Available online www.unicrossjournals.com

JOURNAL OF CONTEMPORARY RESEARCH (JOCRES)

RESEARCH ARTICLE VOL. 2 (3) ISSN:2814-2241

Date Accepted: 31st December, 2023

Pages 125 - 136

WATER QUALITY CONTROL FOR*Clarias gariepinus*RAISED IN CONCRETE TANKSON LOCALLY PRODUCED DIETS AND HISTOPATHOLOGICAL EXAMINATION OF GONADS, LIVER.

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Abstract

Water quality and wellbeing are intimately related and directly proportional. Effect of water in fish tanks on the gonads and liver of the African Catfish (*Clarias gariepinus)* fed on locally produced diets was investigated for 22 weeks. The objective of the study was to assess the physico-chemical quality of the pond water and the histopathology of the gonads and liver of the fish reared in concrete tanks located in Calabar South. The physicochemical parameters were determined in water samples collected from the tanks for a period of 22 weeks (Weekly values of temperature, hydrogen potential, dissolved oxygen and ammonium were respectively between 27.09 ± 0.01 - 32.20 ± 1.06 °C ; 6.00±0.02 - 7.77 ± 0.17 mg/L; 4.76 ± 0.01- 6.98 ± 0.10 mg/L; and 0.00±0.00-0.17 ± 0.04 mg/L. The values of the measured parameters were within the normal values for the culture of *C. gariepinus*. Histological section of the gonads processed by Haematoxylin and eosin techniques showed normal developments of ovaries, testes and liver of fish fed diet A (0% inclusion of chicken offal), B(25% inclusion of chicken offal), C(50% inclusion of chicken offal), D (75% inclusion of chicken offal) and E (100% inclusion of chicken offal).Findings revealed that the water used which was from borehole water source was good and the locally formulated diets had no negative effect on the gonads and liver of the fish studied. The need to carefully consider inputs such as water source and appropriate management of fish tank water will result in high investment cost if high profitability is targeted in fish farming.

Keywords: Water Quality, Control, Clarias gariepinus, Histology, Gonads, Liver.

1.0 Introduction

Water quality and fish health are inextricably linked and directly proportional. It is acknowledged that water quality is the main factor that limits the productivity of aquatic ecosystems and the health of these ecosystems depends on their physicochemical characteristics (Niyoyitungiye *et al.,* 2019). Variations in any of the parameters have a significant impact on the species that live there (Eyo *et al.,* 2014). Fish, unlike other animals, eat and defecate in the same water they live in, and the quality of the water they live in has a direct effect on their wellbeing (Ali *et al.,* 2022). Consumed feed is not adequately processed into body flesh as water quality deteriorates. Poor growth is recorded, fish survival is harmed (Ali *et al.,* 2022. Temperature, dissolved oxygen, pH, and ammonia are all important water quality parameters to control while rearing fish.

Fish farmers should aim to keep the ponds/ tamks dissolve oxygen levels between 4mg/liter and saturation levels as much as possible. When dissolve oxygen levels are constantly too high and water is super saturated too well above 300 percent, fish may develop gas bubble disease (Bhatnagar and Devi, 2013). Fish will be stressed and may die if DO levels are less than 1.5mg/ liter, and the time it takes to reach the target weight of fish will be lengthened, resulting in a loss of investment (Ayim, 2017). Any potentially harmful metabolic waste, such as ammonia (NH_3) broken down into nitrites (NO_2) and then into nitrates, requires DO to be broken down into less harmful forms (NO_3).

Temperature of water has a major impact on the metabolism of fish (Bhatnagar and Devi, 2013). An optimal temperature range for African Catfish is 26°C to 32°C (FAO, 2014).When the water temperature in the ponds is reliably between 16^{o}C and 26^{o}C, feed intake decreases and the rate of fish growth slows dramatically (FAO, 2014).When fish are exposed to temperatures below 15^{o}C on a regular basis, their growth stops and death is imminent (Ayim, 2017).When the water temperature rises above 32°C, however, the impact on the African catfish will be negative (Ayim, 2017). This is due to the fact that oxygen does not dissolve readily in very hot water.

