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APPARENT NUTRIENT DIGESTIBILITY COEFFICIENT OF BREWER'S SPENT GRAIN MEAL BASED DIETS IN CLARIAS GARIEPINUS JUVENILES

Ogechi N. Ugbor[1], Stephen N. Ochang[2], JohnPaul Chinedu Ikwuemesi[1], Okey Alum-Udensi[1] and Chidinma A. Agbakwuo[3]

[1]Department of Fisheries and Aquatic Resources Management, Michael Okpara University of Agriculture Umudike, Nigeria.
[2]Department of Fisheries and Science, Cross River University of Technology, Calabar, Nigeria
[3]Fisheries and Hydrobiology Research Unit, Department of Zoology and Environmental Biology, University of Nigeria Nsukka, Enugu State, Nigeria

ABSTRACT: *The response of catfish fed brewer's spent grain (BSG) based diets was evaluated in the diet of Clarias gariepinus using growth performance, nutrient utilization and apparent digestibility coefficient as indices. Seven iso-nitrogenous diets were formulated with BSG replacing 0,10, 20, 30, 40, 50 and 100% of corn in the diets. A relatively good growth performance and nutrient utilization by Clarias gariepinus fed the test diets were observed. However, the results showed significant difference (p<0.05) in weight gain, SGR, FCR and RGR between treatments, with 0%BSG (control) performing best. There was significant (p<0.05) reduction in apparent digestibility of nutrients with increasing levels of BSG meal in the test diets. The apparent digestibility coefficient for organic matter, protein, lipid, fiber and NFE of the control diet (0% BSG) were highest when compared to the rest of the test diets. However, the digestibility values for protein and lipid in diet A (0% BSG) and diet B (10% BSG) were statistically comparable.*

KEYWORDS: Apparent digestibility, Brewer's spent grain, Growth response, Nutrient utilization and *C. gariepinus.*

INTRODUCTION

Fish and fishery products have been found to be very valuable source of protein and essential micronutrients for balanced nutrition and good health (Reverter *et al*., 2014). Anusiriuka (2002) observed that fish is one of the cheapest and promising sources of animal protein and people can easily digest 93.2% and 93.7% of fish protein and fat, respectively. In 2009, fish accounted for 17% of the world population animal protein intake and 6.5% of all protein consumed (FAO, 2012). Fish farming has been shown to be the main source of fish supply since capture fisheries has been on the decline as a result of pollution and over fishing from the wild (Mgbenka, 2014). However, the soaring costs of conventional fish feed ingredients is a major cause of increase in the production cost and reduction in the profit margin of commercial fish farmers (Eyo and Ezechie, 2004). Mgbenka (2014) reported that between 40 – 60 % of the variable costs is expended on feeding in an intensive fish culture system.

Several attempts have been made by researchers to increase the utilization of nonconventional plant and animal materials to replace conventional feed ingredients like corn and fish meal in fish feed ration (Baruah *et al*., 2003; Eyo, 2004; Eyo and Ezechie, 2004; Abu *et al*., 2010; Jimoh *et al*., 2014). Olurin *et al.* (2006) reported that the digestibility of corn has

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made it the major source of metabolizable energy in most compounded diets for catfish species.

Corn, although widely cultivated has been extensively utilized as human food, feed for livestock and raw materials for industries (Eyo and Ezechie, 2004). In Nigeria, the demand for corn is in excess of production, and the utilization of corn as major basal ingredients in the aquaculture industries will place more constraint on an already over exploited resource. There is therefore, the need to exploit cheaper energy sources to replace expensive corn in fish feed formulation.

Brewery Spent Grains (BSG), a by-product of the brewery industries, constitutes about 85% of the total by-product of the brewing industry (Mussatto *et al*., 2006). Nutritionally, Ozturk *et al*. (2002) reported that the grain has a range of 26 – 30% crude protein level and 13% crude fiber. Spent grain utilization in Nigeria has little attention as a marketable commodity except for its primary use as feeds for ruminant and biomass. It often constitutes a waste and dumping has become the major means of its disposal in developing countries like Nigeria. This is not sustainable because of the different levels of environmental pollution associated with the disposal method (Ajanaku *et al*., 2010). Hence, its successful incorporation in fish feed will not only reduce the cost of fish production but will also serve as a means of waste management.

