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EFFECT OF *Cocos nucifera* WATER COMPOUNDED DIETS ON REPRODUCTIVE PERFORMANCE, HEAMATOLOGY AND GONADAL DEVELOPMENT OF MALE AFRICAN CATFISH (*Clarias gariepinus*) BROODFISH

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Abstract

The effect of *Cocos nucifera* water compounded diets on reproductive performance; heamatology and gonadal development of male African mud catfish (*Clarias gariepinus*) broodfish were investigated. Broodfish (909.63±1.03g) were used for the study. Four isonitrogenous diets (40% crude protein) formulated graded inclusion levels of 4, 6 and 8%, were used for the study of the diets designated as Control (CNR), Diet 1, Diet 2 and Diet 3 respectively. A total of 60 male *C. gariepinus* were randomly selected and distributed in triplicate into twelve (12) outdoor rectangular concrete tanks at a stocking density of five fish per tank for a period of 112 days and were fed at 3% body weight. Reproductive performance indices (% fertilization, % hatchability and % survived) milt volume, milt count motility duration were determined. There was a significant difference in ($p< 0.05$) in weight of gonad, milt count, milt volume, GSI and hatching time, meanwhile there was no significant difference ($p >0.05$) recorded in the performance indices across fertilization, hatchability and survival were Diet 3 of 8%/100 diets fed performed better in terms of % fertilization, % hatchability and % survival. The heamatological changes showed difference across the experimental groups. The histology of individual fed diet 1- 3 shows some level of organized lobules with populated lumen,. The study concluded that *Cocos nucifera* water is efficacious as a proficient agent in fertility for male *Clarias gariepinus.*

Keywords: *Cocos nucifera* water, African mud catfish, egg quality, gonadosomatic index

INTRODUCTION

There are reports that decline in landing from capture fish stocks are far exceeding the point of maximum sustainable yield (MSY) (Danquah, *et. al*., 2021; Anetekhai, A M., 2010). It is in an endeavour to tackle this challenge that the aquaculture industry is expanding so as to satisfy the increasing demand for affordable, safe and high quality fish and fish products (Craig, 2009). Meanwhile, in culturing system, one very important component is the availability of fish feed not just adequate in quantity but nutritional value, good enough for seamless production of seeds which are necessary for the multiplication of the quantity and quality of fish produced. Fish generally require large proportions of high-quality fish feed to produce high-quality eggs and

sperm and to acquire the maximum amount of protein required for healthy development (Hassan, 2001). There is a steady rise in the cost quality feed (Mzengereza *et al*., 2014) that has the potential to maintain good reproductive performance. Nonetheless, the effort to contain the ever rising cost of protein rich feed in order to maintain and optimise the reproductive performance of broodstocks in the production of virile seeds for a sustainable aquaculture practice has prompted research into use of alternative and more readily available plant based sources such as legumes (De Silva *et al*., 1991) jack beans meal (Alegbeleye *et al*., 2001) and *Tetracarpidium conophorum* (Dada and Aguda 2015) either as substitutes, complementaries or enhancers among others. This work looked at the use of *cocos nicufera* as a substitute at varying levels of inclusion for an enhanced reproductive performance of the male African catfish (*Clarias gariepinus*). Clarias is used because it had been found to be one of the most widespread catfish genera in the world (Ng HeokHee, 2001) and according to FAO (2024) Nigeria is the world's largest producer of African catfish, one of the most commercially important freshwater fish species in Africa and therefore with higher potential for sustainability.

MATERIALS AND METHODS

The experiment ware carried out at Department of Aquaculture and Fisheries Management Teaching and Research Farm of Science and Technology Education Post-Basic (STEP. B), Federal University of Agriculture Abeokuta, Nigeria. The healthy coconuts fruit were collected from Osile plantation Sobe, owan west local government Edo state down to Abeokuta and were dehusked and was then carefully cracked open and the liquid content pour into a clean plastic bottle for storage after it had been sieved to ensure particle which fell in accidentally were removed, the extract was then stored in the refrigerator until use.