Hydrogen ion concentration in water is measured by pH, the pH level in the pond should be between 6.5 and 7.5 for the fish. Because of the acidity of the water, fish can die if it falls below 4mg/L. Fish will survive if the pH remains stable between 4 and 6, but they will develop slowly due to stress. According to APHA, AWWA, WEF (2012), low pH in pond water is an indicator of high CO_2 (carbon dioxide) in the water. Fish development is also slowed by high pH levels in pond water, which range from 9 to 11 mg/L (Eyo *et al.*, 2014). When the pH level increases above 11mg/L, the fish will inevitably die. Low pH encourages the development of more ionized ammonia, which is less harmful to fish. High pH in water, on the other hand, has the opposite effect.

Ammonia gas (NH_3), is highly water soluble. It's a natural by-product of organic nitrogen compound (Bhatnagar and Devi, 2013, Abdel-Satar *et al*., 2017). Fish cannot efficiently extract energy from feed when ammonia levels reach toxic levels, the fish will become lethargic and ultimately collapse into a coma and die if the ammonia levels rise too high (Eyo *et al.*, 2014).

2.0 Materials and methods

2.1 Study area

The fish farm (Andem and Sons Fish Farm Limited) is located in Ca1abar South Local Government Area, Cross River State at (latitude 4.9340699, 4°56' 2.6514''N and longitude 8.3282381, 8°19' 41.65716°E).

This is a privately owned farm where the fishes are reared in tanks (concrete, plastic and collapsible). The experimental fish were fed with test diets formulated using chicken offal as replacement for fish meal at varying inclusion levels together with other ingredients (fish meal (FM), wheat offal, soybean meal (SBM), bone meal, lysine, methionine, wheat flour, vitamin premix, sodium chloride (NaCl)). The tanks have an open pipe system completed by buried polyvinyl chloride (PVC) pipes that ensure the water supply. The pipes are equipped with a 1 mm mesh protective mosquito net.

2.2 Source of Water for the Experiment

Water was pumped into the tanks by an electrical pump and piped into the experimental tanks from borehole water (ground water).

2.3 Measurement of physico-chemical parameters

Every four days, water quality parameters including dissolved oxygen (DO), hydrogen ion concentration (pH), temperature (°C), and ammonia (NH_3) were tested. Water temperature was measured in-situ with a mercury in glass thermometer, while hydrogen potential (pH) and oxygen concentration were determined with Jenway meters (model 3050 England for DO and model 9070 for pH), and ammonia (NH_3) was determined ex-situ with a spectrophotometer at Cross River State Water Board's physicochemical laboratory in Calabar.

2.4 Dissection of fish for removal of gonads

The fish were sorted according to sex to avoid mix up. Differentiation of sexes was based on external features (elongated genital papilla for male and around serrated opening for female) and internal features such as gonad. The fish samples were then dissected for removal of gonads. The following body parameters was measured for each specimen: Total length (TL), Total weight (TW) and Gonad weight (GW). Measurements was taken to the nearest 0.1 cm and 0.1 g using measuring board for length and Metlar-2000D electronic weighing balance for weight (Eyo *et al.,* 2014).

2.5 Histopathology of tissues

This was conducted in the Histology Department, University of Calabar Teaching Hospital, Calabar. Tissues (female ovary, male testis and liver) extracted from fish reared with the five experimental diets, were subjected to manual tissue processing by haematoxylin and eosin techniques using the following procedures; fixation, dehydration, clearing, impregnation in wax, blocking out, sectioning, and photomicrography.

3.0 Statistical analysis

Data obtained from this study was expressed in Mean±SD using Microsoft excel statistical tool.