Ighawela *et al*. (2014) noted that information of nutrient digestibility of the several feed ingredients used in formulating fish feeds is necessary. Köprücü and Özdemir (2005) reported that a feed ingredient may appear from its chemical composition to be an excellent source of nutrients but will be of little real value unless it can be digested and absorbed by the target species. Thus, information of nutrient digestibility of the several feed ingredients used in formulating fish feeds is necessary. Therefore, the objective of this study was to evaluate the effect of different dietary levels of BSG on the nutrient digestibility and growth response of *C. gariepinus* juveniles.

MATERIALS AND METHODS

Source and processing of ingredients

Ingredients used in the feeding trial: fishmeal, soybean meal, corn, vitamin/mineral premix, blood meal and palm oil were bought from Ogige market, Nsukka, Enugu State, Nigeria. These ingredients were separately milled, screened with fine mesh net to fine particle size (<250 μm). BSG was sourced at Nigeria Brewery Plc., 9th Mile Corner, Enugu, Nigeria. The spent grain was sun dried for three days until the moisture content were reduced to 10%.

Based on the nutrient composition of the feedstuffs (Table 1), seven isonitrogenous experimental diets were formulated. The experimental diets were formulated to produce diets in which 0% (BSG0), 20% (BSG20), 30% (BSG30), 40% (BSG40), 50% (BSG50%) and 100% (BSG100) of carbohydrate from corn were replaced with that from BSG (Table 2). The feedstuffs were finely grounded and mixed in plastic bowl. With the addition of boiling water, they formed dough. The mixture was then pelleted by passing it through a mincer of 2mm die to produce 2mm diameter size of pellets. The feed was sundried to about 10% moisture content, packed in polythene bags and kept safe in a dry place until used.

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Table 1: Percentage composition of experimental diet fed to *Clarias gariepinus*.

Feedstuffs	0%BSG	10%BSG	20%BSG	30%BSG	40%BSG	50%BSG	100%BSG
Soybean meal	23.44	23.44	23.44	23.44	23.44	23.44	23.44
Blood meal	23.44	23.44	23.44	23.44	23.44	23.44	23.44
Fish meal	23.44	23.44	23.44	23.44	23.44	23.44	23.44
BSG	0.00	2.67	5.34	8.04	10.67	13.34	26.70
Corn	26.70	24.01	21.34	18.62	16.01	13.34	0.00
Vit/min	2.00	2.00	2.00	2.00	2.00	2.00	2.00
Palm oil	1.00	1.00	1.00	1.00	1.00	1.00	1.00

Culture condition

A total number of 420 juveniles of *C. gariepinus* of 6.0 ± 0.83g mean initial weight were procured from East Goshen Farms in Ohanze, Abia State, Nigeria and were transported to Hydrobiology and Fisheries Research Unit, Department of Zoology and Environmental Biology, University of Nigeria Nsukka using an oxygen air bag. They were acclimatized for one week in concrete holding tanks of 7.0 m x 4.0 m x 2.0 m, and with a feed of 40% crude protein.

Fishes were sorted, weighed (using a sensitive weighing balance) and randomly stocked into the experimental tanks at the rate of 20 fish per tank. They were starved overnight before the commencement of the feeding trials (Ayinla, 2007). Fish were offered 5% of their body weight meal per day administered in two equal portions between 8.00 - 9.00 h and 18.00 - 19.00 h. The quantity of feed was adjusted fortnightly based on the weight gain of fish throughout the 20 weeks duration of the feeding trials. The fish were daily monitored for mortality. Dead fish were removed, counted and recorded. Growth response and feed utilization indices were estimated according to Jimoh and Aroyehun (2011).

Acid insoluble ash (AIA) analysis

AIA analyses were carried out on the diets and feces. This was carried out by the addition of 25 ml of 10% HCl to the weighed ash content of a sample. It was covered with a water-glass and boiled gently over a low flame for five minutes. This was then filtered using ashless filters and washed with hot distilled water. The residue from the filter was returned to the crucible and ignited until it was carbon free after which it was weighed. Percentage AIA was calculated as;

$\%AIA = \text{weight of AIA} \div \text{weight of ash} \times 100$

Determination of digestibility coefficient

Digestibility test was carried out as described by Jimoh *et al*. (2013). Feacal sample collection was carried out bi-weekly by siphoning using a pipe (2 cm diameter) three hours after feeding. Uneaten diet was siphoned out 20 min after feeding. Feaces collected were pooled and dried at 60^0C in an electric oven for 24hrs. The proximate analysis of the feedstuffs, diets and faecal matter samples were carried out in three replicates using the procedures described by the AOAC (1990). Digestibility coefficient was calculated on the

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percentage of AIA in feed and in faeces and the percentage of nutrient on diets and feaces as follow:

Digestibility % = 100 – {100 (AIA in feed/AIA in feaces x Nutrient in feaces/Nutrient in feed)}.

