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Research Article

Effects of *Azanza garckeana* on Egg Quality of African Catfish (*Clarias gariepinus*, Burchell 1822) Broodstck

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Abstract: The effect of *A. garckeana* pulp meal on egg quality of *Clarias gariepinus* broodstocks wasinvestigated. Catfish broodstocks (mean weight 450 g) were randomly distributed into concrete tanks (2×2×1.2 m) at nine fish/tank in triplicates. Five difference (0, 50, 100,150 and 200 g/kg) concentration of *A. garckeana* pulp meal (AGPM) were incorporated into the broodstock diet designated as D1, D2, D3, D4 and D5, respectively. The fish were fed for 56 days. The result shows that fish fed the controlled diet had the highest number of eggs. However, among the fish feed AGMP based diet, those fed 200 g/kg of AGPM had higher (158646.0) number of eggs. D_1 had the least percentage fertilization (78%). D3 had the highest hatching rate (98%), followed by D_2 (96%), D_4 (92%), D_5 (76%) and the least D_1 (62%). Survival of hatchlings after two weeks was higher in D_2 (90%), followed by D_3 (88%), D_4 (84%), D_5 (56%) and D_1 (40%). There was no record of deformity of hatchlings in D_2 to D_4 but in D_1 and D_5. *A. garckeana* pulp meal influenced the fecundity, hatching rate and percentage survival of *C. gariepinus* has a potential pro-fertility property which can be harnessed in fish fingerling production.

Keywords: *Azanza garckeana*, broodstock, *Clarias gariepinus*, egg quality

INTRODUCTION

The world's population is constantly growing, especially in urban areas. Indeed, in 2011, the world had 47.5% of the rural population against 52.6% for urban (FAO, 2014). World aquaculture production continues to grow, albeit at a slowing rate. According to the latest available statistics collected globally by FAO, world aquaculture production attained another all-time high of 90.4 million tonnes (live weight equivalent) in 2012 (US$144. 4 billion), including 66.6 million tonnes of food fish (US$137. 7 billion) and 23.8 million tonnes of aquatic algae (mostly seaweeds, US$6. 4 billion). World food fish aquaculture production more than doubled from 32.4 million tonnes in 2000 to 66.6 million tonnes in 2012. In Nigeria, from the recent report aquaculture accounted for 26% of the total production of Capture and aquaculture.

Fish is the major source of protein consumption and sources of food for human, providing a significant portion of nutrients to a large proportion of the people, particularly in the developing world (Onyia *et al*., 2011). Fish require good quality fish feed in high proportion in order to increase the quality of eggs and sperm fertility, so as to achieve maximum protein intake needed for proper growth (Hassan, 2001).

In fish production under intensive method, attempts are made to obtain quality eggs and sperm, to produce the highest possible number of good quality seeds. Several factors affect fish seed quality includes different strains, genetics, nutrition, contents of the feed, pH, temperature and activities of modern aquaculture, which have introduced several substances such as organic matter, chemicals such as fertilizers and insecticides into the water used as a cultured medium (Canyurt and Akhan, 2008). Common practices in hatcheries such as transportation, handling, cleaning, use of chemicals, overstocking; water quality problems are also factors that may negatively influence reproduction (Adeparusi *et al*., 2010). These common factors affect fertilization success in artificial reproduction commonly used for aquaculture production. As a result of these factors, low quality fish, seeds are produced (Adeparusi *et al*., 2010).

The need for high quality fish seed has necessitated research into various ways of improving egg quality and enhancing fertility of sperm to meet the world protein demand. However the continuing expansion of aquaculture requires shifting from synthetic drugs to natural plant. According to Adedeji *et al*. (2006), wild fruits that have potentials in enhancing egg quality and sperm fertility that are not recognized and valued, are

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now being investigated, evaluated and included into fish feeds with little or no side effects on the fish. Hence the use of wild fruits as a medium to improve egg quality and enhance fertility is now receiving some attention. Dada and Ajilore (2009) used an extract of *Garcinia kola* seed to enhance fertility in *Clarias gariepinus* and Adeparusi *et al*. (2010), used *Kigelia Africana* fruit meal to enhance fertility in male *Clarias gariepinus*. The *Clariids* constitute an excellent food fish of high commercial value. It is very important to the sustainability of the aquaculture industry in Nigeria (Owodeinde and Ndimele, 2011).

Azanza garckeana popularly known as "morajwa" (African chewing gum) in Botswana, Azanza is widely known as tree hibiscus, snot apple, wild hibiscus (Orwa *et al*., 2009) and "Gorun Tula" in Northern Nigeria (Gombe State). *Azanza garckeana* is a valuable edible indigenous fruit tree species. In Nigeria *A. garckeana* is found in Tula, Kaltungo Local Government Area of Gombe State.