4.0 Results

4.1 Physico-chemical parameters of the experimental units

Water quality parameters measured in each of the experimental units included; pH, dissolved Oxygen (mg/L), water temperature (°C), and ammonia (mg/L), table 1. The temperature of the water was highest in T_C (32.20 ± 1.06 °C , week 22) and the least mean temperature was recorded in Tc and T_D to be 27.09 ± 0.01 °C (week 2) respectively. All other temperature values recorded fell between these extremes respectively. Results obtained for pH was highest in T_A with a mean value of 7.77 ± 0.17 mg/L (week 18) and the lowest pH was recorded in T_A (6.00 ± 0.02mg/L, week 2), all other pH values recorded fell amid these extremes.

Dissolved oxygen showed a higher mean value of6.98 ± 0.10 mg/L (week 10) in T_D, the least value was likewise recorded in T_B to be 4.76±0.01 mg/L (week 2), every other value fell between these extremes. Ammonia level was lowest in the first week to be 0.00 ± 0.00 mg/L (week 0) in $T_{A,}$ $T_{B,}$ $T_{C,}$ T_D and T_E respectively. The highest ammonia level was recorded in T_C(0.17 ± 0.04 mg/L, week 4), all other values fell between this two extremes. The physicochemical parameters including water temperature (^oC), pH, dissolved oxygen (mg/l) and ammonia (mg/l) were observed to be within the acceptable range for fresh water fish culture.

4.2 Histological sections of *C. gariepinus* tissues fed experimental feeds

The result for the histology of testes, ovaries, and liver of fish fed with the experimental diets showed normal changes in their developments. Plate 1, shows the process of dissection of the experimental fish for removal of gonads and liver. Plates 2a and 2b shows the male gonad (testes) and the female gonad (ovary) extracted from the experimental fish. The interstitial cells of the testes of *C. gariepinus* fed the five experimental diets showed normal testicular cells, normal distribution of the spermatocytes was also observed (Plate 3). The oocytes were distributed normally and fully matured in *C. gariepinus* fed with the five test diets (Plate 4). The result of the histology of the liver of fish fed with the five experimental diets also revealed normal distribution of the liver cell (hepatocytes) with reduced vacuolization (Plate 5).

TABLE 1: Mean Temperature (ºC), pH (mg/L), Dissolve oxygen (mg/L) and ammonia (NH_3 – mg/L) concentration of experimental units

