good water quality, temperature (27.5 to 29.5°C) was recorded with a thermometer, dissolved oxygen (4.5 to 4.8 mgL^{-1}) was determined by Wrinkler's method and pH (7.3 to 8.0) was determined with a pH meter (Hannah E251).

Procurement and processing of feed ingredients

The feed ingredients (fish meal, soybean meal, groundnut cake, noodle waste, fish premix, di-calcium phosphate, vitamin C and salt) including molasses were bought from Soleace Enterprise at Oko-Oba, Agege, Lagos. The ingredients were milled, accurately measured and thoroughly mixed with hot water to form homogenized dough; each diet was pelletized to 2 mm size using a locally made hand pelletizer.

Experimental diets and feeding

The proximate composition of molasses and noodle waste are recorded in Table 1. Four isonitrogenous diets were formulated (Table 2). The control diet was without the test ingredient. The molasses was included in the diet at graded levels; 1.0, 1.5 and 2.0%. The juvenile fish were fed ad libitum by hand twice daily at 10.00 and 17.00 h throughout the experimental period. The feed intake and the weight gained were recorded every week for the duration of the experiment.

Laboratory procedures

Haematological analysis

Blood sample of fish taken at random from each tank were collected in both syringe and heparinized bottles for haematological assay and taken to Bioassay Diagnostic Laboratory Cele-Egbe, Ikotun-Lagos. Haemoglobin (Hb), red blood cells (RBC), white blood cells (WBC) and packed cell volume (PCV) were analyzed using the methods described by Svobodova et al. (1991). Cholesterol and Triglycerides was also determined using enzymatic colometric test. Mean Corpuscular Haemoglobin Concentration (MCHC), Mean Corpuscular Haemoglobin (MCH) and Mean Corpuscular Volume (MCV) were calculated according to the formulae given by Dacie and Lewis (2001).

To calculate the MCHC, expressed as gram of haemoglobin per 100 ml packed cell, the following was used:

$$\text{MCHC} = \frac{\text{Haemoglobin (g/100ml)} \times 100}{\text{Haemocrit (\%)}}$$

To calculate the MCV, expressed in femtolitres (fl or 10^{-15}L), the following was used:

$$\text{MCV} = \frac{\text{Haemocrit (\%)} \times 10}{\text{RBC count (millions/L blood)}}$$

To calculate the MCH, expressed in pictograms (pg), the following was used:

$$\text{MCH} = \frac{\text{Haemoglobin (g/100ml)} \times 10}{\text{RBC count (millions/L blood)}}$$

Histometry analysis

Fish were randomly picked from each tank and dissected using a dissecting kit, each organ were carefully harvested and weighed using an electronic digital scale (KERN770, max.220g, d = 0.0001g). The percentage relative organ weights were calculated as:

$$\text{\% Relative organ weight} = \frac{\text{Organ weight}}{\text{Fish weight}} \times 100$$

Carcass analysis

The proximate composition of fish carcass was taken from each treatment tank after the experiment was analyzed at the Department of Animal Sciences, University of Ibadan. The crude protein, ether extract, ash and dry matter were determined

according to the methodology of AOAC (2004).

Growth, economic and nutrient utilization parameters

Growth was estimated in terms of mean weight, relative weight and specific growth rate (Bagenal, 1978):

Mean weight gain (MWG) = Mean final body weight (g) - Mean initial body weight (g)

$$\text{Relative weight gain (RWG)} = \frac{\text{Average weight gain (g)}}{\text{Number of days (day)}}$$

$$\text{Specific Growth Rate (SGR)} = \frac{(\text{Log}_e W_1\,(g) - \text{Log}_e W_2\,(g) \times 100}{T_2 - T_1\,(\text{day})}$$

Where, e = natural logarithm, $T_2 - T_1$ = experimental period, W_1 = initial weight, and W_2 = final weight.
Nutrient utilization indices were expressed as follows;

$$\text{Feed conversion ratio (FCR)} = \frac{\text{Feed eaten in dry mass (g)}}{\text{Weight gain (g)}}$$

$$\text{Protein efficiency ratio (PER)} = \frac{\text{Mean weight gain (g)}}{\text{Protein intake (g)}}$$

$$\text{Where protein intake} = \frac{\text{Total feed intake}}{\text{Protein content of feed}}$$

Economic evaluation was analyzed in terms of Incidence cost (IC). It was calculated according to Mazid et al. (1997) as follows:

$$\text{Incidence of Cost (IC)} = \frac{\text{Cost of fish (₦)}}{\text{kilogramme of fish produced}}$$

The cost was based on current prices of feed ingredient in the experimental locality as at the time of experiment.