The feed were sourced from a local feed meal and transported to down to federal university of Agriculture Abeokuta, after which was weigh with a top loading balance (Metler Toledo PB 8001, London). The ingredients were grinded together, into a small fine particles size in order to obtain homogenous mass. Cassava starch was added as a binder and the *Cocos nucifera* water pure on the feed and mixed together. The resultant dough press with steam water and Pelleted with a flat pelleting machine (KL 120 Mini, J.K pellet, Henan Sumit, China), using 6mm die hole, and the diet was immediately sun-dried at a temperature between 25-30 0C for days, to prevent mould and mucus throughout the period of the feeding trials, after which the experimental diets were parked in polythene bag labeled accordingly and stored in a cool dry place until use.

Experimental design

A total of sixty (60) sub-adult African mud catfish (*Clarias gariepinus*) weighing between 300 and 350g on average were purchased from a reputable fish farm (Royal valley farm) transported down to federal university of Agriculture Abeokuta and were identified and chosen based on their outward sexual features. The sub-adult fish have noticeable pointed genital papillae that are pink in colour (Olaniyi and Omitogun, 2013). The fish were stocked at five (5) fish in an open concrete holding tanks (1m x 1m x 0.8m) acclimatized for a period of two weeks (14 days) before the start of the experimental feed. Fish in each of the various groups were fed varying diets of *Cocos nucifera* compounded feed at a daily rate of 3% body weight in three equal diets twice daily between 09:00 and 18:00 hrs for a period of twelve (12) weeks, while water quality parameters including temperature, hydrogen ion concentration

(pH) and dissolved oxygen concentration were accessed weekly. At the end of the feeding period various parameters were determine which includes, Weight of fish (g), Weight of gonad(g), Milt count, Milt volume, of fish, Eggs diameter (mm), Hatching time (hr), % Fertilization, % Hatchability and % Survival.

Percentage fertilization: This was determined after twelve hours (12hrs), while the observed white and non-transparent eggs were removed. This was carried out by siphoning the dead and unfertilized eggs that appeared whitish, the fertilize eggs were then counted while the un-fertilized eggs were discarded:

%Fertilization = Total number of stripped eggs – total number of unfertilized eggs)/ (Total number of eggs counted) x 100

(Ayinla and Akande, 1988)

Percentage hatchability: The hatched larval were counted after fertilization, while the un-hatched eggs were discarded and the percentage hatchability estimated as followed

$$\% \text{ Hatchability} = \frac{\text{Total number of eggs hatched}}{\text{Total number of fertilized eggs}} \times 100$$

(Ayinla and Akande, 1988)

Percentage survival: The percentage Survival was estimated through visual means counting of the fries on a daily basis. At the end of the experiment the remaining survived fries were estimated as follow:

$$\% \text{ Survival} = \frac{\text{Total number of fries at the end of the stuidy} \times 100}{\text{Total number of fries at the begining of the study}}$$

(Ayinla and Akande, 1988)

Heamatology: **S**tandard method of Blaxhall and Daisley (1973) were used for the study, blood collected with the aid of needle (syringe 1m capacity) into heamatocrit bottle, the blood withdraw into ethylene Di Tetra Acetatate (EDTA) bottle and shake properly to avoid clotting while parameters such as pack cell volume (PCV), erythrocyte count (Red blood cell), leucocyte count (White blood cell), heamoglobin (HB) were determined.

RESULT AND DISCUSSION

The male reproductive indices observed from the experiment, had an impressive result with a significant performance ($p<0.05$) in weight of gonad, milt count, milt volume and GSI. Approaching the % fertilization, % hatchability and % survival, which had no significant difference ($p>0.05$) with record showed in (Table 3). The experimental diets compounded with *Cocos nucifera* having highest in % fertilization % hatchability and % survival while least in control, this performance can be ascribe to the presence of some trace minerals and vitamin found to be available in Cocos nucifera water, these includes calcium (Ca), copper (Cu), iron (Fe), magnesium (Mg), selenium (Se) and zinc (Zn).