A garckeana is a deciduous shrub or small spreading tree, 3-13 m high, with a diameter at breast height of up to 25 cm (Orwa *et al*., 2009). FAO (1983) reported that *A. garckeana* grows naturally in semi-arid areas receiving lowest annual rainfall of 250 mm and height rainfall of 1270 mm. The objective of the study was to examine the effects of *Azanza garckeana* pulp meal inclusion on egg quality of *Clarias gariepinus*.

MATERIALS AND METHODS

Study area: The experiment was conducted in Teaching and research farm Department of Fisheries Modibbo Adama University of Technology Yola. Nigeria.

Experimental fish: The brood stocks both male and female were collected from fishermen at Lake Geriyo in Adamawa State, Nigeria. The fish were transported in plastic troughs (50 cm diameter×30 cm deep) to teaching and research fish farm of Department of Fisheries, Modibbo Adama University of Technology, Yola Nigeria. The broodstock were acclimatised for 5 days in 10 m^2 earthen pond.

Experimental diet: The experimental diets were formulated to contain 40% crude protein using the Pearson square method. Fish meal, Soybean, wheat bran, vitamin premix, Vitamin C, lysine, methionine cassava starch (binder) was procured in Jimeta market. Soybean was toasted for 10 min using a fabricated manual soybean roasterat 100°C. Fish meal, toasted soybean, wheat bran was grounded separately using hammer mailer. All the feed ingredients were weighed with an electric weighing balance (Model: Metler Toledo PB 8001, London). The feeds ingredients were thoroughly dry mixed manually and packed in a bag until required.

Table 1: Proximate Composition of Formulated Feeds with the varying inclusion level of *Azanza garckeana* pulp meal

	Azanza garckeana pulp meal levels (g/kg)				
Nutrients	D1	D2	D3	D4	D5
Protein	35.50	38.20	40.30	42/8	44.00
Fat	24.90	26.50	27.20	28.90	29.00
Ash	8.50	8.00	7.00	6.50	7.00
Fibre	1.00	1.50	3.50	5.50	7.00
Moisture	10.50	9.00	7.00	10.00	9.50
NFE	19.60	16.80	10.50	6.30	3.00

Collection, processing and preparation of *A. garckeana* meal: The fruit *A. garckeana* was purchased from Tula, in Kaltungo Local Government area, Gombe State, Nigeria and transported by road to Moddibo Adama University of Technology, Yola. The seeds were removed from the pulps and the pulp sun-dried. The dried pulp was pounded into powdered form using pestle and mortar. The pounded pulp was sieved using Bs 0.8 mm mesh sieve, packed in nylon bag and store in a refrigerator until required. The proximate and mineral composition of *A. garckeana* Pulp Meal (AGPM) was analysed according to AOAC (2012).

Experimental design: Various concentrations of AGPM; 0, 50, 100, 150 and 200 g/kg of the fee as D1(control), D2, D3, D4 and D5, respectively were mixed with *Clarias gariepinus* broodstock feed. A total of 135 gravid *C. gariepinus* females and males broodstock were used for the experiment. The broodstock were divided into five equal experimental groups (9 male and female fish per treatment). The AGPM base diets were allotted to the broodstock in replicates in a concrete pond (12×8×1.4 m deep). The broodstock were fed with the experimental diet for the period of 56 days at 30% of their body weight twice daily morning and afternoon (8.00 and 1800 hours) local time). The experimental fish were weighed weekly in order to adjust the daily feed rate of 3% of the total biomass and fed twice daily (8.00 and 1800 h) for 56 days.

At the end of the broodstock treatment with AGPM (56 days) five each of the females and males broodstock were randomly collected from each treatment group and used for evaluating the fecundity, fertilization, hatchability and survival rates of the hatchlings (Table 1).

Fertilization rate: Fertilization rate was determined by randomly selecting the female brood stock from each treatment and induced with Ovatide hormone at 0.2 mL/kg of fish. After 10 h of latency period, the each females were stripped of its eggs into a dry receptacle. Testes from each male were collected after sacrifice. The testes were cut from into two from the lob and the spermatozoa squeezed onto the stripped eggs. Equal volumes of saline water were added to the mixture of the eggs and the milt after 1 min of stirring. Fifty fertilized eggs from each of the treatments were placed

in petri dishes containing well aerated water in replicates. Number of fertilized eggs were recorded 7 h, post fertilization, while the number of hatchlings was recorded after 24-28 h post fertilization. Survival of the hatchlings was recorded 7 days post hatching. The percentage fertilization, hatching, deformed larvae and survival for each treated were estimated as follows:

Fertilization (%) = Number of fertilised eggs/Total number of eggs counted×100

Egg hatchability (%) = Number of whitish broken eggs/Total number of hatchings×100

Deformed Larvae (%) = Number of deformed larvae/Total number of larvae×100

%Survival = Number of hatched eggs up to fry stage/Total number of hatchinglarvae×100

Statistical analysis: Data obtained from the experiment were subject to a one-way-Analysis of Variance (ANOVA). Differences between the means were determine using Turkey's HSD (p = 0. 05) using SPSS version 10.0.