Statistical analysis

The experimental design was complete randomized design and all data collected were subjected to analysis of variance (ANOVA). Comparisons among diets means were carried out by Duncan multiple range test (Duncan, 1955) at significant level of 0.05. All computations were performed using Statistical package SPSS15.0 (SPSS Inc., Chicago, IL, USA).

RESULTS

The mean weight gain (MWG) significantly increased from 1 to 1.5% inclusion of the test ingredients (molasses) before declining at 2% inclusion. The highest MWG (117.47±6.17 g) was recorded at 1.5% inclusion and followed by the control (106.75±6.49 g) while fish on diet with 1% inclusion recorded the least (95.45±4.95 g). Similar pattern was observed in the average feed intake, which were also significant difference ($P<0.05$) across diets. Fish on diet with 1.5% inclusion recorded highest feed intake (100.09±7.12 g) relative to any other group, while fish on 1% consumed the least (81.75±2.33 g). The highest specific growth rate (1.68±0.03%day^{-1}) was recorded by fish on diet with 1.5%, followed by the control (1.62±0.04% day^{-1}), while there was no significant difference ($P>0.05$) between the fish fed diets containing 1 and 2% molasses inclusion. The nutrient utilization parameters (feed conversion ration and protein efficiency ratio) did not show any significant difference ($P>0.05$). Similar result pattern was recorded in the economic parameters studied.

All the haematological parameters tested for showed significant difference ($P<0.05$) across the diets. The packed cell volume and hemoglobin increased from 1 to 2% molasses inclusion, though, no significant difference ($P>0.05$) between the control and the highest inclusion level of 2%. There was also no significant difference ($P>0.05$) in the white blood cell count between the control and 2% inclusion, also no significant difference ($P>0.05$) between 1 and 1.5% inclusion. Similar pattern was recorded in triglycerides. The carcass proximate composition for dry matter, crude protein and ether extract did not record any significant difference ($P>0.05$).

DISCUSSION

The major source of metabolizable energy in most compounded diets of fish and livestock is maize, but it has become eminent that other energy sources should also be used (Aderolu et al., 2009). The increase in the average feed intake, mean weight gain and specific growth rate of fish fed molasses indicated that the diets were adequate to support *Clarias gariepinus* growth. However, the fish perform better at 1.5% inclusion of molasses (Table 3). The highest mean weight gain observed at this point may be due to higher feed intake compared to other levels of inclusion. This suggests a better palatability at 1.5% inclusion than at any other graded levels tested for. According to Glencross et al. (2007), feed intake is a measure of palatability of test ingredient or test diet. The effective utilization of nutrient in the diet up to 1.5% inclusion may be due to the fact that the test ingredient contained no fibre, Aderolu et al. (2009) stated that, excess fibre in fish diet prevent or reduced nutrient utilization along the gastro-intestinal tract (GIT) due to the accumulation of fibre on GIT wall. Curtin (1983) reported that, beside molasses been an energy source, the palatability of molasses makes it an excellent carrier for other feeds especially unpalatable feedstuffs like blood meal. Haematological parameters have been studied with the aim of diagnosing health status of fish before any physical manifestation of diseased condition (Table 4). Some of the factors that have been quoted to affecting haematological indices include food composition (Secombes, 1994), stress

Table 2. Composition of experimental diets.

Ingredient	Control	1% Molasses Inclusion	1.5% Molasses Inclusion	2% Molasses Inclusion
Groundnut cake	15.13	15.13	15.13	15.13
Soybean meal	45.97	45.97	45.97	45.97
Fish meal	27.00	27.00	27.00	27.00
Noodle waste	10.00	9.00	8.50	8.00
Molasses	-	1.00	1.50	2.00
Dicalcium phosphate	1.00	1.00	1.00	1.00
Vitamins C	0.20	0.20	0.20	0.20
Premix	0.50	0.50	0.50	0.50
Salt	0.20	0.20	0.20	0.20
Total	100	100	100	100
Crude protein (%)	46.75	46.63	46.57	46.51
Crude fibre (%)	4.22	4.20	4.19	4.18
Energy (ME/K)	2756.03	2800.40	2822.69	2844.91

Table 3. Growth, nutrient and economic parameters of juvenile African catfish fed graded levels of molasses.