Table 1**:** Gross composition of experimental diets

	Dietary treatment			
Ingredients	CNR	D1	D2	D3
Fish meal	32.9	32.9	32.9	32.9
Soya bean	28.38	28.38	28.38	28.38
White maize	22.56	22.56	22.56	22.56
Brewer waste	6.38	6.38	6.38	6.38
Mineral premix	0.5	0.5	0.5	0.5
Vitamin	1.5	1.5	1.5	1.5
Binder (starch)	4.38	4.38	4.38	4.38
Salt	0.4	0.4	0.4	0.4
Cod liver oil	3	3	3	3
Cocos nucifera	0	4	6	8

Table 2: Proximate composition of the experimental diets

	Experimental diets (Mean ±SE)			
Parameter	CNR	Diet 1	Diet 2	Diet 3
Moisture (%)	9.30 ± 1.20^{a}	10.10 ± 0.70^{b}	9.80 ± 0.81^{b}	9.90 ± 0.22^{b}
Crude protein (%)	40.80 ± 2.20^{a}	40.00 ± 1.7^{a}	40.40 ± 2.21^{b}	40.70 ± 1.15^{a}
Crude fibre (%)	4.65 ± 1.34^{b}	4.25 ± 1.21^{b}	4.35 ± 1.17^{a}	4.75 ± 1.66^{a}
Crude Fat (%)	9.17 ± 1.22^{a}	9.35 ± 1.33^{b}	9.63 ± 1.35^{a}	9.15 ± 0.41^{b}
NFE / CHO (%)	29.13 ± 0.39^{a}	29.64 ± 0.13^{b}	29.32 ± 1.33^{a}	28.7 ± 1.02^{a}
Crude Ash (%)	6.95 ± 0.45^{a}	6.66 ± 0.35^{a}	6.50 ± 0.40^{a}	6.80 ± 0.41^{a}

Mean values with different superscripts are significantly different ($p<0.05$) across the row. Key: CNR- (0%/100 extract in compounded feed); Diet 1- (4%/100 *Cocos nucifera* in compounded feed); Diet 2- (6%/100 of *Cocos nucifera* in compounded feed); and Diet 3- (4%/100 *Cocos nucifera* in compounded feed).

Table 3: Reproductive performance of male *Clarias gariepinus* fed diets containing 4, 6 and 8% of *Cocos nucifera*

	Dietary treatment			
Parameter	CNR	Diet 1	Diet 2	Diet 3
IMW (g)	316.36 ± 3.56^{b}	340.35 ± 5.09^{c}	348.96 ± 6.32^{bc}	$318.88\pm3,09^{b}$
FMW (g)	853.19 ± 0.56^{a}	943.12 ± 3.50^{b}	934.87 ± 4.56^{c}	966.06 ± 1.52^{b}
MWG (g)	536.83 ± 3.51^{b}	602.77 ± 2.06^{b}	585.91 ± 5.50^{c}	$647.18\ \pm3.06^{b}$
SGR(% day $^{-1}$)	479.31 ± 1.71^{a}	538.18 ± 1.36^{a}	523.13 ± 2.51^{b}	577.83 ± 1.51^{c}
Weight of Gonad (g)	4.110 ± 0.11^{a}	5.08 ± 0.60^{a}	5.02 ± 0.23^{b}	5.42 ± 0.00^{b}
Milt Count	221950 ± 2.30^{a}	238650 ± 0.27^{a}	247600 ± 1.70^{b}	243950 ± 3.34^{b}
Milt Volume	0.90 ± 0.40^{a}	0.90 ± 0.43^{a}	1.00 ± 0.63^{b}	1.00 ± 0.37^{b}
G.S.I(%)	0.48 ± 0.33^{a}	0.53 ± 0.60^{a}	0.53 ± 0.31^{a}	0.56 ± 0.00^{b}
Hatching Period	30.00 ± 0.04^{a}	30.00 ± 0.12^{a}	30.00 ± 0.34^{a}	30.00 ± 0.05^{a}
Hatching time(hr	24.48 ± 0.42^{a}	2.56 ± 0.35^{b}	23.24 ± 0.18^{b}	22.60 ± 0.45^{a}
% Ferterlization	65.62 ± 3.00^{a}	74.101 ± 2.04^{a}	80.041 ± 2.64^{a}	85.35 ± 0.762^{a}
% Hatchability	61.20 ± 4.80^{a}	77.011 ± 1.37^{a}	79.201 ± 3.54^{a}	82.642 ± 4.01^{a}
% Survival	50.30 ± 1.20^{a}	62.114 ± 1.73^{a}	63.222 ± 1.90^{a}	66.38 ± 1.33^{a}

