Tropical and Subtropical Agroecosystems

REPRODUCTIVE PERFORMANCE AND SERUM BIOCHEMISTRY OF FEMALE *Clarias gariepinus* BROODSTOCK RAISED IN POND EFFLUENT WATER

[COMPORTAMIENTO REPRODUCTIVO Y BIOQUÍMICA SANGUÍNEA DE HEMBRAS DE *Clarias gariepinus* CRIADAS EN EFLUENTES DE ESTANQUES]

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SUMMARY

The reproductive performance and serum biochemistry of female *Clarias gariepinus* raised in pond effluent water under laboratory condition for 70 days were investigated. Reproductive performance parameters showed that there were no significant differences (P>0.05) in the quantity of egg spawned and percentage fertilization in fish raised in spring and pond effluent water. However, fish raised in spring water had the best percentage hatchability (84.15%) which was significantly different (P<0.05) from fish raised in pond effluent water (67.85%). Survival rate of newly hatched larvae from brood fish raised in spring water was 90.00%, this was significantly different (P<0.05) from 70.50% recorded for hatchlings from brood fish raised in pond effluent water. Fish raised in pond effluent water had higher (P<0.0.5) values of plasma excretory products (urea and creatinine), 29.00mg/dl and 1.30mg/dl respectively, compared with 16.5mg/dl and 0.91mg/dl in fish raised in spring water, which indicated a pathological effect. Serum electrolytes, enzyme activities and plasma proteins of the female brood fish were not significantly (P>0.05) affected by the treatments. It could therefore be concluded that pond effluent water should be treated before it is used for rearing brooders.

Keywords: Reproductive performance, blood parameters, *Clarias gariepinus*, effluent water.

RESUMEN

Se estudio el comportamiento reproductivo y la química sanguínea de hembras de *Clarias gariepinus* criadas en efluente de estanque bajo condiciones de laboratorio durante 70 d. No se encontró diferencia (P>0.05) en la cantida de huevos producidos y el porcentaje de fertilización entre los peces criados en agua de efluente o en agua de manantial. Sin embargo los huevos de peces criados en agua de manantial tuvieron un mejor porcentaje de eclosión (84.15%) y sobrevivencia de larvas (70.5%) comparado con los peces criados en agua de efluente (67.85 y 70.5% respectivamente) (P<0.05).
Los peces criados en efluentes tuvieron valores plasmáticos más altos (P<0.05) de urea y creatinina (29 y 1.3 mg/dl vs. 16.5 y 0.91 mg/dl respectivamente), lo que probablemente indica efectos patológicos. Electrolitos séricos, actividad enzimática y proteína plasmática no fueron afectados (P>0.05). Se concluyó que el agua de efluente puede ser empleada para la cría de *C. gariepinus* pero requiere tratamiento previo.

Palabras clave: Comportamiento reproductivo, parámetros sanguíneos, efluente de estanque.

INTRODUCTION

Commercial production of the African catfish (*Clarias gariepinus)* is becoming popular in Nigeria. This is because it is hardy, grows fast, highly esteemed, commands high market value and according to Hengsawat *et al.* (1997), a valuable specie worldwide. In many arid and semi-arid countries, water is increasingly becoming a scarce resource (Pascod, 1992) due to irregularity of water to fill rain fed ponds, poor quality of water from streams and rivers used by farmers especially in urban areas where domestic sewage and industrial pollution are problems. Whenever good quality water is scarce, Pascod (1992) further observed that water of marginal quality could be used to raise fish and one of such is pond effluent water.

Pillay (1992) reported that pond effluent water increases suspended solids and nutrients of receiving water body; it also reduces the dissolved oxygen content which can lead to production of ammonia, hydrogen sulphide and methane. High level of nutrients can lead to algae bloom which can cause fish mortality. Cripps (1994) observed that waste material from aquaculture effluents occurs in low concentration at high water flow rates compared with other industrial and domestic effluents. In developing countries, there is generally low level of aquaculture production; due to lack of water particularly during the dry season .This could be alleviated by recycling pond effluent water to facilitate all year round production. This study was therefore designed to investigate the use of pond effluent water to raise fish, its effect on reproductive performance and serum biochemistry of female *Clarias gariepinus* broodstock.

MATERIAL AND METHODS

Experimental fish, water and procedures

Twenty four (24) female broodstock of *Clarias gariepinus* (9 month old) with initial mean weight of 293.0±6.2g were used in this study. They were obtained from a commercial fish farm in Ibadan, Nigeria. The fish were transferred into Department of Wildlife and Fisheries Management teaching and research laboratory where they were conditioned for 14 days in eight aquaria of size 60cm x 30cm x 30cm before the experiment.