Parameter	Diets			
	Control	1.0% Molasses	1.5% Molasses	2.0% Molasses
Average initial weight(g)	15.08±0.14	15.25±0.00	15.17±0.29	15.25±0.00
Average final weight (g)	$121.85±6.45^{ab}$	$110.70±4.95^{b}$	$132.63±6.39^{a}$	$115.52±9.98^{b}$
Average feed intake (g)	$92.88±3.69^{ab}$	$81.75±2.33^{c}$	$100.09±7.12^{a}$	$91.44±1.06^{b}$
Mean weight gain (g)	$106.75±6.49^{ab}$	$95.45±4.95^{b}$	$117.47±6.18^{a}$	$100.27±9.99^{b}$
Specific growth rate (%day^{-1})	$1.62±0.04^{ab}$	$1.54±0.03^{b}$	$1.68±0.03^{a}$	$1.57±0.06^{b}$
Feed conversion ratio	0.87±0.08	0.86±0.02	0.85±0.04	0.92±0.10
Protein efficiency ratio	2.45±0.23	2.48±0.07	2.50±0.13	2.33±0.25
Incidence cost (₦/kg)	0.16±0.01	0.15±0.00	0.15±0.07	0.17±0.02

Table 4. Dietary effects of molasses on selected haematological parameters in juvenile African catfish.

Parameter	Diets			
	Control	1.0% Molasses	1.5% Molasses	2.0% Molasses
Packed cell volume (%)	$36.00±3.00^{a}$	$29.50±3.50^{b}$	$34.50±4.50^{ab}$	$37.50±0.50^{a}$
Haemoglobin (g/dl)	$11.65±0.95^{ab}$	$9.65±1.05^{b}$	$11.27±1.46^{ab}$	$12.00±0.36^{a}$
White blood count (g/dl)	$9000±1000^{b}$	$11500±500^{a}$	$12500±1500^{a}$	$9000±1000^{b}$
Cholesterol (g/dl)	$135±5.00^{c}$	$190±0.50^{ab}$	$165±15.00^{b}$	$215±25.00^{a}$
Triglycerides	$211.50±15.50^{b}$	$222.00±26.00^{ab}$	$219.00±19.00^{ab}$	$248.00±0.50^{a}$

(Chen et al., 2004) and toxins (Worle et al., 2007). According to Munkittrick and Leatherland (1983), haematocrit reading is valuable in determining the effect of stressors on the health of fish. The significant increase observed with increased test ingredient showed that the fish were not stressed from the test ingredient inclusion. Haemoglobin is a sophisticated oxygen delivery system that provides the desired amount of oxygen to the tissues under a wide variety of circumstances (Voet and Voet, 1990). According to Blaxhall and Daisley (1973), the determination of haemoglobin can be a good indicator of anaemic conditions in fish, with the recorded values in the present study; it showed that the fish did not suffer from any form of anaemia. The increase observed in the two parameters (packed cell volume and the haemoglobin) may be due to high activity in the fish, according to Weiss and Wardrop (2010); fish with high activity will need more oxygen invariably having the need for increased PCV and haemolobin. The values obtained in this experiment for both the PCV and Hb were within the normal ranges recommended for *C. gariepinus* by Adedeji (2009) who reported PCV values 37.25±2.12 and

Table 5. Carcass composition of juvenile African catfish fed molasses at graded levels.

Parameter	Diets			
	Control	1.0% Molasses	1.5% Molasses	2.0% Molasses
Dry matter (%)	26.33±1.08	25.37±0.62	25.51±0.55	25.47±0.76
Crude protein (%)	75.04±1.07	69.27±0.64	71.96±1.74	69.91±0.12
Ether extract (%)	24.00±1.00	23.33±0.58	22.67±2.08	24.25±0.35

31.17±0.42 and Hb values 10.10±0.214 and 8.40±3.94 for wild and cultured fish, respectively. The fact that molasses may support hematopoiesis in fish cannot be ruled out. The addition of molasses in the diet did not really have effect on carcass proximate composition (Table 5). From these, it could be concluded that the inclusion of molasses in the diet of juvenile African catfish would not have any negative effect on fish growth and health and likewise the health of the consumers.

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