Mean values with different superscripts are significantly different ($p<0.05$) across the row.
KEY: IMW--Initial mean weight, FMW-- Final mean weight, MWG-- Mean weightg gain, SGR-Specific growth rate, GSI -Gonadosomatic index = gonads weight (g) × 100/ fish weight, CNR-(0%/100 extract in compounded feed); Diet 1- (4%/100 *Cocos nucifera* in compounded feed); Diet 2- (6%/100 *Cocos nucifera* in compounded feed); Diet 3- (8%/100 *Cocos nucifera* in compounded feed)

Table 4: Water physiochemical parameters of *Clarias gariepinus* fed diets containing 4, 6 and 8 % varying inclusion of *Cocos nucifera* .

	Experimental group **(Mean ±SE)**				
Parameter	CNR	Diet 1	Diet 2	Diet 3	Standard
Temp. (^{0}C)	25.00±0.16^a	25.30±0.60^a	26.00±0.43^a	25.50±0.92^a	20-35 (FEPA.1991)
pH	6.42±0.43^a	6.00±0.48^a	6.33±0.23^a	6.74±0.40^a	6-9 (Lawson, 1995)
DO(mg/l)	6.22±0.49^a	7.32±0.32^a	7.69±0.42^a	8.26±0.29^a	>1.00(Loyd,(1992)
Nitrate(mg/l)	3.82±0.42^a	3.90±0.14^a	3-04±0.29^a	3.91±0.49^a	<100 (Meade, 1989)

Mean values of the water parameters are not significantly different ($p>0.05$) across the row. Key: Temp.-Temperature., pH- Hydrogen ion concentration., DO-Dissolved oxygen (mg/l).,

Table 5**:** Heamatological Parameters of *Clarias. gariepinus* fed diets containing 4, 6 and 8 % varying inclusion level of *Cocos nucifera*.

	Experimental group (Mean ±SE)			
Parameters	CNR	Diet 1	Diet 2	Diet 3
PCV (%)	30.00±0.56[a]	37.00±0.86[a]	37.50±0.54[a]	38.70±0.42[a]
HB (g/dl)	8.20±0.73[a]	9.55±0.38[a]	9.40±0.39[a]	10.50±0.53[a]
RBC ($x10^{12}$/L)	2.20±0.70[a]	2.60±0.53[a]	2.75±0.96[a]	2.95±0.54[a]
WBC ($x10^{9}$/L)	13.10±0.61[a]	14.30±0.56[a]	14.70±0.65[a]	14.90±0.47[a]
Neu (%)	30.25±0.61[a]	29.50±0.61[a]	29.00±0.61[a]	28.70±0.61[a]
Bas (%)	2.00±0.61[a]	1.12±0.61[a]	0.86±0.61[a]	0.82±0.61[a]

Mean values of the heamatology parameters are not significantly different ($p>0.05$) across the row. CNR- (0%/100 extract in compounded feed); Diet 1-(4%/100 of *Cocos nucifera* in compounded feed); Diet 2- (6%/100 of *Cocos nucifera* in compounded feed); and Diet 3- (8%/100 of *Cocos nucifera* in compounded feed). PCV-Packed cell volume, HB-Heamoglobin count, RBC-Red blood cell, WBC-White blood cell, Neu- Neutrophilis, Bas-Basophils