The fish were randomly divided into two treatments: treatment I Spring water and II pond effluent water, with each treatment having four replicates. Fish in treatment 1 were stocked in fresh spring water obtained from underground water while treatment II were stocked in freshly collected pond effluent water from a commercial catfish farm where fish were stocked at 50,000fish.ha^{-1} and raised for 9months cultured period. Fish were fed at 1% of their body weight daily at 8.30am and 16.30 hours for 10 weeks with 40% crude protein diet (Table 1). Water in each of the treatments was replaced every 48 hours with fresh spring water for the control fish, and pond effluent water for the test fish.

Table 1: Gross composition of experimental Diet

Ingredient	Composition (%)
Fish meal	15.00
Blood meal	6.00
Soya meal	18.65
Groundnut cake	28.00
Maize	28.00
Bone meal	2.50
Oyster shell	1.00
Salt	0.25
Premix	0.60
Total	100

After 70 days of feeding, four female brooders were randomly selected from each treatment for breeding. Selected brooders were injected with fresh *C. gariepinus* pituitary gland extracts intramuscularly above the dorsal fin and stripped separately after 10 hours. One male *C. gariepinus* from each of the treatment was sacrificed respectively for the fertilization exercise. Milt from one lobe of testis was used to fertilize the eggs from each treatment. After fertilization eggs from each treatment were incubated in separate aquaria in a single layer with a flow through system for 36 hours using dechlorinated tap water.

Proximate analysis, growth performance, water quality and hatchability

The experimental diet and two randomly selected female broodstock from each of the treatments were analysed for their proximate composition before and after the feeding trial, using the A.O.A.C. (1995) method. Growth performance data such as weight gain (WG), feed conversion ratio (FCR), feed intake (FT), protein efficiency ratio (PER), specific growth rate (SGR %) and survival rate were determined using the method described by Richter *et al* (2003). Quantity of eggs from brooders from each treatment was estimated using the method described by Viveen *et al* (1985). The percentage fertilization, hatchability, and survival rate were determined by method described by Faturoti *et al* (1992).

Water quality parameters such as dissolved oxygen (DO), biological oxygen demand (BOD), pH, alkalinity, ammonia, nitrate, carbon dioxide (CO_2) and temperature were measured for both the used and freshly introduced pond effluent water (treatment) and spring water (control) every 48hours at 8.00am (Boyd, 1981 and APHA, 1998). Major elements such as Calcium (Ca) and Phosphorus (P) were determined according to A.O.A.C. (1995)

Blood collection and plasma biochemical analysis

3ml of blood was collected from female broodfish in each treatment by the caudal peduncle lateral venipuncture (Stoskpof, 1993) into a lithium heparin anticoagulant tube for plasma biochemical analysis at 8.00am after ten weeks of feeding. Plasma obtained by centrifugation from the lithium heparinised samples was stored at – 25 ºC until analyzed.

The stored plasma samples were later analyzed for sodium and potassium concentration by standard flame photometry method (Uyanik *et al.,* 2001), chloride, calcium, phosphate, bicarbonate, triglyceride, urea, creatinine, alkaline phosphates (ALP), Aspartate (AST) and Alanine (ALT) transaminanses, Cholesterol, total protein and albumin (Toro and Ackermann, 1975; Alkahem *et al.,* 1998; Olayemi, 2001). Globulin was calculated by subtracting albumin from total protein.

Statistical Analysis

Data collected were subjected to one way analysis of variance, statistical difference was established at 95% confidence limit using Duncan multiple range test (Steel *et al.*, 1999).

RESULTS

Proximate compositions of the experimental diet and fish before and after the feeding trial are presented in Table 2, while the growth performance indices are presented in Table 3. The protein content of experimental diet was 40.90%, while there was a slight increase in the percentage protein content of experimental fish from 56.61% to 58.81% and 59.60% in fish raised in spring water and pond effluent water after feeding for 10 weeks (Table 2). Growth performance of fish such as mean weight gain, specific growth rate, feed intake, feed conversion ratio, protein efficiency ratio, and survival rate in both treatments showed that there was no significant difference (P>0.05) in values of all the parameters.

Reproductive performance of female *C. gariepinus* brooders raised in spring water and pond effluent water are presented in Table 4. There was no significant difference (P>0.05) in the mean weight of female brooders, quantity of egg spawned and percentage fertilization in both treatments. However, fish raised in spring water gave the best performance in terms of percentage hatchability with 84.15% which was significantly different (P<0.05) from 67.85% hatchability recorded for fish raised in pond effluent water.

