



Assessment of Toxicity of Inorganic Fertilizers on *Clarias gariepinus* Juveniles Towards Attaining SDG No. 2 and No. 14

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ABSTRACT

This study evaluated the effect of acute toxicity of three commercial inorganic fertilizers NPK 15:15:15, NPK-1; NK 20:20:20, NPK-2; urea and Single super phosphate on *Clarias gariepinus* juveniles in static condition. The concentration of NPK-1, NPK-2, urea and single superphosphate that killed 50% of the African Catfish with 96-h LC₅₀ were 6.25g/l, 7.25g/l, 16.25 and 12.50g/l, respectively. For water quality parameters, there was no significant difference ($p < 0.05$) in the mean of (temperature, Ph and alkalinity, while there was significant difference ($p < 0.05$) in Dissolve oxygen. However, urea treatment parameters were found to have highest numerical value among the treatments in the water treatment parameters. Furthermore, there was significant difference ($p < 0.05$) in the water treatment parameters. Furthermore, there was significant difference ($p < 0.05$) in the haemoglobin percentage among the treatments while no significant difference ($p < 0.05$) was observed in the total count of red blood cell. Planktons were more abundant in urea treatment but more diverse in Single Superphosphate treatment. All the treatments caused damage on the tissues among which are; nuclear pycnosis in the liver tissue and hyperplasia of mucous cell of skin and gill tissue. However, the most damages were observed on the urea treatments. The study concluded that urea is more toxic to the fish than other inorganic fertilizers.

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INTRODUCTION

One of the major environmental issues of our time was the growing concern about water quality suitable for use by humans and animals Calamari and Naeve, (2004). The daily activities of man in one way or the other affect the aquatic environment negatively. These activities, which include the discharge of pollutants into streams and river systems, such as agricultural fertilizers of different types, pesticides, insecticides and industrial effluents, pollute the water bodies and alter ecological balance, EU, (1998). These pollutants influence the quality of these water bodies which is of high importance in the aquatic ecosystem balance and consequently affect the survival of aquatic organisms inhabiting such environments, Odiete, (1999). It was a known fact that water quality conditions are constantly being threatened by pollution. The rivers and coastal water bodies are presently exposed to increasing quantities and concentrations of both natural as well as anthropogenically derived contaminants Ezeka, (2004). Environmental concern about intensive agricultural practices and excessive or inappropriate use of chemical fertilizers calls for some global action among environmentally conscious individuals and other stakeholders, Nychas, (1990).

A fertilizer is any material, organic or inorganic, natural or synthetic, that supplies plants with the necessary nutrients for growth and optimum yield (Addiscott, Whitmore and Powlson, 2001) or a substance added to water to increase the production of natural fish food organisms; (Nwadukwe, 1995). Inorganic (or chemical) fertilizers are fertilizers mined from mineral deposits with little processing for example, lime, potash, or phosphate rock, or industrially manufactured through chemical processes, for example NPK and Urea (Amadi, 2001). Inorganic fertilizers vary in appearance depending on the process of manufacture. The particles can be of many different sizes and shapes (crystals, pellets, granules or dust) and the fertilizer grades can include straight fertilizers (containing one nutrient element only), compound fertilizers (containing two or more nutrients usually combined in a homogeneous mixture by chemical interaction) and fertilizer blends (formed by physically blending mineral fertilizers to obtain desired nutrient rates; (Alexander, (2006). For the sake of this study, NPK, Urea and Single superphosphate will be used. The importance of chemical fertilizers in agricultural production is highly indispensable and is widely acceptable by an ever increasing number of farmers, not only because fertilizers help condition the fragmented and nutrient-depleted soil for further production and boost soil resistance to erosion, but also that it encourages vegetal cover (Almazan and Boyd, 1978).

LITERATURE REVIEW

Inorganic nitrogen (NH_4^- , NO_2^- and NO_3^-) may be present naturally in aquatic ecosystems as a result

of atmospheric deposition, surface and groundwater runoff, dissolution of nitrogen-rich geological deposits, and biological degradation of organic matter, Wetzel, (2001) and Rabalais, (2002). However, activities of human species have substantially altered global nitrogen cycle, increasing both availability and mobility of nitrogen over large regions of the Earth; (Vitousek, Aber, Howarth, Likens, Matson, and Schindler, (2007) and Galloway and Cowling, (2002). Inorganic nitrogen (NH_4^- , NO_2^- , and NO_3^-) enters aquatic ecosystems via anthropogenic sources such as animal farming, urban and agricultural runoff, industrial wastes, and sewage effluents; (Wetzel, (2001) and Rabalais, (2002). Also, the atmospheric deposition of inorganic nitrogen (mainly in the form of NO_3^-) has dramatically increased because of the extensive use of nitrogen fertilizers and huge combustion of fossil fuels, Moomaw, (2002) and Boumans, Fraters and Van Dreht, (2004). As a result, concentrations of nitrate in ground and surface waters are increasing around the world, causing one of most prevalent environmental problems responsible for water quality degradation on a worldwide scale, Wetzel, (2001); Rabalais, (2002) and Smith, (2003).

The use of fertilizer in aquaculture is important for pond fertilization as it increases fish production as a result of nutrient availability for primary production. In aquaculture, fertilizers have been used in various forms and quantities to enhance fish production for greater abundance of fish food organisms, but excessive uses of fertilizer may have adverse effect on water quality and can also cause gill damage, Haygart hand Jarvis, (2002).

Fish and other aquatic animals are subject to a broad variety of stressors because their homeostatic mechanisms are highly dependent on prevailing conditions in their immediate surroundings. The biochemical and histological changes occurring in the body could serve as potent indicators of stress Jhingran, (2002). The Genus *Clarias* is widespread in Africa and South East Asia and its utilization for fish culture has significantly increased during the last few years Bard, *et al.*, (2006) and it was widely cultivated in Nigeria Omotoyin, (2007).

NPK, Urea and SSP which are inorganic sources of fertilizer, are most widely used in agriculture as an economical, balanced and effective fertilizer. Their efficacy in promoting growth of plants and increasing production is incontestable. However, in animals it is responsible for the manifestation of multiple disorders ranging from inhibition of growth to mortality. Their use as fertilizer has been shown to diminish fish production and induce mortality in aquaculture Jhingran, (2002). Applying different fertilizers in fish ponds indicated a negative correlation between fish production and levels of organic nitrogen, Meehean and Marzulli, (2005).

The study was undertaken to monitor the response of fishes to exogenous NPK, Urea and SSP fertilizers. Multiple parameters will be examined including behavioural changes, bio chemical changes, osmoregulation and histological changes which will

help in providing the insight to the generated response.

Sustainable Development Goal 2 (SDG2 or Global Goal 2) aims to achieve “zero hunger”. It is one of the 17 Sustainable Development Goals established by the United Nations in 2015. The official wording is “End hunger, achieve food security and improved nutrition, and promote sustainable agriculture”. Sustainable Development Goal 2 is about creating a world free of hunger by 2030. Source: This sustainable Development Goals Report 2022.

Sustainable Development Goal 14 is about “life below water” and is one of the 17 sustainable Development Goals established by the United Nations in 2015. The official wording is to “Conserve and sustainably use the oceans, seas and marine resources for sustainable development”. Healthy oceans and seas are essential to human existence and life on earth.

The oceans are home to seahorse, dolphins, whales, corals, and many other living creatures. Oceans are our planets life support as they provide water, food and help regulate the weather. Oceans also provide jobs for more than 3 billions people who depend on marine biodiversity for their livelihood.

STATEMENT OF THE PROBLEM

Despite the very useful nature of fishes as a major food nutrient in Nigeria, there has been a growing concern on the environmental impact of water quantity and quality of fishes available to the nation. It has been argued that pollutant discharge such as pesticides, insecticides and other industrial effluents pollute water bodies and thus affecting the quality of healthy aquatic animals available for human consumption. One worrisome situation is the increase in carcinogenic infections in Nigeria. Could this be due to excess chemical in fishes? There has also been decline in quantity of fishes in the aquatic environments in Nigeria, what could be responsible for this? There was an urgent need to investigate toxicities of inorganic fertilizers on *clarias gariepinus* juveniles in Nigeria.

AIM AND OBJECTIVES OF THE STUDY

The present study aims to determine the toxicities of inorganic fertilizers on *clarias gariepinus* juveniles using a static test system.

The specific objectives are to;

- i. Determine the growth rate of *clarias gariepinus* juveniles exposed to different inorganic fertilizers (NPK, Urea and SSP);
- ii. Investigate the effect of different fertilizers on haematological parameters;
- iii. Examine the impact of using different inorganic fertilizers on the plankton abundance and
- iv. Investigate the histo-pathological effect of different inorganic fertilizers to *clarias gariepinus juvenile*

THE SCOPE OF THE STUDY

This study was limited to the use of inorganic fertilizers which are; NPK 15:15:15, NPK 20:10:10, Urea and Single Superphosphate which are the common fertilizers that fish farmers normally use to fertilize their ponds to stimulate the growth of planktons. This study was conducted at the teaching and research farm of the Federal College of Education, Pankshin from September 2022 to September, 2023.