HISTOLOGY OF THE FISH

A

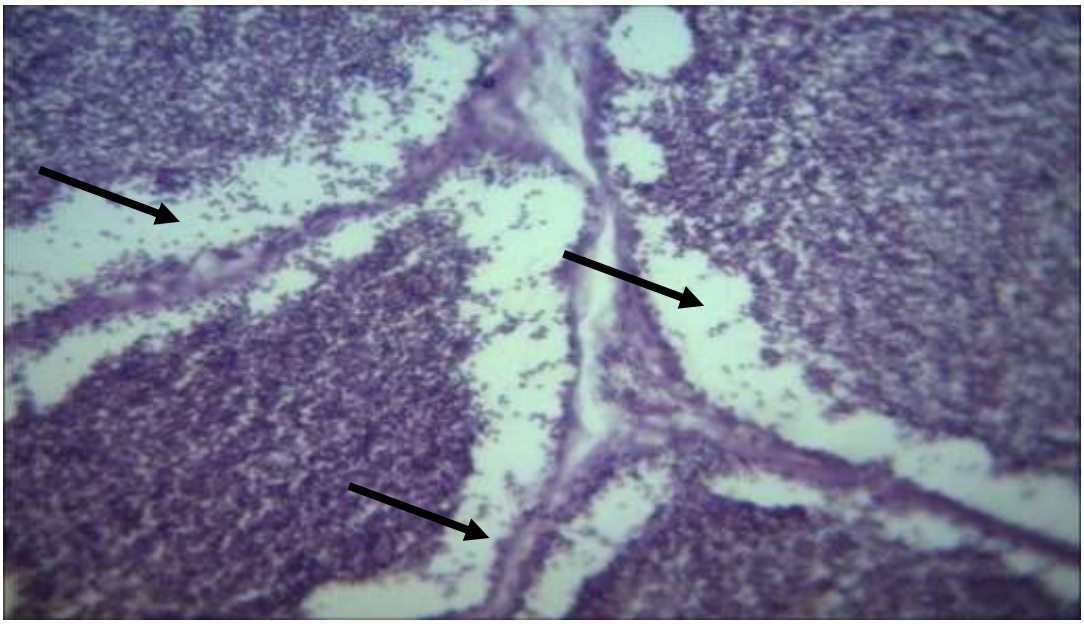

Plate 1: A transverse section through testis of *C.gariepinus* diet fed (0/100 of *Cocos nucifera* water in compounded feed) with empty lobules seen and a bit scanting spermatozoa recorded. X 100