RESULTS

Table 2 shows the proximate and mineral composition of *A. garckeana* pulp meal. The pulp meal contains most of the nutrients required by fish to grow. The crude protein level is 10.9%, while Fat, crude fibre, ash, Nitrogen free extract and moisture were 7.0, 41.3, 11.2, 18.9 and 14.5% respectively.

The mineral compositions of *A. garckeana* pulp meal includes sodium, calcium, iron, magnesium, potassium and phosphorous with 180.3, 17.82, 13.89, 1810.0, 59.50 and 59.50 mg/L, respectively. Ascorbic acid was observed to be 20.5 mg/100 g, while energy value and pH were 2.20 kcal/g and 5.09, respectively.

Fertilization rates were 100% in broodstock treated with 100 g of AGPM/kg of feed, followed by 50 and 150 and 200 g AGPM/kg of diet with 98 and 96, 92 and 76%, respectively (Table 3). There were significant differences ($p<0.05$) between the percentage fertilization values of fish treated with 50 and 100 g/kg of AGPM compared to those of 150 g/kg. Percentage fertilization was significantly lower in fish fed with the control diet.

Broodstock treated with 100 g of AGPM per kg diet had the highest hatchability (98%), followed by fish fed with 50, 150 and 200 g/kg (98, 96 and 92%, respectively). No significant variation ($p>0.05$) was observed among the percentage hatching values of fish treated with 100, 50, 150 and 200 g AGPM/kg. However, significant differences existed between the hatchability values of fish treated with 200 g of AGPM/kg compared to treatments 2, 3 and 4.

The percentage larval survival from broodstock treated 50 g of AGPM/kg was higher (45%) followed closely by those treated with 100 and 150 g AGPM/kg. Lower larval survival was significantly observed in the AGPM free diet followed by those treated 200 g AGPM/kg of feed.

The fecundity rate was significantly higher in fish fed the control diet. Broodstock treated with 200 g of AGPM/kg gave 158646.0 eggs, followed by D4, 3 and 2 with 138234.0, 123711.0 and 103198.0 eggs respectively. Significant variation was observed in the number of eggs among the entire treatments. Fish treated with the AGPM show significant increase in the fecundity rate with an increase in the AGPM dosages.

Table 2: Proximate and Mineral composition of *A. garckeana* pulp meal AGPM)

Nutrients and mineral	Values (%)
Moisture	14.5
Crude protein	10.9
Fat	7.0
Crude fibre	41.3
Ash	11.2
Nitrogen free extract	18.9
Sodium	180.3 mg/L
Calcium	17.82 mg/L
Iron	13.89 mg/L
Magnesium	3.59 mg/L
Potassium	1810.0 mg/L
Phosphorous	59.50 mg/L
Energy value	2.20kcal/g
pH	5.09
Ascorbic acid	20.5 mg/100 g

Table 3: Mean egg fertilization, hatchability and larval survival of *C. gariepinus* broodstock treated with *A. garckeana* pulp meal

	Azanza garckeana inclusion level (g/kg)					
Parameters	D1	D2	D3	D4	D5	SEM
Male (g)	583.0^{a}	550.0^{ab}	600.0^{a}	500.0^{b}	550.0^{ab}	17.48
Female (g)	483.0^{a}	466.7^{b}	466.7^{b}	366.7^{d}	450.0^{c}	36.13
No. of eggs fertilized	39.0^{c}	50.0^{a}	50.0^{a}	49.0^{a}	47.0^{b}	0.48***
% fertilized	78.0^{c}	100.0^{a}	100.0^{a}	98.0^{a}	94.0^{b}	0.97***
No. of hatched eggs	31.0^{c}	48.0^{a}	49.0^{a}	46.0^{a}	38.0^{b}	1.03***
% hatchability	62.0^{c}	96.0^{a}	98.0^{a}	92.0^{a}	76.0^{b}	2.07***
Survival	20.0^{d}	45.0^{a}	44.0^{ab}	42.0^{b}	28.0^{c}	0.86***
% Survival	40.0^{d}	90.0^{a}	88.0^{ab}	84.0^{b}	56.0^{c}	1.71***
Fecundity	192799.0^{a}	103198.0^{e}	123711.0^{d}	138234.0^{c}	158646.0^{b}	27.83***
No. of deformed hatchlings	3.0^{a}	0.0^{b}	0.0^{b}	0.0^{b}	4.0^{a}	0.52***
% Deformed	7.0^{a}	0.0^{b}	0.0^{b}	0.0^{b}	8.0^{a}	1.03***