THE JUSTIFICATION OF THE STUDY/ SIGNIFICANCE

This study will be beneficial to the following set of people; fish farmers, environmental scientist, government agencies, and other researchers.

Fish farmers: This study will benefit fish farmers especially those that are rearing African Cat fish, they will be able to decide the type of inorganic fertilizer that is suitable for the survivability of the fishes. They will also be able to know the fertilizer that can support the growth of the *clarias gariepinus*. Environmental scientist will also find this study informative, this study will x-ray acute toxicity effect of the most common types of fertilizers available to farmers and its impact on the quality of water, water that is not safe for fish, might not be considered suitable for human. More so, this study will also be of benefit to government agencies that are responsible for the regulation of the use of agricultural chemicals. This study will expose the effect of over-using chemicals and how un-safe the residues are.

RESEARCH METHODOLOGY

3.1 Study Area

The study was undertaken in the fish pond of the Department of Agriculture, Federal College of Education Pankshin. The geographical area covered by the study is Pankshin Local Government Area. Pankshin Local Government is one of the oldest local government area in Plateau State. It is about 120 kilometers from Jos and is situated on the West to Mangu Local Government Area to the East, Mikang and Qua'an Pan Local Government Area to the South. Pankshin Local Government Area covers an area of 1.434 square kilometer and has a population of about 168,718 based on the census that was conducted in 2004. The main ethnic and language groups in the Local Government Area are the Ngas, Mupun, Dyis, Kadung, Jing, Chip and Lankan among others. The area is blessed with socio-economic infrastructure and has agrarian economy (Centered upon the production, consumption, trade and sales of agricultural commodities, including plants and livestock) as the mainstay of the people who are also industrious.

3.2 Materials

- Jen way portable Ph meter model 350
- Aqueous bovines solution
- Improved Neubaver heomocytometer slide
- Leishman's stain
- Haemoglobunometer
- Jen way portable dissolved Oxygen meter model 970
- Hcl acid
- Saline solution
- Methyl indication
- Nacl Solution
- Sensitive scale
- Thermometer
- Stopper bottle
- Pipette
- Conical flask

3.3 Management of the Experimental Fish

The juvenile African Catfish were purchased from the local hatchery in Makurdi, Benue state. The juvenile African cat fish were acclimatized in three (3) plastic bamboo containers before the experiment commenced. To mitigate environmental factors as a result of the exposure of the plastic materials and the water used for the experiment to atmospheric temperature, an open shed with rough thatched was place over the water holding vessels with its sides rounded up with wire mesh up to three feet high to prevent the entrance of rodents and human factors. The fish were fed daily with 1.5mm to 4.5mm feed size of the pelleted commercial feeds at 3% body weight throughout the twelve weeks' experimental period. Also measured was the temperature of the water using the thermometer and the pH using the pH meter before the daily feeding practice. The dissolved oxygen was monitored and measured weekly using the dissolved oxygen meter. The volume of the water will be maintained at 0.18m³. The top of the vessels were also covered with 5mm mesh size net to protect the stocks from jumping out while the water in the vessels were changed bi-weekly to avoid the build-up of nitrates and nitrites as effluent.

3.4 Acclimatization

The fishes were acclimatized for 14 days. The 3000 fishes were divided into ten (10) large plastic bamboo containers having 300 fishes each with thirty litres of dechlorinated water. The containers were aerated during this period, and water was renewed daily to discard faecal material as well as left-over food. The fish were fed twice daily with a 45% crude protein diet at 3% of their body weight, half at 08:00 and 16:00 hours, respectively. During this period, dead and abnormal individuals were immediately removed. It was from the acclimated population that healthy test fishes were carefully selected.

3.5 Acute Toxicity Trial Runs

After the acclimation period trial rounds of acute toxicity were done to determine the different concentrations of each of the fertilizers to be used for the experiments, standard methods; (UNEP, 1989) were used in carrying out the static bioassay with continuous aeration experiment. Fishes were fasted 24hours prior to exposure time, the same concentration of the fertilizers was to be delivered into the glass aquaria.

3.6 Experimental procedures

Exposure concentrations of NPK1, NPK2, Urea and SSP fertilizers were prepared at 6.25g/l, 7.25g/l, 16.25 and 12.50g/l respectively. Treatment 1 contained NPK 15:15:15, treatment 2 NPK 20:10:10, treatment 3, Urea, while treatment 4 contained Single superphosphate fertilizer. Each of the treatments was replicated three times. Twelve glass containers were randomly labelled and each filled with dechlorinated tap water up to 8 litres mark for each treatment. The different concentrations were prepared by dissolving directly, different weights of the fertilizers in the dechlorinated tap water, APHA, (2005). The solution was stirred with a glass rod to obtain a homogenous mixture. Within an hour, the containers were randomly stocked with twenty (20) fish each using a scoop net. The test fish were not fed twenty-four hours prior to the experiment and during the ninety-six hours' exposure period. Test solutions from each tank were drained out completely every morning and the fish removed carefully with a scoop net and kept in a thirty litre plastic container. Fresh solutions will be prepared and the fish will be carefully put back. Fresh solutions were prepared daily.

3.7 Water quality parameters

Temperature, dissolved oxygen, pH, and alkalinity of the various test media were determined at 24th, 48th, 72nd and 96th hours intervals during the experimental period as suggested by APHA, (2005) and ASTM, (2004).

3.7.2 Dissolved oxygen

The dissolved oxygen content was assessed with a Dissolved Oxygen Meter.

3.7.3 pH

The pH was determined with a digital pH meter (Hannah product Portugal, Model HA 989).

3.7.4 Alkalinity

The procedure involved the collection of water samples from each Aquarium in stopper bottles. 25ml of the sample were pipetted into a conical flask and 5 drops of methyl red indicator and bromocresol green were added and titrated with standard HCL acid

(0.01N) from a 10ml burette, with continuous shaking until the colour changed from blue to pale pink. The endpoint of pH were read with a pH meter.

3.8 Histological analysis

Two Fish from each aquarium was sacrificed after 15 days of exposure. Blood was collected from the post caudal vein and serum was extracted after centrifugation and stored in -20°C. Liver and muscles (without skin) was quickly extracted, washed with 0.6 % saline weighted, blotted and kept frozen at -20°C for further biochemical analysis. Similarly, kidney, gills, air breathing organs (ABO) and skin will also be preserved in aqueous Bouines solution for histological study at NVRI Jos.

3.9 Haematological Methods

Erythrocyte Count: Total count of RBCs were done with the help of the improved Neubauer Haemocytometer slide and studied under light microscope (Olympus CH₂Oi). The erythrocyte count in fish blood was determined by using 0.85% NaCl diluting fluid. The dilution fluid is 1 part blood: 200 parts diluting fluid. The counting was done in 5 of 25 small square of haemocytometer slide: 4 small squares at four different corners and a central small square.

Calculation: The number of RBCs/mm³ of blood = the total number of cells counted X dilution X 4000 / the number of small squares in which counting was done.

Differential Count of Leukocyte: Blood film was prepared with the Leishman's stain following the standard method used in human blood film preparation. The counting was done in narrow longitudinal strips of the blood film starting from one end of the film to the other end, avoiding lateral edges. While counting the number, different types of leukocytes will be observed. The counting was replicated three times.

Haemoglobin Percentage: The Haemoglobin percentage was estimated by Sahil's Haemoglobinometer. The blood was blown out from the haemoglobin pipette into the haemoglobin tube containing N/10 HCl. The contents of the haemoglobin tube was stirred with glass stirrer and allowed to stand for 10 to 20 minutes. Then N/10 HCl drop by drop was added to the haemoglobin tube while stirring with the glass rod till the colour in the haemoglobin tube match exactly with that of the standard brown plates. Dilution of blood was read off on the haemoglobin tube in terms g/100ml.

3.10 Plankton Identification and Quantification

Plankton identification was done on a light microscope (BRESSER, Germany) 100X magnification using identification keys according to UNESCO (2007). 1 ml of water sample was taken from the collecting bottles (200 ml bottles) using

micropipette and transferred to a Sedgewick Rafter cell (Wild Supply Company, England) then covered with slide at the top and placed under microscope. From 10 randomly selected squares of cell, planktonic organisms were enumerated and numerical abundance was calculated. *Phytoplankton* and *Zooplankton* abundance will be calculated using the following formulas as described by Greenberg *et al.* (1992) and Wetzel and Likens (1991), respectively:

$$\text{Phytoplankton Abundance} = C/F \times V$$

Where C is the number of organisms counted, F is the number of fields counted and V is the volume of sample settled.

$$\text{Zooplankton } l = C \times V_a / N_b \times V_c$$

Where C is the number of *zooplankton* counted, V_a is the volume of the concentrated sample (l), V_b is the volume of counted sample (l) and V_c is the volume of water filtered (l).

In case of plankton diversity, Shannon-Wiener diversity index (H') and evenness (J') were used. Diversity index of plankton was calculated by using the formula as described by Krebs (2007):

$$H = \frac{n \ln(n) - \sum_{i=1}^k \ln(f_i)}{n}$$

Where k is the number of categories, f_i is the number of observations in category i, n is the sample size.