B

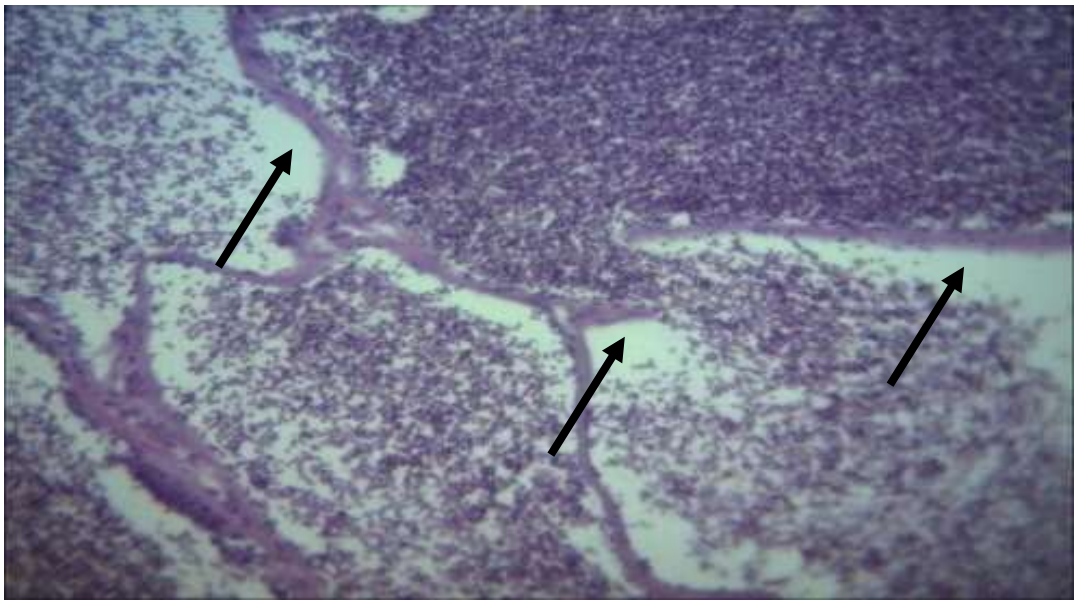

Plate 2: A transverse section through testis of *C.gariepinus* diets fed (4/100 of *Cocos nucifera* water in compounded feed) this showing a partial organized lobules with a lumen recorded. X 100

C

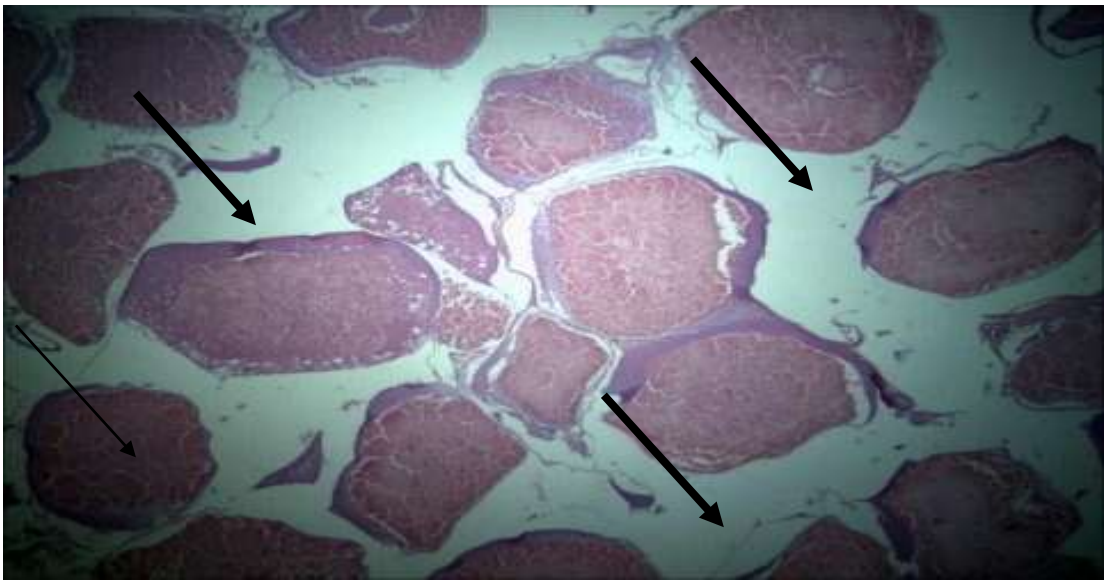

Plate 3: A transverse section through testis of *C.gariepinus* diet fed (6/100 of *Cocos nucifera* water in compounded feed) showing an organized lobules with much spermatozoa in the lumen, recorded. X 100