Survival rate of newly hatched larvae from broodfish raised in spring water was 90.00% which was significantly different (P<0.05) from 70.50% recorded from broodstock raised in pond effluent water. The water quality parameter of the spring water and pond effluent water used for the experiment are presented in Table 5. BOD, CO_2, alkalinity, NO_2, Ca, P, Fe were higher in pond effluent water compared to spring water. Pb and Cu were not detected in any of the treatments.

Table 2: Proximate Composition (%DM) of Experimental feed, and fish before and after feeding.

Sample	Crude Protein	Crude fat	Fibre	Ash	Moisture	*N.F.E
Experimental feed	40.90	5.02	5.98	10.22	10.20	27.68
Fish before experiment	56.61[a]	3.35[a]	1.76[a]	11.64[a]	9.55[a]	17.09[a]
Fish raised in Spring water	58.81[a]	1.59[a]	0.33[a]	10.20[a]	8.70[a]	20.37[a]
Fish raised in Pond Effluent water	59.60[a]	2.50[a]	0.34[a]	9.80[a]	9.00[a]	18.68[a]

Mean with the same superscript in a column are not significantly different (P>0.05). *Nitrogen Free Extract.

Table 3: Growth performance parameters of *C. gariepinus* brooders raised in pond effluent and spring water**.**

Parameters	Spring water	Pond Effluent water
Weight gain (g)	22.5 ± 4.69[a]	17.7 ± 5.48[a]
Specific growth rate (%)	2.10 ± 0.38[a]	2.17 ± 0.99[a]
Feed conversion ratio	1.05 ± 0.16[a]	1.25 ± 0.72[a]
Protein efficiency ratio	0.02 ± 0.002[a]	0.02 ± 0.09[a]
Feed intake (g)	23.28 ± 3.78[a]	19.55 ± 4.27[a]
Survival rate (%)	100.00 ± 0.0[a]	100.00 ± 0.0[a]

Means with the same superscript within each row are not significantly different (P>0.05).
± SD = Standard deviation.

Table 4: Reproductive performance of *C. gariepinus* raised in pond effluent and spring water.

Parameters	Spring water (N=4)	Pond Effluent Water (N=4)
Weight of fish before spawning (gm)	316.9 ± 28.28[a]	309. 3 ± 10.25[a]
Weight of fish after spawning (gm)	211.2 ± 32.31	297.8 ± 10.04
Numbers of eggs	3955 ± 2516.80[a]	5985 ± 2619.83[a]
Percentage fertilization (%)	95.00 ± 0.0[a]	95.00 ± 0.0[a]
Hatchability (%)	84.15 ± 2.051[a]	67.85 ± 3.041[b]
Survival rate %	90.00 ± 2.04[a]	70.50 ± 2.27[b]

Means with the same superscript along the same row are not significantly different (P>0.05)

Table 5: Water quality parameters of pond effluent and spring water used for the experiment.

Parameters	Pond effluent water after 48 h of use	Fresh Pond effluent water (n=4)	Fresh Spring water (n=4)	Used Spring water after 48hrs
Dissolved oxygen mg/L	3.29±0.06	4.50 ± 0.10	6.90 ± 0.07	3.48±0.05
Biological oxygen demand	22.00±0.04	8.20 ± 0.08	1.92 ± 0.02	11.30±0.10
pH	7.5±0.01	7.60 ± 0.02	7.50 ± 0.01	7.40±0.52
Carbon dioxide (CO_2) mg/L	12.00±0.13	11.40 ± 0.12	1.01 ± 0.08	11.60±0.15
Alkalinity (mg/I)	460.0±0.10	289 ± 0.23	150 ± 0.72	380±0.55
Nitrite (NO_2) ppm	0.98±0.0	0.25 ± 0.01	0.01 ± 0.01	0.10±0.0
Temperature (ºC)	25.00±0.0	27.50 ± 0.0	27.50 ± 0.0	25±0.0
Calcium (Ca) ppm	0.32±0.0	1.37 ± 0.01	0.31 ± 0.01	0.03±0.01
Phosphorous (P) ppm	0.05±0.01	0.53 ± 0.02	0.02 ± 0.01	0.01±0.02
Iron (Fe) ppm	0.74±0.02	0.75 ± 0.01	0.10 ± 0.01	0.10±0.0
Lead (Pb) ppm	ND	ND	ND	ND
Copper (Cu) ppm	ND	ND	ND	ND
Ammonia(NH_3) ppm	2.08±0.10	0.82 ± 0.15	0.00 ± 0.0	0.50±0.01

ND = Not Detected

Blood biochemical parameters of broodfish raised in pond effluent and spring water are presented in Table 6. There was no significant different (P>0.05) between the serum electrolytes, enzyme activities and proteins in fish raised in the two treatments. Fish raised in pond effluent water however has a higher values of urea (29.00±2.48mg/dl) and creatinine (1.30±0.07mg/dl) which was significantly different (P<0.05) from those raised in spring water with values of 16.5±0.65mg/dl and 0.95±0.02mg/dl respectively. Cholesterol values was significantly different (P<0.05) with 98.00±4.74mg/dl and 81.25±2.69mg/dl for brooders raised in pond effluent water and spring water respectively. However, triglycerides values was not significantly different (P>0.05) in both treatments.