Species evenness or homogeneity or relative diversity (J') was calculated from the observed species diversity and from the equation of H_{max} as described by Sundar *et al.* [32]. Index of species evenness was measured by using the following formula:

$$J' = \frac{H'}{h_{max}}$$

where, H' = ln(k)

3.11 Data Analysis

Water quality parameter was determined at fixed intervals of 24, 48, 72 and 96 hours respectively. Mortality of the fish species in each tank will be observed and recorded at fixed intervals of 24, 48, 72 and 96 hours, respectively. Dead fishes were removed immediately from the test media, to prevent pollution in test media. A fish was considered dead; when there was lack of movement and reaction to gentle prodding with a glass rod. Other unusual signs of stress were equally monitored, such as uncoordinated and irregular swimming pattern, vertical erection, overturning, and restlessness, jumping out of the tank and gasping for air.

Each set of results obtained from these experiments were analysed using analysis of variance

at 5% probability level among the four treatments. Analysis of the lethal concentration (LC50) values for the 24th, 48th, 72nd and 96th hours with their associated confidence intervals for the various concentrations of NPK and Urea fertilizers were determined by Probit Analysis using Statistical Package for the Social Sciences (SPSS) Data Editor version 25.0.

STATISTICAL ANALYSIS

Data obtained were subjected to analysis of variance and difference between means were separated by Duncan's multiple ranges test (DMRT)

4.0 RESULTS

4.1 Results on growth performance exposed to different commercial fertilizers

Data on the growth performance and body length of African catfish raised in different fertilizer media are presented in Table 4.1. Significant differences ($p < 0.05$) were recorded on the mean weight gain and mean length gain of catfish across the treatment. The heaviest fish was found on T4 (105.6g), followed by

T1(103.7g), the least was found in T3(75.56g) while T2 had 90.75g. The average daily weight recorded were highest in T4(2.85g), followed by T1 (2.80g), while T2 and T3 had 2.45 and 2.03g respectively. The average gain weight recorded were highest in T4 (88.15g), followed by T1 (85.7g), while T2 and T3 had 85.7 and 69.63g respectively. The specific growth rate also showed no significant difference at ($p < 0.05$) with value as follows: T4 (0.49), T1 (0.48), T2 (0.46) and T3 (0.43). No significant difference ($p < 0.05$) was observed on the average final length, however, final mean length was found to be highest in T4 (32.21cm) followed by T1 (29.01cm), the least that was recorded was 24.7cm found in T3 while T2 had 26cm. Furthermore, the mean length gain was also found to also be highest in T4 (25.41cm), followed by T1 (22.50cm), while the least was found in T3 (18.7cm). The highest mortality was recorded in T3 (13.33%), followed by T2 (11.67%), T4 had 10% mortality while the least was found in T1 (8.33%). There was no significant difference ($p < 0.05$) in the feed conversion ratio with a range of 1.04 - 1.61, with the highest found in Urea (1.61), while the least was found in T2 (1.04). Generally, on the growth performance of the fish, Urea was found to be more toxic to the animals and it impeded the growth parameters.

Table 4.1: Growth performance of African Catfish exposed to different fertilizers.

Parameter	TREATMENTS			
	N.P.K-1(T1)	N.P.K-2(T2)	UREA(T3)	SSP(T4)
Initial number stocked	60	60	60	60
Initial mean body weight (g)	18.00	21.12	20.74	17.45
Final mean body weight gain (g)	103.7	90.75	75.56	105.6
Mean body weight gain (g)/fish	85.7 ^a	69.63 ^b	54.81 ^c	88.15 ^a
Average daily weight gain (g)	2.80	2.45	2.03	2.85
Specific Growth Rate	0.48	0.46	0.43	0.49
Initial mean length (cm)	6.51	6.00	6.00	6.80
Final mean length (cm)	29.01	26.0	24.7	32.21
Mean length gain	22.50 ^a	20.0 ^b	18.7 ^c	25.41 ^a
Mortality	5.0	7.0	8.0	6.0
Mortality (%)	8.33	11.67	13.33	10.00
Mean daily feed consumed (g)/fish	2.94	2.55	3.27	3.09
Feed conversion ratio (FCR)	1.05	1.04	1.61	1.08

4.2 Water quality Parameters

The result of the analysis of water quality parameters is presented in Table 4.2, 4.3, 4.4, 4.5. Treatment with inorganic commercial fertilizers caused changes in the water quality parameters. Water temperature inflicts prominent effects in fish life directly or indirectly influencing the aquatic environment. According to Kaur, Masud and Khan (2015), every organism has specific survival range of environmental temperature for its efficient existence and beyond these limits, conditions become lethal. Fish being a cold blooded animal is affected by the temperature of

surrounding water in terms of the body temperature, growth rate, feed consumption, feed conversion and other body function. Jhingran (1982) observed that carps thrive well in the temperature range of 18.3-37.8°C.

Temperature data result is presented in Table 4.2 and Figure 4.1. There was no significant difference down the treatments and across the experimental week. At the first week, the highest temperature was found in T3 (29.0°C) followed by T1 (28.42°C), T2 and T4 had 28.33 and 28.21°C. At the second week, the highest temperature was also found in T3 (27.89°C), followed by T1 (27.46°C) while the

least was observed in T4 (27.20°C). Similarly, after the 3rd week, the highest temperature was also found in T3 (26.81°C), followed by T1 (26.76°C), T2 and T4 had 26.21 and 26.11°C respectively. Similar trend was observed from both 4th and 5th week where the highest of temperature was found in Urea treatment while the lowest temperature was recorded in SSP

treated medium. Generally, all the treatments have temperature (23°C - 29°C) that is optimum for the rearing of *Clarias gariepinus* throughout the period of this experiment, the same range of water temperature as suggested Adeniji and Ovie (2009).

Table 4.2: Effect of different commercial fertilizer on water temperature (°C)

Treatment	Duration in Weeks				
	1 st	2 nd	3 rd	4 th	5 th
NPK-1 (T1)	28.42±7.51	27.46±4.01	26.76±3.31	24.51±3.27	23.24±2.15
NPK-2 (T2)	28.33±6.01	27.21±4.29	26.21±3.26	24.33±3.11	23.27±2.15
Urea (T3)	29.01±7.32	27.89±5.50	26.81±3.67	24.55±3.61	23.77±2.51
SSP (T4)	28.21±6.78	27.20±4.11	26.11±3.29	24.36±3.05	23.03±2.11
LOS	ns	ns	ns	ns	ns
P-value	0.1413	0.9212	0.1136	0.1083	0.9911

SSP= Single Super Phosphate, LOS = level of significance, ns= not significant

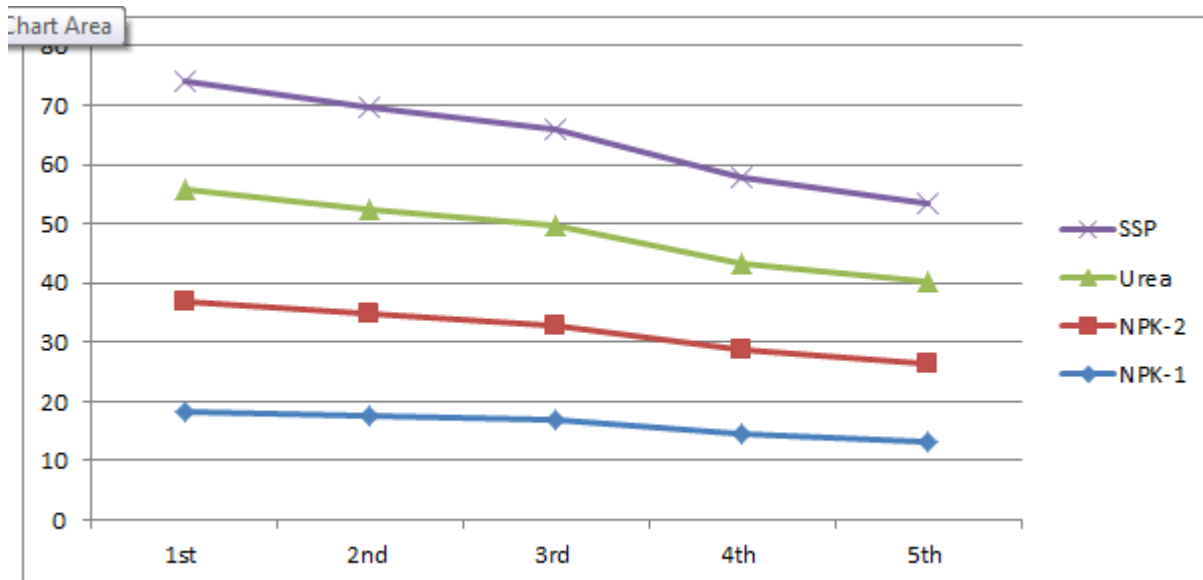


Figure 4.1: Effect of inorganic fertilizers on the temperature

The result of the effect of fertilizer on pH is presented in Table 4.3 and Figure 4.2. There was no significant different at ($p < 0.05$) all though the treatments and across the experimental weeks. At week 1, the highest pH was recorded on T1 (9.08) followed by T2 (9.03), T3 had (9.03) while the least was found in T4 (9.00). Whereas, by the 2nd week, the highest was found in T1(8.95) followed by T4 (8.91), T2 had 8.52 while the least was recorded on T3 (8.33).