Period	Parameters	TA	TB	TC	TD	TE	Period	parameters	TA	TB	TC	TD	TE
Week 0	Temperature (ºC)	27.23 ± 0.00	27.23± 0.00	27.23 ± 0.00	27.23±0.00	27.23 ± 0.00	Week 14	Temperature (oC)	28.6±0.03	28.01±0.02	27.8±0.05	27.00±0.01	27.3±0.07
	pH (mg/L)	6.63±0.00	6.63±0.00	6.63±0.00	6.63±0.00	6.63±0.00							
	Dissolve oxygen (DO)	6.61±0.00	6.61±0.00	6.61±0.00	6.61±0.00	6.61±0.00		pH (mg/L)	6.87±0.13	6.97±0.02	7.98±0.04	6.91±0.09	7.01±0.09
	Ammonia (NH_3)	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00		DO (mg/L)	6.01±0.03	6.96±0.02	6.06±0.05	6.88±0.22	6.01±0.03
								Ammonia (NH_{3}- mg/L)	0.16±0.08	0.03±0.02	0.02±0.02	0.03±0.10	0.17±0.03
Week 2	Temperature (ºC)	27.10 ± 0.02	27.15 ± 0.01	27.09 ± 0.02	27.09±0.01	27.22 ± 0.22	Week 16	Temperature (oC)	28.9±0.03	28.01±0.09	28.4±0.07	27.6±0.05	27.7±0.06
	pH (mg/L)	6.00±0.02	6.18±0.01	6.11±0.02	6.10±0,03	6.92±0.02		pH (mg/L)	7.33±0.76	6.98±0.10	6.97±0.07	6.77±0.33	7.04±0.01
	Dissolve oxygen (mg/L)	5.98±0.02	4.76±0.01	5.98±0.02	5.52±0.06	4.92±0.08		DO (mg/L)	5.06±0.04	6.98±0.10	6.97±0.07	5.02±0.08	5.04±0.01
	Ammonia (NH_3)	0.03±0.01	0.02±0.03	0.01±0.02	0.02±0.002	0.02±0.001		Ammonia (NH_{3}- mg/L)	0.02±0.01	0.04±0.10	0.13±0.07	0.03±0.01	0.13±0.01
Week 4	Temperature (ºC)	27.33 ± 0.73	27.18±0.04	27.37±0.03	28.00±0.02	27.47±0.53	Week 17	Temperature (oC)	29.4±0.04	28.6±0.03	28.5±0.05	28.0±0.08	28.4±0.06
	pH (mg/L)	6.98±0.03	6.76±0.02	6.66±0.04	6.98±0.02	7.08±0.02		pH (mg/L)	7.25±0.63	7.18±0.62	6.94±0.04	7.26±0.18	6.96±0.52
	Dissolve oxygen (DO)	5.83±0.17	5.96±0.04	5.95±0.06	6.15±0.76	5.08±0.02		DO (mg/L)	6.93±0.04	6.96±0.03	6.94±0.04	6.12±0.82	6.96±0.52
	Ammonia (NH_3)	0.02±0.03	0.03±0.02	0.17±0.04	0.04±0.01	0.03±0.002		Ammonia (NH_3)	0.04±0.02	0.13±0.01	0.03±0.02	0.03±0.03	0.13±0.06
Week 6	Temperature (ºC)	28.41±0.05	28.50±0.30	28.71±0.29	29.10±0.05	28.60±0.03	Week 18	Temperature (oC)	28.97±0.49	29.56±0.39	30.26±0.13	29.67±0.34	29.30±0.07
	pH (mg/L)	6.83±0.06	6.91±0.01	7.03±0.01	7.04±0.04	7.19±0.10		pH (mg/L)	7.77±0.17	7.16±0.72	7.00±0.01	6.66±0.06	7.02±0.04
	Dissolve oxygen (DO)	5.03±0.05	6.91±0.01	6.02±0.01	6.66±0.34	6.01±0.10		DO (mg/L)	6.04±0.02	6.96±0.04	6.87±0.13	6.77±0.03	6.22±0.82
	Ammonia (NH_3)	0.13±0.06	0.11±0.01	0.09±0.01	0.13±0.07	0.04±0.03		Ammonia (NH_3)	0.13±0.01	0.02±0.01	0.04±0.01	0.17±0.03	0.13±0.01
Week 8	Temperature (ºC)	28.22±0.32	28.27±0.53	28.33±0.66	28.27±0.06	28.27±0.06	Week 19	Temperature (oC)	30.80±0.40	30.67±0.13	30.27±0.07	31.03±0.31	31.37±0.12
	pH (mg/L)	7.13±0.08	7.96±0.03	7.12±0.08	7.25±0.06	7.02±0.02		pH (mg/L)	6.66±0.34	6.15±0.05	6.33±0.66	7.01±0.00	6.98±0.02
	Dissolve oxygen (DO)	6.92±0.06	6.96±0.03	6.92±0.01	6.00±0.01	6.02±0.02		DO (mg/L)	6.25±0.75	5.18±0.62	5.18±0.22	5.68±0.32	5.71±0.04
	Ammonia (NH_3)	0.17±0.03	0.20±0.06	0.13±0.01	0.13±0.03	0.13±0.03		Ammonia (NH_3)	0.03±0.06	0.03±0.06	0.02±0.08	0.03±0.00	0.13±0.01
Week 10	Temperature (ºC)	28.80±0.02	27.66±0.04	27.8±0.02	28.01±0.03	27.7±0.03	Week 20	Temperature (oC)	28.60±0.30	27.53±0.06	27.30±0.00	28.01±0.09	28.42±0.39
	pH (mg/L)	7.08±0.02	7.09±0.04	7.09±0.01	7.03±0.06	7.10±0.01		pH (mg/L)	6.98±0.04	7.04±0.94	7.05±0.33	7.10±0.02	6.96±0.52
	Dissolve oxygen (DO)	6.04±0.08	6.05±0.04	6.97±0.03	6.98±0.02	6.93±0.01		DO (mg/L)	6.01±0.03	6.82±0.18	6.00±0.02	5.98±0.04	5.14±0.02