Apparent organic matter digestibility (AOMD) was calculated as follow:

AOMD = 100 – {100 (AIA in feed/AA in feaces)}

Statistical analysis

All determinations were conducted in triplicates and the means were subjected to Analysis of Variance, where, the ANOVA revealed a significant difference, Fishers Least Significant Difference (LSD) was used to compare differences among individual treatment means using SPSS version 17.

RESULTS

Table 1 shows the percentage composition of the various ingredients used in the feed formulation of the experimental diets. The results of the proximate composition of the corn meal (CM) and brewers spent grain (BSG) used in this feeding trial revealed that BSG had significantly ($p < 0.05$) higher crude protein (21.26%) and fiber (17.6%) than CM (9.78% cp) and 4.05% fiber. However, CM had statistically ($p < 0.05$) higher crude fat (4.16%) and carbohydrate (66.28%) than BSG.

Table 2: Proximate composition of feed ingredients (%)

Feedstuffs	Protein	Fiber	Ash	Moisture	Fat	NFE
Fishmeal	64	1.45	11.35	14.36	2.23	7.50
Blood meal	78.65	0.45	7.10	0.85	0.48	8.46
Soybean meal	42.31	2.14	1. 05	5.35	30.12	6.49
Corn	9.78[b]	4. 05[b]	4.20[a]	11.30[a]	4.16[a]	66.28[a]
BSG	21.26[a]	17.60[a]	4.87[a]	2.14[b]	1.79[b]	51.39[b]

Mean within the same row with different superscripts are significantly different (P<0.05); SEM = Standard Error of Means

Table 3: Proximate composition of the experimental diet

	Diets							
	A	**B**	**C**	**D**	**E**	**F**	**G**	**SEM**
Crude protein	40.85[a]	40.64[a]	40.59[a]	40.73[a]	40.39[a]	40.81[a]	40.9[a]	0.35
NFE	24.38	25.95	23.23	23.52	23.09	22.66	20.07	1.82
Lipid	3.70	4.20	4.20	4.30	4.12	4.30	4.60	0.73
Ash	5.50	5.62	5.92	5.98	6.37	5.79	5.50	0.11

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Moisture	3.91	1.76	2.71	2.47	1.28	2.46	2.64	0.08
Fiber	4.08	4.15	4.55	4.80	5.25	6.45	6.80	0.86
AIA (%)	2.98	3.13	3.05	3.11	3.24	3.08	3.29	0.66

Mean within the same row with different superscripts are significantly different (P<0.05); SEM = Standard Error of Means

Growth performance and feed utilization of *C. gariepinus* fed BSG based diets

The results for the feed utilization and growth parameters are presented in table 4. Fish fed the control diet gained 552.06 g, while the fishes fed 10% BSG diet gained 518.43 g. Weight gain of 486.40 was recorded in fishes fed diet containing 20% BSG meal. The fishes fed control diet and 10% BSG diet did not show statistical significant (p>0.05) difference but were significantly different (p<0.05) from values obtained in fishes fed diet containing 20%, 30%, 40%, 50% and 100% BSG based diets.

The results for the specific growth rate (SGR) and relative growth rate (RGR) showed that fishes fed with control diet had the highest values of 2.60 and 10,616.54; and lowest values of 2.24 and 5573.20 were recorded in fishes fed with 100% BSG respectively. The SGR and RGR were observed to be decreasing with increase in the amount of BSG meal in the test diets. Significant differences (p<0.05) were observed in the feed conversion ratio (FCR) of fishes fed the control diet and the rest of the test diets.