Furthermore, at week 3, the highest pH was recorded in T1 (8.77) followed by T4 (8.74) while T2 and T3 had pH 8.65 and 8.70 respectively. More so, at week 4, the highest pH was recorded in both T1 and T2 (8.75), while T3 and T4 had 8.70 and 8.71 respectively. At the expiration of the experiment, the highest pH was found in T2 (8.83) followed by T4 (8.79) while T2 and T3 had 8.83 and 8.75 respectively.

Table 4.3: Effect of different commercial fertilizer on water pH

Treatment	Duration in Weeks				
	1 st	2 nd	3 rd	4 th	5 th
NPK-1(T1)	9.09±0.21	8.95±0.10	8.77±0.11	8.75±0.08	8.72±0.05
NPK-2 (T2)	9.08±0.32	8.52±0.05	8.65±0.04	8.75±0.07	8.83±0.10
Urea (T3)	9.03±0.19	8.33±0.02	8.70±0.06	8.70±0.06	8.75±0.07
SSP (T4)	9.00±0.27	8.91±0.03	8.74±0.08	8.71±0.09	8.79±0.15
LOS	ns	Ns	Ns	ns	ns
P-value	0.1311	0.2214	0.1413	0.1136	0.2121

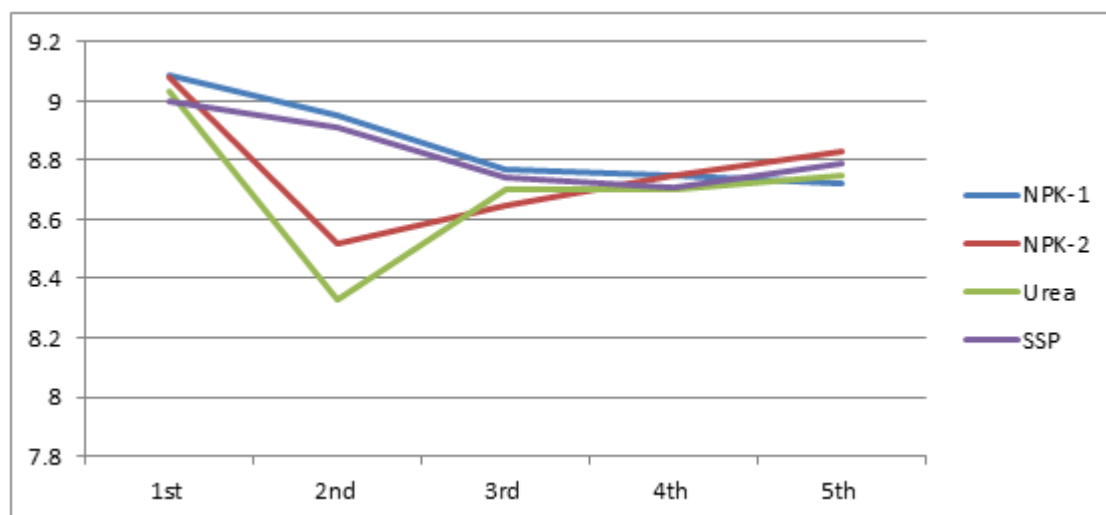


Figure 4.2: Effect of inorganic fertilizers on pH

The result on the dissolve oxygen (DO) is presented in Table 4.4 and Figure 4.3. The result revealed that there is no significant difference ($P < 0.05$) between the different means of the treatments except at week five where the treatment had significant impact on the DO level. At the end of the first week, the highest DO was found in T4 (6.53mg/L) followed by T3 (6.21mg/L) whereas, T1 and T2 had 5.71 and 5.31 ppm respectively. Similarly, at the 2nd week T4 also had the highest DO (4.56mg/L) followed by T3 (4.41mg/L),

the lowest was found in T1 (3.45mg/L) while T2 had (4.12mg/L). Furthermore, it was observed at 3rd week that the highest DO was found in T4 (4.87mg/L) followed by T2 (4.00mg/L), the least was found in T3 (3.40mg/L) while T1 had (3.45). On the 4th week, the T4 (5.28) followed by T2 (4.11mg/L), the least was found in T3 (3.07mg/L) while T1 had (3.08mg/L). At the end the experimental period, T4 had the highest (5.25mg/L) followed by T2 (4.58mg/L), T1 had (3.35mg/L) while T4 had the least 2.31mg/L.

Table 4.4: Effect of different commercial fertilizer on Dissolve Oxygen (mg/L)

Treatment	Duration in Weeks				
	1 st	2 nd	3 rd	4 th	5 th
NPK-1 (T1)	5.71±0.07	3.45±0.05	3.45±0.05	3.08±0.01	3.35±0.06 ^a
NPK-2 (T2)	5.31±0.10	4.12±0.06	4.00±0.06	4.11±0.02	4.58±0.05 ^b
Urea (T3)	6.21±0.03	4.41±0.03	3.40±0.03	3.07±0.01	2.31±0.02 ^c
SSP (T4)	6.53±0.02	4.56±0.02	4.87±0.02	5.28±0.02	5.25±0.01 ^b
LOS	Ns	ns	ns	ns	*
P-value	0.0711	0.2214	0.1311	0.1136	0.02

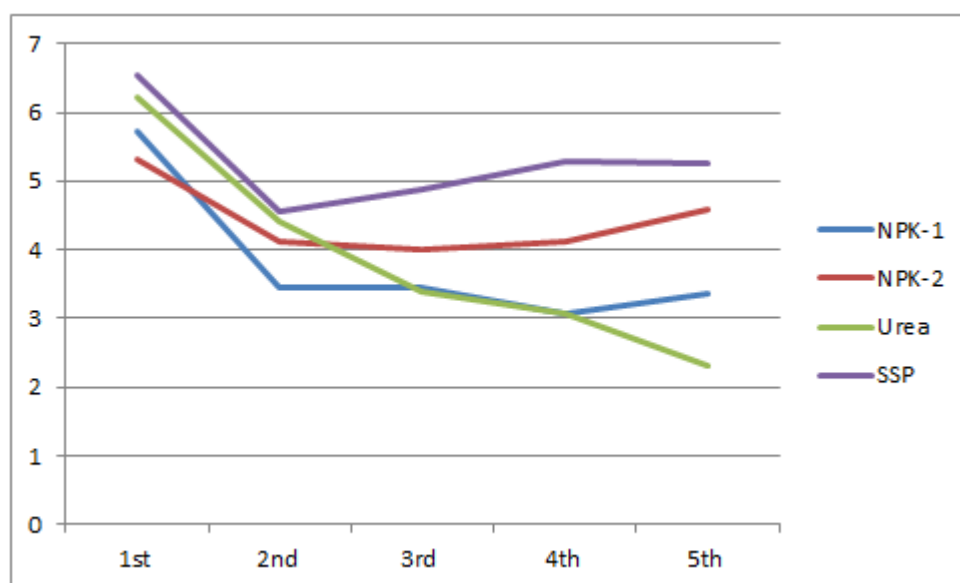


Figure 4.3: Effect of inorganic fertilizers on the Dissolve Oxygen

The result of the effect of fertilizers on alkalinity of medium is presented in Table 4.5 and Figure 4.4. There is no significant difference ($p < 0.05$) in the mean of the treatments across the experimental weeks. At the 1st week, highest value of alkalinity was observed in T3 (51.21) followed by T2 (44.54), T1 had 40.61 while T4 had the lowest alkalinity 38.00. Similarly, at the 2nd week, the highest alkalinity was also found in T3 (54.11) followed by T2 (46.21), T1 (40.01) while the least was found in T4 (41.50). It was also found

that the alkalinity is also highest in urea treatment (54.11) followed by T2 (49.42). T4 had (44.00), the least is recorded on T1 (41.51). At the 4th week, T3 was also the highest (54.11) followed by T2 (52.01), T4 had (46.00) while the least was observed in T1 (42.51). At the expiration of the experiment, the highest level of alkalinity was recorded in T3 (59) followed by T2 (55.11), T3 had (47.51) while the least was found in T1 (44.51).