	Ammonia (NH_3)	0.10±0.00	0.13±0.03	0.13±0.03	0.13±0.03	0.10±0.01		Ammonia (NH_3)	0.17±0.03	0.13±0.02	0.14±0.06	0.03±0.07	0.02±0.01
Week 12	Temperature (ºC)	27.7±0.05	27.6±0.04	27.8±0.02	28.01±0.03	27.7±0.03	Week 22	Temperature (oC)	30.93±0.64	32.11±0.94	32.20±1.06	32.11±0.94	31.10±0.06
								pH (mg/L)	7.16±0.74	7.17±0.32	7.13±0.03	7.15±0.75	7.13±0.76
	pH (mg/L)	6.92±0.04	7.02±0.02	6.90±0.05	7.01±0.91	6.98±0.03		DO (mg/L)	5.77±0.03	6.92±0.04	6.40±0.01	5.33±0.73	6.65±0.05
	Dissolve oxygen (DO)	5.90±0.10	5.25±0.76	5.90±0.05	5.02±0.98	5.98±0.03		Ammonia (NH_3)0	0.03±0.06	0.13±0.03	0.02±0.02	0.03±0.06	0.12±0.06
	Ammonia (NH_3)	0.03±0.02	0.10±0.02	0.13±0.05	0.13±0.06	0.17±0.03							

* Values represents the mean of the triplicate experimental unit

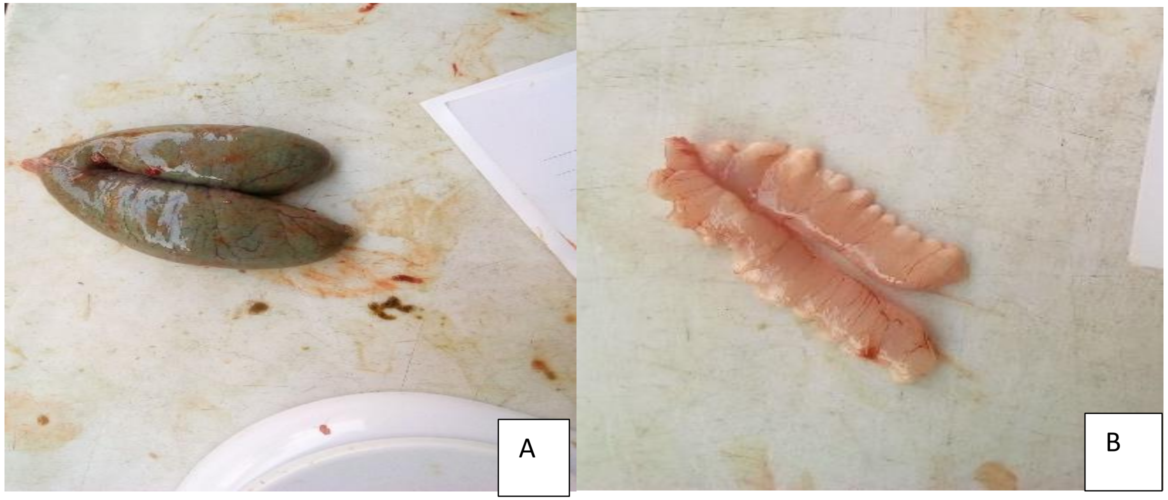

Plate 1: Dissection of fish for organ removal

Plate 2: Extraction of gonads in male and female*C. gariepinus*(A: Ovaries and B: Sperm)