Table 6: Mean serum biochemical parameters of *C. gariepinus* broodstock raised in pond effluent and spring waters.

Parameters	Brooders raised in Spring water (n=4)	Brooders raised in Pond Effluent water (n=4)
Serum electrolytes		
Sodium (mmol/1)	130.25 ± 1.265 [a]	130.25 ± 1.70 [a]
Potassium (mmol/I)	5.75 ± 0.39 [a]	5.00 ± 0.11 [a]
Chloride (mmol/I)	99.25 ± 0.86 [a]	99.75 ± 1.32 [a]
Biocarbonate (mmol/1)	20.75 ± 0.48 [a]	21.75 ±0.25 [a]
Calcium (mg/ldl)	8.70 ± 0.07 [a]	8.35 ± 0.03 [a]
Phosphate (mg/ldl)	5.00 ± 0.39 [a]	4.90 ± 0.16 [a]
Enzymes activities		
Alkalinephosphatates (AP) (iu/dl)	186.75 ± 30.92 [a]	167.25 ± 4.03 [a]
Aspartatetransferase (AST) (iu/l)	21.75 ± 4.33 [a]	34.75 ± 2.06 [a]
Gamma glutamyl transferase (iu/l)	6.75 ± 2.50 [a]	7.00 ±0.41 [a]
Plasma excretory products		
Urea (mg/dl)	29.00 ±2.48 [a]	6.50 ± 0.65 [b]
Creatinine (mg/dl)	1.30 ± 0.07 [a]	0.95 ±0.02 [a]
Plasma proteins		
Total proteins (g/dl)	5.68 ±0.12 [a]	5.87 ±0.01[a]
Albumin (g/dl)	2.72 ± 0.06 [a]	2.70 ± 0.9 [a]
Globulin (g/dl)	3.18 ± 0.02 [a]	3.18 ± 0.09 [a]
A-G Ratio	0.72 ± 0.15 [a]	0.85 ± 0.05 [a]
Plasma lipids		
Cholesterol(mg/dl)	98.00 ± 4.74 [a]	81.25 ± 2.69 [a]
Triglycerides(mg/dl)	78.50 ± 6.69 [a]	63.00 ± 3.58 [a]

Means with the same superscript along the same row are not significantly different (P>0.05).

DISCUSSION

The proximate analysis of experimental diet showed that it was suitable for conditioning *C. gariepinus* brooders as recommended by Ayinla (1988) and Hengsawat *et al.* (1997). The slight increase in carcass protein of the brooders in both treatments from the initial values showed that feed given was utilized not only for egg development but also for growth. This is in agreement with the findings of Cho *et al.* (1985) and Richter *et al.* (2003). Fish raised in spring water showed a better feed conversion ratio, higher weight gain, hatchability and survival rate of larvae. This might be due to better water quality condition, since Boyd (1981) reported that good water quality enhanced better growth and reproductive performance of fish.

The higher values of BOD, CO_2, NH3, NO_2, Ca^+, P^+, and Fe^{2+} observed in this study was similar to the findings of Ayinla (1999). He reported the potential impacts of such untreated water discharged on the external environment with adverse effect on BOD, COD, DO, solid accumulation, nutrient enrichment leading to hyper-nutrification and eutrophication.

The higher values of plasma excretory products of urea and creatinine in brooders raised in pond effluent water further confirmed the effect of water quality on the performance of the brooders. This is because high levels of these parameters constitute stress on the brooders, leading to the inability of the brooders to effectively utilize feed, thereby affecting reproduction and early life activities of the offspring. This is in agreement with the findings of Schreck *et al.* (2001) and Ogunsanmi (1994a, b) on the effect of stress on reproductive activities of fish. The urea level observed in catfish raised in pond effluent water was about three times higher than those reported by Agbede *et al*. (1999) and Oyelese *et al.* (1999) for adult catfish. While brooders raised in spring water had similar urea levels to those reported by Agbede *et al. (*1999) and Oyelese *et al.* (1999) for adult catfish. The level of creatinine in fish raised in pond effluent water was also higher than those reported by Oyelese *et al.* (1999) for adult catfish.

CONCLUSION

Pond effluent water might be used for raised Catfish, however it should be treated before is used for rearing brooders.

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