Table 4.5: Effect of different commercial fertilizer on Alkalinity (mg/L)

Treatment	Duration in Weeks				
	1 st	2 nd	3 rd	4 th	5 th
NPK-1	40.61±49.05	40.01±48.16	41.51±47.23	42.51±47.23	44.51±48.07
NPK-2	44.54±55.45	46.21±50.11	49.42±51.01	52.01±48.16	55.11±51.14
Urea	51.21±45.21	52.17±46.21	54.11±45.31	57.14±45.41	59.11±45.31
SSP	38.00±52.14	41.50±51.37	44.00±52.14	46.00±52.14	47.00±52.14
LOS	ns	ns	ns	ns	ns
P-value	0.1141	0.1311	0.1413	0.2157	0.2214

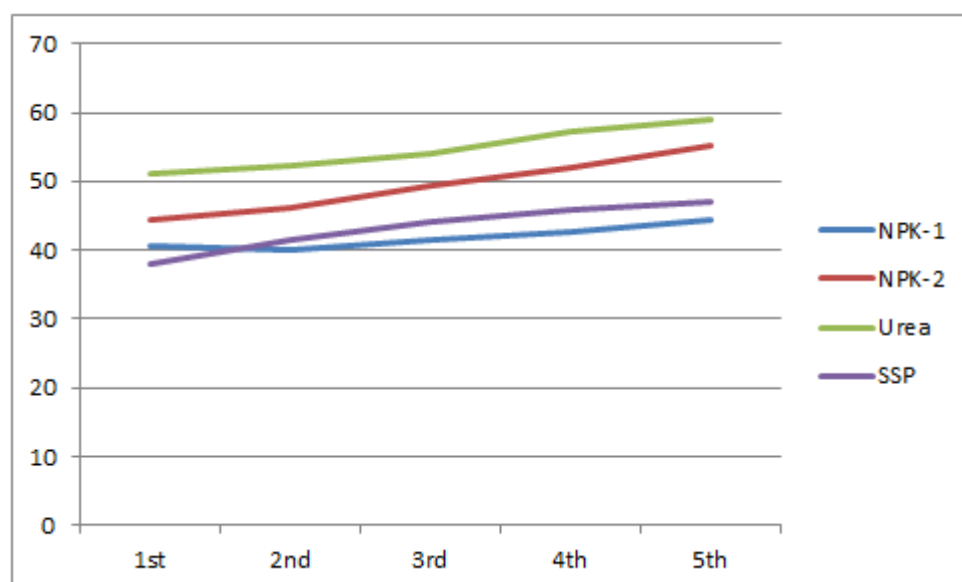


Figure 4.4: Effect of inorganic fertilizers on the Alkalinity

4.3 Lethal Concentration

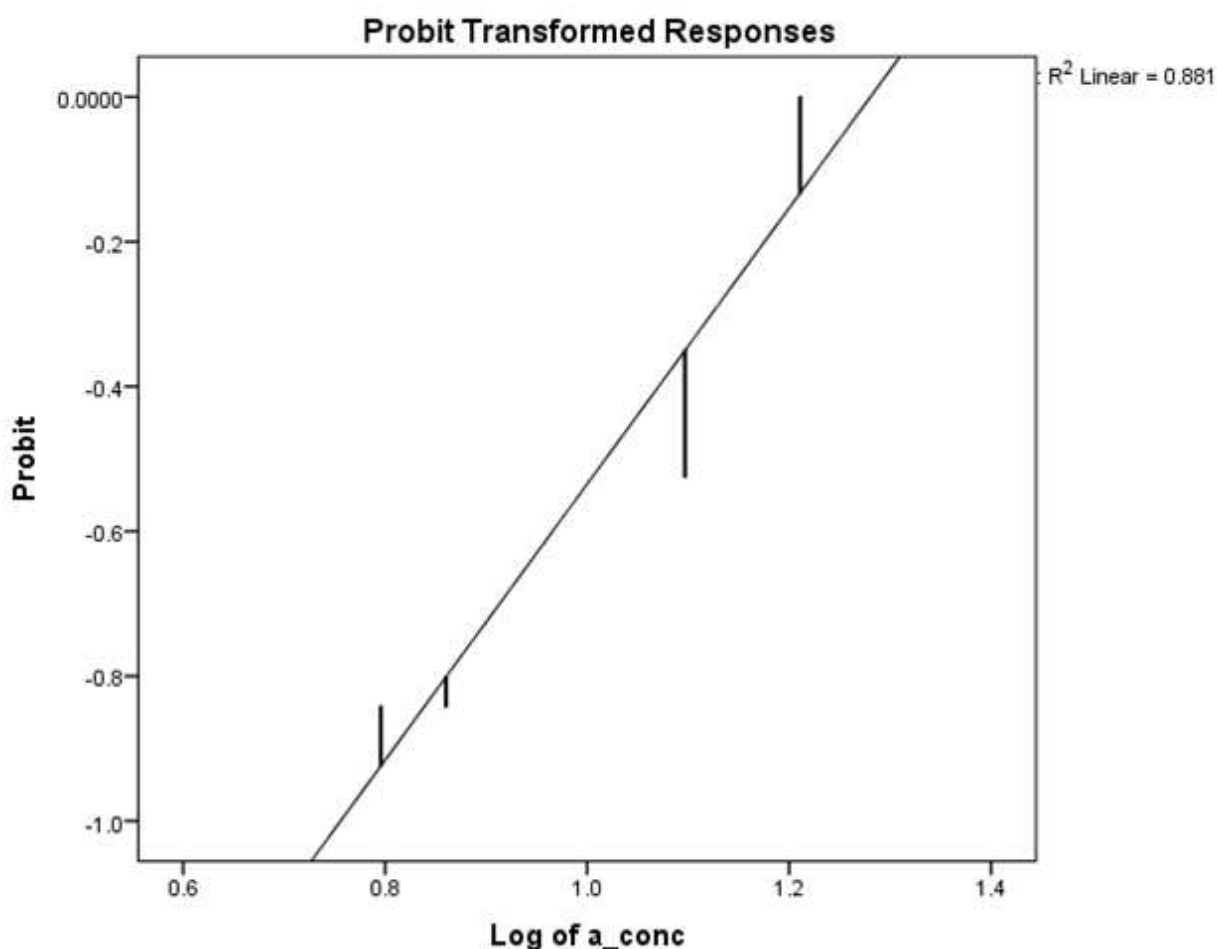
The table 4.6 above displays the lethal concentration of various inorganic fertilizers (N.P.K-1, N.P.K-2, Urea, and Single Super Phosphate (SSP)) at 50% mortality rate among Juvenile of African Cat Fish

(*Clarias gariepinus*). It was observed that the concentration of N.P.K 15:15:15, N.P.K 20:10:10, Urea and SSP that killed 50% of the African Cat Fish (*Clarias gariepinus*) within 96h, was 6.25g/l, 7.25g/l, 16.25g/l, and 12.50g/l, with 6.25g/l and 14.20g/l as the lower and upper limit respectively.

Table 4.6 Lethal concentration of different commercial fertilizers Analysis

Treatment	Conc. (g/l)	Log Conc.	No of Fish	Mortality	Mortality (%)	Probit
N.P.K-1	6.25	0.796	10	2	20	1.0806
N.P.K-2	7.25	0.860	10	2	20	2.034
Urea	16.25	1.211	10	5	50	4.691
SSP	12.50	1.097	10	3	30	3.479

Confidence interval: 6.25g/l and 14.25g/l for lower and upper limit



4.4 Impacts of Fertilizers on the Hematological Parameters

The results on total count (TC) of haemoglobin percentage (Hb) and red blood cell (RBC) is presented in Table 4.7. It was observed that there is significant difference in the Hb among the treatments ($p < 0.05$) while significant difference was observed in RBC. The highest value of Hb percentage was found in T3 (10.40) followed by T2 (9.14), T1 had 9.26 while the lowest was found in T4 (8.11). Whereas, the count of RBC was highest in T3 (3.21 mm^{-3}) followed by T1 (2.36), T2 had (2.117 mm^{-3}) while the least was also found in T4 (2.13 mm^{-3}). This result shows that Urea had significantly higher Hb and RBC count among all the treatment (Figure 4.5 and 4.6).

From Table 4.8 below the highest percentage of lymphocyte was found in T4 (30) followed by T3 (24), T2 had 21 while the least was recorded in T1 (18). Heterophil was found most abundant in T4 (15) followed by T3 (11), T2 had 10 while T1 had the least value (7). More so, T1 had the highest eosinophil (12) followed by T2 (10), both T3 and T4 had 9%. Basophil was found highest in T1 (17) followed by T2 (15), T3 and T4 had 15 and 7 % respectively. For monocytes, the highest value was recorded in T4 (18) followed by T3 (15) while T1 and T2 had 10 and 13 respectively. Neutrophil was highest on T3 (14) followed by T1 (13) while T2 and T4 had 10 and 6% respectively. Finally, the highest value of thrombocyte was recorded on T1 (14) followed by T3 (12) whereas T2 had (6), while the least was recorded in T4 (4).