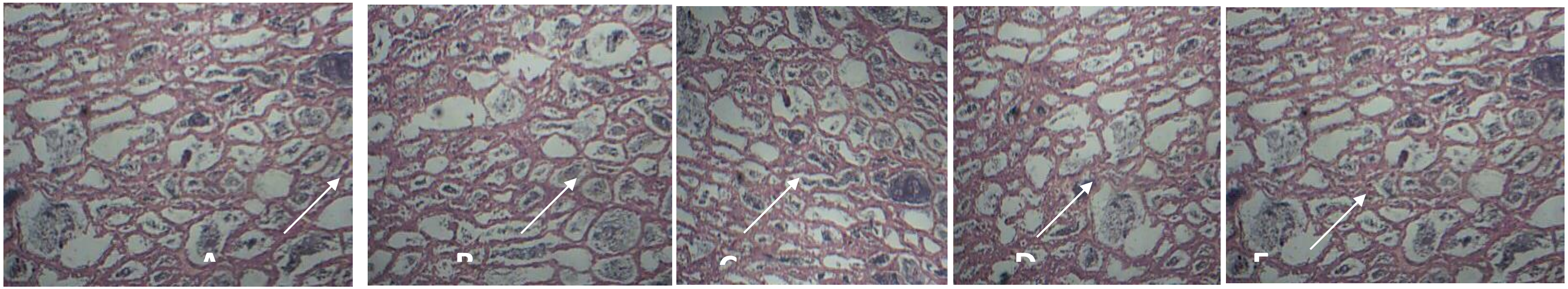

PLATE 3: Histological sections of *C. gariepinus* testes fed the five experimental diets. Arrow shows a normal distribution of the spermatocytes(spermatozoa) (X 40, H and E stains).

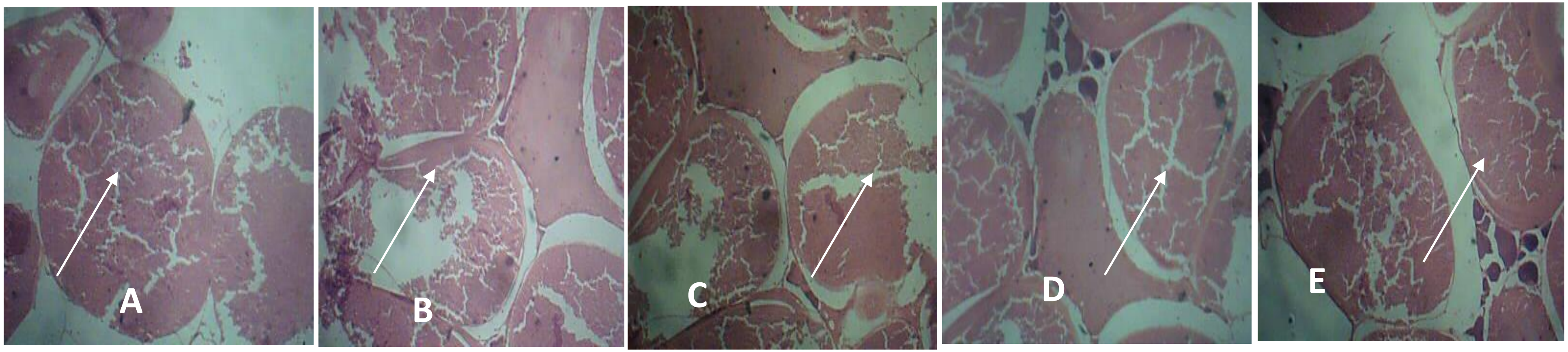

PLATE 4: Histological sections of *C. gariepinus* ovary fed the five experimental diets. Arrow shows a normal distribution of the oocytes (X 40, H and E stains)