Table 4: Growth response and nutrient utilization of feeding *C. gariepinus* with BSG based diets for 20 weeks

	Diets							
Parameters	**A**	**B**	**C**	**D**	**E**	**F**	**G**	**SEM**
Wi (g)	5.20^{a}	6.50^{a}	5.60^{a}	5.60^{a}	5.70^{a}	5.40^{a}	5.00^{a}	0.46
Wf (g)	557.26^{a}	524.93^{ab}	492.10^{b}	424.00^{c}	402.70^{c}	353.30^{d}	283.66^{e}	23.77
Wg (g)	552.06^{a}	518.43^{ab}	486.40^{b}	418.40^{c}	397.00^{d}	347.9^{e}	278.66^{f}	19.61
RGR	10616.54^{a}	7975.85^{c}	8687.50^{b}	7471.43^{c}	6964.91^{d}	6442.60^{d}	5573.20^{e}	13.12
SGR	2.60^{a}	2.44^{b}	2.44^{b}	2.40^{b}	2.36^{c}	2.32^{c}	2.24^{d}	0.44
MGR	8.46	8.31	8.22	7.95	7.86	7.64	7.28	0.28
FCR	2.04^{e}	2.45^{d}	2.64^{c}	2.91^{c}	3.37^{b}	3.78^{b}	4.62^{a}	0.77
Survival (%)	96	100	100	97	96	100	100	

Mean within the same row with different superscripts are significantly different (P<0.05); SEM = Standard Error of Means.

Wi = Mean intial weight; Wf = Mean final weight; Wg = Mean weight gain; RGR = Relative growth rate; SGR = Specific growth rate; MGR = Metabolic growth rate; FCR = Food conversion ratio.

Apparent nutrient digestibility coefficient of BSG based diets in *C. gariepinus*

Table 5 revealed the proximate composition of the fecal sample. A general reduction in the nutrients of the feces as compared to the diets showed that some percentages of nutrients were absorbed and made available for metabolism. Significant (p<0.05) differences exist in the crude protein, fiber and Nitrogen Free Extract (NFE) of the fecal samples tested. Diet G

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(100% BSG) had the highest crude protein (22.91%), crude fiber (27.76%) and NFE (5.46%) in the feaces while diet A (0% BSG) had the least values.

Table 5: Proximate composition (%) of faecal samples of *C. gariepinus* fed BSG based diets

	Diets							
Parameters	**A**	**B**	**C**	**D**	**E**	**F**	**G**	**SEM**
Protein	15.21^{d}	15.48^{d}	16.98^{c}	16.72^{c}	18.10^{b}	19.82^{b}	22.91^{a}	1.83
Lipid	1.96	2.10	2.01	2.06	1.98	2.20	2.18	0.04
Fiber	13.60^{e}	15.21^{d}	17.92^{c}	18.34^{c}	20.67^{b}	21.85^{b}	27.76^{a}	2.71
Ash	2.71	2.41	2.21	2.25	2.11	1.89	1.73	0.91
Moisture	9.54	9.67	9.77	9.83	9.45	9.99	9.61	0.32
NFE	49.86^{e}	52.16^{d}	52.88^{d}	53.61^{c}	53.66^{c}	55.18^{b}	57.67^{a}	3.75
AIA	3.70^{c}	3.79^{c}	3.89^{c}	4.08^{c}	4.32^{b}	4.57^{b}	5.46^{a}	0.21

Mean within the same row with different superscripts are significantly different (P<0.05); SEM = Standard Error of Means

Table 6: Apparent digestibility coefficients of BSG based diets fed to *C. gariepinus*

	Diets							
Parameters	**A**	**B**	**C**	**D**	**E**	**F**	**G**	**SEM**
AOMD	81.56^{a}	77.56^{b}	74.17^{b}	73.37^{b}	69.79^{c}	66.90^{c}	59.80^{d}	3.68
APD	92.62^{a}	89.81^{a}	86.72^{b}	82.40^{b}	80.84^{b}	77.72^{c}	68.61^{d}	4.03
ALD	86.65^{a}	85.88^{a}	85.69^{a}	83.81^{b}	80.91^{bc}	78.86^{c}	74.00^{d}	2.92
AFD	51.65^{a}	43.20^{b}	43.21^{b}	41.10^{c}	38.80^{d}	36.30^{e}	30.80^{f}	5.81
ANFED	50.86^{a}	39.67^{b}	37.92^{b}	37.41^{b}	35.66^{c}	34.71^{c}	29.70^{d}	5.17

Mean within the same row with different superscripts are significantly different (P<0.05); SEM = Standard Error of Means; AOMD = Apparent organic matter digestibility; APD = Apparent protein digestibility; ALD = Apparent lipid digestibility; AFD = Apparent fiber digestibility; ANFED = Apparent Nitrogen Free Extract digestibility.