Table 4.7: Total count of RBC and haemoglobin percentage (Hb)

Treatment	HB	TC of RBC (mm^{-3})
N.P.K-1 (T1)	9.26 ± 0.34^a	2.46 ± 0.23
N.P.K-2 (T2)	9.14 ± 0.21^a	2.17 ± 0.17
Urea (T3)	10.40 ± 1.23^b	3.21 ± 0.21
SSP (T4)	8.11 ± 0.14^c	2.13 ± 0.81
P-Value	0.031	0.071
LOS	*	ns

Note: *=significant at $p < 0.05$, ns= not significant, abc= means within the same column with no superscript in common differed significantly

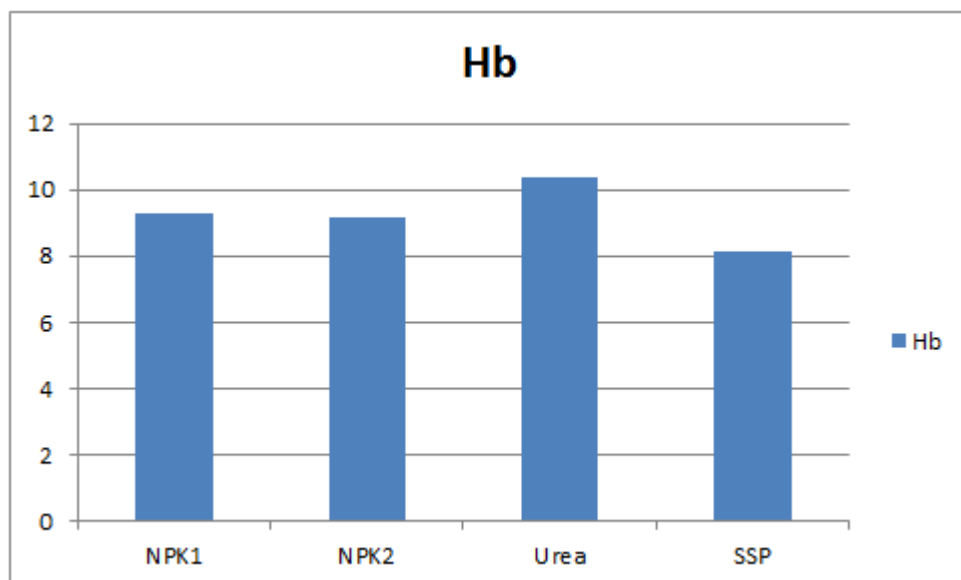


Figure 4.5: Haemoglobin percentage

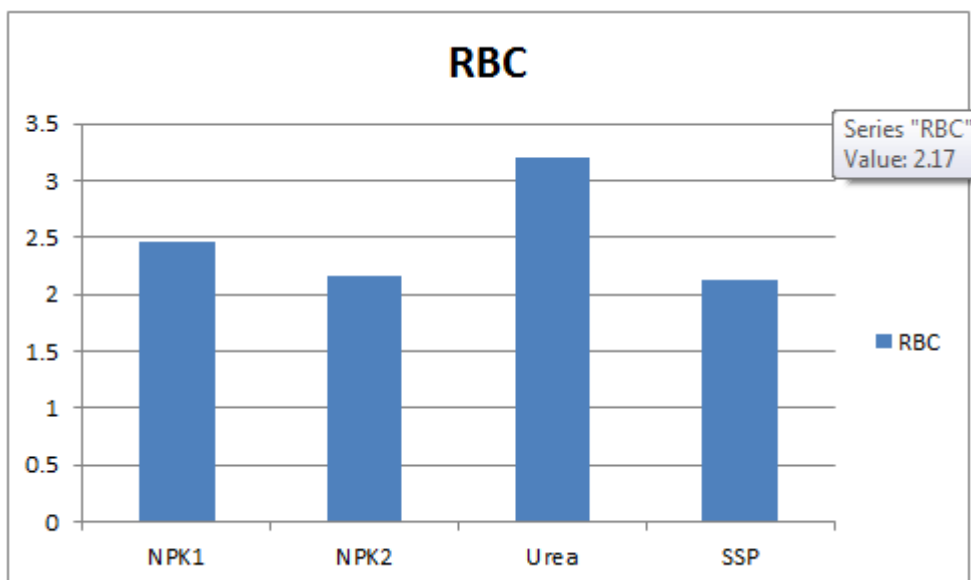


Figure 4.6: Total count of Red blood cell

Table 4.8 Differential count of White Blood Count (WBC)

Leucocytes	N.P.K-1(T1)	N.P.K-2(T2)	Urea(T3)	SSP(T4)
Lymphocyte	18	21	24	30
Heterophil	7	10	11	15
Eosinophil	12	10	9	9
Basophil	17	14	15	7
Monocytes	10	13	15	18
Neutrophil	13	10	14	6
Thrombocyte	14	9	12	4

4.5 Effect of fertilizers on plankton abundance

Table 4.9 shows the abundance of planktons in each of the treatments. Treatment 4 (T4) had the highest plankton diversity (p phytoplanktons and 5 zooplankton), this could account for the high growth rate of *Claria garieoinus* recorded in this study (Table 4.1) while treatment 1 and T2 had the lowest

of the treatments. Treatment 4 (T4) had the highest plankton diversity (6 phytoplanktons and 3 zooplanktons). The phytoplanktons count for urea treatment is the highest (11.80) followed by T2 (8.41), T1 had (8.22) while the least count was recorded in

T4 (6.36). *Scenedesmus acuminatus*, *Oedogonium Sp* and *Oscillation Sp* were more abundant in SSP treatments with respective value 0.61×10^3 , 0.33×10^3 and 0.24×10^3 . While *Chaetoceros decipiens*, *Anabaera Sp* and *Microcystis Sp* were more abundant in urea treatment with value 3.51×10^3 , 2.31×10^3 and 3.51×10^3 respectively. For zooplanktons, urea treatment had the highest count of *Thermocyclops Sp* and *Rotifer* with 0.49×10^3 and 0.31×10^3 respectively. Zooplanktons was most

abundant in T4 (1.97) followed by T3 (0.94), T2 had (0.57) while the least was observed in T1(0.35). The result also revealed that T4 triggered the production of phytoplankton *Cyclotella sp* (0.22×10^3) and *Cosmarium sp* (0.15×10^3) was absent in all other treatments. Similarly, T4 also induced the production of *Trichocora obtusidas* (0.75×10^3) and *Daphnia* (0.20×10^3) which was also absent in all the other treatments.

Table 4.9: Plankton composition and abundance ($\times 10^3$) in individual treatments

Component	N.P.K-1 (T1)	N.P.K-2 (T2)	Urea (T3)	SSP(T4)
Phytoplankton				
<i>Chaetoceros decipiens</i>	2.31±0.34	2.29±0.20	3.11±0.47	2.50±0.22
<i>Anabaera Sp.</i>	-	-	2.31±0.22	2.20±0.19
<i>Scenedesmus accuninatus</i>	1.04±0.03	1.12±0.10	0.52±0.02	0.61±0.01
<i>Cyclotella Sp.</i>	-	-	-	0.22±0.02
<i>Microcystis Sp.</i>	3.37±1.22	3.41±1.31	3.51±1.42	-
<i>Englena viridis</i>	0.31±0.03	0.33±0.02	0.21±0.02	0.11±0.01
<i>Oedogonium Sp.</i>	0.18±0.1	0.22±0.04	1.02±0.01	0.33±0.03
<i>Melosira Sp.</i>	-	-	-	-
<i>Cosmarium Sp.</i>	-	-	-	0.15±0.03
<i>Oscillation Sp.</i>	1.01±0.01	1.04±0.01	1.12±0.02	0.24±0.02
<i>Pandorina Sp.</i>	-	-	-	-
Total Abundance	8.22±1.74	8.41±1.68	11.80±2.18	6.36±0.53
Zooplanktons				
<i>Saccodern sp</i>	-	-	0.14±0.02	0.72±0.03
<i>Thermocyclops Sp</i>	0.12±0.01	0.31±0.02	0.49±0.03	0.30±0.02
<i>Trichocora obtusidas</i>	-	-	-	0.75±0.10
<i>Rotifer</i>	0.23±0.04	0.26±0.04	0.31±0.03	--
<i>Daphnia</i>	-	-	-	0.20±0.01
Total Abundance	0.35±0.05	0.57±0.06	0.94±0.08	1.97±0.16

4.6 Histological Effect of fertilizers on *Clarias gariepinus*

The inorganic fertilizers used in this study altered the histology of various organs of the fish as shown Figure 4.7, 4.8 and 4.9. Disorganization of the gill and dilation of the blood capillary is clearly seen in treatments 1, 2 and 3, although more marked in SSP treatment (Figure 4.7). Further changes in the histology of skin were observed in the mucus and club cell. Hyperplasia of mucous cell and sloughing of club cells of skin occurred at treatment with SSP.

Exfoliation of epithelial cell lining and disorganization of epithelial cell layer was also seen. The club cell in the middle layer of skin shows vacuolization and necrotic sections were obvious in urea NPK 2 treatment, this could be because the level of ammonia is higher in those fertilizers than in other. Liver is a centre for metabolism, detoxification of xenobiotic, excretion of harmful substances, exfoliation of epithelial cells in the lining of secondary lamellae was observed in both urea and NPK 2 than in the other treatments.