Means in rows having different superscripts are significantly different ($p<0.05$)

Table 4: Mean water quality parameters during the experiment

A. garckeana inclusion levels (g/kg)	Water quality parameters			
	Temp. (°C)	pH	DO (mg/L)	Conductivity
D1	25.4	7.96	5.88	31.02
D2	25.7	8.15	5.87	32.05
D3	25.68	8.09	5.5	33.06
D4	25.73	8.14	5.73	34.10
D5	25.66	9.15	5.81	31.10

Deformed hatchlings were observed in the fish fed control diet and those feed with higher (200 g of AGPM/kg of feed (8.0 and 7.0%, respectively). There were no significant differences between the number of deformed hatchling in fish treated with 0 g/AGPM compared to those fed 200 g AGPM/kg of feed.

Water quality parameters: The water quality during the study is presented in Table 4. Temperatures were in the range of 25-25.5°C, while pH, dissolved oxygen and conductivity were between 7-9.15, 5-5.88 mg/L and 31-34.10, respectively.

DISCUSSION

The moisture content and crude protein of AGPM recorded in this study was close to 13.54 and 10.05%, respectively reported by Suleiman *et al*. (2012) for the *A. garckeana* fruits from Southern Danfur of Sudan. However, Saka *et al*. (1994) and Orwa *et al*. (2009) reported 12% crude protein level fruits of *A. garckeana* from Botswana. Iron, sodium and magnesium content of AGPM in this study were comparable to those in the fruits reported by Suleiman *et al*. (2012). This variation could be due to the fact that only the pulp was used in this study.

From the results of this study, *A. garckeana pulp*influenced the egg quality of *C. gariepinus* such as rate of fertilisation, hatchability, survival and fecundity rate. This agrees to the fact that wild fruits have potentials in enhancing egg quality and sperm fertility (Adedeji *et al*., 2006). The AGMP has high contents of minerals that could have influenced the egg quality.

The higher percentage fertilization, recorded in this study was higher than 90.88% reported by Adeparusi *et al*. (2010) for *C. gariepinus* broodstock treated with various dosages of *Kigelia Africana* seed powder and 48-51.71% reported by Dada and Ogunduyile (2011) for broodstock of *C. gariepinus* treated with different dosages of Velvet bean (*Mucuna pruirens*) dietary seed meal. The variation in the fertilization rate could be due to the differences in plant species used.

Diyaware *et al*. (2010) reported 73-83.90% fertilization rate when *C. anguillaris* female was fertilized with milt obtained from *C. Anguillaris* through male ablation (without sacrifice the male). Diyaware *et al*. (2012) observed 68-71% fertilization rate after fertilizing female *C. gariepinus* with milt obtained from male *C. gariepinus* treating with *Garcinia kola* meal.

The hatching rate recorded in this study was lower than 63-88.27% reported by Diyaware *et al*. (2010) in *C. anguillaris* female fertilized with milt collected from *C. anguillaris* through (male ablation) and 58% hatching rate of *C. gariepinus* by Diyaware *et al*. (2012) after using milt from male treated with 0.3 g/kg of *Garcinia kola* seed meal.

Dada and Ajilore (2009) observed high fecundity rates when *C. gariepinus* female broodstock were fed 0.25 g of *G. Kola*seed powder per kg of feed and larger egg size when fed higher (2.0 g/kg of feed) dosage of *G. kola* based diet.

It was observed that fecundity of *C. gariepinus* increased with increasing inclusion level agree with the work done by other authors that extracts from fruits can enhance fecundity in *C. gariepinus* (Dada, 2012; Dada and Ajilore, 2009). The increase in fecundity of *C. gariepinus* in this present study could be attributed to the presence of flavonoid and carotenoids in the plant. Newbrey and Reed (2011) reported that carotenoids can be important antioxidant and immunostimulant for developing embryos and adult birds. Adesanya *et al*. (2007) reported that biflavonoids and Xanthone are potent anti-oxidants which are capable of increasing the production of oestrogen, the key hormone involved in the production and maturation of eggs in the ovary.

CONCLUSION

A. garckeana in the diets of *C. gariepinus* at 150 and 200 g/kg improved the reproductive performance of *C. gariepinus* broodstock. This could be usedas a reliable ingredient for propagating *C. gariepinus* fingerling production. This study established the efficacy of *A. garckeana* as fertility enhancer in *C. gariepinus* broodstock and should be encouraged as it will minimize the dependence on synthetic drugs as fertility enhancing agents.

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