D

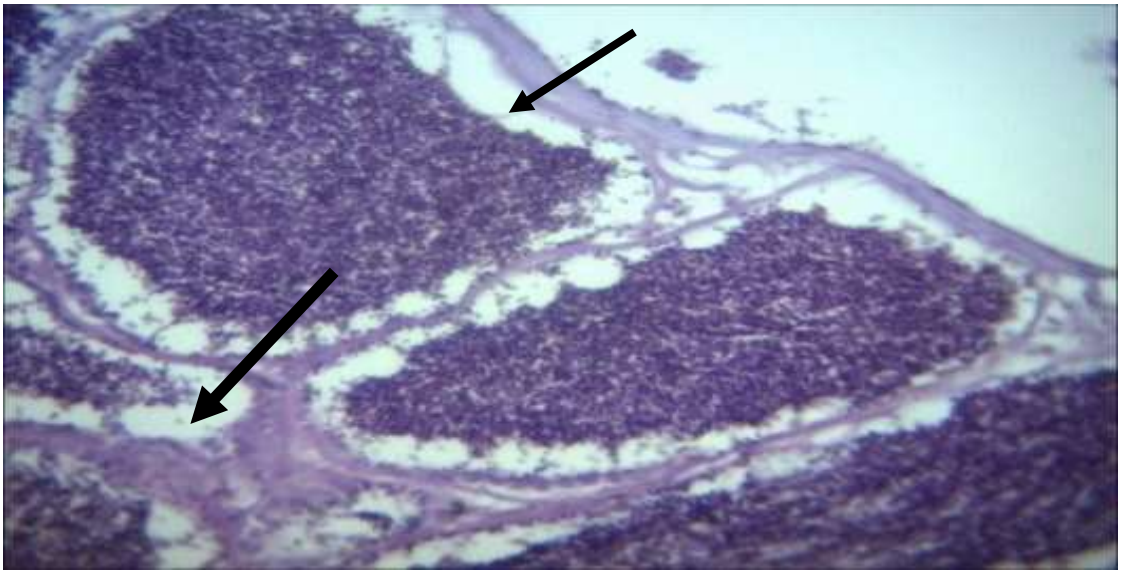

Plate 4: A transverse section through testis of *C.gariepinus* diet fed (8/100 of *Cocos nucifera* water in

compounded feed) showing an organized lobules with very much spermatozoa in lumen recorded. X 100

Histological description

A. **A.** Plate 1 Control (CNR) (0%/100% fed diet containing (*Cocos nucifera*) water compounded feed testis: No visible lesions seen. Spermatozoa found in the lumen, though appear scanty
B. Plate 2 (4 %/100% fed diet containing (*Cocos nucifera*) water compounded feed testis: seminiferous tubular show varying degree of a bit abundant spermatocytes (AS)
C. Plate 3 (6 %/100% fed diet containing (*Cocos nucifera*) water compounded feed testis: No visible lesions seen, matured spermatozoa present in the lumen (arrowheads)
D. Plate 4 (8 %/100% fed diet containing (*Cocos nucifera*) water compounded feed testis: Seminiferous tubuler show varies degree of abundant spermatozoa (AS).

while that of vitamins such as B1, B2, B3, B6 and vitamin E which had been investigated in relation to male fish fertility due to their role in spermatogenesis, sperm maturation motility which is also associate with increase sperm count and this could be attributed to the findings of Adewumi (2006) who reported 35 and 72% performance on egg and sperm quality fed with heated soyabeans based. The value of GSI obtained for male *C. gariepinus* broodstocks in this study falls within the range reported by Gamal Abd El-Nasser *et al.*, (2001). The parked cell volume (PCV), white blood cell WBC ($x10^9$/L), HB (g/dl), recorded highest mean value in the experimental diets compounded with

Cocus nucifera which apparently most important of heamatological tools that provide information about the health and physiological status of fish (Fazio *et al*., 2013a) as against the Control (CNR). The increase in growth and survival of the experimental fish *Clarias gariepinus* observed in the experiment could be as a result of the presence of potent antioxidant which are capable improving growth in fish (Venkatramalingam *et al*., 2007).

Meanwhile the physico-chemical parameter result observed fall within tolerable range when compare with FEPA (1991), (Lawson, 1995), (Loyd,(1992) and (Meade, 1989) standard values in fish culture. The histology of the fish *Clarias gariepinus* show no visible lesions, with matured spermatozoa present in the lumen of the experimental fish fed *Cocus nucifera* compounded feed.

CONCLUSION AND RECOMMENDATION

The result of the study revealed that the *Cocos nucifer*a had a mark effect in development of fish in terms of growth performance, gonad development and hematological changes in aquaculture. This conclude that *Cocos nucifera* water has the capacity if embraced by the aquaculture industry in reducing the challenges of milt quality in fish.

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