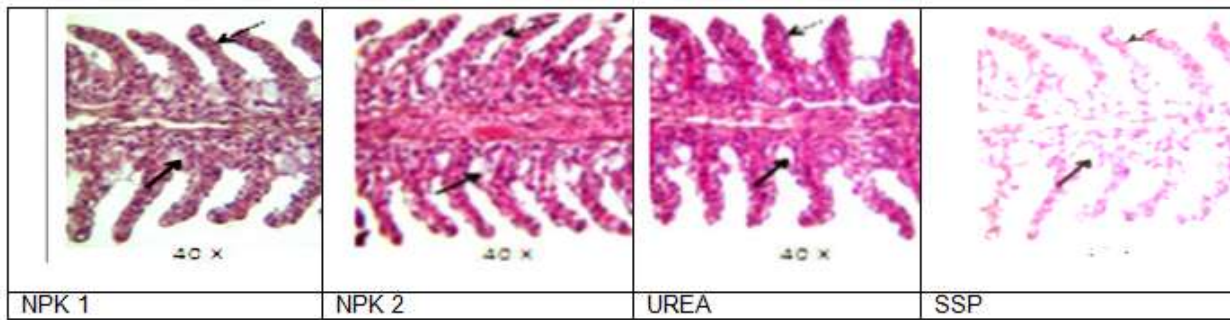


Figure 4.7: Histopathology of gills in *C. gariepinus* in response to different fertilizers. Thick arrows indicate hyperplasia of mucous cells. Excessive mucous secretion was observed with NPK 2 and Urea in the gills. Thin arrows indicate exfoliation of epithelial cells in the lining of secondary lamellae in both NPK1 and SSP

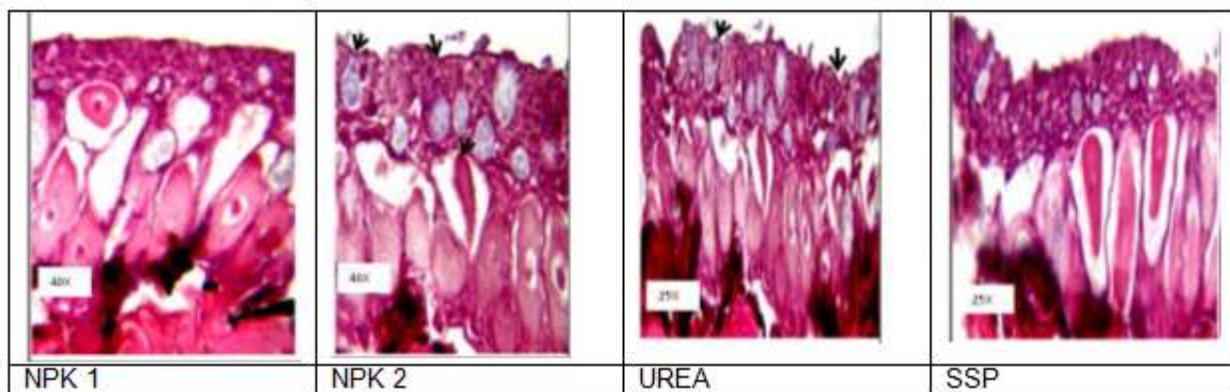


Figure 4.8: Histopathology of skin epidermis in response to different fertilizers showed hyperplasia of mucous cell, sloughing of club cells and exfoliation of epithelial cell lining and disorganization of epithelial cell layer especially at the urea treatment

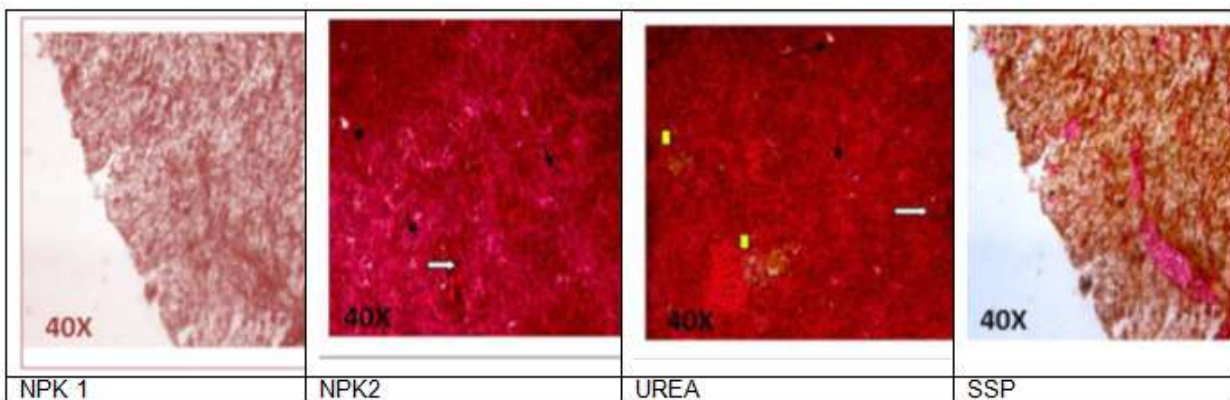


Figure 4.9: Histopathology of liver in response to different fertilizers in *C. gariepinus*. Urea alter liver tissue organization and resulted in nuclear pycnosis (white arrow) fatty degeneration (yellow arrow) necrosis & vacuolar degeneration (black arrow) in liver.

5.0 DISCUSSION

5.1 Results on growth performance exposed to different commercial fertilizers

The growth parameters (average weight gain and average length gain) show significant difference ($p < 0.05$). All the fertilizers impeded the growth of the fish, but remarkably, Urea had the greatest impact as the fishes in that treatment had poor growth. Several authors have reported toxicity of fertilizers on the

water pond for different fish species (Ofojekwu, Nwani, and Ihere, 2008; Erol, Sevki, Halis and Ilhan, 2010; Essien, Asuquo and Ekpo, 2014) which had led to high mortality. This result of this present study is consistent with the report of Ofojekwu *et al.* (2008) and Essien *et al.* (2014) who confirmed urea as a toxicant at lethal concentration which resulted not just in poor growth but also remarkably higher mortality with prolonged usage. The difference in the toxicity rate of the test medium could be because urea has more nitrogen when compared with NPK 15:15:15

and NPK 20:10:10 while SSP has no nitrogen content. This could be understandable because urea is known to hydrolyze easily in water to give ammonium carbonate, which is volatile and thus releases ammonia (Onusiriuka and Ufodike, 1992). The amount of ammonia present at a particular time would affect the toxicity of urea as reported by Ufodike and Onusiriuka 1990.

5.2 Water quality Parameters

5.2.1 TEMPERATURE

The result on the temperature implies that the type of fertilizer does not have significant impact ($p < 0.05$) on the temperature of media. This result is similar to the findings of Kaur *et al.* (2015) and Asuquo, Essien-Ibok and Abiaobo (2016) who reported that fertilizers does not affect the thriving temperature of *Cyprinus carpio* and *Heterobranchus bidorsalis* respectively. Similarly, Ofojekwu *et al.* (1990) and Ufodike, *et al.* (2008) who exposed *Tilapia zilli* and *Clarias gariepinus* fingerlings respectively to acute concentrations of inorganic fertilizers; NPK, urea, calcium hydroxide ($\text{Ca}(\text{OH})_2$), potassium phosphate ($\text{Na}_3\text{PO}_4 \cdot 12\text{H}_2\text{O}$) and sodium nitrate (NaNO_3); and reported there was no significant difference between the various mean values of temperature and pH ($P > 0.05$).

5.2.2 pH

The study revealed that there was no significant difference between the various means value of pH. This study also agrees with the work of Ufodike and Onusiriuka (1990), Ofojekwu *et al.*, (2008) and Kauret *al.* (2015). The pH during the experiment ranged between 8.33 ± 0.02 – 9.09 ± 0.21 in all the treatments which is within the optimum limit for growth and health of most fresh water is in range 6.5 – 9 (Boyd, 1998). The suboptimal pH can cause stress, increased susceptibility to disease and poor growth in fish. The findings were also in agreement with Sahu, Jena, Das, Mondal and Das (2007) who reported that pH was found to be higher under the influence of the application of organic and inorganic fertilization alone or in combination of both Qin, Culver, and Yu (1995) also observed that inorganic fertilizer enhance the primary productivity, dissolved oxygen, pH than organic fertilization.

5.2.3 Dissolve Oxygen

Fertilizers treatments had no significant impact on the dissolve oxygen level except at the 5th week. However, the at the first week, the values of DO were at the desirable limit as fresh water fish requires $\text{DO} \geq 5 \text{mg/L}$ for good growth and development as stipulated by Swingle (1969). According to Boyd (1998), oxygen concentration of less than 3.5 ppm is fatal to carps within duration of 24 hours. However, near fatal and fatal level of DO was recorded from 2nd to the 5th week. The most fatal DO was recorded on Urea treatment and this could account for the high mortality on this treatment. This result of this study

negates the findings of Kaur *et al* (2005) who reported that inorganic fertilizer application has no impact on the DO level of fish medium; the discrepancy could be because of the differences in the types of fertilizer used and the duration of the experiments. Shevgoor *et al.* (1994) also reported that increasing level of fertilization raise all the water quality parameters insuitable range except dissolved oxygen which showed the variation at dawn by the application of high manuring rate.