There was significant (p<0.05) reduction in apparent digestibility of nutrients with increasing levels of BSG meal in the test diets. The apparent digestibility coefficient for organic matter, protein, lipid, fiber and NFE of the control diet (0% BSG) were highest when compared to the rest of the test diets. However, the digestibility values for protein and lipid in diets A (0% BSG) and diet B (10% BSG) were statistically comparable (Table 6).

DISCUSSION

Brewer’s spent grain (BSG), a by-product of brewery industry, is high in nutritional value. Previously, BSG had been considered to be an industrial waste that required special treatment. The results of the present study showed that the inclusion level of BSG up to 20 % had no negative effect on growth response as represented by final weight, weight gain, SGR and survival compared to Diet 1. However, FCR was affected by feeding different inclusion levels of BSG. A similar growth response, but poor feed efficiency, in experimental feed containing high BSG can be explained by the less palatability of the diets when the feed contains a high level of BSG (Eyo and Ezechie, 2004). It has also been reported that different

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starch and protein sources affected the physical quality of extruded fish diets which can in turn affect its palatability and acceptability (Øverland *et al*. 2009; Sørensen *et al*., 2010). Another plausible explanation to the growth differentials among the treatment groups may be the imbalance in the digestible energy of the test diets (Eyo, 2004), since energy of the test diets were observed to decrease with increase in the level of BSG in the diets.

Carbohydrates are generally cheaper dietary energy sources than protein and lipids. However, fish species show different ability to digest and metabolize alternative dietary components, in particular the carbohydrate fraction (Dabrowski and Guderley, 2002; Hemre *et al.*, 2002). The digestion and metabolism of feed ingredients is dependent on fish species, source, inclusion level and treatment of the ingredient (Krogdahl *et al.*, 2005).

The bioavailability of nutrients and energy in feedstuffs for catfish may be defined mainly in terms of digestibility or, in the case of energy, metabolizability. Digestibility describes the fraction of the nutrient or energy in the ingested feedstuff that is not excreted in the faeces. Metabolizability describes the fraction of the digested energy that is not excreted in the urine and through the gills. Both digestible energy and metabolizable energy have been used to express feedstuff energy values for fish, but many researchers use and report only digestible energy values because of difficulties in obtaining metabolizable energy values for fish (NRC, 1983).

The values of apparent digestibility of protein and lipid for the control diet and diets containing BSG obtained during the experiment ranges from 68.61 – 92.62 and 74.00 – 86.65 respectively. These results were similar to those obtained by Mukhopadhyay (2001) and Jimoh *et al*. (2014). The Apparent protein digestibility values in various ingredients of plant origin have been reported. Hossain *et al*. (1997) reported 76.2% - 94.0% in rohu , 78.9% - 85.8% in common carp (Mukhopadhyay, 2001), 52.5% - 94.1% in catla (Jafari and Anwar, 1995) and 88.06% - 91.10% in *C. gariepinus* (Jimoh *et al*., 2014). The low carbohydrate digestibility recorded in this study was similar to that reported by Adeparusi and Jimoh (2002) for *Oreochromis niloticus* fed lima bean and Jimoh *et al*. (2014) for *C. gariepinus*. The digestibility of carbohydrate has been known to vary with the complexity of carbohydrate, source treatment and level of inclusion in the diet. A plausible explanation for the lower digestibility coefficient recorded for crude fibre and carbohydrate in the *C. gariepinus* fed for both control and test diets maybe as a result of the physiological requirements of the experimental fish as *C. gariepinus* are reported to be carnivorous (Jimoh *et al.,* 2014). Fagbenro *et al*. (2013) had similar report when they fed *C. gariepinus* with sunflower and sesame seed meal.

The organic matter digestibility coefficient reported in this study was slightly higher than the values reported by Fagbenro *et al*. (2013) and Jimoh *et al.* (2014). The variation may be as a result of many factors includes processing methods, experimental methodology, different test ingredients etc (Krogdahl *et al.*, 2005).

The present results indicate that the nutrient components and energy in the BSG were relatively well digested by the experimental fish (*C. gariepinus*). This is in line with the view that the African catfish should be classified as an omnivorous fish that has the potential to utilize all dietary components, including carbohydrates, more efficiently than many other fish species (Nematipour *et al.*, 1992; Jantrarotai *et al*., 1994). It could therefore be suggested that BSG meal could be used for partial replacement of corn meal in the diets for *C. gariepinus,* as they have a fairly digestible protein and carbohydrate.

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