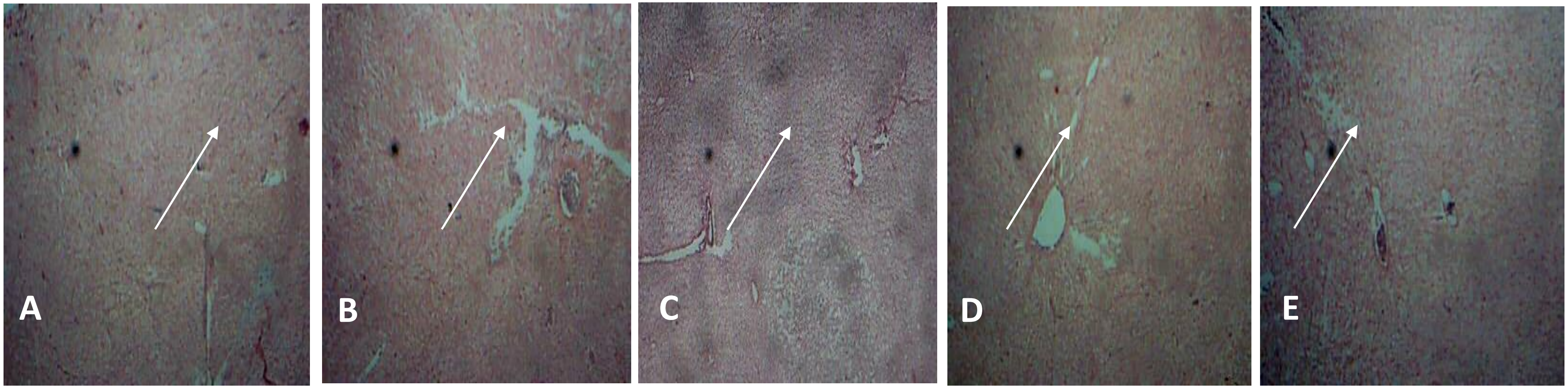

PLATE 5: Histological sections of *C. gariepinus* liver fed the five experimental diets. Arrow shows a normal distribution of the hepatocytes with reduced vacoulization(X 40, H and E stains)

5.0 Discussion

The measurement of the physico-chemical parameters of water gives a first estimate of its quality because they have an influence on several physical, chemical and biological processes (Coulibaly, 2013). They therefore help to assess the quality of the water and to know whether or not it is suitable for aquatic life (Noumon *et al*., 2015) in general and that of fish in particular. Results of this study showed slight variations in the physico-chemical parameters of the water in the tanks (tank A, tank B, tank C, tank D, and tank E), measured for twenty-two (22) weeks.