5.2.4 Alkalinity

Generally, the present study revealed that the total alkalinity ranged between 38-59mg/L in all the treatments during the 5 experimental weeks which is within the suitable range for fish production (Kaur, 2015). Alkalinity was relatively stable throughout the experiment, with narrow fluctuations in urea treated medium. This result is similar to the report of Essien *et al.* (2014) who reported alkalinity of 38-40mg/L of pond treated with chemical fertilizer of various concentrations. Furthermore, the level of alkalinity increased with increase in the number of weeks. Boyd *et al.* (1998) stated that total alkalinity is an important environmental variable in aquatic ecosystem because it interacts with other variables that affected the health of aquatic animals or the fertility of the ecosystem. Boyd and Lichtkoppler (1979) suggested that water with total alkalinities of 20 to 150mg/l contains suitable quantities of carbon dioxide to permit plankton production for fish culture. According to Wurts and Durborow (1992), alkalinity between 75 to 200 mg/L, but not less than 20 mg/L is ideal in an aquaculture pond. According to Santhosh and Singh (2007) the ideal value for fish culture is 50-300 mg/L.

5.3 Lethal Concentration

The results from the study on the lethal concentration were lower than the values reported by Ufodike and Onusiriuka (2008) and also Mac Kinlay and Buday (1997) for N.P.K-1 and N.P.K-2. Ufodike and Onusiriuka (2008) estimated that the 96-h LC50 value of composite fertilizers for African catfish (*C. gariepinus*) ranged from 33.9 mg/L for $\text{Ca}(\text{OH})_2$ to 1.25 g/L for NaNO_3 . In another study, Ofojekwu *et al.* (2008) reported that the 96 hr LC50 of urea fertilizer for *Tilapia zilli* fingerlings to be 15.85 g/L with lower and upper confidence limits being 8.85 and 28.46 g/L respectively. In the present study, the 96-h LC50 value of urea fertilizer for *C. Gariepinusi* was 26.54 g/L with 25.99 g/L and 27.00 g/L as the lower and upper limits respectively. This is higher than that reported by Ofojekwu *et al.* (2008) for *Tillapia zilli* while similar to the report of Ufodike and Onusiriuka (2008) for *C. gariepinus*. The difference between this current report and that of Ofojekwu *et al.* (2008) might be related to differences between the fish species. *Clarias* species is a well-known hardy fish. The difference might be related to fertilizer composition, fish and physicochemical characteristics of the test water (Saha *et al.*, 2002; Palanivelu *et al.*, 2005).

5.4 Impacts of Fertilizers on the Hematological Parameters

In the present study, all the fertilizers applied to *Claria gariepinus* exhibited distinguishable response of haematological variables. It was suggested that haematological parameters reflects the ecological conditions of the habitat of the fishes (Goel, Mishra, Gupta and Wadhwa, 1998; Maitra and Nath, 2014). The result of this study falls in line with the works of Akinrotimi *et al.* (2011b) on acute haematological study of cichlid fish. *Sarotherodon melanotheron* exposed to toxicants. The rise in WBC, neutrophils and monocytes shows an immune response to the toxicants. The result is in agreement with work of Akinrotimi and Gabriel (2012) on submission of remarkable richness of toxicants on the fish blood, where they found that more of these white blood cells and its components are recruited to combat the stressor in the blood stream of the fish. The result shows the values of RBC, Hb, lymphocyte and neutrophil were higher in the urea group than in other treatments which was in agreement with the studies of Maitra and Nath (2014) who reported that sub-lethal concentration of urea increase RBCs count as well as Hb at exposure for 14 days. Similarly, this is also in conformity with the observation of Sasikala *et al.* (2011) who observed significant changes in haematological parameter in *Channa striata*. Initial increase in both studied parameters and then gradual decrease with the increase of doses, indicating slow recovery from adverse condition in the fishes. Roy and Nath (2001) reported almost similar observation in case of Thiamethoxam treated *Oreochromis niloticus*. Then a gradual decrease in total count of RBCs and haemoglobin percentage indicates anaemia that could be due to break down and destruction of RBCs triggered by influx of urea into erythrocytes as in case of phenol-dosed fishes Maitra and Nath (2014). Haematological parameters have been considered as indicator of stress, induced by pesticides and variation in RBCs count and the haemoglobin concentration was due to the deleterious effect of pollutant on the erythropoietic tissue of *Mystus vittatus* (Verma, Sarita and Dable, 2002). Goger and Sawant (2009) suggested that differential count (DC) of leukocytes is a reliable proof the negative effect of urea on these leukocytes, though its haematological index is used to study the change in environmental conditions.

5.5 Effect of fertilizers on plankton abundance

The result on the effect of fertilizers on the planktons abundance revealed that SSP treatment is has more phytoplanktons and zooplanktons diversity more than all other treatment and also more zooplanktons count. However, urea had more phytoplanktons count. Furthermore, zooplankton *Trichocora obstusidas* and *Daphnia* which was available in SSP treatments but absent in other treatment has been reported to be among the preferred food for *Clarias gariepinus* during early life (Oyin, 2013). Hence the observed higher survival of juvenile in T4 as against other

treatments, similarly Boyd and Massaut (1999) reported that inorganic fertilizers have much higher concentrations of nutrients such as nitrogenous compounds than manures.

5.6 Histological Effect of fertilizers on *Clarias gariepinus*

The gill of *C. gariepinus* helps in survival under desiccation and hypoxic condition in water. The important histopathological alterations observed in the gill of *C. gariepinus* due to the effect of inorganic fertilizer include detachment and lifting of the respiratory epithelia from the underlying vascular components of the secondary lamellae and sloughing off viable epithelial cells (Fig. 4.7), which results in haemorrhage into the lumen fuse causing reduction of volume of the lumen. The result correlate with findings of Rajan and Banerjee (1993) for histopathological damage to the air sac of *H. fossilis* exposed to a lethal concentration of mercuric chloride. The skin is a primary defensive organ of any organism. In *C. gariepinus* exposed to NPK fertilizer, the club cells in the middle layer of skin show vacuolization and many other symptoms of necrosis. The disorganization of epithelial cell layer due to hyperplasia and breakdown of a crust of a dead layer of cells increases the barrier distance between dissolved oxygen in the media and blood in sub-epidermal blood vessels. Further, liver which is the primary organ for metabolism, detoxification of xenobiotics and excretion of many harmful substances was monitored for histological changes under NPK induced stress. The liver has the ability to degrade toxic components, but its regulating mechanisms can be overwhelmed by elevated concentrations of these compounds, and could subsequently result in histological changes and structural damage (Brusle, *et al.*, 1996). High dose of NPK showed larger lesion area in liver than low dose.

Further, the histopathological study of gill under different sub-lethal doses of showed hyperplasia of primary and secondary lamellae, degeneration of epithelium, fusion of adjacent secondary lamellae, increased mucus production, secondary lamellae appeared thickened and shortened with extremely rough surface and considerable mucus in both low and high dose treatment. A similar observation was also made by Tandjung *et al.*, and Lamchumchang *et al.*, in brown trout, *Salmo trutta* and *Oreochromis niloticus*.

In conclusion, the present study provides a detailed insight of the responses mounted by fishes to exogenous NPK fertilizer. It further provides an understanding of the impact of employing chemical fertilizers on aquatic life.

6.0 CONCLUSION AND RECOMMENDATION

6.1 Conclusion

Sequels to the discussion of the findings in this study, the following conclusion were made;

- i. The various fertilizer (N.P.K-1, N.P.K-2, Urea, and SSP) use in the experiment impeded the growth of the fish
- ii. There is no significant variation among the fertilizer on the water parameter over the period of experiment, except water for DO.
- iii. Water temperature, pH, and Alkalinity were suitable for the survival of the fish within the period of experiment
- iv. Dissolve oxygen of the water was within the toxic value over the period of experiment
- v. The lethal concentration that killed 50% of the fishes within 96h were 6.25g/l, 7.25g/l, 16.25g/l, and 12.50g/l
- vi. There was significant variation among the fertilizer in the total count of Haemoglobin in the fishes
- vii. All fertilizer treated on the fishes exhibited distinguishable response of Haematological variables
- viii. Single super phosphate had highest white blood count in lymphocyte, heterophil, and monocytes.
- ix. Urea had the highest white blood count in neutrophil
- x. N.P.K-1 and 2 had the highest white blood count in eosinophil, basophil, and thrombocyte.
- xi. Single super phosphate had the highest plankton diversity
- xii. N.P.K-1 and 2 had the lowest plankton diversity
- v. When toxicities of inorganic fertilizer are minimised in our environment and water bodies' biodiversity will thrive thereby providing sufficient food to the increasing human population.

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6.2 RECOMMENDATION

Recommendation

From the result of this study, the following are recommended:

- i. Fish farmers using inorganic fertilizers should endeavour to monitor the time/duration of the application, and not exceed the days, so as to avoid acute toxicity
- ii. It is thus recommended that the application of these fertilizers in aquatic ecosystems either in ponds, irrigations or farms should be carefully controlled or monitored, such that concentrations that are lethal to aquatic life could be avoided.
- iii. Single super phosphate seems to be least toxic inorganic fertilizer, so fish farmers can use it for fertilization
- iv. There is also a great need to provide further baseline data on inorganic fertilizer. Such studies should be concerned with providing information on research such as, the effects of sub-lethal concentrations of fertilizer on the; serum/plasma enzymes, metabolites and hormones of *C. gariepinus*.

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