Temperature is a key factor that impacts the activities of fish (Ipungu *et al.*, 2015). It plays an essential role in fish reproduction (Ali *et al.*, 2022), in their metabolism and their growth. The monthly temperature variations measured in the different tanks as presented in table 1 fluctuate between 27.09 ± 0.01 °C and 32.20 ± 1.06 °C during this study and were within the normal ranges for the culture of *C. gariepinus*, however, the temperature values obtained are higher than those recorded by Makori *et al.* (2017) in Busia County in Kenya (24 to 26 °C). But the temperatures recorded in the water of the studied farm are favorable to aquatic life (Noumon *et al.*, 2015). Hydrogen potential (pH) is an essential parameter for maintaining life in the aquatic environment and its stability is very important in fish farming (Ali et al., 2022). Usually, a pH between 7 and 8.5 is ideal for biological productivity. Fish can be stressed in waters with a pH ranging from 4 to 6.5 and 9 to 11 (Kane *et al.* 2015). The pH values of the tanks monitored were between 6.10 ± 0.03mg/L to 7.77 ± 0.17 mg/L. Although, the basic nature of the water in most areas in Calabar is from (6.00 - 8.07). This basic character can also be due to the quality of the substrate (Faurie, 2011 and Ayim, 2017). On the other hand, the acidity of the waters of the tanks during the study period which was considerably from March to August is essentially due to the biological activity of the environment. It could also be explained by the mineralization of organic particles and the discharge of runoff containing chemicals from human activities. The pH values recorded during the sampling period are within the range recommended for the breeding of *C. gariepinus*; values between 5 and 11 (Eyo *et al.*, 2014 and Ayim, 2017). Our values are near to those reported by Ouattara *et al.* (2005) in Côte d'Ivoire (6.73 to 8.77) and Osman and El-Khateeb (2016) in Egypt (6.8 to 8.5). On the other hand, the minimum and maximum values recorded during the present study are lower than the minimum and maximum values obtained by Ghannam and Aly (2018) which are 7.00 and 8.90.

Dissolved oxygen is an environmental parameter that is not toxic to fish but an insufficient concentration of dissolved oxygen in the water can cause breathing difficulties for fish which can lead to their death (Chouti *et al.*, 2010). Dissolved oxygen concentrations recorded in this study were within the normal ranges for the rearing of fish, the different tanks waters were appropriately oxygenated throughout the sampling period. This is probably the result of the processes of dissolution of atmospheric oxygen by air/water contact (Martel *et al.*, 2013). The dissolved oxygen values observed in the present study are higher than those recorded by Coulibaly *et al.* (2018) which vary from 3.63 ± 0.60 to 4.70 ± 1.23 mg/L in a fish farm in Offoumpo in the department of Agboville (Cote d'Ivoire). Our results meet the required standard for rearing *C. gariepinus*. Ammonium levels observed in the different experimental tanks were suitable for fish farming however, the value recorded may have come from the decomposition of waste and excrement from fish and from uneaten food by the fish and aligns with the report of Bahri, (2009). The ammonium concentrations determined in the present study are similar to those reported by Ntumba *et al.* (2016) in the ponds in Kinshasa which are between 0.1 and 0.4 mg/L. Nonetheless, ammonium contents remained below threshold (2.3 mg/L) for fish farming.

Histopictures of testesfed the five experimental diets show a normal distribution of the spermatocytes (spermatozoa) (X 40, H and E stains) in all the experimental treatments studied. Theovaries and liver of *Clarias gariepinus* also showed normal developments of the gonads and liver across the fish studied in the different treatments (A to E), which implies that the locally formulated feed used in feeding the fish were good for consumption by fish. Finding from this study differed from the observations of Lawrence and Temiotan (2010) whose work on the histopathological effect of Gammalin 20 on African catfish (*Clarias gariepinus*) revealed histological changes in the gills and liver of the fish (hydropic degeneration of the gill rays, degeneration of the gill lamellae and necrosis) which are usually related to gills function disorders.

5.1 Conclusion

In the present study, we analyzed the water quality parameters of the waters of fish tanks reared with *C. gariepinus* when fed with a formulated diet containing Chicken Offal at varying concentrations. Four parameters, namely temperature, pH, dissolved oxygen, ammonium examined were observed to be within the normal range for the culture of fresh water fish species. The results from the histopathological analysis revealed normal development of the gonads and liver of the experimental fish and indirectly shows that *C. gariepinus* could tolerate up to 100% replacement level of fish meal and other sources of animal protein with Chicken offal without any negative influence on the *C. gariepinu s*health. This study has provided baseline information for extensive, semi-intensive and intending fish farmers who are challenged by the high cost of fish feeds could alternatively formulate their fish feed using locally available ingredients. And with good management of physicochemical parameters of their ponds, fishes will grow well without any disorder of any organs such as; gonads and liver.

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